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# Module 1

## Microscopy

* Smaller limit of resolution of a microscope, the greater its **resolving power**.
* Phase contrast/differential interference exploits differences in the phase of light passing through a structure with a refractive index different than the surrounding medium.
* **Fluorescent microcopy**: detects fluorescent dyes to how location of substances in the cell.
* **Confocal scanning**: uses a laser beam to illuminate a single plane of a fluorescently labeled specimen (3-D reconstruction).
* **The electron microscope**: uses an electron beam rather than light – limit of resolution: 0.1-0.2nm- magnification 100,000X than light microscope.
* **TEM**: transmission electron microscopy. Electrons are transmitted through the specimen.
* **SEM**: scanning electron microscopy. The surface of a specimen is scanned by a beam of electrons deflected from specimen ‘surface.

## Biochemistry: Important Advance

* **Chromatography**: techniques used for sample preparation which let separate molecules by size, charge or binding affinity.
* **Electrophoresis**: the cell is loaded into a gel and then an electric field is applied to the gel. This Electric field moves the molecules through the gel differentially
* Since DNA molecules are negatively charged, when the electric field is applied to the gel, the DNA molecules moved towards the positive charges. But larger molecules move slowly and run through the argos matrix and run next to a sample of known molecular weight called the DNA ladder. Uses an electrical field to move proteins, DNA or RNA molecules through a medium based on size/charge.
* **Mass spectrometry or MassSpec**: determine size and composition of protein by measuring mass to charge ratio of ions in a sample.

## Genetic: Information Flow and inheritance

Humans have 23 pairs of chromosomes.

Uses ultracentrifugation and electrophoresis to separate DNA and RNA molecule.

Recombinant DNA technology, restriction enzymes cut DNA at specific places to create recombinant DNA molecules with DNA from different sources.

DNA sequencing: methods to determine base sequences of DNA molecules.

Possible to sequence entire genome (entire DNA content of a cell).

Cell membrane: a barrier which maintains physical integrity.

**Covalent bonds**: sharing of a pair of electrons between 2 atoms.

## Carbon-containing molecules are stable

* Stability is expressed as bond energy.
* Bond energy is expressed as cal/mol, amount of energy required to break one mole (6 x 10^23) of bonds.
* A calorie is the required energy to increase the T of 1g of water by 10C.

## Bond Polarity

Polar bond result from a high electronegativity of O2 and sulfur compared to carbon and hydrogen.

Water molecules are polar: electrons drawn by oxygen, partial negative charge at the end of O2, a partial positive charge around hydrogen molecules, bent shape.

## **Water is cohesive**: network of hydrogen-bonded molecules, hydrogen bond is weak compared to covalent bonds.

* Combined effect of many hydrogen bonds accounts for water’s high
  + **Surface tension**: vast number of hydrogen bonds.
  + **High Specific heat** gives water its T stabilizing capacity.

Specific heat: amount of heat a substance must absorb to raise its T by 10C.

* Water changes temperature very slowly.
* Water is an excellent solvent: due to its polarity.
* Takes part in many chemical reactions.

## Selectively Permeable Membranes

A barrier such

* Impermeable to much of the cell contents.
* Insoluble in water.
* Permeable to water.

## Cellular membrane is a hydrophobic barrier

* Consists of: **phospholipids**, **glycolipids**, and **membrane proteins, sterols** – cholesterol.

## Membrane lipids Are Amphipathic

* **Phospholipid** have a polar head, due to negatively charged phosphate group linked to a positively charged group, and two non-polar hydrocarbon tails.

A Membrane is a Lipid Bilayer with Proteins embedded within it

Polar heads of membrane phospholipids face outward toward aqueous environment.

Hydrophilic tails are oriented inward.



## Membranes are Selectively Permeable

* Cellular constituents are mostly polar or charged and are prevented from entering or leaving the cell.
* Because of the hydrophobic interior, impermeable to most polar molecules and very impermeable to ions.
* Non-polar and very small molecules diffuse.
* The rate at which a molecule diffuse across lipid bilayer depends on its size and solubility.



# Module 2- Macromolecules of the cell

4 major classes of macromolecules:

* Nucleic Acids
* Proteins
* Polysaccharides
* Lipids

## Proteins

### Cell processes

**Transcription** takes DNA and makes RNA out of it. Nature makes an RNA copy of DNA and by post-transcription makes mRNA out of it. **Translation** is the process where small molecules are added to the mRNA to build up first **polypeptides**, amino-acids in small repeat units. Then there is a process to convert polypeptides into a protein: post-translation. And when it takes a 3D shape it takes protein activity to effect particular functions like an effector molecule and then activate more functions.

### Small molecules

* **Amino acid**: monomeric components of proteins.
* **Aromatic bases** (purines and pyrimidines): components of nucleic acids: DNA and RNA.
* **Sugars** (monosaccharides):
  + Ribose: components of nucleic acids
  + Glucose: used in metabolism to make energy
* **Lipids**: components of phospholipids.

### Levels of organization in Protein structure

* **Primary**: Amino acid sequence based on covalent peptide bonds.
* **Secondary**: fold alpha-helix, beta-sheet or random coil based on hydrogen bonds.
* **Tertiary**: 3D folding of a single polypeptide chain based on hydrogen bonds, disulfide bonds, electrostatic interactions and hydrophobic effect.
* **Quaternary** (macromolecule): association of two or more polypeptides with same interactions seen in tertiary structure.

### 4 Protein major classes

* enzymes, (**catalysts**) that greatly increase rates of chemical reactions in cells.
* **Structural proteins**: provide support and shape to cells and organelles, giving cells their characteristic appearances.
* **Motility proteins**: play key roles in the contraction and movement of cells and intracellular structures.
* **Regulatory proteins**: are responsible for control and coordination of cellular functions, ensuring that cellular activities are regulated to meet cellular needs.
* **Mono-functional proteins**: have a single function: catalytic, structural, motility, or regulatory.
* **Bi-functional proteins** plays two different roles.

### The monomers are amino acids

* Proteins are linear polymers of amino acids.
* 60 different kinds of amino acids, but only 20 are used in protein synthesis.
* **Every amino acid has the basic structure with a carbonyl group, an amino group, a hydrogen atom, and a R-group all attached to a single carbon atom.**
* The R group determines the characteristics (size, polarity and pH) for each type of amino acid.
* Amino acids such as valine, methionine, and alanine are nonpolar (hydrophobic), while amino acids such as serine, threonine, and cysteine are polar (hydrophilic). The R groups of lysine and arginine are positively charged so these amino acids are also known as basic (high pH) amino acids. Proline is an exception to the standard structure of an animo acid because its R group is linked to the amino group, forming a ring-like structure.
* Except for glycine, for which the R group is a hydrogen atom, all amino acids have at least one asymmetric carbon atom. Therefore, most amino acids exist in two isomeric forms, L and D-amino acids.

### The structure of 20 amino acids

* **Group A**: hydrophobic and nonpolar R groups.
* **Group B**: hydrophilic and polar R group, uncharged.
* **Group C**: hydrophilic, polar R group, and protonated or ionized at cellular pH.

### The polymers are Polypeptides and Proteins

* Stepwise addition of new amino acid to a growing chain of amino acids by **a dehydration (condensation) reaction**: formation of polymers and water molecule.
* The reaction could be reversed by adding back water molecules; used by cells to excrete water waste.
* -H and -OR groups are removed as water comes out and the covalent bond between the carboxyl group and an amino group is called **peptide bond**.

### Peptide bond formation

Always an N-terminus at one end and C-terminus at the other end.

### Polypeptide and Proteins

* Product of amino acid polymerization is a polypeptide (polymers of peptides).
* Protein is a polypeptide or polypeptides that have folded properly, combined with other additional components needed for proper functioning, and is now functional.
* Protein is a polypeptide chain that have attained a unique stable, 3-D shape and it biologically stable.
* Monomeric protein consists in a single polypeptide vs. multimeric proteins, two polypeptides: a dimer, 3 polypeptides: a trimer.
* **Ribonuclease** is a monomeric protein.
* Hemoglobin is a multimeric protein. It contains 4 polypeptides, (2 alpha-subunit and 2 beta-subunits), a chain and II chain.
* Each subunit contains a **heme group** with an iron atom. Each heme iron can bind a single oxygen molecule.
* **Homomeric**, and **heteromeric**: protein made up at least of two different polypeptides chains.
* Protein structure is determined by its amino acid sequence which drives the folding and intramolecular bonding of the linear amino acid chain, which ultimately determines the protein's unique three-dimensional shape.
* Protein synthesis.

### Primary structure

* Primary structure is the amino acid sequence of the constituent polypeptides.
* Amino acids are always written from the N-terminal to the **C-terminal**, direction in which the polypeptide is synthesized.
* Once incorporated into a polypeptide chain, individual amino acids are called **amino acid residues**.
* **Disulfide bond**: very stable bond between two sulfur atoms of 2 cysteine amino acid residues.

### Secondary structure

Because of the folding groups of amino acids are close to each other.

The group interactions result in two structural patterns: the alpha-helix and beta sheet conformations.

### Categories of Proteins

* **Fibrous proteins**: have extensive secondary structure (either a helix or beta sheet) giving them a highly ordered and repetitive structure.
* **Globular proteins**: most of the proteins.
  + The polypeptide chain is folded in a compact structure. It is folded locally into alpha-helical or Beta-sheet structures. These regions are folded on one another to give the protein its compact, globular shape.
  + The folding is possible because the interspersed random coils allowing the polypeptide to loop and fold.
  + Have unique tertiary structures
  + They consist of a number of segments called domains. A **domain** is a discrete, locally folded unit of tertiary structure. A domain typically contains 50-350 amino acids, and usually has a specific function.

### Tertiary Structure

3-D folding of a single polypeptide chain.

### Quaternary structure

* Level of organization concerned with subunit interactions and assembly.
* Association of 2 or more polypeptides to form a multimeric protein.
* Applied only to multimeric proteins.
* The bonds and forces that maintain quaternary structure are the same as those responsible for tertiary structure: **hydrogen bonds, electrostatic interactions, hydrophobic interactions, and covalent disulfide bonds**.

### Disulfide Bond formation in Insulin

It could be reversed.

### The Primary structure of Insulin

Insulin consists of two polypeptides, A and B chains. The two chains are covalently linked by two inter-chain disulfide bonds.

### Structure of Hair

Alpha keratin protein: 3 helices of a-keratin wrap into protofibrils which then bond together to form microfibrils. Microfibril= 9 + 2 protofibril based structure. Microfibrils aggregate to form macrofibrils.

### The roles of DNA and RNA in Protein synthesis

**mRNA**: directs amino acids sequence of polypeptides.

**tRNA**: binds to amino acids and directs them to proper locations within the growing polypeptide chain.

**rRNA**: components of the ribosomes that serve as the site of protein synthesis.

## Nucleic Acids

### Transcription and Translation

* **Transcription**: DNA molecule is transcribed into an RNA molecule.
* **Translation**: takes RNA and converts into protein.

### Nucleic Acids

* Nucleic acids are macromolecules critical in the storage, transmission and expression of genetic information.
* Are linear polymers of nucleotides, strung together in a genetically determined order.
* Two major types are DNA and RNA.
* DNA contains the sugar deoxyribose, RNA contains 5-carbon sugar ribose in each of its nucleotides.
* DNA plays as the repository of genetic information, whereas RNA molecules play several different roles in the expression of that information during protein synthesis.
* **mRNA**: directs amino acids sequence of polypeptides that is during polypeptide synthesis.
* **tRNA**: binds to amino acids and directs them to proper locations within the growing polypeptide chain.
* **rRNA**: components of the ribosomes that serve as the site of protein synthesis.

### The Monomers are Nucleotides

* Nucleic acids are informational macromolecules that contain non-identical monomeric units in a specified sequence.
* The monomeric units of nucleic acids are called nucleotides.
* DNA and RNA each contain only four different kinds of nucleotides.
* Each nucleotide consists of **a five-carbon sugar, a phosphate group, and a nitrogen-containing aromatic base. The sugar is either D-ribose (for IRNA) or D-deoxyribose (for DNA).**
* **The phosphate is joined by a phosophoester bond to the 5' carbon of the sugar (-C-O-P-), and the base is attached at the 1' carbon. The base maybe either a purine or a pyrimidine.**
* DNA contains the **purines**: **adenine** (A) and **guanine** (G) and the **pyrimidines**: **cytosine** (C) and **thymine** (T). RNA also has **adenine**, **guanine**, and **cytosine** but contains the pyrimidine **uracil** (U) in place of thymine.
* ATP is the energy-rich compound used to drive a variety of reactions in the cell, including the activation of monomers for polymer formation.
* Nucleotides plays two roles in the cell:
  + Monomeric units of nucleic acids
  + Serve as intermediates in various energy transferring reactions.

### Polymers: DNA and RNA

* Nucleic acids are linear polymers formed by linking each nucleotide to the next through a phosphate group.
* The result of a condensation reaction with the -H and -OH groups come off from the sugar and the phosphate group respectively is **a 3’,5’ phosphodiester bond**.
* Incoming nucleotides must be added in a specific, genetically determined sequence. The **template to specify nucleotide order is DNA for both DNA and RNA synthesis.**
* **Purines: Adenine, Guanine**
* **Pyrimidines: Thymine, Uracil, Cytosine**
* Purine and pyrimidine bases have carbonyl groups and nitrogen atoms capable of hydrogen bonds formation under appropriate conditions.
* Paring of A with T (or U) and G with C

### A DNA molecule is double-stranded Helix

* The double helix consists in two complementary chains of DNA twisted together around a common axis to form a right-handed helical structure.
* The two chains are oriented in opposite directions along the helix, one in 5’3’ direction and the other in 3’-5’ direction.
* The sugar phosphate backbones of the two strands could be envisioned as the sides of a circular staircase where each step corresponds to a pair of bases held in place by hydrogen bonding.
* The right-handed helix is an idealized version of the B-DNA, the main form of DNA.
* Z-DNA is a left-handed double helix, with a longer, thinner sugar phosphate backbone.

### RNA Structure

* Secondary and tertiary structures are well understood only for tRNA molecules.
* **A nucleotide is composed of three components, namely a nitrogenous base, phosphate group, and sugar. A nucleoside is composed of two components, namely a nitrogenous base and sugar.** This is the basic difference between a nucleotide and a nucleoside.

### The Phosphorylated Forms of Adenosine

* Adenosine occurs as the free nucleoside, the monophosphate (AMP), the diphosphate (ADP), and the triphosphate (ATP).
* The bond that links the first phosphate to the ribose of adenosine is a low-energy **phosphoester** bond, whereas the bonds that link the second and third phosphate groups to the molecule are higher-energy **phosphoanhydride** bonds.

### Hydrogen Bonding in Nucleic Acid Structure

A-T pair held together by two hydrogen bonds, whereas the C-G pair has three hydrogen bonds.

## Polysaccharides

### Polysaccharides

* No known informational role in the cell.
* They are the storage polysaccharides starch and glycogen and the structural polysaccharide cellulose.
* Each of these polymers contains the 6-carbon sugar glucose and its single repeat unit.

### The Monomers are Monosaccharides

* The repeats are single sugar called monosaccharides.
* A sugar can be an aldehyde or ketone that has two or more hydroxyl groups.
* 2 categories of sugars: aldosugars, with a terminal carbonyl group and the ketosugars, with an internal carbonyl group.
* Sugars are classified as triose (3 carbons), a tetrose (4), a pentose (5), a hexose (6), or a heptose (7).
* Most common: aldhohexose D-glucose, C6H12O6.

### Polysaccharides

Glucose also occurs in disaccharides consisting of 2 monosaccharide units linked covalently.

* **Maltose**: 2 glucose units linked together.
* **Lactose**: glucose linked to a galactose.
* **Sucrose**: glucose linked to a fructose.

### Polysaccharide Polymers are Storage and Structural

* Polysaccharides perform either storage or structural functions in cells. The most familiar storage polysaccharides are the starch of plant cells and the glycogen of animal cells. Both of these polymers consist of alpha-d-glucose units linked together by a glycosidic bond.

### Glycogen (storage)

* Glycogen is highly branched, with linkages occurring every 8 to 10 glucose units along the backbone and giving rise to short side chains of about 8 to 12 glucose units.
* Glycogen is stored mainly in the liver and in muscle tissue.  
  In the liver it is used as a source of glucose to maintain blood sugar levels, whereas in muscle it serves as a fuel source to generate the ATP needed for muscle contraction.

### Starch (storage)

* Starch occurs both as un-branched amylose and as branched amylopectin.
* Like glycogen, **amylopectin** has a (1 -> 6) branches along the backbone and give rise to longer chains.
* Starch deposits are about 10-30% amylose and 70-90% amylopectin.

### Cellulose (structural polysaccharide)

* Cellulose is an important polymer quantitatively; more than half of the carbon in higher plants is present in cellulose.
* Like starch and glycogen, cellulose is also a polymer of glucose, but the repeating monomer is **beta-d-glucose** and the linkage is therefore beta (1 -> 4).
* Cellulose forms rigid, linear rods. These aggregate into **microfibrils**.
* Plant and fungal cell walls consist of these rigid microfibrils of cellulose embedded in a **noncellulosic matrix**.
* Mammals do not possess an enzyme that utilize cellulose as food (cannot cleave glycosidic bonds).

## Lipids

* The distinguished feature of lipids is their **hydrophobic nature**.
* They resemble one another more in their soluble properties than in their chemical structures.
* **Not the result of stepwise polymerization found for proteins, nucleic acids, and polysaccharides.**
* Rich in nonpolar hydrocarbon regions and have relatively few polar groups.
* Some lipids are amphipathic having both a polar and a nonpolar region.
* They play at least 3 main roles in the cell:
* Energy storage
* Membrane structure
* Transmission of chemical signals into and within cells.
* The six main classes of lipids are **fatty acids, triacylglycerols, phospholipids, glycolipids, steroids, and terpene**s.

### Fatty Acids Are the Building Blocks of several classes of Lipids

* A fatty acid is a long, unbranched hydrocarbon chain with a carboxyl group at one end.
* It is amphipathic; the carboxyl group renders one end (“head”) polar whereas the hydrocarbon, “tail” is nonpolar.
* Fatty acid yields a great deal of energy upon oxidation.
* **Fatty acids without double bonds are saturated fatty acids: every carbon atom in the chain has the maximum number of hydrogen atoms attached to it.**
* **Unsaturated fatty acids contain one or a few double bonds.**
* General formula: n carbon atoms Is **CnH2nO2**.

### Triacylglycerol Are Storage Lipids

* Triacylglycerols (triglycerides) consist of a glycerol molecule with 3 fatty acids linked to it.
* Glycerol is 3-carbon alcohol with a hydroxyl group on each carbon.
* Fatty acids are linked to glycerol by ester bonds, formed by the removal of water.
* Triglycerides are synthesized stepwise, with one fatty acid added at a time.
* Monoglycerides contain a single esterified fatty acid, diglycerides have 2, triglycerides have 3.
* Triglycerides are usually solid or semi-solid at room temperature and are called fats.
* In plants, mots triglycerides are liquid at room temperature – vegetable oils.

### Phospholipids Are Important in Membrane Structure

* Critical to the bilayer structure found in all membranes.
* Phospholipids are phosphoglycerides or sphingolipids.

### Steroids Are Lipids with a Variety of Functions

* Are derivatives of a 4-membered ring compound called phenanthrenes which makes them structurally distinct from other lipids.
* Only property that links to other classes of lipids: relatively nonpolar and therefore hydrophobic.
* Cholesterol is an amphipathic molecule, with a polar head group and a nonpolar hydrocarbon body and tail.
* Cholesterol found primarily in membranes.
* Cholesterol is the starting point for the synthesis of all the steroid hormones, which include the male and female sex hormones, the glucocorticoids and mineralocorticoids.

### Terpenes are Formed from Isoprene

Terpenes, synthesized from 5-carbon compound isoprene, also called isoprenoids.

Isoprene and its derivatives are joined together in various combinations to produce vitamin A1, carotenoid pigments.

# Module 3 – Introduction to Cells and Organelles

## Types of Cells and Their Properties

* The main distinction between two cell types (Prokaryotes vss. Eukaryotes) is the membrane bound nucleus of eukaryotic cells.
* Prokaryotic cells can be divided into bacteria and archaea.
* Phylogenetic Tree of Life: ancestral cell -> bacteria, Archaea, Eukarya.
* Gram’s stain: staining to distinguish bacteria:
  + Gram-positive microorganisms have higher peptidoglycan content, whereas gram-negative organisms have higher lipid content.

### There Are Three Limitations on cell size

* *Need to maintain adequate surface are to volume ratio*

Cells that are specialized for absorption have characteristics to maximize surface area/volume.

* *Diffusion rates of Molecules*

Eukaryotic cells avoid the problem of slow diffusion rates by using carrier proteins or vesicles.

* *The Need for adequate local concentration of essential substances*

The larger the cell is, the more difficult to maintain these higher concentrations required for different type of reactions to occur.

### Eukaryote Cells use Organelles to compartmentalize Cellular Function

* As cell size increases, the number of molecules increase proportionately with volume.

The challenges of diffusing macro-molecules across the cell or accumulating higher concentrations in certain regions of cells can be mitigated by eukaryote cell development of organelles.

### Chromosome

DNA is tightly packed into gene and into chromosome and contained in the nucleus.

### Genetic Information

Eukaryotic cells replicate DNA and then distribute their chromosomes into daughter cells by **mitosis and meiosis, followed by cytokinesis, division of the cytoplasm**

Bacterial and archaeal cells replicate their DNA and divide by binary fission.

<https://biologydictionary.net/difference-binary-fission-mitosis/>

## The Eukaryotic Cell – Plasma Membrane, Organelles, And the Endosymbiont Theory

* A typical eukaryotic cell has: a **plasma membrane**, a **nucleus**, a membrane bounded **organelle**, and the **cytosol** supported by a **cytoskeleton**.
* The Plasma Membrane Defines Cell Boundaries and Retains Content
* Membrane proteins are also amphipathic, with polysaccharides attached to them: **glycoproteins**.
* Plasma membrane is selectively permeable membrane: only certain compounds can move across this membrane, tight control of transport across in either direction for the cell.

### Mitochondrion

* Site of aerobic respiration.
* Oxidation of sugars and other fuel molecules extract energy from food and stores it as ATP.
* Most molecules for mitochondrial functions, are localized on the cristae or the matrix.
* Tissues with high demand for ATP have many mitochondria, located within the cell at the site of greatest energy needs (e.g., sperm and muscle cells).
* We inherit mitochondrial DNA (mtDNA) only from our mothers.

### The Chloroplast

* Site of photosynthesis in plants and algae.
* Large and quite be numerous.
* Like mitochondrion, contain own ribosomes, and a small circular DNA molecule.

### The nucleus is the information center of the Eukaryotic cell

* The nuclear envelope has numerous pores controlled by various proteins constituting a **nuclear pore complex**.
* The nucleus includes the nucleolus, the site of ribosomal RNA synthesis and ribosome assembly.
* The **endosymbiont theory**: mitochondria and chloroplasts and bacteria are similar; it suggests that mitochondria and chloroplasts originated from prokaryotes.
* Similarities:
* Circular DNA molecules without associated histone.
* rRNA sequences
* ribosome size
* sensitivities to inhibitor of RNA and protein synthesis.
* Type of proteins used in protein synthesis.
* Surrounded by double membranes
* Inner membrane has bacterial-type lipids

### The Endoplasmic Reticulum

* **Cisternae**: is the lumen; tubular membranes and flattened sacs.
* **Lumen:** internal space of ER.
* **Rough ER**:
  + Rough ER studded with ribosomes on cytoplasmic side.
  + In rough ER, free ribosomes not associated with ER.
* **Smooth ER**:
  + No role in protein synthesis.
  + Involved in synthesis of lipids and steroids (cholesterol and its derivatives).
  + Responsible for inactivating and detoxifying substances.
* **Sarcoplasmic reticulum** has critical functions in contraction.

### The Golgi Complex

* Packaging station or the *post office*; processing and packaging secretory proteins and in complex polysaccharide synthesis.
* Role of sending off proteins to distant locations within the cells through vesicles.
* Accepts vesicles that bud off of the ER.
* The contents of vesicles from the ER are modified and processed in the Golgi complex.
* Secretory and membrane proteins are mainly **glycosylated** (addition of short-chain carbohydrates).

## Cell Vesicles, Structural Components, and Examples of Cellular Invaders

**Endomembrane system**: ER, Golgi, secretory vesicles and lysosomes.

The cytoplasm contains the cytosol and cytoskeleton

### Ribosome

* Ribosomes read or translate mRNA to link amino acids together and form proteins.
* Very small, can only be seen under microscope electronic.
* Small sedimentation coefficients.

### Cytoplasm

* Interior of the cell not occupied by the nucleus.
* Cytosol is the semifluid substance in which organelles are suspended.
* Cytosol is permeated by the cytoskeleton.
* Synthesis of fats and proteins and initial step of releasing energy from sugars take place in the cytosol.

### Cytoskeleton



* The cytoskeleton is a 3-D array of interconnected **microfilaments**, **microtubules**, and **intermediate filaments**.
* Gives cell its definitive shape and internal organization.
* Plays also a role in cell movement and cell division.
* Serves as a framework for positioning and moving organelles, and macromolecules within the cell.

### Microtubules

* Are critical to mitosis.
* They form the **mitotic spindle fibers** that separate chromosomes prior to cell division.
* Play a role in the organization of the cytoplasm: overall shape, organelle organization, movement of macromolecules, distribution of microfilaments.
* Cylinders of longitudinal arrays of **protofilaments** with a hollow center called a **lumen**.
* Each protofilament is a linear polymer of **tubulin** with polarity.
* **Tubulin** is a dimeric protein (alpha-tubulin and beta-tubulin).

### Microfilaments

* Smallest components of the cytoskeleton.
* Form connections with plasma membrane to give structure and affect movement.
* But also help to move cell in specific way during cell division.
* Are polymers of the protein **actin**.
* Actin is synthesized as a monomer called **G-actin**.
* Subunits are polymerized into **F-actin**.
* Have a polarity.

### ECM and Cell Wall Are “Outside” the cell

* For plant and fungal cells, cells walls consist in cellulose microfibrils.
* Bacterial cell walls are composed of **peptidoglycans**, long chains of GlcNAc and MurNAc
* These are held together by peptide bonds between a small number of amino acids, forming a netlike structure.

### The ECM

* Support role
* In animal cells: a network of **proteoglycans** surrounds **collagen** fibers.
* Regulate processes:
  + Cell motility and migration.
  + Cell division.
  + Cell recognition and adhesion.
  + Cell differentiation during embryonic development.

### Vacuoles

* Membrane containers for temporary storage and movement of compound.
* Plant vacuole large to keep the plant upright.

### Secretory Vesicles

* After being processed by the Golgi complex, materials are exported from the cell into secretory vesicles.
* They move to the plasma membrane and fuse with it, releasing their content outside the cell.
* Endomembrane system of the cell: ER, Golgi, secretory vesicles and lysosomes.

### Lysosome

* Single membrane organelles that store **hydrolases**, enzymes that can digest any biological molecules.
* A special carbohydrate coating on the inner lysosome membrane protects it from digestion.
* All of the lysosomal enzymes are **acid hydrolases**, are active at low pH, but not at higher pH (7.2), pH of the inner cell.

### The Phagolysosome

* **Phagocytosis**: ingestion of bacteria by phagocytes.
* **Phagolysosome**: merge of phagosomes with lysosomes to destroy bacterial pathogens.

### Peroxisome

* Similar to lysosome but contains peroxide.
* Helps to break down fatty acids.

### Hydrogen Peroxide

* H2O2 highly toxic to cells but can be formed into water and oxygen by the enzyme catalase.
* These reactions are confined to peroxisomes that contain **catalase**, so that cells are protected from the harmful effects of peroxide.
* Peroxide production is increased during cellular stress (infection, disease, UV exposure) and can serve as a useful biomarker for early infections.

### Viruses

* Small.
* Invade and infect cells, using synthetic machinery to produce more viruses’ particles.
* No cytoplasm, organelles, or ribosomes and consist of only a few different molecules of nucleic acid and protein.
* Consist of a coat (capsid) or protein surrounding a core, containing DNA or RNA.
* Viruses that infect bacteria are **bacteriophages** or **phages**.  
  Each virus has a characteristic shape, defined by its protein capsid
* Some viral capsids consist of a single type of protein, while more complex viruses have capsids with a number of different proteins
* Some viruses are surrounded by a membrane, and are called **enveloped viruses**; HIV.

### Bacteriophage

* In theory, bacteriophage exist for every type of bacterium.
* Present in every ecosystem.
* Can be highly specific for their hosts.

# Module 4 -Enzymes

## Enzyme Structure

* Catalysts lower the temperature which a chemical reaction occurs and make it easier to happen.
* All reactions occur at 98.6 degrees F.
* An extremophile is an organism that thrives in extreme environments.
* Activation Energy = Transition state energy – Reactant state energy
* With enzyme, we drop the activation energy.
* In the reaction sequence with catalyst, number of molecules increases for same amount of time and energy.
* The **shape** and the chemical environment inside the **active site** permit a chemical reaction to proceed more easily. Active site is where the reaction happens converting reactants to products.
* Molecule coming into is reacted by the **substrate**, has to fit within the shape, electronic bonding happens with the chemical environment, pulls the molecules apart to form the product.

## Factors Affecting enzymes

* **Substrate concentration**: more enzymes, more reactions
* **pH**: most of enzyme react at cellular pH
* **Temperature:** specific T at which enzyme works.
* **Inhibitor**: can slow down the rate of the chemical reaction, even can stop the enzyme to function.
* Denature of the enzyme: pull off water molecules from the enzyme.
* Enzyme substrate complex: when the substrate locks into the enzyme.
* At the end the enzyme can go back at the reactance side of the reaction coordinate diagram: **reversibility**.
* More material present, the more reaction takes place as long there is enough reactant present.
* Reaction velocity does not increase beyond **Vmax**.

# Module 5 - Membranes and the Endomembrane System

## Membrane Characteristics and Composition

### The functions of the membrane

* Serves as a permeability barrier between the cell and outside environment.
* Localizes and organizes different functions within the cell.
* Facilitates transport of different molecules within the cell between organelles and also its outside environment: nutrients, ions or water, and wastes.
* Helps the cell to perceive its external environment and respond appropriately thru receptor mediated signal transduction, transmission of signals from outer surface to cell interior.
* Mediate interactions with other cells.

### Membranes are sites of specific proteins and specific functions

* Different functions associated with membrane proteins: act as enzymes, integral proteins, signaling molecules
* **Differential centrifugation**: purify or tagging proteins based on localization and molecular weight
* **Immunostaining**: tag a specific protein with an antibiotic of an epitope of a protein. Allows different, assessment of different regions within the cell by microscopy.

### Regulation of Transport across the cell is a main function of membrane protein

* Receptors are specific proteins which by binding trigger changes in cell function allowing signals to be transmitted from the outer of the cell to its interior.
* Chemical signal molecules usually bind to membrane proteins, receptors, on the outer surface of the plasma membrane.
* Growth factor stimulates the cell to continue to replicating.

### Membrane Proteins Mediate Cell Adhesion and Cell-to-Cell Communication

**Cadherins** promote adhesion between similar types of cells in a tissue.

**Adhesive junctions**, **Tight junctions**, **Gap junction**.

### Membrane protein play role in other cell functions

* Uptake from the cell: **endocytosis** and secreting of substances: **exocytosis**.
* Autophagy: self-recycling.

### Membrane Structure: Fluid Mosaic Model

Model has 2 key features:

* A fluid lipid bilayer.
* A mosaic of proteins attached to or embedded in the bilayer.

### Three classes of membrane proteins

* **Integral** membrane proteins (hydrophobic segments)
* **Peripheral** proteins (hydrophilic locate on surface of the bilayer).
* **Lipid-anchored** proteins attached to the bilayer by covalent attachments to lipid molecules.

### Main class of membrane lipids: phospholipids, glycolipids and sterols

### Phospholipids

* Includes glycerol-based **phosphoglycerides** and sphingosine-based **sphingolipids**.
* **Amphipathic**: can easily form lipid bilayers.

### Glycolipids

* Glycerol-based and sphingosine-based **glycosphingolipids**
* Most common of glycosphingolipids are **cerebrosides** and **gangliosides**.

### Gangliosides

Expressed on the surface of the plasma membrane, can be involved in immune reactions.

### Sterols

* Main sterol in animal cell membrane is cholesterol needed to stabilize and maintain membranes, adds firmness and integrity to the plasma membrane and prevents it from becoming overly fluid.

### Membrane Asymmetry: most lipids are distributed unequally between two monolayers

* Refers to the difference in the kind of lipids and degree of saturation of fatty acids in the phospholipids.
* Once established membrane asymmetry does not change much.
* Movement of lipids from one monolayer to another requires their hydrophilic heads to move through the hydrophobic interior of the bilayer.

### Lipids move freely within their monolayer

* Rotation, lateral diffusion, transverse diffusion
* Movements are rapid and random.

### Membrane fluidity is measured using fluorescence recovery after photobleaching (FRAP).

### Membrane function properly only in the fluid state

* Membrane has an optimal temperature: more fluid with increase of T and vice versa.
* Below Tm, any functions that rely on membrane fluidity will be disrupted.
* Long-chain of acid chains and saturated fatty have higher Tm (saturated with H2 and no double bonds; they pack together well in the membrane.

## Membranes Functions and the Endomembrane System

### Membrane Composition

* Most membrane fatty acids vary in chain length and degree of saturation: helps those membranes to be fluid at physiological temperatures.
* Cholesterol molecules are rigid and can act as spacers within the hydrocarbon chain of phospholipids to prevent a tightly packed layer and helps to reduce the tendency of the membrane to gel.
* **Fluidity buffer**: help to maintain the correct amount to fluidity.
* **Sterols** decrease the permeability of membranes to ions and small polar molecules.

### **Lipid rafts** localized regions involved in cell signaling

* Regions with concentrated lipids: lipid microdomains.
* Dynamic and changing composition

### Function of the lipid rafts

* Thought to have roles in detecting and responding to extracellular signals.
  + transport of nutrients and ions across membranes
  + binding of activated immune system cells to their microbial targets
  + transport of cholera toxin into intestinal cells

### Receptors in lipid rafts

* Can precipitate the downstream cellular signaling cascade that organized with lipid rafts interior to the membrane., for ex. in phosphorylation event.
* Cholera toxin binds to a receptor which is associated with lipid microdomains.

### The membrane consists of a mosaic of proteins: evidence from Freeze-Fracture Microscopy

* Bilayer is frozen and then hit with diamond knife.

### Membrane Proteins are Oriented Asymmetrically Across the Lipid Bilayer

* Once in place proteins cannot move across the membrane

### DNA Sequencing

* Gives an idea of different protein regions and an idea of which portions of the protein are likely transmembrane regions: identification of likely structure and orientation of protein in membrane.

### The Endomembrane System

* Membranes define cellular borders and organelles but also involved in transport, signaling and adhesion: this system is called endomembrane.

### Variations in Amount of Rough and Smooth ER

### The Golgi Complex

* In ER, glycoproteins are sorted and packaged for transport via the **trans-Golgi** network or TGN.
* Materials to be exported from the cell are packaged into secretory vesicles.
* These move to the plasma membrane and fuse into it releasing their contents outside.
* ER, Golgi, secretory vesicle and lysosomes make up the **endomembrane system**.
* Many of the proteins trafficked through the membrane go through a process called **glycosylation**: addition of carbohydrates side chains to proteins.
* This forms **glycoproteins**.
* Initial Glycosylation occurs in the ER.
* All carbohydrate side chains have a common **core oligosaccharide**.

### Roles of the ER and Golgi Complex in Protein Trafficking

* Proteins synthesized in the rough ER must be directed to a variety of locations.
* Each protein contains a chemical tag, targeting to a specific transport vesicle.

### Protein and Lipid Tags

* A tag could be an amino acid sequence, a hydrophobic domain, or **oligosaccharide** side chain or some other feature (depending on the protein and destination).
* Membrane lipids can also be tagged to help vesicle to reach their destinations.
* Lipid tags can be one or more phosphate group.

### Exocytosis and Endocytosis: Transporting Material Across the Plasma Membrane

Endocytic vesicle.

Phagocytosis

* Ingestion of large particles up to and including whole cells or microorganisms.
* “Professional phagocytes”: neutrophils, macrophages, and dendritic cells.

### Receptor-Mediated Endocytosis

* A receptor-mediated drives endocytosis (or **clathrin-dependent endocytosis**), to ingest growth factors, hormones, serum proteins, enzymes, cholesterol, antibodies, iron, viruses, bacterial toxins.

### Process of receptor-mediated endocytosis

1. The receptor-ligand complexes diffuse laterally into coated pits.
2. Additional proteins on the cytosolic surface of the membrane: adaptor proteins: **clathrin**, **dynamin**, induce curvature and invagination of the pit.
3. Eventually the pit pinches off forming a coated vesicle.
4. The **clathrin** coat is released leaving an uncoated vesicle
5. Coat proteins and dynamin are recycled to the plasma membrane and the uncoated vesicle fuses with an endosome.

# Module 6 – Membrane Transport

## Transport Across Membranes: Overcoming the Permeability Barrier.

### Cell Transport

* Cell transport is the ability of the cell to move ions and organic molecules across membranes selectively.
* Most substances that move across the membranes are not macromolecules or fluids but dissolved ions and small organic molecules-solutes.
* Common ions transported: **sodium**, **potassium**, **calcium**, **chloride**, and **hydrogen**.
* Most of the molecules are metabolites-substrates, intermediates and products in the various metabolic pathways: **sugars**, **amino acids**, and **nucleotides**.
* More than 2/3 of the energy your body expends in the resting state Is used to maintain gradients of ions such as H+, K+, Na+, and Ca2+.
* **Electrochemical gradient**: concentration gradient + membrane potential.
* Stored energy gradient used to drive uptake of other solutes, synthesis of ATP.
* In nerve cells, gradients of K+ and Na+ responsible for transmission of nerve impulses.

### Simple diffusion: unassisted movement down the gradient.

* Because of the hydrophobic interior of the membrane, simple diffusion relevant only for small, nonpolar molecules.
* **Facilitated diffusion mediated by carrier protein**: **GLUT1** – glucose transporter.

**Anion** exchange protein Cl- in – HCO3 out.

* **Facilitated diffusion mediated by channel protein**. Aquaporin channel proteins.
* **Active transport** – Na+/K+ pump: 3 Na+ out – 2 K+ in, driven by hydrolysis of ATP, electrochemical potential across membrane.

### Osmosis of water across membrane

* Water molecules not charged; concentration similar on opposite sides of membrane.
* Water tends to move from regions of lower solute concentration (higher free energy) to regions of higher solute concentration (lower free energy).
* Diffusion always moves solutes toward an equilibrium.
* For most cells, water will move inward because the concentration of solutes is almost always higher inside a cell than outside.
* **Osmosis**: movement of water in response to differences in solute concentration.

### Second Law of Thermodynamics

* Diffusion always proceeds from regions of higher energy to lower free energy: molecules flow down their concentration gradient, and ions flow down their electrochemical gradient.
* 3 main factors affecting diffusion: size, polarity and size.

### Solute size

* Size rule holds up to about the size of glucose (ethanol and glycerol are able to diffuse, glucose not).
* Water, O2 and Co2 can diffuse across a bilayer by simple diffusion.

### Solute Polarity

* The more hydrophobic or nonpolar, a substance is, the more readily and rapidly it can move across the membrane.

### Ion permeability

* Lipid bilayer very impermeable to ions.
* Impermeability very important: cells must maintain an ion gradient across its plasma membrane in order to function: either a gradient of sodium ions (animal cells) or protons (mitochondria, chloroplasts).
* Proteins that facilitate ion transport provide hydrophilic channels.

### Rate of simple diffusion directly proportional to concentration gradient

* Simple diffusion thermodynamically always an **exergonic process** (no energy required)
* Simple diffusion is **a linear relationship** between the inward flux of the solute across the membrane and the concentration gradient of the solute, with **no saturation at high concentrations**.

### Facilitated diffusion: protein-mediated movement down the gradient

* **Facilitated diffusion is subject to saturation and follows Michaelis-Menten kinetics**.
* **Facilitated diffusion** **or passive transport** does not require energy, **process is exergonic**:
* Example movement of glucose across the plasma membrane of an erythrocyte. Concentration of glucose is higher in blood than in erythrocytes, so transport of glucose across the plasma membrane is passive.

### Carrier and channel proteins facilitate transport by different mechanisms

* Channel proteins form **hydrophilic channels** through the membrane.
* **Pores**: large and nonspecific channels. Formed by transmembrane proteins called **porins**, allow molecules weight up to about 600Da to diffuse across the membrane.
* Most channels are small and highly selective: **ion channels** – **more rapid** no need for a protein to change its shape and capture a solute.
* Carrier proteins are called **permeases**.
* Like enzymes very specific.
* Carrier proteins differ in number of salutes transported (**uniport**), and the direction in which they move.
* Glucose carrier protein is a **uniporter**.
* When two solutes are transported simultaneously and their transport coupled: **co-transport**.
* Same direction: **symport** – opposite direction: **antiport**.

### Glucose transporter: a uniport carrier

* The erythrocyte is capable of glucose uptake by facilitated diffusion, in erythrocyte: GluT1 (glucose transporter).
* GluT1 provides a hydrophilic channel for n-glucose molecules alternating between T1 and T2 conformations.

### Active transport: protein-mediated movement up the gradient.

* Always moves solutes away from thermodynamic equilibrium (up a concentration or electrochemical gradient), therefore always requires energy (ATP -> ADP).
* Process **endergonic**, occurs only when coupled to an exergonic process.
* Performs 3 major functions:

1. Makes possible the uptake of essential nutrients from environment or ssurroundingss of the cell.
2. Allowed secretory products and waste materials to be removed from the cell or organelles.
3. Enables the cell to maintain constant, non-equilibrium intracellular concentration of specific ions: K+, Na+, Ca2+, H+.

### Pumps

* Membrane proteins involved in active transport.
* Passive transport: inherently nondirectional w.r.t membrane. Active transport has directionality: unidirectional or vectorial process.
* Direct or primary active transport: coupled to an exergonic chemical reaction, most commonly hydrolysis of ATP.
* Indirect or secondary active transport: driven by the co-transport of cations-protons down the electrochemical gradient: exergonic inward movement of protons provides energy to move the solute against its concentration gradient.

### Na+/K+ pump

* Uses ATP for energy, example of a transport ATPase.
* Directional: 2 K+ in, 3 Na+ out.
* E1, E2 conformational changes.

# Module 8 - DNA, Chromosomes, the Nucleus

## Information and DNA

* **Replication**: 2 DNA copies are distributed to daughter cells when cell divides.
* **DNA replication**: DNA synthesis.
* **Mitosis**: cell division.
* **Transcription**: involves the use of selected segments of DNA as templates for the synthesis of mRNA and other RNA molecules.
* **Translation**: amino acids are joined in a sequence dictated by the sequence of nucleotides in mRNA. protein synthesis (cytoplasm).
* **Expression of genetic information**: transcription and translation.

## DNA

* Helix is right-handed.
* Contains 10 nucleotide pairs per turn and advances 0.34nm per nucleotide pair.
* Each complete turn of the helix adds 3.4nm to the length of the molecule.
* Diameter of helix: 2nm.
* Pyrimidine-Purine pairing.
* Two chains of DNA complementary to each other.
* Two chains create a **major groove** and **minor groove** which play significant roles in the interactions of variety of molecules.
* **Antiparallel orientation** of the two DNA strands.
* Nucleotides linked by **phosphodiester bonds**.
* **5’-3’ orientation- 3’-5’ in opposite strand**: 5’ carbon linked to 3’ carbon.
* **Supercoiled DNA**: DNA double helix twisted upon itself.
* **Circular DNA** molecules are **negatively supercoiled**.
* Raise temperature to denature DNA: **DNA melting temperature** TM.
* G-C: 3 H2 bonds, A-T: 2 H2 bonds.
* **Strand separation** can be readily achieved because the two DNA strands are bound together by relatively **weak, non-covalent bonds**.
* Strand separation is integral part of DNA replication and RNA synthesis, transcription.
* By raising slowly, the temperature: **DNA denaturation happens** (two strands separated), when lowering the temperature, the reverse happens and **is DNA renaturation**.
* Maximum absorption of UV lights **around 260nm**. As strands separate, absorbance of solution increases rapidly.
* Ability to renature nucleic acids forms the basis of **nucleic acid hybridization**.

## DNA in the genome

* Genome of an organism or virus: complete copy of all the genetic information.
* For many viruses or prokaryotes, genome resides in on DNA molecule.
* Eukaryotic cells have a nuclear genome, a mitochondrial genome.
* Nuclear genome consists of multiple DNA molecules.
* Genome size increases with complexity of the organism with exceptions.
* Genome size is less important than the number and identity of functional genes.
* DNA must be efficiently packaged into cells and yet remain accessible to cellular machinery for DNA replication and transcription.

## Restriction Enzymes

* Cut DNA molecules in places where it encounters a specific recognition sequence, called a **restriction site**.
* Cleave DNA into fragments ranging from a few hundred to a few thousand base pairs
* More amenable to manipulation.
* Gel electrophores is the technique used to separate the fragments to each other, their number and lengths.

## The Nucleus

* Site where the chromosomes are localized and replicated and where DNA is transcribed.
* Both the repository of most cell’s genetic information and control center for expression of the information.