# Haalim Study

**What is Life? Follow us on Youtube for short lectures**
**https://www.youtube.com/playlist?list=PLP4jqZZBeTPpOiQ9TpXV23-sm9yuuY3Nl**
Anything is living if:

- It can acquire energy from the environment, e.g., plants acquire energy using sunlight and carbon dioxide and animals gain energy by eating plants like goats eat plants.
- It is capable of reproducing itself, e.g., all animals produce young ones like lions produce cubs. Plants also reproduce seeds to give rise to new plants.
- Mutating / changing itself: all organisms have a property of mutations, i.e., their heredity material – DNA changes itself during division or other times and the result is change in any characteristic of the organism. This characteristic may be beneficial or harmful; organisms survive better if mutation is beneficial and may die if it is harmful

**Biology – The Study of Life**

Biology (Bio – life; logos – study, reasoning); biology is hence the study of life or living organisms. Biology is about exploring the living part of the world, e.g., studying about animals, plants and even microorganisms is biology. Biology have many subdivisions; for example, anatomy – the study of structures, physiology – the study of functions, microbiology – the study of microorganisms and many more. The exploration of life helps in understanding the phenomena of nature and effective utilization and management of natural resources. We can find solutions to various problems for example treatment for various diseases could be discovered, methods for energy production from biological materials may be found, and e.g., few bacteria can produce fuel from grasses.

**Levels of Organization in Life**

- Atoms
- Molecules
  - Micromolecules
  - Macromolecules
- Cells
- Tissues
- Organs
- Organ-systems
- Organisms
- Populations
- Communities
- Ecosystems
- Biosphere

**Atoms**

Greek: a, not; tom, to cut: The smallest component of an element that have all the properties of that element. In nature, 92 kinds of elements are present, out of which only 16 make the living organisms, called bioelements. Bioelements are Oxygen (O), Carbon (C), Hydrogen (H), Nitrogen (N), Calcium (Ca), and Phosphorus (P). These elements make 99% of living mass. Others ten elements make 1 % of total living mass named Potassium (K), Sulfur (S), Chloride (Cl), Sodium (Na), Magnesium (Mg), Iron (Fe), Copper (Cu), Manganese (Mn), Zinc (Nz), and Iodine (I).

All living things consist of atoms, like all other forms of matter. Atoms consist of "subatomic particles"; charged or not charged. These include electrons which are negatively charged particles, protons which are positively charged and neutrons which have no charge. Protons and neutrons are present inside the center and the electrons revolve around these in orbits. Atoms do not live in isolation but join together to make molecules (compounds).

**Molecules**

Atoms join together by a process called "bonding" to each other to construct molecules.

Bonding is of two types.

- Ionic bonds
- Covalent bonds

In ionic bonding one atom gives one or more of its electron to the other atom which is called a donor and the other receive the electron called recipient. The donor atom then becomes positively charged and the recipient becomes negatively charged. In covalent bonding, however, the atoms share one or more of their electrons and these electrons revolve in the orbit of both atoms. Covalent bonding is more strong form of bonding.

On the basis of their size molecules are categorized into micromolecules and macromolecules. Micromolecules are the molecules with low molecular weight, e.g., glucose, water. Macromolecules are the molecules with high molecular weight, e.g., proteins, carbohydrates and lipids. An organism consists of enormous number of biomolecules different types. Though some organisms are unicellular, i.e., consist of one cell only. Many other organisms are multicellular, i.e., these consist of many cells.

***Figure 1.1 Covalent Bond Vs. Ionic Bond***

**Organelles**

Molecules make organelles. Organelles are sub-cellular structures, assemble together to make cells – the units of life, e.g., mitochondria, lysosomes, Golgi bodies, nucleus. For example, mitochondria of a cell (Singular: mitochondrion) is called "powerhouse" of the cell. This organelle is present in the cytoplasm of the cells and makes energy for the cells hence called "power house". These are found in both plants and animals. Another example is nucleus present in almost all cells.

***Figure 1.2 Organelle: Nucleus***

**Topic No 02**

**Cells – the basic unit of life**

All the living organisms consist of cells. Cells are called the basic units of life. Cells are specialized in their structure and functions. There are different types of cells present in the bodies of multicellular organisms. But some organisms like amoeba consist of only one cell. Cells are categorized based upon the placement of their nuclear material into prokaryotic and eukaryotic cells. Prokaryotic (Pro: old, Karyotic: related to nucleus) cells are those cells that do not have a true nucleus – it means that their nuclear material is not covered by a membrane. While eukaryotic cells have a true nucleus, i.e., their nuclear material is covered by a membrane called nuclear membrane. Sometimes a cell makes a whole unicellular organism, like Prokaryotes and Protists. A variety of cells makes a single, multi-cellular organism.

***Figure 2.1 Animal & Plant Cell***

**Stem Cells**

These are the undifferentiated cells in which most of the genes are switched on and these have a potential to make almost all cells of the body. These cells are present in a few places in adult organism or present in the embryos. These are useful for human beings because these can be used in making organs of any type which may be damaged by for example a disease.

**Tissues**

These are the groups of similar kind of cells which perform a same function. The tissues perform a common function, specialized to the tissue. For example, epithelial tissue that makes the skin.

*Figure 2.2 Onion Epidermis (Tissue)*

**Organs and organ systems**

Tissues group and work together to make a unit called organ. Different tissues in an organ work differently to perform collective function of the organ. For example in stomach, there are muscle tissues that contract and relax for grinding and there are secretory cells which secrete gastric juices to digest the food. Collective action is the secretion of gastric juices and its mixing.

*Figure 2.3 Human Digestive System*

In the simple to complex organisms, many organ-systems are present, for example, in humans digestive system, cardiovascular system, respiratory system and many more are present that work for a specific purpose. Digestive system consists of oral cavity, esophagus, stomach, intestine, pancreas, liver and rectum. Cardiovascular system consists of heart, vessels and blood.

**Organism**

Organ-systems join together to constructs organisms. In an organism, the organs and organ-systems coordinate to perform the activities of the whole organism. For example, in human's brain control the activities of most of the organs and organ-systems. If a person is running; cardiovascular system provides it oxygen and nutrients, muscles contract and relax for movement and nervous system coordinate all of these functions.

**Topic No 03**

**Population:**

All organisms of a species living in an area at a particular time are called population, like all deer in a forest. Biologists study populations to explore the interactions between organisms. For example, interactions between male-male, female to female or else.

*Figure 3.1 Ducks in a local park*

**Community & Ecosystem**

Different populations living in an area in a particular time, for example, in a forest plants, animals, algae, fungi live together are called a community. Populations interact with each other and also to the abiotic factors of the area to make Ecosystem, for example, a lake ecosystem.

*Figure 4 Plant community in a forest*

**Biosphere**

The part of the world covered or inhabited by the living organisms is called biosphere. This is also called zone of life on Earth. Biosphere includes all ecosystems, like forests, lakes, oceans and valleys where biotic components exist.

**Topic No 04**

**What is Cell Theory?**

Cell theory in its modern form states:

- All living organisms are composed of one or more cells.
- Cells are the smallest living things, the basic unit of all living organisms.
- Cells arise only by division in pre-existing cells.

**How Cell Theory Developed?**

- Cells were first described by Robert Hooke (Curator for instruments of Royal Society of London) in 1665.
- Leeuwenhoek (Textile Merchant) observed tiny organisms in pond's water, called them animalcules.
- In 1809, de-Lamark proposed that nobody can have life if its parts are not cellular tissues.
- Robert Brown discovered nucleus in the cell.
- In 1838, Schleiden stated that all plants are aggregates of individual cells.
- In 1839, Schwann stated that all animal tissues consist of cells.
- In 1855, Virchow proposed that cells arise from pre-existing cells.
- In 1862, Pasteur provided experimental proof for the above.

**Cellular Components**

Cells consist of several components. Some important and common ones are discussed

below:

- Cell membrane
- Cell wall
- Cytoplasm
- Cytoskeleton
- Organelles (e.g. nucleus, ribosomes)

*Figure 4.1 A Typical Animal Cell*

**Cell Membrane**

- Cell membrane is the external most layer that covers the cell from outside.
- Functions of the cell membrane are:
  - It acts as a barrier, i.e., it separates the cell from environment.
  - It provides protection to the inner parts of the cells including all the organelles.
  - Another important function of cell membrane is transport of materials. Cell membrane manages the transport in and out of the cell.

**Figure 4.2 Structure of Plasma Membrane**

**Topic No 05**

**Structure and Function of Cell Membrane**

Structure of cell membrane is described by Fluid Mosaic Modal. It consists of lipid bilayer, proteins and carbohydrates. Lipid bilayer provides it with fluidity, flexibility and transport of lipid like substances. Proteins are integrated inside the membrane or present on its peripheries called **integrated proteins** and **peripheral proteins**, respectively.

Some proteins are trans-membrane, i.e., these are integrated and their ends (domains) are exposed from both intracellular and extracellular side of the membrane. These make channels for transport of materials, e.g., aquaporins are the protein channels

for transport of water. Proteins and glycoproteins (carbohydrates attached to proteins) make receptors for message transmission. Cells carry out their message transmission with other cells or environment with the help of these glycoproteins mostly, we call these **receptors**.

**Movements across Cell Membrane**

- Some molecules can pass directly e.g. few lipids.
- Other molecules need channels to pass through; channels are made up of proteins.

There are different types of channels for different molecules like water channels and ion channels (Sodium channels, Calcium channels).

**Topic No 06**

**Cell Wall**

It is the outermost covering in many organisms surrounding the cell membrane.

Prokaryotic cells, fungi and plant cells have a cell wall around their cell membrane. Cell wall makes the outermost covering in these organisms. Cell wall is tough in comparison to cell membrane; it is a rigid structure. Cell wall in plants consists of cellulose, hemicellulose, and pectin. Fungal cell walls consist of a long polymer called, chitin.

Prokaryotic cell wall consists of a polymer, called peptidoglycan. Functions of the cell wall are protection, shape, strength and support. Plant cells have 2 types of cell wall, primary cell wall and secondary cell wall. Primary wall consists of mainly cellulose, hemicellulose and pectin. Secondary cell wall contains cellulose and some other molecules like lignin which make it stronger structure.

*Figure 6.1 Primary Cell Wall*

*Figure 6.2 Secondary Cell Wall*

**Cytoplasm**

Cytoplasm is a semitransparent substance present between plasma membrane and the nucleus. It contains water in which organic (e.g. proteins, carbohydrates) and inorganic materials (e.g. salts), which are partially or fully dissolved in this solution. Cytoplasm provides space for metabolic reactions (e.g. glycolysis). It also provides space for functioning of organelles and metabolic reactions.

**Topic No 07**

**Cytoskeleton**

Cytoskeleton is the skeletal framework of the cell – a network of filaments and tubules.

There are three types of cytoskeletal elements called microfilaments, intermediate filaments and microtubules.

Microfilaments are the smallest in their diameter. These help in movement of organelles and the cell. These consist of helical chains of a protein called actin e.g. in muscle cells these are highly modified. Intermediate filaments are intermediate in size. These consist of different proteins belong to a protein family called keratins. These filaments help in maintaining shape and placement of the cell and its various parts. These also provide protection to various parts of the cell particularly to the nucleus. Microtubules are largest in diameter, these filaments consist of a protein called tubulin which makes dimers and then long and large hollow tubes. These help in movement of the organelles inside the cells and also in movement of the cell itself. Cilia and flagella consist of microtubules.

These filaments help in maintaining shapes of organelles and cell. For example these make the nuclear lamina, a layer that maintain the shape of the nucleus and give it support.

*Figure 7.1 Cytoskeletal fibers*

**Topic No 08**

**Cell Organelles**

These are sub-cellular structures that perform a particular function. These include nucleus, mitochondria, endoplasmic reticulum and many more.

**Nucleus**

It is the organelle that contains genetic material. It is present in the center in animal cells usually. In plant cells, it is present on a side due to presence of a large vacuole. Nucleus is covered by nuclear membrane with nuclear pores. It is filled with a fluid called nucleoplasm. It also contains a denser body called nucleolus which is involved in ribosomal RNA production. Genetic material is present inside the nucleus in most of the eukaryotic cells, though, some cells have extra nuclear DNA. DNA is present in the form of chromosomes, which are visible during cell division.

**Ribosomes:**

Ribosomes are the protein making machinery of the cells. These are present free in cytoplasm or attached to endoplasmic reticulum. A large number of ribosomes are present in cells. Eukaryotic ribosomes are slightly different than prokaryotic ones in their size.

*Figure 8.1 Structure of Ribosome*

**Mitochondria**

Mitochondria are called power house of the cell. These make energy for the cells in the form of ATP (Adenosine Tri Phosphate). ATP is the biological or chemical form of energy. Mitochondria have a double membrane, one is called outer and the other is inner membrane. Mitochondria are filled with matrix containing circular DNA molecule and other molecules including the enzymes. Mitochondria are self-replicating organelles.

*Figure 8.2 Structure of Mitochondria*

**Plastids**

Plastids are the double membrane bound organelles, present in plants and in the other organisms which are producers such as algae. These are of three main types:

- Chloroplasts are present in the green parts of plants. These are green in color and their color is due to chlorophyll, the green pigment. These help in photosynthesis.
- Chromoplasts are the organelles present in the fruits and flowers of the plants. Beautiful colors of fruits and flowers are due to presence of Chromoplasts which contain red, yellow, orange and more colored pigments.
- Lecuoplasts are the plastids present in the roots and tubers. These are colorless pigments and their function is to store various materials in the roots and tubers, e.g., potatoes.

Plastids have a double membrane system. Their membranes are called outer membrane and inner membrane. They have a membrane system called thylakoid. Stacks thylakoids are called grana. They have a matrix inside inner membrane which is called stroma.

These organelles like mitochondria have their own circular DNA. These are self-replicating organelles.

*Figure 8.3 Structure of a Chloroplast (A kind of Plastid)*

**Endoplasmic Reticulum**

Endoplasmic reticulum (ER) is a network of interconnected channels present inside the cells. This is of two types: Rough ER and Smooth ER. Rough endoplasmic reticulum is the rough due to the ribosomes present on its surface. This type is involved in protein modification. The other type is free of ribosomes so that shape of this one is smooth giving it its name. SER is involved in metabolism of lipids and carbohydrates.

*Figure 8.4 Endoplasmic reticulum*

**Golgi Apparatus**

This is also called Golgi bodies or Golgi complex. This is also very important organelle of the cells. It was discovered by Camillo Golgi. Golgi apparatus consist of flattened disks called cisternae which are associated with endoplasmic reticulum. These are called the post office of the cell because these pack materials in the form of vesicles. For example the proteins formed by the ribosomes and modified by the endoplasmic reticulum enter in the cisternae and here these are packed in the vesicles and transferred to the part of the cell where these are required or secreted out of cells.

**Centrioles**

These are hollow and cylindrical bodies present near the nucleus of the animal cells. It is also present in some lower plants. A pair of Centrioles is collectively called centrosome.

Their function is during cell division. These make the spindle fiber during cell division in animal cells.

**Vacuoles**

Vacuoles are membrane bound organelles present in most of the cells. Their major function is storage of various materials including food materials to waste materials. If these store food then these are called food vacuoles. Their size is from small to very large in different cells according to the requirements of the cells. In mature plant cells a single large vacuole is present. Contractile vacuole in unicellular fresh water organisms helps in removal of water from the body.

**Lysosomes**

Lysosomes are membranous sacs filled with enzymes. Lysosomes are spherical bag like structures that are bound by a single layer membrane. These are found in all eukaryotic cells and act as 'garbage disposal' or the 'digester' of the cell. These act as disposal system of the cell. They break down complex proteins, carbohydrates, lipids and other macromolecules into simpler compounds. These simple compounds are returned to the cytoplasm and are used as new cell building materials. They are used for digestion of cellular waste products, dead cells or extracellular material such as bacteria.

**Topic No 09**

**Genetics**

Genetics is a branch of biology in which we study the modes of inheritance that how genetics information is passed from one generation to next.

**Historic Perspective**

It has been long recognized that children inherit features from their parents. Humans were breeding 5000 years ago, these people recognized the genetic information is passed from one generation to next children offspring take features and characters from their parents. The prevailing view of hereditary was blending which basically implies that information once it is passed on to next generation it does not separate. For example we mix two dyes blue and red we will get purple dye. Once we formed the purple dye we cannot take out red or blue color separately. Mendel did a lot of experiments which proved that the principle of blending is not valid.

Like If purple dye wants to reproduce purple dye can only contribute purple color to its offspring. Mendel proved that it was incorrect when the purple dye is going to reproduce it will give again the two characteristics the Red and Blue color will separate in the offspring.

**Important Definitions:**

A character is an observable feature such as flower color.

A trait is a particular form of a character such as white flower.

A heritable charter trait is one that is passed from parent to offspring.

**Topic No 10**

**What is Genetics?**

- Genetics is the study of genes, heredity and variation.
- It is considered as a field of Biology.
- The principles of heredity were explained by Gregor Mendel in 1866.

*Figure 10.1*

*Figure 10.2*

Gregor Mendel used garden pea as experimental plant for formulating the laws of heredity, following were the properties of garden pea.

- Seed in a variety of shapes and colors.
- Self, cross pollinate.
- Takes up little space.
- Short generation time
- Produces many offspring.

**Topic No 11**

**Common Genetics Terminologies**

What is Character: A heritable feature (skin color, height etc). What is Trait: variant for a character (i.e. brown, black, white etc). What is True-breed: all offspring of same variety.

Different generations of a cross can be P generation (parents)

F1 generation (1st filial generation) F2 generation (2nd filial generation)

**Pure Cross:** A cross between a true breed plant/animal with another true breeds plant/animal is called pure cross

**Hybrid Cross:**

True breeding X True breeding

WW X ww

F1 generation X F1 generation

Ww X Ww

Genotype and Phenotype: Genetic make-up of an organism is called Genotype while physical appearance of an organism is called Phenotype.

*Figure 11.1 Genotype & Phenotype*

Dominant and Recessive: when one characteristic expresses itself over the other i.e. round over wrinkled was dominant in Gregor Mendel experiments while the trait that does not show through in the first generation is called as recessive trait i.e. wrinkled.

**Topic No 12**

In this lesson we look at Mendel's experiment and what was the Mendel's question, how he designed the experiment and how he was able to conclude the experiment.

**Mendel's Subject:**

Mendel was working on pea plants (Garden pea plants), easy to cultivate, short life-span and several other characteristic. Flowers are reproductive organ of these plants, as most flowers have both male and female reproductive organs.

The ovary is the female sex organ as shown in Figure 12.1. The stigma is the part of the flower where pollen lands and extends a tube, pollen is sperm equivalent of the plants.

Anthers are the male sex organ, these are attached to stamen which are filamentous structures and anthers produce pollen which can then land on stigma and fertilize the egg inside the ovary.

So, Mendel use these plants it was easy to grow them as mentioned.

***Figure 12.1 Anatomy of a pea flower***

**Mendel's Technique:**

Mendel's used a paint brush he rubbed it on the anthers and he collected pollen in the tip of the brush and later dusted on the stigma of the female part of the flowers, as a result the pollen fertilize the egg and resulted in seed which he could grow and look as the next generation of the cross of the breeding experiment between two types of flowers.

**Monohybrid Cross:**

"A monohybrid cross is the hybrid of two individuals with homozygous genotypes which result in the opposite phenotype for a certain genetic trait."

"

Monohybrid cross is responsible for the inheritance of one gene. It can be easily shown through a Punnett Square.

Monohybrid cross is used by geneticists to observe how homozygous offspring express heterozygous genotypes inherited from their parents.

**Topic No 13**

**Mendel's First Law of Segregation**

Mendel developed his genetic laws in 1866, using pea plants, but they were not rediscovered in the scientific literature until 1900. Mendel stated his laws in terms of "chance" or probability. In modern terminology, Mendel's First Law states that for the pair of alleles an individual has of some gene (or at some genetic locus), one is a copy of a randomly chosen one in the father of the individual, and the other if a copy of a randomly chosen one in the mother, and that a randomly chosen one will be copied to each child. He also said that each allele has an equal chance to be the one copied, and that the copying of alleles to different offspring or from different parents are independent. This very basic set-up underlies all of genetics.

Since a parent has two alleles of each gene, the parent has 0.5 chance of passing one of the alleles to the offspring. For example, if a parent has a normal CF gene, and a mutant CF gene, he or she has a 0.5 chance of passing the mutant gene to the offspring. Likewise, he or she has a 0.5 chance of passing the normal gene to the offspring.

Segregation of the sex chromosomes works the same way. In the case of X-linked genes, the mother has two alleles for each X-linked gene, therefore she has 0.5 chance of passing one of them to an offspring. The father, on the other hand, only has one X, and he only passes it to his daughters. Therefore, the chance that he will pass an allele of an X-linked gene on to a daughter is 1, and that he'll pass it to a son is 0. He passes his Y chromosome to each son.

**Experiment:**

***Figure 13.1 Mendel's Experiment (1st Law of Segregation)***

**Topic No 14**

**Mendel's Second Law OR Mendel's Law of Independent Assortment**

Mendel's 2nd law states that during gamete formation the segregation of each gene pair is independent of other pairs. Mendel's 2nd law is often referred to as the principle of independent assortment. Both of Mendel's laws are about segregation, which is the separation of allele pairs. The law states that the separation of one pair of alleles isn't related to the separation of other pairs of alleles, and so is very important in Mendelian genetics. The only time there is an exception to this rule is when linkage is involved.

**Reference:**

Hartl D.L. and Ruvolo M. (2011) Genetics: Analysis of genes and genomes, page 91, 8th Edition, Published by Jones and Bartlett Learning

**Topic No 15**

**Diversity of Cells and Mitosis**

**Animal Vs. Plant Cell**

**Animal Cell**

- Cell membrane is outer most layer
- Many small vacuoles
- Nucleus in the center
- Roughly round and/or have
- variable shapes
- Plastids are absent

- Cilia present
- Centrioles present

**Plant Cell**

- Cell wall is present outside the cell membrane
- Have a large central vacuole
- Nucleus on a side
- Roughly rectangular shape due to presence of cell wall
- Plastids are present
- Cilia rarely present
- Centrioles absent in most of the plants

*Figure 15.1 Animal and Plant Cell*

**Prokaryotic Cell Vs. Eukaryotic Cell**

**Prokaryotic Cell**

- No defined nucleus but a nucleoid region
- No membrane bound organelles
- Small in size (e.g. 1-2 microns in bacteria)
- Cell wall consist of peptidoglycan (a polymer of amino acids and sugars)

**Eukaryotic Cell**

- Defined nucleus with nuclear membrane
- Membrane bound organelles are present
- Larger than prokaryotic cells on average (20 microns of animal cells)

Cell wall consist of cellulose (plants) and chitin (fungi)

**Topic No 16**

**Cell Division**

Division is the property of cells. Cells have to divide for various purposes, for reproduction or for growth and repair of the tissues. There are two types of the cells called somatic cells and germ line cells. Somatic cells are those which make all the tissues of the body except for some cells which are involved in reproduction. Few cells involved in reproduction and are meant for making gametes for reproduction.

There are two types of the cell divisions called Mitosis and Meiosis. Mitosis is the division of the somatic cells and also serves as means of asexual reproduction. Meiosis is however involved in division of the germ line cells.

*Figure 15.2 Stages of Mitosis in Onion Root Tip Cells*

**Cell Cycle**

Cells go through a cyclic process in which they pass by various phases over time. These phases include a phase of rest, high metabolic activity and division. It is also defined as the sequence of events or a cyclic process between divisions of the cell. The cell cycle consists of the following phases:

- Interphase (consist of Gap 1 phase, Synthesis Phase, Gap 2 phase)

- M Phase (Mitosis or Division Phase)
- Resting or G0 Phase (Gap 0, read as Gap not)

**Interphase**

Interphase consist of following phases. This is normally called as rest phase but actually it is a phase of high metabolic activity.

**Gap 1**

Cells increase in size in Gap 1. They produce materials required for DNA synthesis. The *G1 checkpoint* control mechanism ensures that everything is ready for DNA synthesis.

**Synthesis phase**

DNA replication occurs during this phase. An S phase check point checks that whether the synthesis of DNA is correctly done or not. If everything is correct cell continue to the next phase and if not then cell wither have to die or correct its errors.

**Gap 2**

During the gap between DNA synthesis and mitosis, the cell will continue to grow. Cell prepares all the materials required for division, e.g. microtubule proteins. *The G2 checkpoint control mechanism ensures that everything is ready to enter the M (mitosis) phase and divide.*

**Cell Division or M- Mitosis Phase**

Cell growth stops at this stage and cellular energy is focused on the orderly division into two daughter cells. A checkpoint in the middle of mitosis (called *Metaphase Checkpoint*) ensures that the cell is ready to complete cell division.

**Resting or G0 (Gap 0)**

A resting phase in which the cell has leaves the cycle and has stopped dividing. G0 starts from G1 and cell may sustain in G0 may be for years.

**Topic No 17**

**Mitosis**

Mitosis is the cell division that results in two daughter cells which are like each other. Though, mitosis is a term that is used to describe the nuclear division.

Cell division consists of following phases:

**Karyokineses** – division of nucleus

- Divided into Prophase, Metaphase, Anaphase and Telophase

**Cytokinesis** – division of cell

- Different in animal and plant cells

**Karyokinesis**

**Prophase**

- Chromatin material condenses and chromosomes becomes visible
- Each chromosome is replicated (duplicated) and consist of two sister chromatids

- At the end of the stage nuclear membrane disappear
- Centrioles move towards poles of the cell and microtubules starts forming

**Metaphase**

- Chromosomes arrange themselves at the equator
- Kinetochores are attached to the microtubules

**Anaphase**

- Chromosomes starts moving towards the poles
- The sister chromatids separate from each other, so each sister chromatid move towards a pole

**Telophase**

- Chromosomes (sister chromatids) reaches at the poles
- Nuclear material starts ***de-condensing*** again
- Nuclear membrane starts forming

The nuclear division is complete, next is the **Cytokinesis or division of the cell.**

**Cytokinesis**

Cytokinesis in plant and animal cells is different from each other. In plant cells, a **cell plate** starts forming in the center of the cell and moves towards sides. The cell plate divides the cell into two cells. The cell plate consists of material produced by Golgi bodies in vesicles. This material contains the cell membrane and cell wall components.

Cytokinesis in animal cells occurs in a different way. In animal cells a cleavage furrow is formed by invagination of cell membrane. This process occur with the help of cytoskeleton (microfilaments particularly). The furrows divide the cell into 2 daughter cells.

**Figure 17.1 Cleavage in Animal Cells**

**Topic No 18**

**Importance of Mitosis**

Mitosis is the cell division which helps in development and growth processes of the cells and hence the tissues. Development of new cells is a requirement in many parts of the body like epithelia usually keep growing. Replacement of cells and wound healing is another requirement of the organisms which also need new cells. Regeneration is a capability of some organisms. They came make their lost parts. This process also needs mitosis. Mitosis also serves as a means of asexual reproduction in various organisms.

**Errors in Mitosis**

Cells divide correctly most of the times because of many check points at various phases but sometimes it may go wrong. If it happens due to any reason the result is usually serious problems. Uncontrolled division of the cells may result into abnormal tissue growth and cancer.

**Topic No 19**

**Cell Division – Meiosis (Reduction Division)**

**MEIOSIS - CONSIST OF TWO DIVISIONS**

Meiosis occur in germ line cells to make gametes, gametes are formed for sexual reproduction. Meiosis is also called reduction division because it results in four daughter cells which are haploid. We know that chromosomes occur in pairs e.g. human have 46 chromosomes in 23 pairs which is called a diploid number. The chromosomes in each pair are called homologous because these are like each other and are complementary to each other. Meiosis makes cells that have a half number of the chromosomes in each daughter cell called a haploid number also.

**Diploidy and Polyploidy**

The condition of having two sets of chromosomes is called diploidy (2N number of chromosomes). The gametes formed by meiosis, hence, are called haploid (N number of chromosomes). For example, in humans 2N is 46 and N is 23 in each gamete, when gametes combine in fertilization the chromosome number is retained to 2N. Some plants have more than two sets of chromosomes and are called polyploids. This characteristic is called polyploidy.

*Figure 19.1 Homologous Chromosomes (A) Before Replication; (B) After Replication*

**Phases of Meiosis**

- Meiosis I

  - The reduction division
  - Chromosome number becomes half in each daughter cell (N)

- Cytokinesis I
- Meiosis II

  - Just like mitosis
  - Chromosome number remains same in daughter cells

- Cytokinesis II

**Topic No 20**

**Meiosis I**

- **Meiosis I consist of following phases:**

  - Prophase I
  - Metaphase I
  - Anaphase I
  - Telophase I

**Stages of Meiosis I**

**Prophase I** is a long phase in meiosis. It consist of following stages:

- It is marked by the pairing of homologous chromosomes (synapses) and recombination (exchange of the parts of chromosomes.
- Paired chromosomes are called **Bivalents or Tetrads.**

*Figure 20.1 Chiasmata formation to Recombination*

**Metaphase I**

- Chromosomes attach to the spindle fibers by kinetochore, one kinetochore per a chromosome and not per chromatid.
- One homologue on each side so there is a 50-50 chance to get each parents chromosome.

**Anaphase I**

- The chromosomes move towards the poles.
- A homologue, consist of two chromatids moves and sister chromatids do not separate.

The result is half number of chromosomes towards each pole.



**Telophase I and Cytokinesis**

- Chromosomes reach at poles, half on each side.
- Cell divides and then starts meiosis II.

*Figure 21.1 Prophase I, Metaphase I & Anaphase I*

**Events of Meiosis II**

- Karyokinesis

- Prophase II
- Metaphase II
- Anaphase II
- Telophase II

- Cytokinesis

- Each cell divides into two cells.

At the end of meiosis each cell divides into **4 haploid cells.**

**This division is different in males and females.**

*In females*, after first meiotic division (meiosis I), cytoplasm is unequally distributed and one large and other small cell are produced. The small cell is called a polar body. Then both of these cells carry out meiosis II. Polar body divides into 2 polar bodies. The large cell however is divided into one ovum (large) and another polar body. So that meiosis results into 1 ovum and 3 polar bodies.

*In males*, both meiotic divisions' results into equal sized cells called sperms.

**Comparison of Mitosis and Meiosis**

**Mitosis**

- Cell divides into 2 daughter cells
- Alike in males and females
- Chromosome number remains equal (2N) in daughter cells

**Meiosis**

- One cell divides into 4 daughter cells
- Different in males and females

Chromosome number becomes half (N) in daughter cells

**Topic No 22**

**Importance of Meiosis**

Major advantage of meiosis is the genetic variations by recombination (crossing over).

During prophase I of the meiosis crossing over takes place which result in genetic recombination. When gametes combine to make a zygote, more variations arise. This variation assures new combinations resulting in increase in adaptability of the organism.

**Errors in Meiosis**

Meiosis is a well regulated process but sometimes errors may arise which may lead to mostly serious disorders. The common cause of disorders in non-disjunction of the homologous pairs of chromosome abnormally. This may results into unequally distributed chromosomes in the gametes and when these fertilize, they give rise to individuals with disorders.

For example, in Down's syndrome the affected individual have 3 homologues in the 21$^{st}$ pair of chromosomes.

*Figure 22.1 Non-disjunction results into abnormal gametes*

Non-disjunction results into abnormal gametes, e.g., in above diagram N and N are

normal and N +1 or N -1 are abnormal gametes

**Twins**

There are two types of twins.

- **Fraternal twins**

  - These are produced by separate eggs (also called dizygotic twins)/. These are produced if two eggs are released and fertilized.
  - These are genetically different from each other and may be both males, females and both male and female.

- **Identical twins**

  - These are produced by division in the same egg (also called monozygotic twins). These are produced by division in the zygote.
  - These are genetically same / identical and have same characteristics. Both of these are either males or females.

**Environment and twins**

Environment may affect even the identical twins. Identical twins may also have different characteristics if they are brought up in different environments. We know that the genes interact with environment to produce various characteristics. This characteristic also affects the twins.

**Topic No 23**

**Molecular Biology** is the study of biological molecules related to genes, gene products and heredity. In the present age, world is in the midst of two scientific revolutions. One is information technology and the other is Molecular Biology. Both deal with the handling of large amounts of information. Molecular Biology has revolutionized the biological sciences as well especially in the fields of Health Sciences and Agricultural Sciences.

**Contribution of Molecular Biology**

- The almost complete sequence of the DNA molecules comprising the human genome was revealed in the year 2003. So, in theory, science has made available all of the genetic information needed to make a human being. However, the function of most of a human's approximately 35,000 genes remains a mystery.
- The other main arena where molecular biology has a massive impact is agriculture. New varieties of genetically engineered plants and animals have already been made and some are in agricultural use.
- So you can well imagine that how much important is this subject for you and for the economy of Pakistan.

**Topic No 24**

**Heredity Information Flow**

Reproducing itself is a property of life. Transferring characteristics to next generation is a property of living organisms. Heredity information flow in living organisms is carried out by the genes. The "Chromosome theory of heredity" states that the genes are present on chromosomes and are responsible for the transfer of characteristics from generation to generation. Genes are present in the form of DNA molecules, organized in a structure called chromosome (chromatin material).

**Chromatin - the Genetic Material**

Genetic material is present in the nucleus of the cell in eukaryotes and in the nucleoid region in prokaryotes. These are called chromatin material. Chromatin material is not visible during interphase (non-dividing state) of the cell. These become visible during cell division due to condensation of chromosomes.

**Functions of Genetic Material**

There are some important properties of genetic material, which are following:

- It replicates itself.
- It regulates the growth and development of the organism.
- It allows the organism to adapt to the environmental changes.

**Chromosomes - DNA – Genes**

Chromosomes consist of DNA molecule associated with proteins. In chromosomes, DNA is wrapped around proteins. Few of these proteins are called histones and few others. DNA is associated with histone and non-histone proteins in a chromosome.

**Introduction of DNA and Gene**

**DNA** is a macromolecule (large molecule) organized in structure chromosome. In prokaryotes DNA is a circular molecule. In eukaryotes it is a long linear molecule.

Mitochondria and chloroplast also have their own circular DNA molecules. these are plasmids. present in prokarotes

**Gene** is a length of DNA that codes for a peptide or protein. So that gene is a part or length of DNA.

**Condensation of Genetic Material**

Chromatin material condenses during prophase of mitosis in the form of chromosomes.

Chemical analysis shows that chromosome consist of DNA and proteins. DNA is a long molecule about 2nm thick running continuously within each chromosome. Chemical analysis shows that DNA is acidic in nature.

**The Structure of Chromosomes**

Chromosome consists of a DNA molecule wound around proteins. DNA in human cell

(All chromosomes) is about 6 feet long, packed in a microscopic nucleus of a cell. In one human chromosome, it is 1.7-8.5 cm long. How is this possible? The answer is “coiling” and “super-coiling”. The chromosome consists of highly condensed structure. If we can open this like a thread, then the long thread will appear like a flower like structure called solenoid which consists of many smaller units. These small units are called “nucleosomes”. A nucleosome is a length of DNA coiled around a set of proteins. The

DNA coils around histones twice, which is up to 200 base pairs long. Two nucleosomes are connected to each other by a length of DNA, which is called “linker DNA” (up to 80 base pairs long).

*Figure 24.1 Chromosome coiling and nucleosome*

**Figure 24.2 Two nucleosomes**

**Topic No 25**

**Chemical Composition of DNA**

DNA is a complex macromolecule (large molecule). DNA stands for Deoxyribose

Nucleic Acid. The smallest unit of DNA is called a “nucleotide”; nucleotides join to make polynucleotide. We can say that DNA consists of nucleotides joined together.

**Nucleotides**

Each nucleotide consists of:

I. Deoxyribose sugar
II. Phosphate group
III. Nitrogenous base

*Figure 25.1 Structure of Nucleotide*

There are four nucleotides based upon four different nitrogenous bases attached to them.

Nitrogenous bases are of two types: purines and pyrimidine. Purines include two bases Adenine and Guanine which have a double ringed structure. Pyrimidine bases include the other ones called Thymine and Cytosine that have single ringed structure.

**Topic No 26**

**Mechanism of Gene Action**

Genes express themselves by making proteins. Making the proteins by DNA occur by two processes called transcription and translation. **Transcription** is formation of a form of RNA from DNA called messenger RNA (mRNA). mRNA is formed inside the nucleus in eukaryotes and in nucleoid region in prokaryotes. The next process is **translation**, which is formation of a protein or peptide by mRNA with the help of another organelle called ribosome.

**Replication** is another function of DNA. It is doubling of DNA molecule to make two copies of itself. Replication occurs before cell division to make copies of DNA for the daughter cells.

Genetic code is a term used for the parts of DNA that code for proteins. A codon is a 3 nucleotides code for an amino acid, i.e., codon is a 3 nucleotide set of DNA molecule that codes for a protein.

**Transcription and Translation**

- Transcription = DNA mRNA
- Translation = mRNA Protein

The following scheme is called the central dogma of molecular biology / genetics

DNA □ RNA □ Protein

**Transcription**

DNA □ mRNA

The process of transcription involves an enzyme called RNA polymerase. One strand of

DNA act as the template strand which is actually coded into the mRNA.

**Steps of transcription**

- RNA polymerase identifies and attaches to a region called promoter on the DNA upstream the gene.

RNA polymerase open the double helix chain which results in the formation of transcription bubble.

**Topic No 27**

**Transcription Process**

- RNA polymerase moves on the gene, the helix unwinds and make a complementary strand of RNA. This strand of mRNA protrudes out of transcription bubble.
- At end of the gene there is a stop sequence. Usually it is a series of GC base pairs followed by a series of AT base pairs.
- These sequences make a hair pin loop like structure which stops RNA polymerase from transcribing.
- Thymine is coded as uracil in mRNA.

**Transcription in Prokaryotes and Eukaryotes**

In prokaryotes, the mRNA directly moves into cytoplasm and its translation starts because there is no nuclear membrane, nucleoid region is continuous with cytoplasm. In eukaryotes, mRNA formed moves out of nucleus through nuclear pores and then it is translated in the cytoplasm with the help of ribosomes.

**Modification of mRNA in eukaryotes**

mRNA in eukaryotes has to travel from nucleus to cytoplasm, to protect it from the action of nucleases (the DNA cutting enzymes) and proteases (protein cutting enzymes), it is modified. On its 5' end a cap of 7 methyl GTP is added; while on the 3' end a poly A tail is added. Introns are also removed. Introns are DNA sequences in the eukaryotes which are non-coding and should be removed from the mRNA. The coding regions are called exons.

**Figure 27.1 mRNA in eukaryotes have regions – exons and introns**

**Topic No 28**

**Translation**

mRNA  Protein

The process of translation consists of three major steps: initiation, elongation, and termination.

**Steps of Transcription**

- In prokaryotes, translation starts while transcription is going on because there is no barrier between nucleoid and cytoplasm.
- In eukaryote, first introns are removed.

**Process of Translation**

**Initiation:**

- The mRNA binds to the small unit of ribosome.
- The large ribosomal subunit has 3 binding sites called E (Exit), P (Peptidyl), and

  A (Aminocyl).

- When the first codon (triplet code) is aligned at the P site then the large ribosomal subunit attaches to the small subunit.

- A tRNA carrying the amino acid methionine attaches to the start codon (AUG) on the messenger RNA.

**Elongation:**

- A tRNA with its amino acid attaches to the A binding site.
- Peptide bond formation occurs between the methionine and the amino acid carried at the A binding site.
- Ribosome moves in the 3' direction down the messenger RNA by three bases, shifting the tRNA and polypeptide chain to the P Binding site.
- The A binding site is open and a vacant tRNA (without amino acid) is in the E binding site.
- Now, the next tRNA brings another amino acid and bind to A site.
- A peptide bond is formed between the second and this new (thirs) amino acid.
- Ribosome moves in 3’ direction and the vacant tRNA is released from the E site.
- This process continues until a stop codon arrives on mRNA.

A Releasing factor comes and binds to the A site in place of stop codon. The polypeptide chain separates from tRNA and ribosome. Then ribosomal unit disassemble again. mRNA molecule also released which has been coded.

**Topic No 29**

**Nucleotide**

Nucleic acids are important group of biomolecules which are responsible for storage & transmission of hereditary information. Like proteins and polysaccharides, nucleic acids are also polymeric compounds.

The repeating units in the nucleic acids are Nucleotides. There are two main types of nucleic acids, Deoxyribonucleic acids (DNA) and ribonucleic acids (RNA)

**Figure 29.1 DNA Vs. RNA**

**Topic No 30**

**The chemical structure of DNA**

DNA is a polymer of Deoxyribonucleotides. It is composed of three components: Deoxyribose, Nitrogenous Base, Phosphoric acid.

*Figure 4 Detailed about Chemical Structure of the DNA*

**Topic No 31**

**Chemical Composition of RNA**

RNA (Ribonucleic acid) is a polymer of ribonucleotides. Each ribonucleotide is composed of three components:

I. A ribose sugar
II. A Nitrogenous Base
III. A Phosphoric acid

**Figure 31.1 Chemical Composition of DNA and RNA**

**Topic No 32**

**Chemical Composition of Protein (Summary)**

- Proteins are polymers of amino acids
- They range in size from small to very large
- All the proteins are made up of twenty different types of amino acids. So these amino acids are called standard amino acids
- In a protein molecule, each amino acid residue is joined to its neighbor by a specific type of covalent bond which is called Peptide Bond
- Amino acids can successively join to form dipeptides, tripeptides, tetrapeptides, oligo peptides and polypeptides

**Figure 32.1 Amino Acid Structure**

**Topic No 33**

**Carbohydrates: Sugar Polymers**

They act as source of energy that can be transported and have structural role as Monosaccharides, Disaccharides, Oligosaccharides and Polysaccharides.

**Monosaccharides**

Monosaccharides, also called simple sugars, are the simplest forms of sugar and the most basic units from which all carbohydrates are built. They are usually colorless, water-soluble and crystalline solids. Monosaccharides are produced by plants, all living cell have glucose.

**Figure 33.1 Structure of Glucose, Fructose and Galactose**

**Topic No 34**

**Carbohydrates: Glycosidic Linkages**

h
stereoisomers found in nature. Stereoisomerism in sugars is biologically significant because the enzymes that act on sugars are strictly stereospecific. It is as difficult to fit the wrong sugar stereoisomer into an enzyme's binding site as it is to put your left glove on your right hand.

Monosaccharaides are colorless, crystalline solids that are freely soluble in water but insoluble in nonpolar solvents. Most have a sweet taste. In the open-chain form, one of the carbon atoms is double-bonded to an oxygen atom to form a carbonyl group; each of the other carbon atoms has a hydroxyl group. If the carbonyl group is at an end of the carbon chain (that is, in an aldehyde group) the monosaccharide is an aldose; if the carbonyl group is at any other position (in a ketone group) the monosaccharide is a ketose. The simplest monosaccharaides are the two three-carbon trioses: glyceraldehyde, an aldotriose, and dihydroxyacetone, a ketotriose. The aldopentoses D-ribose and 2-deoxy-D-ribose are components of nucleotides and nucleic acids.

All the monosaccharides except dihydroxyacetone contain one or more asymmetric (chiral) carbon atoms and thus occur in optically active isomeric forms. The simplest aldose, glyceraldehyde, contains one chiral center (the middle carbon atom) and therefore has two different optical isomers, or enantiomers.

One of the two enantiomers of glyceraldehyde is, by convention, one of these two forms is designated the D isomer, the other the L isomer. As for other biomolecules with chiral centers, the absolute configurations of sugars are known from x-ray crystallography. To represent three-dimensional sugar structures on paper, we often use Fischer projection formulas. In Fischer projection formulas, horizontal bonds project out of the plane of the paper, toward the reader; vertical bonds project behind the plane of the paper, away from the reader.

The carbons of a sugar are numbered beginning at the end of the chain nearest the carbonyl group. Two sugars that differ only in the configuration around one carbon atom are called epimers; D-glucose and D-mannose, which differ only in the stereochemistry at C-2, are epimers, as are D-glucose and D-galactose (which differ at C-4).

Some sugars occur naturally in their L form; examples are L-arabinose.

In aqueous solution, aldotetroses and all monosaccharides with five or more carbon atoms in the backbone occur predominantly as cyclic (ring) structures in which the carbonyl group has formed a covalent bond with the oxygen of a hydroxyl group along the chain. The formation of these ring structures is the result of a general reaction between alcohols and aldehydes or ketones to form derivatives called hemiacetals or hemiketals. Two molecules of an alcohol can add to a carbonyl carbon; the product of the first addition is a hemiacetal (for addition to an aldose) or a hemiketal (for addition to a ketose).

Addition of the second molecule of alcohol produces the full acetal or ketal, and the bond formed is a glycosidic linkage.

The reaction can produce either of two stereoisomeric configurations, denoted $\alpha$ and $\beta$. Isomeric forms of monosaccharides that differ only in their configuration about the hemiacetal or hemiketal carbon atom are called anomers, and the carbonyl carbon atom is called the anomeric carbon.

**Topic No 35**

**Lipids**

Lipids are organic compounds that contain hydrogen, carbon, and oxygen atoms, which form the framework for the structure and function of living cells."

**Properties of Lipids**

- Lipids are oily or greasy nonpolar molecules, stored in the adipose tissue of the body.
- Lipids are a heterogeneous group of compounds, mainly composed of hydrocarbon chains.
- Lipids are energy-rich organic molecules, which provide energy for different life processes.
- Lipids are a class of compounds characterized by their solubility in nonpolar solvents and insolubility in water.
- Lipids are significant in biological systems as they form a mechanical barrier dividing a cell from the external environment known as the cell membrane.

Lipids are also the building blocks of many hormones and are an important constituent of the plasma membrane. Lipids include fats, oils, waxes, phospholipids, and steroids.

**Examples of Lipids**

**Waxes**

Waxes are another biologically important category of lipids. Wax covers the feathers of some aquatic birds and the leaf surfaces of some plants, where its hydrophobic (water-repelling) properties prevent water from sticking to, or soaking into, the surface. This is why water beads up on the leaves of many plants, and why birds don't get soaked through when it rains.

**Steroids**

Steroids are another class of lipid molecules, identifiable by their structure of four fused rings. Although they do not resemble the other lipids structurally, steroids are included in lipid category because they are also hydrophobic and insoluble in water. All steroids have four linked carbon rings and several of them, like cholesterol, also have a short tail. Many steroids also have an –OH functional group attached at a particular site, as shown for cholesterol below; such steroids are also classified as alcohols, and are thus called sterols.

**Cholesterol**

The most common steroid, is mainly synthesized in the liver and is the precursor to many steroid hormones. Cholesterol also serves as the starting material for other important molecules in the body, including vitamin D and bile acids, which aid in the digestion and absorption of fats from dietary sources. It's also a key component of cell membranes, altering their fluidity and dynamics.

Of course, cholesterol is also found in the bloodstream, and blood levels of cholesterol are what we often hear about at the doctor's office or in news reports. Cholesterol in the blood can have both protective effects and negative effects on cardiovascular health.

**Topic No 36**

Fatty acids may be **saturated or unsaturated.**

In a fatty acid chain, if there are only single bonds between neighboring carbons in the hydrocarbon chain, the fatty acid is saturated. **Saturated fatty acids** are saturated with hydrogen; in other words, the number of hydrogen atoms attached to the carbon skeleton is maximized.

When the hydrocarbon chain contains a double bond, the fatty acid is an **unsaturated fatty acid**. Most unsaturated fats are liquid at room temperature and are called oils. If there is one double bond in the molecule, then it is known as a monounsaturated fat (e.g., olive oil), and if there is more than one double bond, then it is known as a polyunsaturated fat (e.g., canola oil). Saturated fats tend to get packed tightly and are solid at room temperature. Animal fats with stearic acid and palmitic acid contained in meat, and the fat with butyric acid contained in butter, .

| Saturated Fats | Unsaturated Fats |
|---|---|

| | |
|---|---|
| Contains a single bond. | Contains at least one double bond. |
| Not to be consumed more than 10 percent of total calories per day. | Not to be consumed more than 30 percent of total calories per day. |
| Excessive consumption leads to heart diseases. | Good for consumption, but excessive may increase cholesterol. |
| Increases low- density lipoproteins (LDL), which is called as bad cholesterol. | Increases High-density lipoprotein (HDL), which is commonly known as good cholesterol and also reduce low-density lipoproteins (LDL). |
| Would not spoil quickly. | Spoil quickly. |
| Foods sources of saturated fats are whole milk, butter, cheese, margarine, coconut oil, vegetable oil, meat, peanut, fried foods, etc. | Foods sources of unsaturated fats are walnuts, flax, avocado, sunflower oil, soybean oil, fish oil, canola oil, red meat, etc. |
| High melting point. | Low melting point. |
| Solid state in room temperature. | Liquid state in room temperature. |

**Topic No 37**

**Study of Omics**

- Genomics: all the genes
- Pharmacogenomics choice of personalized medicine
- Nutri-genomics choice of best diet
- Toxicogenomics prediction of toxicity
- Epigenomics: all epigenetic changes in genome
- Transcriptomics: all the mRNAs
- Proteomics : all the proteins
- Interactomics : all interactions between all proteins
- Metabolomics (or metabonomics): all metabolites

**Genome and Genomics**

The complete set of DNA found in each cell is known as the genome and study is called as genomics.

**Proteome and Proteomics**

The complete set of proteins found in each cell is known as the proteome.

Proteins concentration (and activity) may be different than gene expression due to post-translational modification

**Metabolomics**

The complete set of metabolites found in each cell is known as the metabolome.

Use of high-throughput mass spectrometry to analyze the metabolic components of cell.

**Metabolomics**

Useful for determining the effects of the environment or gene transformation on the metabolism of the plants/animals.

***Figure 37.1 Important branches of Omics***

**Conclusion**

Genomics, proteomics and metabolomics will give an integrated, wholistic view of the cell.

**Topic No 38**

**Genomics, Proteomics and Metabolomics**

**Genome and Genomics**

The complete set of DNA found in each cell is known as the genome and study is called as genomics.

**Proteome and Proteomics**

The complete set of proteins found in each cell is known as the proteome.

Proteins concentration (and activity) may be different than gene expression due to post-translational modification

**Metabolomics**

The complete set of metabolites found in each cell is known as the metabolome.

Use of high-throughput mass spectrometry to analyze the metabolic components of cell.

**Metabolomics**

Useful for determining the effects of the environment or gene transformation on the metabolism of the plants/animals.

**Conclusion**

Genomics, proteomics and metabolomics will give an integrated, wholistic view of the cell.

Can be used to monitor or modify organisms in a comprehensive way. Bioinformatics - the key to understand the plethora of information and modeling the cell.

**Topic No 39**

**Genome Informatics**

**Introduction**

Genome sequencing provides the sequences of all the genes of an organisms. A major application of bioinformatics is analysis of full genomes that have been sequenced.

**Genomics:** It is the study of all of a person's genes (the Genome), including interactions of those genes with each other and with the person's environment.

**Genome Informatics:** Genome Informatics is the field in which computational and statistical techniques are applied to derive biological information from genome sequences.

Genome Informatics includes method to analyze DNA sequence information and to predict protein sequence and structure.

**Genome Analysis:**

- Sequencing
- Assembly

- Repeat identification and masking out
- Gene prediction
- Looking for EST and cDNA sequences
- Genome annotation
- Expression analysis
- Metabolic pathways and regulation studies
- Functional genomics
- Gene location/gene map identification
- Comparative genomics
- Identify clusters of functionally related genes
- Evolutionary modeling
- Self-comparison of proteome

**Conclusion**

Sequencing and analysis of full genomes paves the way for future discoveries. Different model organisms can help explore our Genome and what matters most for us.

**Topic No 40**

**Prokaryotic Genome**

Prokaryotes are the organism whose genetic materials (DNA) is not enclosed in nuclear membrane (No membrane bound organelles).

***Figure 40.1 Prokaryotic Genome***

First prokaryotic genome sequence was that of *Hemophilus influenza*e, which paved the way for sequencing of many other organisms.

***Figure 40.2 Features of representative prokaryotic genomes***

**Conclusion**

Prokaryotes are simple genomes, easy models to study biochemistry and molecular biology of life processes. Sequencing is done on economically important organisms.

**Topic No 41**

**Eukaryotic Genome**

Eukaryotes have larger genome as compared to prokaryotes. Eukaryotes have tandem repeats, introns in their protein-coding genes, heterochromatin and euchromatin region.

***Figure 41.1 Eukaryotic Genome (An animal cell)***

**Conclusion**

Eukaryotes are distinguished by the presence of prominent nuclei, have larger genome, tandem repeats and introns in their protein-coding genes.

**Topic No 42**

**Viral Genomes**

Genomes of Viruses

- 
- Viral genomes can be
- ssRNA
- dsRNA
- ssDNA
- dsDNA
- Linear
- Ciruclar

**Viruses Genomes**

- A viral genome is the genetic material of the virus.
- Also termed the viral chromosome.
- Viral genomes vary in size -few thousand to more than a hundred thousand nucleotides.

**Viruses with RNA Genomes**

- Almost all plants viruses and some bacterial and animal viruses
- Genomes are rather small (a few thousands nucleotides)

**Viruses with DNA Genomes**

- Often a circular genome
- lambda = 48,502 bp

**Replicative form of Viral Genomes**

- All ssRNA viruses produce dsRNA molecules
- Many linear DNA molecules become circular

**Viruses and Kingdoms**

- Many plants viruses contain ssRNA genomes.
- Many fungal viruses contain dsRNA genomes.
- Many bacterial viruses contain dsDNA genomes.

***Figure 42.1 DNA, RNA & RNA-DNA Viruses***

**Genomes in Virions:** The genomes of viruses can be composed of either DNA or RNA, and some use both as their genomic material at different stages in their life cycle. However, only one type of nucleic acid is found in the virion of any particular type of virus.

***Figure 42.2 Viruses with number of genes***

**Genome of Pox Virus**

- Linear dsDNA 130-375 kbp; covalently closed termini.
- Large hairpin structure at eah terminus - up to 10 kb total at each end is repeat sequence.
- Encode 150-300 proteins.
- Coding regions are closely spaced, no introns.

Coding regions are on both strands of genome, and are not tightly clustered with respect to time of expression or function.

**Topic No 43**

**Bacterial Genomes**

**Genomes of Bacteria**

- Small organisms carry high coding density (85-90%)
- 1 gene per 1000 bases in prokaryotes
- Large variation in genome size between bacteria

**Genomes of Bacteria – Large Variation**

- Tremblaya princeps 140kb, 121 coding sequences
- Sorangium cellulosum
- 14000kb
- 11599 coding sequences

***Figure 43.1 Comparison of regulatory genes in bacterial genomes***

***Figure 43.2 Distribution of genes among selected bacterial genomes and their sizes***

**Conclusion**

- Small organisms carry high coding density.
- Large variation in genome size between bacteria

**Topic No 44**

**Polymerase chain Reaction**

Molecular technology has become a crucial tool for identifying new genes with importance in medicine, agriculture, animal production and health, environment and the industry related to these areas. Among the applications of molecular techniques is important to highlight the use of the Polymerase Chain Reaction (PCR) in the identification and characterization of viral, bacterial, parasitic and fungal agents. This technique was developed by Kary Mullis in the mid 80's and since then it has been considered as an essential tool in molecular biology which allows amplification of nucleic acid sequences (DNA and RNA) through repetitive cycles in vitro. The mechanisms involved in this methodology are similar to those occurring in vivo during DNA replication. The amplification of specific nucleic acid sequences, even in the presence of millions of other DNA molecules, is achieved by thermo-stable DNA polymerase enzyme (as the name of this technique suggests: “polymerase chain reaction”) and specific primers. Primers are short sequences of DNA or RNA (oligonucleotides) that initiate DNA synthesis.

**Reference:** Polymerase Chain Reaction Edited by Dr. Patricia Hernandez-Rodriguez, Chapter # 8: Polymerase Chain Reaction: Types, Utilities and Limitations, Page 157-158.

**Topic No 45**

**Steps of PCR**

PCR is a biochemical process capable of amplifying a single DNA molecule into millions of copies in a short time.

A typical PCR consists of:

**Initial Denaturation:** The reaction temperature is increased to 95 °C and the reaction is incubated for 2–5 min (up to 10 min depending on enzyme characteristics and template complexity) to ensure that all complex, double-stranded DNA (dsDNA) molecules are separated into single strands for amplification.

**Cycling:**

1.
    1. **Denaturation:** The reaction temperature is increased to 95 °C, which melts (disrupts the hydrogen bonds between complementary bases) all dsDNA into single-stranded DNA (ssDNA).
    2. **Annealing:** In annealing short DNA molecules called primers bind to flanking regions of the target DNA. The temperature is lowered to approximately 5 °C below the melting temperature ($T_m$) of the primers (often 45–60 °C) to promote primer binding to the template.
    3. **Extension:** The temperature is increased to 72 °C, which is optimum for DNA polymerase activity to allow the hybridized primers to be extended.DNA polymerase extends the 3′ end of each primer along the template strands.

**Repeat:** Steps 1–3 repeated ("cycled") 25–35 times to exponentially produce exact copies of the target DNA.

**Final elongation:** Single step is performed at a temperature of 70-74°C for 5-15 minutes after the last PCR cycle to ensure that any remaining single-stranded DNA is fully extended.

**Final hold temperature:** This step at 4-15°C for an indefinite time may be employed for short-term storage of the reaction.

**Figure 44.1 Three steps of PCR—denaturation, annealing, and extension—as shown in the first cycle, and the exponential amplification of target DNA with repeated cycling.**

**Topic No 46**

**Types of PCR**

**Multiplex PCR**

Multiplex PCR is an adaptation of PCR which allows simultaneous amplification of many sequences. This technique is used for diagnosis of different diseases in the same sample. Multiplex PCR can detect different pathogens in a single sample. Also it can be used to identify exonic and intronic sequences in specific genes and determination of gene dosage.

**Nested PCR**

This PCR increases the sensitivity due to small amounts of the target are detected by using two sets of primers, involving a double process of amplification. The first set of primers allows a first amplification. The product of this PCR is subjected to a second PCR using the second set of primers. These primers used in the second PCR are specific to an internal amplified sequence in the first PCR. Therefore, specificity of the first PCR product is verified with the second one. The disadvantage of this technique is the probability of contamination during transfer from the first amplified product into the tube in which the second amplification will be performed.

**Reverse Transcriptase PCR (RT-PCR)**

This PCR was designed to amplify RNA sequences (especially mRNA) through synthesis of cDNA by reverse transcriptase (RT). Subsequently, this cDNA is amplified using PCR. This type of PCR has been useful for diagnosis of RNA viruses, as

well as for evaluation of antimicrobial therapy. It has also been used to study gene expression in vitro, due to the obtained cDNA retains the original RNA sequence. The main challenge of using this technique is the sample of mRNA, because this is considered difficult to handle by low level and concentration of mRNA of interest and low stability at room temperature together with sensitivity to action of ribonucleases and pH change.

**Semi-quantitative PCR**

This technique allows an approximation to the relative amount of nucleic acids present in a sample, as mentioned above. c . Then, internal controls (that are used as markers) are amplified. The Amplification product is separated by electrophoresis. Agarose gel is photographed after ethidium bromide staining, and optical density is calculated by a densitometer. The disadvantage of the technique is possibility of nonspecific hybridizations, generating unsatisfactory results. Control of specificity is performed using highly specific probes for hybridization.

**Topic No 47**

**Applications of PCR**

During the past 30 years molecular techniques have been under development, however these have had a rapid and tremendous progress in recent year. Among molecular techniques, PCR and its different variations are highlighted as the most commonly used in laboratories and research institutes. Thus, these have contributed to identification and characterization of several organisms and understanding of physiopathology of diverse diseases in human, animal and plant. Also these have provided clues for future research directions in specific topics with impact in public health such as genetics and biochemistry of antimicrobial resistance. The following describes some applications of PCR and its variants in studies in human medicine, forensic sciences, and agricultural science and environment.

**Medicine**

Molecular biology techniques, particularly PCR, have had a major impact on medicine. The versatility of molecular techniques has allowed advances and changes in all fields of medicine. The following is an overview of the main impacts generated for molecular biology in medical sciences. Clinical microbiology has been transformed with the use of molecular technology because it has generated a benefit to the patient affected by infectious diseases. Molecular biology has allowed the development of clinical microbiology because it has been possible to identify microorganisms that are difficult to culture, that have many requirements of laboratory or dangerous for laboratory personnel. These problems have been reduced with the implementation of molecular diagnosis that provides high sensitivity, specificity, precision and speed with one small sample. These applications are transforming and complementing the work of biochemists, immunologists, microbiologists and other health professionals who see in the molecular tools new alternatives for a rapid diagnosis of microorganisms as well as for the determination of multiple factors associated with antibiotic resistance thus expanding the knowledge of microbial epidemiology and surveillance at the genetic level. The usefulness of PCR in identification of microorganisms has led to the selection and quality assurance of blood that blood banks are using for patients with different pathologies. The incorporation of molecular techniques has been of great importance in the identification and characterization of many viruses, including influenza, which through a rapid, sensitive, and effective molecular diagnosis has allowed inclusion of early treatment to benefit patients and control of a high impact infection.

**Forensic science**

In forensic pathology, classic morphology remains as a basic procedure to investigate deaths, but recent advances in molecular biology have provided a very useful tool to www.intechopen.com Polymerase Chain Reaction: Types, Utilities and Limitations 165 research systemic changes involved in the pathophysiological process of death that cannot be detected by morphology. In addition, genetic basis of diseases with sudden death can also be investigated with molecular methods. Practical application of RNA analysis has not been accepted for post-mortem research, due to rapid decomposition after death. However, recent studies using variants of conventional PCR (qPCR and RT-PCR) have suggested that relative quantification of RNA transcripts can be applied in molecular pathology to research deaths ("molecular autopsy"). In a broad sense, forensic molecular pathology involves application of molecular biology in medical science to investigate the genetic basis of pathophysiology of diseases that lead to death. Therefore, molecular tools support and reinforce the morphological and physiological evidence in research of unexplained death.

**Topic No 48**

Blotting is used in molecular biology for the identification of proteins and nucleic acids and is widely used for diagnostic purposes. This technique immobilizes the molecule of interest on a support, which is a nitrocellulosic membrane or nylon. It uses hybridization techniques for the identification of the specific nucleic acids and genes. The blotting technique is a tool used in the identification of biomolecules such ad DNA, mRNA and protein during different stages of gene expression. Protein synthesis involves expression of a DNA segment which gets converted to mRNA to produce the respective protein. Molecules such as DNA, RNA and proteins are subjected to biochemistry analysis which are separated using blotting techniques. In the case of a cell, these molecules are present altogether and hence with the help of blotting scientists are able to recognize a specific molecule out of all others. Blotting is performed by allowing a mixture of molecules of interest pass through a block of gel which separates the molecules based on their molecular sizes  The hence processed molecules are required to be hard-pressed against a suitable membrane which will in turn transfer the molecules from the gel onto a suitable membrane (nylon, nitrocellulose or PVDF) via capillary action. After the molecules are transferred to the membrane their position does not change.

Southern blotting was introduced by Edwin Southern in 1975 as a method to detect specific sequences of DNA in DNA samples. The other blotting techniques emerged from this method have been termed as Northern (for RNA), Western (for proteins), Eastern (for post-translational protein modifications) and South-western (for DNA-protein interactions) blotting.

Subtypes of blotting such as northern, western & southern depend upon the target molecule that is being sought. When a DNA sequence is the foundation or code for a protein molecule, the particular DNA molecule of interest can be blotted using Southern Blotting technique. During gene expression, when the DNA is expressed as mRNA for a protein production, this process can be identified by Northern blotting. Finally, the coded mRNA produces the concerned protein, this protein identification can be done by Western Blotting.

Blotting approaches are viewed as an aide to the gel electrophoresis which is generally applied for separation of DNA/RNA/protein and yields reproducible results attributed to their excellent resolving power. Thereby, specific molecules can be detected amid the combination of [illegible]lecules that are subjected to the separation. Most of the methods have a general step wherein the molecules of interest are tra[illegible]red once separated are transferred from the gel to a solid membrane phase which is accomplished by drenching in a solution[illegible]s the gel and the membrane via penetrable paper. Many types of multifaceted apparatus are also provided from a lot of sup[illegible]blotting which is more specifically useful for transfer initiated from less porous polyacrylamide gels compared to commonly use porous agarose gels. In case of DNA and RNA the detection of specific sequences in the membrane are carried out via hybridization with nucleic acid labeled probes which in the case of proteins is replaced by the use of labeled antibody probes. The initially developed protocols applied radioactive probes labeled with, radioactive isotopes for detection purposes via implementation of autoradiography procedures. In this process using the pattern of decay emissions radiated from a radioactive material is applied to produce an image on an x-ray film which can also be made available as a digital image by application of scintillation based gas detectors or systems based on phosphorimaging. Keeping in mind the harmful effects of exposure to radioactivity other kinds of labeling systems have been developed which includes fluorescent and chemiluminescent reagents. Luminescence/fluorescence based methods are considered to be sensitive and background free thus generates more accurate results. Also, there are several other modifications that have been implemented in contrast to the original method like DNA probes are more commonly applied rather than RNA, the nylon membranes have replaced the traditional nitrocellulose, to avoid the renaturation of DNA sequences during transfer the transfer is carried out in alkaline which was supposed to be in neutral solution as per the original protocol.  The DNA is subjected to acid treatment for reducing its size inorder to increase the transfer rate of larger fragments. The gel strips and tube gels applied in the original protocol are no longer used, instead gel formats are applied. Though there have been many changes in the original protocol still the modern day protocols retain most of the fundamental features of the original protocol.

Southern blotting was applied in many important studies like the genetic mapping of the human genome which was based on blotting based detection of restriction fragment length polymorphisms. Also, the DNA fingerprinting was first developed via hybridization of the human DNA restriction digestion products with minisatellite probes. However, most of the primary applications of this method have been now substituted by DNA sequencing and polymerase chain reaction (PCR) as they provide more extensive facts or data and moreover are easy to implement. Nevertheless, blotting is still applied in a lot of arenas like in the measurement of copy number, analysis of long stretches of DNA that is difficult to be amplified or sequenced using PCR or DNA sequencing and in case of structural analysis of DNA wherein the physical forms of DNA are separated using two-dimensional gel electrophoresis and thereafter it is detected using blotting of specific components.

**Southern Blotting**

The first of these techniques developed was the Southern blot, named after Dr. Edwin Southern who developed it to identify specific DNA sequences. Southern blotting is a detection technique used to find the target DNA sequences in the DNA sample in the field of molecular biology. The process starts from electrophoresis of DNA molecules which are hybridized in a blotting membrane followed by a transfer step where DNA from gel is transferred onto the blotting membrane.

**Topic No 49**

**Principle**

Restriction endonucleases, which is an enzyme, is used to break the DNA into small fragments. These fragments are then separated using electrophoresis. The fragments achieved is then classified according to their size (kDa). Thus, DNA fragments are transferred to the blotting paper where it is incubated with probes. Probes used in the Southern blotting can be highly selective. They can selectively bind with a resolution of 1 in a million and the characteristics to bind to the intended target fragments.

**Materials Required**

**Reagents**

- The buffer used for electrophoresis is TAE or TBE.
- The agarose preferred should be of electrophoresis grade.
- For staining the DNA, ethidium bromide (0.5 µg ml–1, dissolved in H2O) is used. But there are other DNA staining dyes such as SYBR green which can replace ethidium bromide for safe handling.

Note: Ethidium bromide being a mutagen requires careful handling. It is advised to wear gloves during its usage and follow the pertinent regulations for disposal of pipette tips. Avoid touching any objects with ethidium bromide contaminated gloves.

- 2Xand 20X SSC (the composition of 20X SSC includes 3.0 M NaCl and 0.3 M sodium citrate)
- 6X DNA loading buffer composed of 0.25% bromophenol blue, 0.25% xylene cyanol FF and 30% glycerol in water.
- Suitable DNA markers of varying molecular weight also referred to as DNA ladders are used as standards for reference.
- The prehybridization and hybridization mixture consists of 0.5% SDS, 6X SSC, 5X Denhardt's solution and 100 mg ml–1 sheared, denatured salmon sperm DNA or yeast tRNA.
- Denhardt's solution widely used for hybridization is made up of 0.02% Ficoll, 0.02% polyvinylpyrrolidone and 0.02% bovine serum albumin (BSA)
- Paraffin oil
- Cellulose nitrate or nitrocellulose membrane filter with uniform porosity. (e.g., Millipore 25 HAWP; nylon membranes used for blotting protocols are available under various trade names from commercial suppliers)
- RNase A (20 pg ml–1 in 2X SSC) for the specific cleavage is needed.
- Restriction enzyme and an appropriate buffer is used.
- Radioactively labelled RNA as a probe for specific detection where autoradiography is done.

Note: Handling of radioactive probes must be careful which necessitates reliable safety measures and legal regulations.

- For detection of RNA labelled with tags such as 3H, 35S, 125I or 14C, there is a requirement of 2,5-Diphenyloxazole (PPO) in toluene at a concentration of 20 % wt/vol.

**Equipment**

- The transfer from narrow strips of gel can be achieved by using three pieces of glass or Plexiglas having a size of 5 cm × 20 cm with the thickness similar to the thickness of the gel.
- There is a requirement of thick, dry filter paper (four to five in numbers) or paper towels of 10 cm × 18 cm in size.
- Hybridization vessel possess larger dimension (0.8 mm deep × 2 cm high × ~1 cm longer) than that of membrane used for hybridization and the material used for developing hybridization vessel is Perspex (note: Several alternative procedures are followed for hybridization)
- Four narrow pieces of Perspex possessing thickness similar to that of gel. The length of Perspex is sufficient to surround the gel at a spacing of ~3 mm
- A tray having 20–50 mm (approx.) depth and 20 mm (approx.) length and width larger than the gel

- A glass sheet with length sufficient to be placed on the tray and narrower to have a gap of 10 mm on each side
- Several thick pieces of filter paper having a large area as compared to gel. The length of filter paper is adequate to cover the glass sheet and can be dipped within the tray.
- A moistened piece of nitrocellulose membrane, having a wider area which can cover the entire gel. The nitrocellulose membrane is placed on the top of four strips made of Perspex. The moistening of nitrocellulose membrane is done using 2× SSC.
- Paper towels which are stacked one on top of other.
- Apparatus for casting gel.
- Gel tank for carrying out electrophoresis is necessary.
- Power supply for the entire device set up is required.

**Reagent Setup**

- DNA: The entire procedure is initiated by employing enzyme digested DNA of varying concentrations which will quantify the optimum DNA concentration and specify restriction enzyme to be used. Generally, an amount of 1 µg of DNA derived from clones (e.g. from plasmid or bacteriophage clones) is adequate enough for plasmids having low-copy-number. We require larger amounts for carrying out the separation of complex DNA (e.g. genomic DNA). The advisable range to be considered would be 5–10 µg.
- The electrophoresis buffer used is TAE which has a composition of 40 mM Tris, 20 mM acetic acid, 1 mM EDTA with pH range of 7.4–8.2 which is normally made as stock concentration of 20X or TBE made up of 89 mM Tris, 2.5 mM EDTA, 89 mM borate, normally made as a 10 × stock).
- TAE is recommended to be best when we run gels for a shorter interval of time and when the recovery of DNA fragments from gel is to be carried out.
- TBE is considered to be a better buffer especially when we have to run the gels for a time period exceeding 2 h.

**Procedure**

### Step 1: DNA purification

To extract the DNA present inside the nucleus of a cell, we must first lyse the entire cell to enable the expulsion of the DNA. Incubating the cell culture with detergent lyses the entire cell. Now the lysed sample contains DNA, protein, and debris. Protein is lysed by adding the proteinase enzyme and incubated. DNA is purified and separated by alcohol precipitation and fibers are removed by using a buffer.

Apart from standard manual isolation procedures, there are commercially available kits like GenElute™ for the isolation of DNA from a variety of sources such as mammalian cells, plants, bacteria, fungi. High purity of the isolated DNA is ensured in case of use of commercial kits.

### Step 2: Fragmentation

The long nucleotide sequences should be broken into smaller fragments for the purification or identification process. This is done by the restriction endonuclease enzyme.

All the reagent necessary for the digestion process should be kept on ice before setting up a restriction digestion reaction with suitable enzyme and appropriate DNA concentration. The components are added to the multiwall plates or microcentrifuge tubes (PCR tubes) and mixed by aspirating the contents with the help of a pipette slowly to avoid formation of any bubbles. It is to be taken care that the enzyme has to be added at the last step and until then it should be stored at −20 °C. To ensure complete digestion of the DNA a surplus amount of enzyme is supposed to be added as the fragments produced due to partial digestion can cause ambiguity in the results leading to inaccurate analysis of the subsequent blot to be analyzed. Nevertheless, the concentration of the enzyme added in total should not surpass one-tenth of the total volume of the digest, because the glycerol content generally found in stock of the enzymes has the capacity to impede digestion process when present at high concentration. Also, in order to minimize the probable errors due to pipetting master mixes should be prepared accordingly when there is a requirement of analysis of a large number of samples.

The incubation of the digests is carried out at 37 °C either in an incubator or a water bath. A water bath is generally preferred. For DNA samples obtained from cloning 1–2 h deemed sufficient. In case of genomic DNA there is a requirement of overnight digestion wherein there is a chance that the enzyme is over in between. To ascertain that the overnight reaction is successful

half of the enzyme is to be added at the beginning of the digestion reaction and the second half can be added in the morning and the incubation can be continued for an hour.

Once the digestion is over it may be the case where a concentration step is required to ensure that suitable volume of DNA is present for loading into the gel which is generally fixed at 20 µl per well. This makes the total volume to be in 24 µl after the addition of 6X loading buffer. A standard method for concentrating the DNA samples involve precipitation in presence of ethanol after which the DNA sample is resuspended in ddH2O. The traces of ethanol should be removed completely which otherwise would lead to spilling of the samples out of wells once the samples are loaded.

### Step 3: Gel Electrophoresis

Nucleic acids are negatively charged molecules. So they move towards the anode in an electrophoresis chamber. The movement of the DNA fragments differentiates the rate of the transport thus enabling the separation by size.

The percentage of the gel that is to be used and size of the gel has to be determined. The percentage will decide on the size of the fragments that will be separated and the size of the gel on the other hand will give the range of fragments that are feasible to be resolved. Longer gels are generally needed in the case of separation of genomic DNA or multiple fragments that are having similar sizes in order guarantee appropriate separation. Generally, a 0.7–2% gel is considered to be adequate for most of the applications. However in case of some genomic DNA samples it may be required that a low percentage of gel is supposed to be run for the necessary separation of the fragments. When a less than 0.8% gel is required to be run a high base gel percentage is required (upto 2%) to provide support as low percentage gels are very fragile. It is necessary that the base gel is dispensed before the comb is positioned. As soon as the base gel is set the low percentage gel is poured on top and the comb is placed in a way that it does not touch the base gel at the bottom.

- A 1X electrophoresis buffer is made after dilution of the prepared stock solution in ddH2O and agarose is added to it in a conical flask which is kept under constant stirring to avoid formation of clumps. Thereafter, the agarose is melted in a microwave or on a stirrer with heating under constant stirring. In case a microwave is used the gel should be swirled after every 30s to ensure even consistency is maintained. In most of the standard microwave the gel completely melts within 3 min.

Note: the agarose should be checked regularly during heating as it is susceptible to boiling in a short time. To ensure there is no spillage it is recommended that a conical flask sufficiently larger in volume is to be used during agarose preparation.

- The flask containing the molten gel is required to be transferred to a magnetic stirrer and the gel is allowed to cool down slowly to around 60ºC under gentle but constant stirring. The ethidium bromide is supposed to be added to the gel at this stage, at a concentration of 5 µg ml–1.
- By the time the gel is cooled down the 1X electrophoresis buffer is prepared in an appropriate amount to fill the tank reservoir upto the level of few millimetres above the gel slab to ensure proper immersion,
- The gel casting tray is prepared with a comb of suitable teeth size depending on the sample volume to be loaded. The casting tray should be in a straight platform before pouring the gel onto it. The formation of bubbles is supposed to be avoided as far as possible as they render the DNA to run in an irregular manner. The bubbles in case formed, can be by burst by using a pipette tip and this is to be done as soon as the gel has been poured.
- As soon as the gel is set it can be understood by observing its opalescent appearance and the comb should be gently removed. The gel is then transferred to the tank and 1X running buffer is added to it inorder to immerse the gel. The buffer should be added in sufficient amount keeping in mind the maximum capacity of the tank because it increases the buffering effect as well as protects the gel from melting down due to generation of heat particularly if the gel is to be run for longer period of time. One has to be careful while removing the combs so as to not tear the wells. In case the gel apparatus in use provides for removing the comb after the gel is placed in the tank and immersed in the buffer the comb should be removed in the final step to avoid breakage of the wells. When a gel of lower percentage is used, before removal of comb a brief refrigeration could help in sustaining the wells in proper shape.
- The DNA samples are prepared by adding 1X loading buffer for each 5 µl of sample. The loading dye and the sample should be mixed well without formation of any bubbles which will eventually lead to spilling of the samples from the wells once they are loaded.
- The samples are then loaded carefully into the wells and one or preferably two of the wells are left blank for the molecular weight marker to be loaded which generally comes in a ready to load formulation. While loading the pipette tip is supposed to be placed at below the corner at the top of the well so that the pipette tip does not pressurise against the bottom of the well which may also lead to perforation of the well. The sample is to be loaded slowly to avoid spillage out of the wells and mixing of samples with the contents of the next well.

- The lid is then placed on top and connected to the power pack with the voltage set at an appropriate level. Inorder to ascertain if the setup is working fine small bubbles can be seen popping up as the power supply is switched on. In case of genomic DNA, the voltage should be set at low preferably not exceeding 20 V and for plasmid DNA it can be around 100V. The electrodes should be located in correct position for the DNA to run in the appropriate direction. The positive socket is generally red and the negatively charged one is black in color.

### Step 4: Denaturation

DNA thus attained are double stranded in nature. For our purpose of probe hybridization, we need a single stranded DNA. DNA is therefore denatured in an alkaline solution. This results in the formation of denatured DNA.

### Step 5: Blotting

Blotting is the transfer of the fragmented DNA sequence to the nitrocellulose membrane or nylon membrane. The process is done by either electroblotting or capillary blotting. The DNA molecule is saturated using a NaCl solution and permanently fixed using either UV radiation or drying.

**Choice of the membrane for blotting**

In the original protocol nitrocellulose membrane have been used for the blotting in case of Southern blot but in recent times nylon membranes have been implemented for the blotting process due to their ability to bind more amount of DNA efficiently which allows the Southern blot to be carried out with less amount of target DNA.

- The preparation time for transfer takes approximately 2–3 h
- In case the gel is run without ethidium bromide, it needs to be immersed electrophoresis buffer mixed ethidium bromide for 0.5–2 h.
- Thereafter, the gel is transferred carefully to a gel documentation system for capturing the photograph of the gel under ultraviolet light of 256 nm although when DNA is present in high amounts it can also be detected at 310 or 365 nm. If a scale is placed along with the gel it helps in co-relating the fluorescence photograph with the final autoradiograph that will be generated.
- If sufficient separation of the DNA has not occurred then the gel can be run for a longer time after returning to the tank. Distinct bands generally appear in case of clonal DNA and genomic
- DNA mostly yields a smear in which the repetitive elements of the DNA are represented by the appearance of brighter bands. Thereafter the portion of the gel to be subjected to blotting can be cut apart using a blade. In the experiments where large fragments needs to be transferred the gel should be subjected to a dilute acid bath for approximately 10 min inorder to bring in depurination.
- In the next step the gel is denatured completely by submerging it for 15–30 min in the presence of 1.5 M NaCl, 0.5 M NaOH in a tray that is placed in a rocking platform.
- The above mentioned solution is then substituted with 0.5 M Tris HCl (pH 7) and 3 M NaCl. It was further incubated for 15-30 min which leads to neutralization of the gel.

The time required for transfer is about 15 min.

- In case of the transfer of strips the constituents of gel transfer are prepared as such that there should be a portion of thick filter paper of appropriate dimension preferably 20 cm × 18 cm dipped in 20X SSC buffer along with a nitrocellulose filter which is equal to the length of the strip of the gel having a width of 1 cm dipped in 2X SSC. The filter paper is required to be immersed and the membrane should be transferred by initially floating them on the solution surface to avoid uneven transfer due to the trapped air.
- The large filter paper is laid on a glass or plastic platform after being soaked in 20× SSC in case of narrow strips. Utmost care should be taken so that no air is trapped below. In case of wider strips, the tray is first filled with 20× SSC before a glass plate is laid on to it. Thereafter, a compact arrangement of filter paper soaked in 20× SSC is placed on it before being dipped into the tray.
- A glass or a Perspex sheet is placed on uppermost and towards one of the sides of the wet paper.
- The gel is then removed from the above neutralizing solution and is laid parallel to the glass approximately 2–3 mm apart. Similarly, on the opposite side of the gel, the second glass is laid around 2–3 mm apart.
- The nitrocellulose membrane is then laid on the top portion of the gel with the ends balanced on the glass forming a between the air space. Air should not be trapped between the membrane and the gel hence the alignment has to be carried out with care. The membranes should not be shaken once it is placed.

- The absorbent paper is finally placed on the top. It is to be noted that placing a heavy weight on the top may disorient the gel in turn affecting the final results.
- The transfer process is then started for approximately 3 h which vary depending on the concentration of the gel as well as the size of the fragment in question. Inorder to avoid drying during the transfer 20× SSC must be replenished to avoid shrinkage of the gel. During this process of the addition of buffer, it must not fill the gap between the gel and the glass or sheets.
- Once the transfer is completed the nitrocellulose membrane is removed carefully in a way that the gel should remain attached.
- Then the nitrocellulose membrane is turned and the boundaries of the gel is traced using a pencil
- The gel is then removed from the nitrocellulose membrane and if some amount of DNA is left out during the transfer it can be visualised under UV light as explained before.
- The portion of the nitrocellulose membrane which was touching the gel should be removed using a blade.
- The cut portion is then kept immersed in 2X SSC for approximately 10–20 min before it is subjected to baking at 80 °C for 2 h in a vacuum oven. In another way the DNA can also be fixed via UV crosslinking mediated by exposure to short wavelength UV light in a commercial set up. If desired the dry membranes can also be stored at room temperature for future reference

Once the transfer process is completed the preparation for hybridisation reaction is to be carried out. For this a blocking step that usually takes around an hour is mandatory for eliminating non-specific reactions. This step is also known as the prehybridization step.

The membrane is incubated in standard Denhardt's solution for 1 h or for certain cases more time is required depending on the type of reaction. Salmon sperm DNA is also widely applied as a blocking agent. Commercially available prehybridization solution like PerfectHyb™ Plus buffer is used for preparing blocking solution containing Salmon sperm DNA. Briefly after incubating the prehybridization solution to 42°C the heat snap chilled salmon sperm DNA is added to it at a concentration of 50 µg/mL. The prehybridization solution containing the sperm DNA is allowed to interact with the blotted membrane inside a hybridization chamber for up to 5 h.

### Step 6: Hybridization

Labeled probe is added to the membrane buffer and incubated for as It takes several hours for the probe to find the exact target sequence.

The time required for hybridization usually 1–16 h depends on factors like complexity of the probe and concentration

The radioactive RNAs are present in limited amount as it is derived from cells making it an important factor for the original protocol. Due to this the volume of the hybridisation solution used is supposed to be less so that the desired RNA concentration can be achieved. There can be two approaches for hybridisation experiment where one uses smaller volume and the other utilizes a vessel destined to hold the membrane in low volume of liquid. The suitable set up possess a plastic bag that can be sealed using heat and it consists of optimum amount of hybridisation mix and can be dipped in a water bath or a hybridisation tube with capped ends. In case of the tube it must be ensured that the membrane is wet when the tube is subjected to rotation cycles in a rotisserie oven. The probes applied can be either RNA or DNA. The cells act as a source for the RNA or alternatively, they can also be obtained from clones or PCR products via in vitro transcription In case of DNA they can be either clones or PCR products. The procedure adopted for the labeling method relies on the on the source from which the probe is derived.

- In case of hybridization by immersion in paraffin oil a drop of solution containing the probe RNA of appropriate concentration depending on the membrane size is placed on a plastic sheet.
- One of the ends of the nitrocellulose membrane is allowed to soak the liquid from the drop by slowly moving the membrane over the drop surface. Once it is wet the same is repeated for the other side of the membrane.
- The membrane is then dipped in hybridization solution saturated with paraffin oil at a temperature of 40–65 °C which depends on the type of the solvent in use. Here it is to be taken into account that when the membrane is baked in presence of 2X SSC that leads to the introduction of salt which in turn will be the deciding factor for the solvent in which RNA will be dissolved. This process yields good results but suffers from disadvantages like uneven background which can be solved if the hybridization is processed at 40 °C in presence of 2X SSC and 40% formamide. This method also works well in case of hybridization involving large fragments.
- In case of hybridization using a vessel destined to place the membrane in presence of a small quantity of liquid.
- The vessel is initially filled with the solvent that is required for hybridization to take place.

- The membrane is then fed into it through a thin aperture at the top.
- The solvent is then drained off and the probe solution is added. The amount of the solution needed depends on the size of the membrane. For wider membranes this type of vessel is not suitable instead they can be hybridised inside a close-fitted cylindrical tube for transferring several gels are too wide to be hybridized in this type of vessel. In case the hybridization is carried out inside a water bath the top is sealed as the water bath contains a lid. Moreover another advantage is that in this case RNA can be reused.
- The membrane is then subjected to hybridisation for a suitable period of time depending on the concentration and purity of RNA as well as other conditions. It is appropriate to leave it overnight for most of the cases.
- Once the hybridisation period is over the membranes are removed and the blotted against filter papers
- Thereafter, it is washed for 20–30 min using large amount of solvent used in hybridisation and at the same temperature at which hybridisation is carried out.
- The excessive amount of the probe is washed using the sodium chloride and buffers comprising of detergents. In case the salt concentration is lowered the wash is considered to be more stringent and more amount of such nonspecific probe that is bound to the membrane is removed with a gradual increase of the number of stringent washes. Application of a high temperature while washing also adds up to the stringency of the wash. Once the wash is completed only the specific hybrids remain on the blot and the non-specific ones are removed.
- The membrane is checked for background using a radiation monitor. In case it is high the membrane is treated with RNase for 30 min at 20 °C and then rinsed using 2X SSC.
- Finally the membranes are air dried.

Detection time is approximately 1–48 h, which mostly depends on the nature of the probe

- In detection of radioactivity of RNA there are two options namely 32P-labeled RNA detection and 3H-, 35S-, 125I- or 14Clabeled RNA detection.
- For the detection of 32P-labeled RNA the membrane is wrapped in plastic with no air bubbles trapped in it and laid on X-ray film. With the application of slight pressure the membranes are flattened.
- For Detection of 3H-, 35S-, 125I- or 14C-labeled RNA fluorography is implemented. The membranes are soaked in a solution of PPO in toluene and then the membranes are air dried. Thereafter, they are laid on the X-ray film and maintained at –70 °C

The approximate timings for all the processes are crucial in obtaining good results.

- The restriction digestion usually takes 2–24 h which can comprise of overnight digestion.
- Similarly in case of electrophoresis the timing can vary between 1–16 h and can be implemented overnight
- The preparation for the transfer takes around 0.5–3 h
- For setting up the transfer apparatus approximately 15 min is required.
- Transfer time can be between 3–16 h including overnight transfer.
- Blocking to reduce non-specificity can be done for upto 1 h
- The hybridisation process takes 1–16 h and as per the probe nature and amount it can continue overnight.
- The detection takes around 1–48 h which is affected by the yield in hybridisation and probe specificity.

**Topic No 50**

**Northern Blotting**

Northern blot is a method used for detection of a specific RNA sequence in RNA samples.

**Procedure**

- Isolation of intact mRNA
- Separation of RNA according to size (through a denaturing agarose gel
- Transfer of the RNA to a solid support
- Fixation of the RNA
- Hybridization of the immobilized RNA to probes complementary to the sequences of interest.
- Removal of probe molecules that are nonspecifically bound to the solid matrix.
- Detection, capture and analysis of an image of the specifically bound probe molecules.

**Conclusion**

Northern blot is a method used for detection of a specific RNA sequence in RNA samples

**Topic No 51**

**Northern Blot – Applications**

**Applications**

- Study of gene expression in eukaryotic cells.
- To measure the amount & size of RNAs transcribed from eukaryotic genes.
- To estimate the abundance of RNAs.
- To equalize the amounts of RNA loaded into lanes of gels.
- Use of housekeeping gene (endogenous constitutively-expressed gene).
- Normalizing samples according to their content of mRNAs of the housekeeping gene.

**Conclusion**

Northern blot is used in many ways while studying RNAs.
Topic No 52

**Western blotting**

A technique used to detect the presence of a specific protein in a complex protein mixture.

- To determine the molecular weight of a protein.
- To measure relative amounts (quantitation) of the protein present in complex mixtures of proteins that are not radiolabeled.

Western blots have become one of the most common analytical tools for the detection of viral proteins. Characterization of monoclonal and polyclonal antibody preparations and in determining the specificity of the immune response to viral antigens.

A technique used to detect the presence of a specific protein in a complex protein mixture.

**Topic No 53**

**Western blotting procedure**

Procedure

I. Sample preparation
II. Gel Electrophoresis
III. Blotting (or transfer)
IV. Blocking
V. Antibody probing
VI. Detection

1. The choice of extraction method depends primarily on the sample and whether the analysis is targeting all the proteins in a cell or only a component from a particular subcellular fraction.
2. Samples are loaded into separate wells. A protein marker is also loaded. The separated protein mixtures are transferred to a solid support for further analysis.
3. Transfer can be done in wet or semi-dry conditions. Semi-dry transfer is generally faster. Wet transfer is recommended for large proteins, >100 kD.
4. Blocking is a very important step in the immune detection phase of Western blotting because it prevents non-specific binding of antibody to the blotting membrane.

***Figure 53.1 Electrophoretic transfer***

Protein of interest is detected and localized using a specific antibody. Western blotting protocols utilize a non-labeled primary antibody directed against the target protein.

A species-specific, labeled secondary antibody directed against the constant region of the primary antibody is then used. The most common antibody label used in Western blots is HRP.

The signal is detected when HRP is exposed to a substrate solution in the final step of the immune detection procedure.

**Topic No 54**

**Western blotting procedure**

Procedure

I. Sample preparation
II. Gel Electrophoresis
III. Blotting (or transfer)
IV. Blocking
V. Antibody probing
VI. Detection

1. The choice of extraction method depends primarily on the sample and whether the analysis is targeting all the proteins in a cell or only a component from a particular subcellular fraction.
2. Samples are loaded into separate wells. A protein marker is also loaded. The separated protein mixtures are transferred to a solid support for further analysis.
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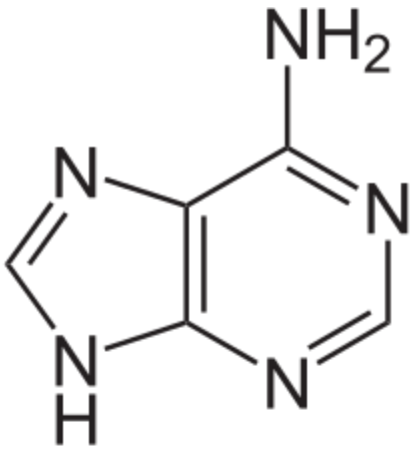
The signal is detected when HRP is exposed to a substrate solution in the final step of the immune detection procedure.

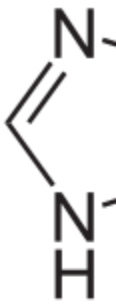
**Topic No 55**

**Nature of Mutations**

- DNA mutations may be very simple (single base change) or very complex and including several thousands of nucleotides. The simplest mutations are switches of one base for another.
- There are two kinds of such mutations which include:- Transitions and Transversions
- Transitions are pyrimidine-to-pyrimidine and purine-to-purine substitutions, such as thymine (T) to cytosine (C) and adenine (A) to guanine (G). Transversions are pyrimidine-to-purine and purine-to-pyrimidine substitutions, such as T to G or A and A to C or T.
- Other simple mutations are insertions or deletions of a nucleotide or a small number of nucleotides.

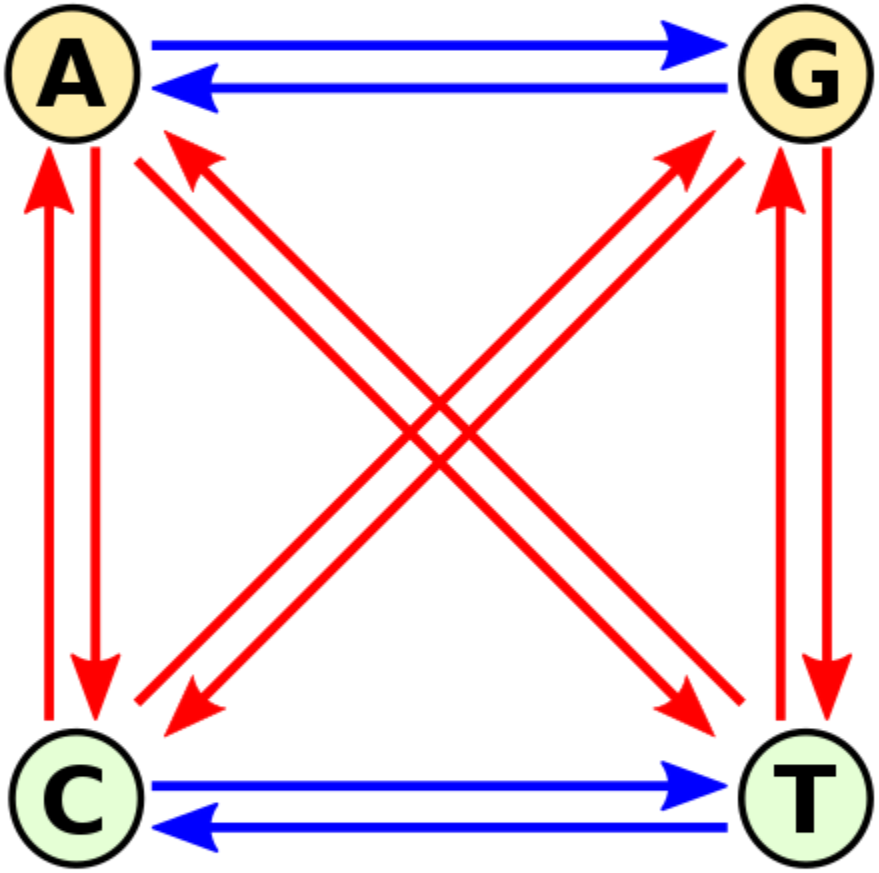



*Purines*




**A**denine

**Transitions**

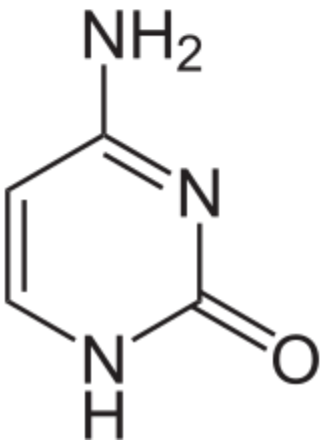



**Transversions**

**Tr**

**C**ytosine

**Transitions**




*Pyrimidines*

*Figure 55.1 Transitions Vs. Transversions Mutations*

All such mutations that alter a single nucleotide are called point mutations. Other kinds of mutations cause more drastic changes in DNA, such as extensive insertions and deletions and gross rearrangements of chromosome structure.

Such changes might be caused, for example, by the insertion of a transposon, which typically places many thousands of nucleotides of foreign DNA in the coding or regulatory sequences of a gene.

**Topic No 56**

**Introduction to Biotechnology**

Biotechnology is the use of microbes, animal/plant cells and their products to synthesize, break down or transform materials. Primarily it includes the use of recombinant DNA technology and genetic engineering techniques to improve upon the quality of processes.

**Branches of Biotechnology**

**Traditional biotechnology** refers to the conventional techniques that have been used for many centuries to produce beer, wine, cheese etc.

**Modern Biotechnology** embraces all methods of genetic modification by recombinant DNA & cell fusion techniques together with the modern developments of traditional biotechnological processes.

**White Biotechnology** refers as development of processes and microorganisms for Industrial processes. Example Enzyme Production.

**Topic No 57**

**Biotechnology – a key technology of the 21st century**

Biotechnology is the use of cell components, cells and organisms for many different purposes such as producing goods and services, providing services or for research and development. Biotechnology techniques makes gentler and more effective individualized therapies possible. It can also contribute to optimizing industrial processes and helping agriculture adapt to climate change. In short, Biotechnology has great potential to influence and benefit agriculture, forestry and fishery. In conjunction with conventional technologies, modern biotechnology holds promise of increased and sustained food production.

**Growing Teeth:**

The 21st century has brought important biotechnological advances in the field of medical science research, such as the application of knowledge and new biological techniques to improve human health. Biotechnology is essential in dentistry to replace the artificial materials that are used today by biological materials based on stem cells, which have the ability to mimic replicas of a tooth or another stomatognathic system tissue. Stem cells have the capacity for continuous renewal and can differentiate into many cell types. Recently, the following stem cells have been identified: dental pulp stem cells (DPSCs), periodontal ligament stem cells (PDLSCs), apical papilla stem cells (SCAPs), stem cells from human exfoliated deciduous teeth (SHED), periapical follicle stem cells (PAFSCs), mesenchymal stem cells derived from adipose tissue (AD-MSCs), and bone marrow mesenchymal stem cells (BMSC).

**Reference: Ayala Escandón CL & Cortes RamírezJ M. Dentistry facing the biotechnological advances of the 21st century. J Oral Res 2018;7(7):216-218.doi:10.17126/joralres.2018.06**

In this current pandemic, we are all aware of the importance and enormous utility of biotechnology, the discipline that produced the modern techniques used to develop the long-awaited COVID-19 vaccine. Not only has biotechnology produced vaccines in record time, but this field will be crucial to respond to the complex challenges that lie ahead, as laid out by the United Nations in 2015 in its Sustainable Development Goals (SDG), the aim of which is to eradicate poverty, protect the planet, and ensure prosperity for all. These ambitious goals entail profound transformations of our production systems, as well as measures to combat hunger, achieve food security and better nutrition, promote sustainable agriculture, combat climate

change, and keep terrestrial and marine ecosystems safe from pollution, among others. Global well-being and quality of life depends on being able to achieve these objectives.

In the broad sense, biotechnology is any technological application that uses biological systems, living organisms, or parts/derivatives thereof to create or modify products and processes for specific uses. For centuries humanity has used biotechnological processes to make products such as beer, wine, cheese, and bread, all of which are produced by the action of live microorganisms. However, in recent decades biotechnology has undergone spectacular advances, increasingly contributing to fundamental daily activities, from pharmaceutical development to food production to the treatment of contaminants. In the Department of Microbial and Plant Biotechnology at the CIB Margarita Salas, 13 research groups are seeking to understand how plants, arthropods, and microorganisms interact and respond to their environment in order to develop biotechnological applications for agricultural , environmental, and industrial sectors. Our aim is to improve the resilience of plants to pathogens and new environmental conditions associated with climate change, develop novel strategies for the control of pests and diseases, accelerate breeding of crops, and harness the potential of microbiological systems for bioremediation of pollutants and the production of chemicals, biofuels, and biopolymers (plastics) from biomass or industrial waste, all via sustainable and circular economy systems. The transfer of technology and knowledge to productive sectors is a key objective of the department's research groups, as

well as groups from other departments that participate in health-related biotechnology projects

**Reference: Pilar S. Testillano, CSIC Investigator at CIB Margarita Salas Deputy Director of the Margarita Salas Center for Biological Research**

**Topic No58**

**Immunity**

It is defined as the state of protection from infectious disease.

It has both a less specific and more specific component.

1. The less specific component, innate immunity, provides the first line of defense against infection.
2. The specific component, adaptive immunity, does not come into play until there is an antigenic challenge to the organism.

*Figure 58.1 Overview Diagram of Immunity and its types*

**The Immune system**

Immune system is the defense system that has evolved to protect animals from invading pathogenic microorganisms and cancer.

It is able to generate an enormous variety of cells and molecules capable of specifically recognizing and eliminating an apparently limitless variety of foreign invaders.

*Figure 58.2 The Immune System*

**Immune response**

An immune response can be divided into two related activities

- Recognition
- Response

Immune recognition is remarkable for its specificity. The immune system is able to recognize subtle chemical differences that distinguish one foreign pathogen from another.

Furthermore, the system is able to discriminate between foreign molecules and the body's own cells and proteins.

Once a foreign organism has been recognized, the immune system recruits a variety of cells and molecules to mount an appropriate response, called an effector response, to eliminate or neutralize the organism.

In this way the system is able to convert the initial recognition event into a variety of effector responses, each uniquely suited for eliminating a particular type of pathogen.

Later exposure to the same foreign organism induces a memory response, characterized by a more rapid and heightened immune reaction that serves to eliminate the pathogen and prevent disease

**Immunology**

The discipline of immunology grew out of the observation that individuals who had recovered from certain infectious diseases were thereafter protected from the disease. The Latin term immunis, meaning "exempt," is the source of the English word immunity, meaning the state of protection from infectious disease.

**Reference**

Kuby Immunology, 6th Edition, Authored by Kindt, Goldsby and Osborne, Chapter 1, Pages 1, 4

Please follow the given links to learn about the basic structure of human immune system, its function and to know about innate and adaptive immunity. To open the link, please copy the URL and paste it into new tab.

https://www.youtube.com/watch?v=k9QAyP3bYmc

https://reverehealth.com/live-better/brief-overview-immune-system/

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2923430/

**Topic No 59**

**Innate Immunity**

I. Less specific component of Immune system
II. First line of defense against infection
III. Most components of innate immunity are present before the onset of infection and constitute a set of disease-resistance mechanisms that are not specific to a particular pathogen but that include cellular and molecular components that recognize classes of molecules peculiar to frequently encountered pathogens.
IV. Phagocytic cells, such as macrophages and neutrophils, barriers such as skin, and a variety of antimicrobial compounds synthesized by the host all play important roles in innate immunity.
V. In contrast to the broad reactivity of the innate immune system, which is uniform in all members of a species,

**Adaptive immunity**

- the specific component
- Does not come into play until there is an antigenic challenge to the organism.

Because adaptive immune responses require some time to marshal, innate immunity provides the first line of defense during the critical period just after the host's exposure to a pathogen. In general, most of the microorganisms encountered by a healthy individual are readily cleared within a few days by defense mechanisms of the innate immune system before they activate the adaptive immune system

Adaptive immunity is capable of recognizing and selectively eliminating specific foreign microorganisms and molecules (i.e., foreign antigens). Unlike innate immune responses, adaptive immune responses are not the same in all members of a species but are reactions to specific antigenic challenges. Adaptive immunity displays four characteristic attributes:

- Antigenic specificity
- Diversity
- Immunologic memory
- Self/nonself-recognition

The antigenic specificity of the immune system permits it to distinguish subtle differences among antigens. Antibodies can distinguish between two protein molecules that differ in only a single amino acid.

**Reference**

Kuby Immunology, 6th Edition, Authored by Kindt, Goldsby and Osborne, Chapter 1, Pages 4,5,9

Please follow the links to learn about innate and adaptive immunity in detail. To open the link, please copy the URL and paste it into new tab.

I. https://www.khanacademy.org/test-prep/mcat/organ-systems/the-immune-system/a/innate-immunity
II. https://www.khanacademy.org/test-prep/mcat/organ-systems/the-immune-system/a/adaptive-immun

**Topic No 60**

**What is the immune system?**

Your immune system is a complex network of cells, tissues, and organs. Together they help the body fight infections and other diseases.

*When germs such as bacteria or viruses invade your body, they attack and multiply. This is called an infection. The infection causes the disease that makes you sick. Your immune system protects you from the disease by fighting off the germs.*

Parts of the immune system:

The immune system has many different parts, including:

- Your skin, which can help prevent germs from getting into the body
- Mucous membranes, which are the moist, inner linings of some organs and body cavities. They make mucus and other substances which can trap and fight germs.
- White blood cells, which fight germs
- Organs and tissues of the lymph system, such as the thymus, spleen, tonsils, lymph nodes, lymph vessels, and bone marrow. They produce, store, and carry white blood cells.

**How does the immune system work?**

Your immune system defends your body against substances it sees as harmful or foreign. These substances are called antigens. They may be germs such as bacteria and viruses. They might be chemicals or toxins. They could also be cells that are damaged from things like cancer or sunburn.

When your immune system recognizes an antigen, it attacks it. This is called an immune response. Part of this response is to make antibodies. Antibodies are proteins that work to attack, weaken, and destroy antigens. Your body also makes other cells to fight the antigen.

Afterwards, your immune system remembers the antigen. If it sees the antigen again, it can recognize it. It will quickly send out the right antibodies, so in most cases, you don't get sick. This protection against a certain disease is called immunity.

*Figure 60.1 The Immune System (Organization)*

**Types of immunity:**

There are three different types of immunity:

- Innate immunity is the protection that you are born with. It is your body's first line of defense. It includes barriers such as the skin and mucous membranes. They keep harmful substances from entering the body. It also includes some cells and chemicals which can attack foreign substances.

- Active immunity, also called adaptive immunity, develops when you are infected with or vaccinated against a foreign substance. Active immunity is usually long-lasting. For many diseases, it can last your entire life.

- Passive immunity happens when you receive antibodies to a disease instead of making them through your own immune system. For example, newborn babies have antibodies from their mothers. People can also get passive immunity through blood products that contain antibodies. This kind of immunity gives you protection right away. But it only lasts a few weeks or months.

**Reference: National Library of Medicine**

**Topic No 61**

**What can go wrong with the immune system?**

Sometimes a person may have an immune response even though there is no real threat. This can lead to problems such as allergies, asthma, and autoimmune diseases. If you have an autoimmune disease, your immune system attacks healthy cells in your body by mistake.

Other immune system problems happen when your immune system does not work correctly. These problems include immunodeficiency diseases. If you have an immunodeficiency disease, you get sick more often. Your infections may last longer and can be more serious and harder to treat. They are often genetic disorders.

There are other diseases that can affect your immune system. For example, HIV is a virus that harms your immune system by destroying your white blood cells. If HIV is not treated, it can lead to AIDS (acquired immunodeficiency syndrome). People with AIDS have badly damaged immune systems. They get an increasing number of severe illnesses.

**Topic No 62**

**Lymphatic System**

A network of fine tubes throughout your body collects fluid called lymph from tissues. Part of its job is to pick up dead cells and germs. Waste is filtered out at small bean-shaped lymph nodes, and the liquid goes back into your bloodstream. An infection can make the nodes swell. You may have felt them in your neck when you had a sore throat or cough.

*Figure 32.1 The Human Lymphatic System*

**Role in immunity**

In addition to serving as a drainage network, the lymphatic system helps protect the body against infection by producing white blood cells called lymphocytes, which help rid the body of disease-causing microorganisms. The organs and tissues of the lymphatic system are the major sites of production, differentiation, and proliferation of two types of lymphocytes—the T lymphocytes and B lymphocytes, also called T cells and B cells. Although lymphocytes are distributed throughout the body, it is within the lymphatic system that they are most likely to encounter foreign microorganisms.

**Topic No 63**

**Vaccines**

**Introduction to Vaccination (Immunization)**

The ways of providing specific protection against many common and damaging pathogens after stimulation of organism's or individual's immune system is called as vaccination or immunization.

For vaccination, there is stimulation of humoral immunity for production of antibodies against pathogen. These antibodies has the ability to neutralize specific antigens of pathogens. This is depending upon the nature of pathogenesis and the site of infection by pathogen. These antibodies are also called as Antitoxin which neutralize the toxin production from pathogens. Those toxins get masked by antitoxins and not able to bind its specific receptor on target cell. Moreover, antibodies against pathogens also bind complement and lead to lysis of pathogen which is also termed as complement mediated intracellular killing of pathogen. However, intracellular pathogens cannot be neutralized by antibodies for which cell mediated immunity is activated by providing specific T-cells to host. Immunization against intracellular pathogen is provided through cell-mediated immunity

**Topic No 64**

**Types of Vaccination**

There are two modes of providing specific protection against mostly common & damaging pathogens. These modes of immunization can be natural or artificial in their nature. Following are the types of immunization

1.
    1. Active Immunization
    2. Passive Immunization

- Active Immunization

  It is the induction of immunity after exposure of host to antigen. In this case, antigenic exposure is mandatory. Active immunization can occur naturally by exposing to microbe or other antigen when there is no prior exposure before the entry of antigen. Similarly there is also no pre made antibodies in the host's serum. In this form of immunization, immune system develops antibodies against the microbe after its exposure. This process of antibodies generation is considered slow. Moreover a memory response is also generated in which antibodies remained in use for longer time. This form of immunization can be artificial in nature which is achieved by either injection of microbe before natural exposure or treated microbes or toxins.

- Passive Immunization

  It is the induction of active humoral immunity. In this kind of immunization, antibodies are mandatory. Passive immunization can occur naturally by transfer of maternal antibodies to fetus through placenta. However, the artificial induction can be done by injecting gamma globulins from other individuals or animals to host.

**Natural vs Artificial Immunization**

Natural active immunization

Natural immunization is acquired immunization which is achieved by natural exposure to antigen or acquiring infection by an infectious agent or microorganism e.g measles virus. This is also termed as natural active immunization. This form of natural exposure to antigen stimulate the humoral immunity as a result specific antibodies are generated against exposed microbe.

Moreover, there is also activation of cell mediated immunity for intracellular pathogen.

**Artificial Active Immunization**

Artificial immunization is the kind of immunization which is acquired by artificial exposure to antigen or microorganism which is administrated to body in less virulent or attenuated form like heat killed forms of microbes. This artificial exposure with antigen stimulate the humoral immunity in the form antibodies generation. Moreover, artificial activation of cell mediated immunity for intracellular pathogen for example BCG for Mycobacterium tuberculosis. Artificial immunization: acquired immunization by artificial exposure. This artificial exposure to antigen or microbe which is by administration of less virulent (attenuated), heat killed microbes to organism. Artificial exposure to antigen stimulate the humoral immunity for antibodies

Natural Passive Immunization

On the other hand, natural immunization can be acquired immunization which can be obtained by isolating gamma globulins (antibodies) from other's individual. This is called as natural passive immunization. Furthermore, the transfer of gamma globulins (IgG) from mother's blood to fetus via placenta. The placental cells contains FcRγ for IgG transfer to fetal circulation. Similarly, the natural transfer of IgA can be occur through breast milk to new-born baby.

Artificial Passive immunization

This form of artificial immunization is the acquired immunization which is achieved by administrating gamma globulins or antibodies from others individual's or immune animal's serum for example antibodies against tetanus

**Topic No 65**

**Novel Vaccines**

There are also various novel ways of designing vaccines in order to reduce in toxicity of vaccines.

These have the potential to provoke both humoral & cell-mediated immunity. These are mainly used in experimentally for research purpose. However, these would be available for clinical use in

future

- DNA vaccines

  These are cloned viral peptides which are transfected into host cell. These vaccines can generate both humoral and cell-mediated response like live attenuated vaccines. Similarly, Anti HIV-DNA vaccines are used in experimental stage which have very low efficacy so far.

- Immunodominant Peptides

  These are simple and easy to prepare. Moreover, these peptides are incorporated with MHC to induce both humoral & cell-mediated responses.

- Anti-Idiotype antibodies

These antibodies are used against polysaccharide antibodies. These have long lasting immune response with memory

**Topic No 66**

**Virus name**

The name "virus" has been derived from a Latin word meaning "slimy liquid" or "poison." Viruses are microscopic parasites that lack the capacity to thrive and reproduce outside of a host body. These can multiply only in living cells of animals, plants, or bacteria. Latin word meaning poison or venom given by Beijerinck (1897).

**WHAT ARE VIRUSES?**

Viruses are submicroscopic, obligate intracellular parasites. Most are too small to be seen by optical microscopes, and they have no choice but to replicate inside host cells. This simple but useful definition goes a long way toward describing viruses and differentiating them from all other types of organism. However, this short definition is not completely adequate.

Viruses are unique in nature. They are the smallest of all self-replicating organisms, historically characterized by their ability to pass through filters that retain even the smallest bacteria. In their most basic form, viruses consist solely of a small segment of nucleic acid encased in a simple protein shell. Viruses have no metabolism of their own but rather are obliged to invade cells and parasitize subcellular machinery, subverting it to their own purposes.

**Virus Size**

- Submicroscopic

  Smaller than bacteria

  Smaller than the smallest cell

***Figure 66.1 (A) Sizes of animal and plant cells, bacteria, viruses, proteins, molecules, and atoms are indicated. The resolving powers of various techniques used in virology, including light microscopy, electron microscopy, X-ray crystallography, and nuclear magnet***

**Reference:**


- Cann, A. J. (2016). Principles of molecular virology (standard edition). Academic press. 6th edition
- Knipe, D. M., Howley, P. M., Griffin, D. E., Lamb, R. A., Martin, M. A., Roizman, B., & Straus, S. E. (2013). Fields Virology, Volumes 1.


**Topic No 67**

**UNIQUE PROPERTIES**

- Living as well as non-living characteristics
- Unable to cultivate in laboratory easily
- Simplest structure
- Complex interaction with host
- Replication within host

**To study an important class of pathogens**

Many types of pathogens cause disease in humans. The most familiar are viruses and bacteria. Viruses cause diseases ranging from AIDS and smallpox to the common cold. They are essentially fragments of nucleic acid (DNA or RNA) instructions, wrapped in a protective shell of proteins and (in some cases) membrane. They use the basic transcription and translation machinery of their host cells for their replication.

Of all the bacteria we encounter in our lives, only a small minority are dedicated pathogens. Much larger and more complex than viruses, bacteria are usually free-living cells, which perform most of their basic metabolic functions themselves, relying on the host primarily for nutrition.

Some other infectious agents are eukaryotic organisms. These range from single-celled fungi and protozoa, through large complex metazoa such as parasitic worms.

Some rare neurodegenerative diseases, including mad cow disease, are caused by an unusual type of infectious particle called a prion, which is made only of protein. Although the prion contains no genome, it can nevertheless replicate and kill the host.

Even within each class of pathogen, there is striking diversity. Viruses vary tremendously in their size, shape, and content, and the same is true for the other pathogens. The ability to cause disease (pathogenesis) is a lifestyle choice, not a legacy shared only among close relatives

**Virus life cycle**

Bacteria, fungi, and eukaryotic parasites are cells themselves. Even when they are obligate parasites, they use their own machinery for DNA replication, transcription, and translation, and they provide their own sources of metabolic energy. Viruses, by contrast, are the ultimate hitchhikers, carrying little more than information in the form of nucleic acid. The information is largely replicated, packaged, and preserved by the host cells. Viruses have a small genome, made up of a single nucleic acid type—either DNA or RNA. The genome is packaged in a protein coat.

Viruses replicate in various ways. In general, replication involves (1) disassembly of the infectious virus particle, (2) replication of the viral genome, (3) synthesis of the viral proteins by the host cell translation machinery, and (4) reassembly of these components into progeny virus particles.

**To study viral cultivation in lab**

- Tissue culture
- Embryonated egg
- Live animal

**Reference :**

Flint, S. J., Racaniello, V. R., Rall, G. F., Skalka, A. M., & Enquist, L. W. (2015). Principles of virology 4th edition. ed.

**Topic No 68**

**VIRUS**

A virus is a sub-microscopic particle that can infect living cells. Viruses are much smaller than prokaryotes, ranging in size from about 20–300 nanometers (nm), though some can be larger. Prokaryotes are typically 0.5–5.0 micrometers (μm) in length. For example, if a virus was about the size of three soccer balls lying side-by-side, then a prokaryote would be about the size of soccer field.

| Virus | Size |
|---|---|
| **Foot and Mouth Disease Virus** | 27 nm |
| **Pox Virus** | 300 nm |

**Structure**

An individual virus is called a virion. It is a tiny particle much smaller than a prokaryotic cell. Because viruses do not consist of cells, they also lack cell membranes, cytoplasm, ribosomes, and other cell organelles. Without these structures, they are unable to make proteins or even reproduce on their own. Instead, they must depend on a host cell to synthesize their proteins and to make copies of themselves. Although viruses are not classified as living things, they share two important traits with living things. They have genetic material, and they can evolve.

**Simple Viruses**

- a protective protein coat
- a nucleic acid genome

Deoxyribonucleic Acid (DNA)

or

Ribonucleic Acid (RNA)

**WHY VIRUSES NEED HOST?**

- No enzymes of their own
- No metabolism
- Require host machinery

**COMPLEX VIRUSES**

Simple viruses only contain protein coat and nucleic acid genome. Complex viruses also contains some additional contents e.g.

- Lipid
- Polysaccharides
- Trace elements

**Topic No 69**

Structure of enveloped and non-enveloped viruses. Non-enveloped viruses are composed of capsid protein and nucleic acid (DNA or RNA), viz. nucleocapsid. Whereas enveloped viruses are composed of an envelope and nucleocapsid. Some viruses may have spikes to help attach to the host cell. Most viruses infect only specific host cells.

*Figure 69.1 Naked Nucleocapsid Virus Vs. Enveloped Virus*

**MORPHOLOGY**

- Viruses come in a variety of shapes.
- Helical Viruses: Protein subunits interact with each other and with the nucleic acid to form a coiled, ribbon like structure: e.g. tobacco mosaic virus, influenza virus, rabies virus. Ebola virus. Structures with helical symmetry.
- Polyhedral shapes: The capsid shell is made of repeating subunits of viral protein (may be one kind of subunit or several, according to the virus). - All faces of the icosahedron are identical. like the influenza virus
- Complex shapes: Most viruses have either icosahedral or helical capsids, but some viruses do not fit into either category. Poxviruses and large bacteriophages are two important examples.

**Reference:**

Maclachlan, N. J., & Dubovi, E. J. (Eds.). (2011). Fenner's veterinary virology. Academic press. 4th edition

**Topic No 70**

**Bacteriophages**

**Introduction**

Even bacteria can get a virus! The viruses that infect bacteria are called bacteriophages, and certain bacteriophages have been studied in detail in the lab (making them some of the viruses we understand best).

We'll take a look at two different cycles that bacteriophages may use to infect their bacterial hosts:

- The lytic cycle: The phage infects a bacterium, hijacks the bacterium to make lots of phages, and then kills the cell by making it explode (lyse).
- The lysogenic cycle: The phage infects a bacterium and inserts its DNA into the bacterial chromosome, allowing the phage DNA (now called a prophage) to be copied and passed on along with the cell's own DNA.

**A bacteriophage is a virus that infects bacteria**

A bacteriophage, or phage for short, is a virus that infects bacteria. Like other types of viruses, bacteriophages vary a lot in their shape and genetic material.

- Phage genomes can consist of either DNA or RNA, and can contain as few as four genes or as many as several hundred.
- The capsid of a bacteriophage can be icosahedral, filamentous, or head-tail in shape. The head-tail structure seems to be unique to phages and their close relatives (and is not found in eukaryotic viruses).

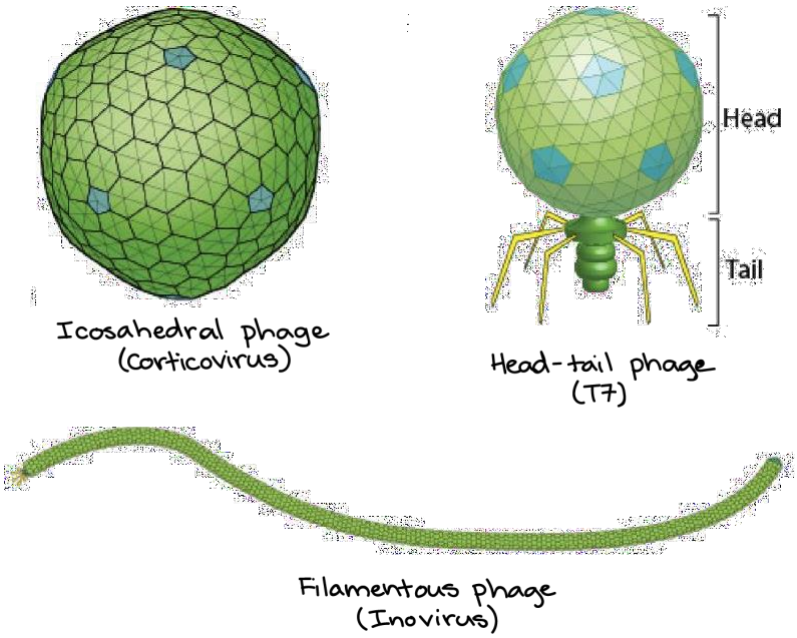



**Figure 70.1 Icosahedral phage, head-tail phage, and filamentous phage**

**Bacteriophage infections**

Bacteriophages, just like other viruses, must infect a host cell in order to reproduce. The steps that make up the infection process are collectively called the lifecycle of the phage.

Some phages can only reproduce via a lytic lifecycle, in which they burst and kill their host cells. Other phages can alternate between a lytic lifecycle and a lysogenic lifecycle, in which they don't kill the host cell (and are instead copied along with the host DNA each time the cell divides).

Let's take closer look at these two cycles. As an example, we'll use a phage called lambda (\lambdaλlambda), which infects E. coli bacteria and can switch between the lytic and lysogenic cycles.

**Topic No 71**

BACTERIOPHAGES

Properties

A **bacteriophage**
also known informally as a ***phage*** is a virus that infects and replicates within bacteria and archaea

General Characteristics

Virus architecture

- Virus particle called virion
  - Consists of nucleic acid surrounded by protein coat
    - Capsid
- Shapes
  - Isometric
  - Helical
  - Complex
- Two types of virion
  - Naked – without envelope
  - Enveloped – surrounded by lipid membrane

SIZE

VIRUS INTERACTIONS WITH HOST CELLS

LYTIC CYCLE

**Topic No 72**

**MICROBES**

They are minute, unicellular organisms that are also called as germs or microorganisms. They are too small to be seen with the unaided eye. Therefore, a microscope is used to observe them. They are found all around us. They live in water, soil, and in the air. The human body is home to millions of these microbes too, also called microorganisms.

Some microbes make us sick; others are important for our health. The most common types are bacteria, viruses and fungi. There are also microbes called protozoa. These are tiny living things that are responsible for diseases such as toxoplasmosis and malaria.

**MICROBIOLOGY**

It is the study of microorganisms. The field is concerned with the structure, function, and classification of such organisms and with ways of both exploiting and controlling their activities.

**CLASSIFICATION OF MICROBES**

Classification of microorganisms has been a challenge. In the currently accepted scientific classification of Life, there are three domains of microorganisms:

- Eukaryotes
- Bacteria
- Archaea

**EUKARYOTIC VS PROKARYOTIC CELL**

***Table 72.1 Differences between prokaryotic and eukaryotic cells***

| Cell Parts | Prokaryotes | Eukaryotes |
|---|---|---|
| Nucleus | **No** | **Yes** |
| Cell Wall | **Yes** | **Yes – Plant and fungus cells**<br><br>**No – Animal cells** |
| Cell membrane | **Yes** | **Yes** |
| Cytoplasm | **Yes** | **Yes** |
| Membrane bound organelles | **No** | **Yes** |
| Ribosomes | **Yes** | **Yes** |

*Figure 72.1 Differences between prokaryotic and eukaryotic cells*

**ANIMATION:**

https://www.youtube.com/watch?v=iDVq2etiknU

**Reference:**

Microbiology: An Introduction, 10th Edition, authored by Tortora, Funke, and Case _ pages 2, 6, and 274 (Chapter 10).

**Topic No 73**

**Introduction**

There is a need to classify microbes in order to study them conveniently, to identify them easily, to establish relationship between organisms and to establish the evolutionary relationships.

**Bases of Classification:**

- Similarities among them
- Morphology used to be the main feature prior to the advent of molecular biology
- Biochemical similarities
- Genetic similarities
- Evolutionary relationships

**Five Kingdom Classification of Organisms**

The first person to divide living things into five broad kingdoms was North American ecologist Robert Whittaker. This researcher proved in 1959 that fungi were not plant organisms - previously it was thought that they were - and a decade later he proposed the creation of the fungi kingdom to differentiate them from plants. Whittaker's theory was widely accepted, and the scientific community thereby added a new group to the previous four-kingdom system, established by the American biologist Herbert Copeland in 1956.

**Animal Kingdom**

The kingdom Animalia is the most evolved and is divided into two large groups - vertebrates and invertebrates. These animals are multi-celled, heterotrophic eukaryotes with aerobic respiration, sexual reproduction and the ability to move. This kingdom is one of the most diverse and comprises mammals, fish, birds, reptiles, amphibians, insects, molluscs and annelids, among others.

**Plant Kingdom**

Trees, plants and other species of vegetation make up part of the Plantae kingdom - one of the oldest, and characterized by its immobile, multicellular and eukaryotic nature. These autotrophic things, whose cells contain cellulose and chlorophyll are essential for life on Earth since they release oxygen through photosynthesis. As regards their method of reproduction, this may be either sexual or asexual.

**Fungi Kingdom**

This name is used to designate the fungi kingdom which includes yeasts, moulds and all species of mushrooms and toadstools. These multicellular aerobic heterotrophic eukaryotes have chitin in their cell walls, feed off other living things, and reproduce through spores.

**Protista Kingdom**

This group is the most primitive of the eukaryotics and all the others are descendants of it. The Protista kingdom is paraphyletic - it contains the common ancestor but not all its descendants - and it includes those eukaryotic organisms that are

not deemed to be animals, plants or fungi such as protozoa. As it is so heterogeneous it is difficult to categorise it, since its members have very little in common.

**Monera Kingdom**

This is the kingdom of microscopic living things and groups together the prokaryotes (archaea and bacteria). This group is present in all habitats and is made up of single-cell things with no defined nucleus. Most bacteria are aerobic and heterotrophic, while the archaea are usually anaerobic and their metabolism is chemosynthetic.

The classification of the five kingdoms of nature remains the most accepted today, although the latest advances in genetic research have suggested new revisions and reopened the debate among experts. Such is the case for the sixth kingdom of Carl Woese and George Fox, who in 1977 divided bacteria into two types (Archaea and Bacteria), and the seventh kingdom of Cavalier-Smith, who added a new group to the previous six for algae called Chromista.

**Three Domain Classification**

It was proposed by Carl Woese in 1977. According to this system, organisms are divided into following domains.

**1. Bacteria:** These are prokaryotes that are usually single-celled organisms. Their cell wall is made up of peptidoglycan.

**2. Archea:** These are prokaryotes. Their cell wall has no peptidoglycan but has special lipids. They also vary in rRNA sequencing and have capability to live in extreme conditions. They include thermophiles and halophiles

**3. Eukarya:** They include the following kingdoms:

- ime molds, protozoa, algae):
- Algae contain cellulose cell wall and are photosynthetic in nature, as they can produce oxygen.
- Protozoans are motile and unicellular organism.
- Slime molds are Protozoa like at one stage and fungus like in another.

Fungi

- Unicellular form is called yeast.
- Multicellular form is known as mold or mushroom

Plants

Animals

**Other Microbes**

Viruses: These are acellular organisms that consist of proteins and nucleic acids

Viroids: These are composed only of circular ssRNA, for example, potato spindle tuber viroid

Virusoids: These contain circular ssRNA and need helper viruses for their replication and encapsidation. These are also called satellite viruses

Prions: These are defined as infectious protein particles.

**Naming Organisms: Binomial Nomenclature**

Carl Linnaeus established the system of scientific nomenclature in 1735. According to this system, each organism has two names: the genus and specific epithet (descriptive word)

**Scientific Names:**

Scientific names of microorganisms are italicized or underlined. The genus is capitalized, and the specific epithet is lowercase. Scientific names are "Latinized" and used worldwide. These names may be descriptive or honor a scientist.

For example, the scientific name of Escherichia coli honors the discoverer, Theodor Escherich and describes the bacterium's habitat—the large intestine, or colon.

The name of Staphylococcus aureus, describes the clustered (staphylo-) spherical (cocci) cells and the gold-colored (aureus) colonies.

After the first use, scientific names may be abbreviated with the first letter of the genus and the specific epithet:

Escherichia coli and Staphylococcus aureus are found in the human body. E. coli is found in the large intestine, and S. aureus is on skin.

**ANIMATION:**

https://www.youtube.com/watch?v=fU0XO1X1tAE

**Reference:** Microbiology: An Introduction, 10th Edition, authored by Tortora, Funke, and Case_pages 3-6, 281, 393-394.

**Topic No 74**

**INTRODUCTION**

An unaided human eye can only distinguish two objects only if these are 0.2 mm apart from each other. Thus, the resolving power of the human eye is 0.2mm or 200µm. On the other hand, objects to be studied by biologists are too small to be seen with naked eye. For instance, most cells have a diameter of about 10µm only. For observing such tiny objects, scientists make use of microscopes.

**MICROSCOPE**

A microscope is an instrument that magnifies objects otherwise too small to be seen, producing an image in which the object appears larger. It was first invented by Antonie Van Leeuwenhoek. Thus, he was called as the "Father of Microbiology".

Most photographs of cells are taken using a microscope, and these pictures can also be called micrographs.

**HISTORY**

Microscope was firstly invented by Antonie van Leeuwenhoek. For the same reason he is known as the Father of Microbiology

USES OF MICROSCOPE

- Microscopes are useful in
- Diagnostics
- Identification of organisms
- Histology – Tissue analysis
- Examining Forensic Evidences
- Studying cells and Micro-organisms
- Scientific Research

**PARTS OF COMPOUND MICROSCOPE**

**Eyepiece** or ocular lens having10x magnification power, using which you can observe the magnified image of object.

**Arm** of microscope, which connects eyepiece to base of microscope.

**Stage** where we place our specimen slide to be observed.

**Stage clips** to hold the slide in proper place during observation, there are stage clips which fix the slide in its particular position.

**Objective lenses** of 4X, 10X, 40X,100X magnification powers that are mounted on a rotating turret called as nosepiece. Nosepiece can be rotated to change the lens, which clicks into position when the lens is set. The four lenses are color coded as well to give an idea of their magnification power.

- The lowest power objective lens is coded red and has a magnification of 4x.
- The lens coded yellow has a magnification power of 10x.
- The high-power magnification lenses, coded blue and green respectively, have a power of 40X and 100x. However, 100x is also called as oil immersion lens and its reason will be explained shortly.

**Condenser:** Whose function is to focus the light rays onto the specimen under observation.

**Diaphragm** along with its control lever. This is used to control the size of aperture through which light passes. Alternately the intensity of light can also be controlled through another knob known as the illumination control knob.

**Light source/Illuminator** from where the light emits. Light is then passed to the condenser lens to be focused on the specimen.

**Coarse adjustment knob** that moves the stage up and down and for focusing the specimen.

**Fine adjustment knob:** As you know every person have specific vision power so while observing every individual focus the specimen at different position. The Fine adjustment knob here is used to deal with such differences and helps to focus the object without moving the specimen or slide. This in simple terms is used for someone who otherwise wears glasses.

**Base** provides basal support to the microscope.

*Figure 74.1 Parts of compound microscope*

**Reference:**

Microbiology: An Introduction, 10th Edition, authored by Tortora, Funke, and Case_Read pages 55-59 (Ch 3).

**Topic No 75**

**MAGNIFICATION POWER**

Magnification is the ability of lens to enlarge the object. Magnification power of microscope is determined by the focal lengths of the two lens systems. To get the total magnification take the power of the objective lens used that may be 4X or 10X or 40x or 100X and multiply it by the power of the eyepiece that is 10X. For example, if you are using objective lens of power 40X, then multiply 40 with 10, because power of eyepiece is always 10x. As a result, total magnification of microscope will be 400X.

**REFRACTIVE INDEX**

It is defined as the ability of a medium to bend the light when passing from one medium into another. Light rays move in a straight line through a single medium

**RESOLUTION POWER**

It is the Ability of the lenses to distinguish between two closely lying objects as separate.

Light microscope resolving power is 0.2 µm.

*Figure 75.2 Resolution Power, n = refractive index, 1.5 for immersion oil*

**ANIMATION:**

https://www.youtube.com/watch?v=--dIBinUdeU

**Topic No 76**

**Compound Microscope**

In a compound microscope, magnifying powers of two convex lenses, eyepiece and objective lens, are used to produce a magnified image of very small objects. Light from a light source pass through a thin object. The objective lens produces a magnified 'real image' of object. This image is again magnified by the ocular lens eyepiece to obtain a magnified 'virtual image' that is a final image and can be seen by eye through the eyepiece. Thus, the light passes directly from the source to the eye through the two lenses, the field of vision is brightly illuminated. That is why; it is a bright-field microscope. Transparent specimens can be stained before their observation. These stains usually color different parts of the cells.

*Figure 76.1 Compound/Bright-Field/Light Microscope*

Dark Field Microscope- It is used for the Examination of unstained microbes. It contains special kind of condenser, with an opaque disc obstructing the path of light from the light source centrally but allowing a peripheral ring of light to pass. As a result, bright specimen can be viewed against a dark background.

*Figure 76.2 Working Principle of Dark Field Microscopy*

*Figure 76.3 Dark-Field Microscope*

Phase Contrast Microscope- that is applicable in Cell culture. It is used to observe living cells in their natural state without any previous fixation or labeling. Its principle is such that, when light passes through cells, small phase shifts occur. These phase shifts are converted into changes in amplitude, which can be observed as differences in image contrast.

*Figure 76.4 Image of live cells produced by Phase Contrast Microscopy*

Fluorescent Microscope – is another variation of microscopes in which specimens are formerly stained with Florescent dyes, for example DAPI(4′,6-diamidino-2-phenylindole) and Hoechst stains.

During microscopy, specimen is illuminated with light of a specific wavelength that is absorbed by fluorophores and converted into a light of longer wavelength. As a result, fluorescent specimen can be easily observed.

*Figure 76.5 Principle of fluorescent microscopy*

**Figure 76.6: Image of cells produced by fluorescent microscope**

**Topic No 77**

**Electron Microscope:** It has high magnification and resolution. It uses a beam of accelerated electrons as a source of illumination.

**Figure 77.1: Electron Microscope**

**Figure 77.2: Image produced by Electron Microscope**

There are two main types of electron microscope including:

- Transmission Electron Microscope (TEM)
- Scanning Electron Microscope (SEM)

***Table 77.1 Main differences between a SEM and a TEM***

| | SEM | TEM |
|---|---|---|
| Type of electron | Scattered, scanning electrons | Transmitted electrons |
| High tension | ~1–30 kV | ~60–300 kV |
| Specimen thickness | Any | Typically, <150 nm |
| Type of info | 3D image of surface | 2D projection image of inner structure |
| Max. magnification | Up to ~1–2 million times | More than 50 million times |
| Max. FOV | Large | Limited |
| Optimal spatial resolution | ~0.5 nm | <50 pm |
| Image formation | Electrons are captured and counted by detectors, image on PC screen | Direct imaging on fluorescent screen or PC screen with CCD |
| Operation | Little or no sample preparation, easy to use | Laborious sample preparation, trained users required |

**ANIMATION:**

https://www.youtube.com/watch?v=0BMVXwahWfI

**REFERENCE:**

Microbiology: An Introduction, 10th Edition, authored by Tortora, Funke, and Case_ Read pages 59-66.

**Topic No 78**

**Digestive System**

**Nutrition**

Acquiring energy from environment is one essential property of life. Plants acquire energy from sunlight and prepare their food. We call them producers. Animals, however, acquire energy from either plants or other animals or organic materials. We call the consumers or decomposers.

**Common Modes of Nutrition in Animals**

- Herbivores
  - Plant eaters, e.g., goat
- Carnivores
  - Flesh, meat eaters, e.g., lion
- Omnivores
  - Mixed food, e.g., bear, crows
- Detritivores
  - Dead organic matter, e.g., earthworms

Dentition is according to the mode of nutrition

*Figure 78.1 Dentition in animals; incisor, canine, premolar and molar teeth*

Mouth Parts also are also according to the mode of nutrition Animals have different types of tongues according to their mode of nutrition. For example, frogs have forked, inverted and sticky tongue to catch insects. Tongue of chameleon is also sticky and very long. Beaks of birds make another example. Birds have a variety of beaks; long, small, curves and many other. All of these are present in birds according to their mode of nutrition for example parrots have long curves beak for cutting and breaking nuts. Pelican have a very long and wide for catching fish from water. Snouts in mammals make another example. Mammals have different snouts according to their food.

*Figure78.2 A parrot: observe the long and curved beak*

**Topic No 79**

**Nutrition, Ingestion, Digestion, Absorption, Assimilation, Elimination**

- After acquiring food and ingestion, animals have to digest it.
- Digestion is actually, breaking down food into small ingredients. For example, carbohydrates to glucose.
- Absorption is movement of food into blood after digestion.
- Assimilation is utilizing those broken small molecules for getting energy for organism's functions.
- Elimination is removal of the undigested matter from the body.

**Nutrition and Digestion in Human beings**

Humans are omnivores, i.e., we eat plants (vegetables) and meat (chicken, beef). Digestive system of humans hence is adapted accordingly.

**Topic No 80**

**Basic Components of Human Digestive System**

- Alimentary canal

  - From mouth to anus

- Alimentary canal in humans consist of following parts:

  - Oral cavity
  - Pharynx
  - Esophagus
  - Stomach
  - Intestine (small and large) Rectum and anus

- Accessory glands

  - Liver, Pancreas

*Figure 79.1 Digestive System in Human beings*

**Sensing the Food**

We look at the food and smell when we get a food. These are called sensory qualities of food. This is the first part of food selection and foods which are improper are rejected. If the food looks inappropriate or it smells bad, it is rejected. This is a human adaptation just like other animals.

**Alimentary Canal –** From mouth to anus

**The Oral Cavity**

Food selection is one function of the oral cavity. Oral cavity has tongue with taste buds. Food selection takes place by taste buds if food tastes bad it is rejected. Tongue converts the ground food mass to a bolus. Grinding the food is the other important function of oral cavity. Oral cavity contains teeth for this purpose. Dentition is according to the food types. Teeth are of four types: incisors, canine, premolars and molars. Humans have all four types of teeth; few animals have others. Lubrication and chemical digestion is another function of the oral cavity. Salivary glands secrete saliva which contains amylase for digestion of starch. It also contains mucous for lubrication of food.

**Pharynx**

Pharynx is a part of digestive system. It lies just behind the mouth and nasal cavity and above the esophagous and larynx. Food bolus passes through pharynx to enter the esophagus. Pharynx helps swallowing by closing the trachea and nasal cavity pathways.

**Esophagus**

It is a long tube starts from pharynx and enters stomach. From pharynx, food enters esophagus. In esophagus, food moves down to stomach by a series of muscular contractions called “peristalsis”. Bolus moves down by alternate contraction and relaxation of muscles of esophagus. Sometimes an antiparistalsis occur, i.e., movement from stomach to mouth that result in vomiting.

**Stomach**

Stomach is mainly a storage organ. It also helps in digestion of proteins. Stomach has guarded openings on either side. There is a sphincter (group of muscles) which is pyloric sphincter that controls movement of food from stomach to intestine. The other sphincter is present where esophagus meets stomach. Gastric glands in stomach produces gastric juice, which contain mucus, hydrochloric acid and an enzyme called pepsinogen. Pepsinogen is converted into pepsin which is its activated form.

Pepsin converts proteins to peptides. Protection of the stomach walls from the acid particularly is important, mucus secreted by the stomach walls cover the stomach lining to protect it from acidic damage.

Stomach is also designed to mix up the food with enzymes and acid. Churning movements of stomach walls thoroughly mixes the food with gastric juice. Churning converts food to chyme, small part of which enters the small intestine by pyloric sphincter

**Topic No 81**

**Small Intestine**

It consists of three parts: duodenum, jejunum and ilium. Duodenum (first 25 cm of intestine) is the first part where most of the digestion occurs. Bile enters here from liver and helps emulsification and digestion of lipids. Pancreatic juice from pancreas also enters here, which contains trypsin, pancreatic amaylase and lipase for digestion of protein, carbohydrates and lipids. Intestinal juice is also released, which help in digestion of all foods.

Jejunum is the next 2.4 m of the small intestine; remaining digestion takes place here. Ilium is the last 3.5 m long part where absorption takes place. Its lumen is highly folded. It have extensions of the epithelium called villi and microvilli, which increases the surface area to a greater extent to increase absorption.

***Figure 81.1 A single villus in small intestine***

**Topic No 82**

**Large Intestine**

Undigested food comes in the large intestine. It consists of various parts including caecum, colon, and rectum. Absorption of water from the undigested matter occurs in colon. Water is returned to blood vascular system. Remaining semi solid mass is called feces that are are stored in rectum, when rectum is full it gives rise to a reflex for defecation and feces are excreted out. Intestinal Motility is an important factor. If the motility in large intestine is increased, then there is less water reabsorption and result is a situation called diarrhea. On the other hand, if motility is decreased then constipation is the result which means that feces will have less than normal water.

**Liver and Pancreas**

Liver produces a secretion called bile which helps in fat digestion and emulsification. Bile is stored in gall bladder and is released by its contraction. Liver performs various functions called deamination, converting ammonia to urea, destroys old blood cells and manufactures fibrinogen, which is blood clotting factor.

**Topic No 83**

**Transport System & Blood - The Circulatory Fluid**

**Why transport systems?**

Organisms have to exchange materials with the environment. They also need to distribute

nutrients, gases and metabolic products within their body.

**Living organisms**

- acquire energy or food from the environment; they have to distribute energy and

  food within the body.

- need an intake of oxygen and removal of carbon dioxide from their bodies.
- need to remove toxic materials and waste products from their bodies.

- have to distribute hormones and other materials in the body.
- have to exchange materials with the environment

For all that, they require a Transport System.

**Transport systems in organisms**

A transport system should fulfill the requirements of the organism for all of the above functions like nutrient distribution or gas exchange.

We take some examples here:

- Unicellular organisms
  - e.g., amoeba
- Simple multicellular organisms
  - Small organisms, e.g., sponges, hydra
- Plants and animals

**Unicellular organisms**

Unicellular organisms have a simple body, just one cell. Their requirements are also few.

For example, amoeba performs the transport via cytoplasmic streaming which is the movement of their cytoplasm and few organelles. Food vacuoles distribute the food in the body. Contractile vacuoles remove the water from the body. Cytoskeleton helps in the movement of all the above parts.

*Figure 83.1 Amoeba*

**Simple multicellular organisms**

Simple multicellular organisms transport materials by simple methods. Like hydra have a mouth and a body cavity; water enters through mouth and transport of materials takes place in the body cavity. Body wall also exchange materials with the water outside as organism is aquatic. Hence, water act as a medium of transport. Few cells help in transport, which are present inside the body towards body cavity.

*Figure 83.2 Hydra: Transport System*

**Transport in plants**

Transport in plants occurs by roots and conducting tissues called phloem and xylem. Roots absorb water and materials from soil. Plants remove water through a process of transpiration. The leave has guarded openings called stomata, which open and close to remove water.

*Figure 83.4 Stomata: Opened & Closed*

**Transport in animals**

Animals have a system called circulatory system for transport. There are two kinds of this system called an open circulatory system and closed circulatory system. In an open circulatory system, the circulatory fluid is freely moving in the body cavity bathing tissues and there is/are pumping heart/s but these only send blood to some vessels which are then connected to body cavity. In closed circulatory system, the blood never leaves vessels and heart and is always present inside the vessels.

**Topic No 84**

**Basic components of human circulatory system**

Blood is the circulatory fluid which is the medium for transport. Blood vessels are the tubular system for transport of blood within the body. Heart is the pumping organ that pumps the blood toward the body.

**Composition of blood**

Human blood consists of plasma (fluid) and cellular content (cells). Plasma is the fluid with dissolved and un-dissolved materials. It consists mainly of water, which have many inorganic and organic materials dissolved in it. Cellular content consists of Red Blood Cells (RBCs), White Blood Cells (WBCs) and Platelets. Red blood cells are also called erythrocytes and white blood cells are also called leukocytes. Plasma consists of 55% of the blood. Cells constitute 45% of the blood Average human body has about 5liters of blood. Blood contents could be separated based upon their densities and weights by various methods including centrifugation and sedimentation.

*Figure 84.1 Components of blood*

**Functions of the main components of blood**

**Blood plasma**

Plasma consists of 55% of the blood, It consist of 92% water. 0.9% of it consists of salts – NaCl, Mg, Zn ions etc. The salts maintain the pH of the blood. Plasma has 7-9% part consist of proteins. Important proteins include antibodies, fibrinogen and albumin. Plasma also contains digested food, nitrogenous wastes, hormones, respiratory gases etc.

**Topic No 85**

**Blood cells and cell-like bodies**

Cellular components of blood consist of erythrocytes, leucocytes, thrombocytes. Erythrocytes – Red Blood Cells carry oxygen with the help of a protein called hemoglobin. In men 5-5.5 million cells/mm3 RBCs are present. This number is 4- 4.5 million/mm3 in women. Men have usually larger bodies and more muscle mass, so that their demand for oxygen is more than women. Leukocytes – White Blood Cells are categorized in to two kinds: Agranulocytes and granulocytes.

Agranulocytes are those white blood cells which have nongranulated cytoplasm, these include monocytes and lymphocytes. Granulocytes are those WBCs, which have granulated cytoplasm. These include basophils, eosinophils and neutrophils. Platelets are the third type of cells, or we can call these, cell like bodies. These are formed by fragmentation of a single cell called megakaryocyte.

Erythrocytes (RBCs) do not have a nucleus at maturity. That is why these look concave under microscope.

Blood cells include RBCs and WBCs (lymphocytes, monocytes, neutrophils, eosinophils, and basophils).

Figure 85.1 Platelets

**Topic No 86**

**Blood groups in human beings and blood transfusion**

Blood cells have some specific protein present on their surfaces called antigens, which are meant for the identification of the blood cells. About 29 blood groups are identified. The most important blood group systems are, however, two named as ABO and Rh blood group systems. There are four main types of blood groups present in human beings according to ABO blood group system. These blood groups are based upon presence or absence of two antigens on surface of RBCs called antigen A and antigen B (Table 1). Other important blood group is based upon presence or absence of Rh antigens on the surface of RBCs. The people have Rh antigen on the surface of their RBCs are called Rh positive and those who do not have it are called Rh negative. ABO system works with the help of Rh system. So that we talk of the blood groups based upon ABO system with Rh positive or negative. For example, A +ve means that the person has an A blood type with Rh antigens present.

*Table 1 Blood groups, antigens and antibodies*

Blood transfusion Blood transfusion is required in case of some diseases, surgery or in case of injuries. Blood groups of donor and recipient should match, otherwise could cause agglutination reactions leading to even death of the patient. Blood groups are matched using antigen antibody reactions. A blood group O -ive is called Universal donor because it has not antigens, hence, it cannot react with any of the antibodies in recipient's blood. Universal acceptor is AB+ blood group which has all antigens.

**Topic No 87**

**CIRCULATORY SYSTEM IN HUMANS – BLOOD VESSELS & HEART**

**Circulatory System in Humans**

Circulatory system in humans consists of blood, vessels and heart. Blood is the circulatory fluid. We can call it a liquid tissue. Blood consist of plasma – the watery fluid and cells – the cellular content. Blood has been described in detail in last lecture. Here we talk about heart and blood vessels. Heart is a pumping organ and is highly muscular. Function of the heart is to pump blood with pressure towards lungs and body. Vessels are of three main types; arteries, veins and capillaries. Arteries carry blood from the heart to body. Veins return blood from body to heart. Capillaries connect arteries to veins. Function of the capillaries is the exchange of materials.

**Structure and functions of blood vessels**

Arteries and veins are elastic in their structure. These also have a layer of muscles. These consist of 3 layers:

- External layer - connective tissues layer
- Middle layer, consist of smooth muscles
- Inner layer, consist of endothelium

Lumen of both of these is meant for blood flow. Arteries have a lumen smaller in diameter and veins have large.

*Figure 87.1 Artery, vein and capillaries*

*Figure 87.2 Comparison of arteries, vein & capillaries*

**Arteries** carry blood from heart to body. These face maximum pressure. Their function is to distribute blood to all the body tissues. Blood is pumped in the arteries by the heart with very high pressure that is why arteries are thick walled to withstand high pressure

**Topic No 88**

**Veins** have to collect blood from the body and return it to the heart. Veins face less pressure, pressure almost negligible in major veins. Blood flow in major veins against gravitational pull and with low pressure so that veins have valves that prevent backflow of blood. In many parts of the body like arms and legs there are muscles that contract and relax to move the blood towards heart. Capillaries have to exchange materials with tissues; these originate from arteries and pass from tissues then joins to make veins.

**Capillaries** have to carry out exchange of materials with the tissues that is why their walls are just one celled thick. Capillaries when enters tissues makes branches, where they branch groups of muscles are present which may open or close to increase or decrease the blood flow towards tissues.

*Figure 88.1 Comparison of an artery and a vein*

Main arteries and veins in human arterial and venous systems

- head, shoulders, arms

other body organs like kidneys, gonads, liver etc

**Topic No 89**

**Structure of human heart**

Heart in human beings is a muscular pump to push blood towards body and lungs. Heart in humans is of the size of a clenched fist. Heart consists of four chambers including 2 atria and 2 ventricles. Atria receives blood from body and lungs. Ventricles push blood from heart to body and lungs. These chambers (atria and ventricles) are connected to each other. At their junction valves are present, which are responsible to prevent backflow of blood. Left side of the heart, particularly ventricle is stronger because it has to push blood to the whole body in comparison to right side.

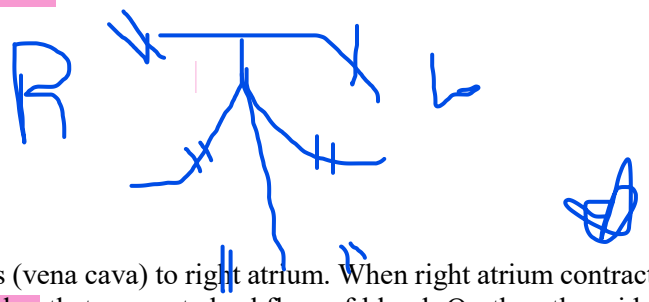



*Figure 89.1 Structure of Human Heart; ® Michigan Medicine*

**Double pumping action of heart and role of valves**

\Deoxygenated blood from body enters the heart through major veins (vena cava) to right atrium. When right atrium contract, blood enters the right ventricle. There is a valve called tricuspid valve that prevents backflow of blood. On the other side, oxygenated blood from lungs enters the left atrium and when left atrium contract it enters the left ventricle. At the junction of left atrium and ventricle another valve is present called bicuspid valve that prevents backflow of blood. Tricuspid and bicuspid valves are collectively called atrioventricular vales.

In fact, both atria are filled at the same time and also contract at the same time to push blood in the ventricles. Both ventricles also contract at the same time to push blood toward body (left one) and lungs (right side). Right ventricle pushes blood in the pulmonary artery towards lungs. At this junction a valve is present called pulmonary valve that prevent backflow of blood from pulmonary arteries to right ventricle. Left ventricle pushes blood towards aorta, which is the main artery distributing blood the body. This junction is also guarded by a valve called aortic valve, which prevents backflow of heart from the aorta to left ventricle. Pulmonary and aortic valves are collectively called as semilunar valves.

Self-excitatory system of the heart

Heart has self-excitatory system. Heart has s group of muscles called sinoatrial node which produces impulse (electrical activity) that spread in the heart to contract its various chambers systematically. Impulse spread toward atria first and causes the atria to contract. Then it enters another node called atrioventricular node where its magnitude is increased and then it enters the ventricles and the ventricles contract. The self-excitatory group of muscles of the heart is called pacemaker.

**Cardiac Cycle**

Cardiac cycle is the time period from when the blood enters the heart to the time when blood is pushed by the heart. We can also say that it is the time period from artial and ventricular diastole to ventricular systole. Cardiac cycle consists of following steps:

- Both atria and ventricles are relaxed. This is the time when both atria are filled with blood from body and lungs. All heart muscles are relaxed, which means that these are at diastole.
- Then both atria contract and blood are pushed into ventricles. This is called atrial systole. At this time, both AV valves are open and both semilunar valves are closed. Then the ventricles contract and push blood into aorta and pulmonary artery. This is called ventricular systole. With this, a cardiac cycle is complete.
- Then again, both atria and ventricles are relaxed, i.e., come back to the initial state.

**Topic No 90**

**Double Circulation**

Human circulatory system is called a double circulatory system because heart receives and sends blood from and to lungs (pulmonary circuit) and body (systemic circuit). Look at the diagram below for a detailed view.

- Systemic circulation

  - From heart to body and vice versa
  - Head, shoulders and arms
  - Lower body organs

- Pulmonary circulation : From heart to lungs and vice versa

**Topic No 91**

**Why respiration?**

- Living organisms needs energy for their activities.
- Energy is extracted by complex metabolic processes from food or photosynthesis.
- Respiration is the process of exchange of gases to produce energy in biological form.

**What is respiration?**

Respiration consist of processes involves exchange of gases in living organisms or

metabolic reaction to produce energy. Respiration is one of the most important metabolic

activities. It is of two types:

- Organismic respiration consists of breathing or ventilation, i.e., inhalation and exhalation movements and gas exchange.
- Cellular respiration consists of metabolic reactions to produce energy by utilization of oxygen, production of carbon dioxide and extracting and conserving energy from food molecules in biological form, such as ATP.

**Cellular respiration**

Cellular respiration is of two types:

- Aerobic, in which oxygen is utilized to produce energy.

Example is electron transport chain in mitochondria.

- Anaerobic, in which oxygen is not involved but energy is produced using other molecules.

  Example is fermentation in some bacteria.

**Organismic respiration**

Organismic respiration is the ventilation or breathing, i.e., the movements which causes inhalation and exhalation. Air contains gases and exchange of gases occurs in the specialized organs, for example, lungs in humans.

**Difference between cellular and organismic respiration**

- Cellular respiration is a series of events of metabolic reactions for production of energy.
- Organismic respiration is the process of coordinated movements of body that results in inhalation and exhalation of air. We also call it breathing or ventilation.

**Respiration in air and water**

Respiration in air is easier in comparison to water because air is lighter, and water is denser. Also, air is much more saturated with oxygen than water. So that gas exchange in air is easier in the water than in water.

**Respiration in plants**

Gas exchange in plants occurs by stomata in mesophyll cells. Stomata are openings guarded by guard cells; these are used for transpiration in plants through water vapors. Roots of land plants get the oxygen required by the plants from air present in soil. Aquatic plants acquire their oxygen from oxygen dissolved in water.

**Respiration in animals**

In animals, respiration occurs by means of specialized organs, for example gills in fishes and lungs in terrestrial animals. The most important property of the respiratory organs is respiratory surfaces that support and facilitate the process of respiration.

**Properties of Respiratory surfaces**

Properties of Respiratory surfaces include a large and moist surface area, a thin epithelium, ventilation (air availability) and a capillary network.

**Respiration in some small animals**

In small animals, respiration occurs by means of general body surface or specialized systems. Hydra is an aquatic organism, which have a mouth and body cavity through which water moves in and out. Cells of the inner body wall exchange gases with that water. Cells of the epidermal layer exchange gases with the water.

In some other organisms like earthworms which have a closed circulatory system the exchange of gases takes place by the capillaries, which are present in closed vicinity with the body wall.

In the arthropods, especially insects like cockroach a very special system of gas exchange exist called the tracheal system. Tracheal system is a system of tubes which are extensions of the body wall and makes a network of tubes called tracheae, tracheoles and air sacs in the body. These have openings to the exterior called spiracles. Air enters in through spiracles and enters the trachea and tracheoles, which are present close to the tissues. Gas exchange is easier because in trachea only air is present, which is a very

lighter medium. Tracheal system is a very efficient system of respiration.

**Respiration in fish, frog and birds**

**Fishes**

Fishes are aquatic animals; these have respiratory organs called gills. Water enters through mouth into the gills where gas exchange occurs and then water move out from gills. Heart is fishes is called a single circuit heart, which pumps blood towards gills where gas exchange occurs and then blood moves towards body.

**Frogs**

Frogs are called a transition between aquatic and land animals. These live in water and land both. These have three different types of respiratory system at different stages of life. When a frog is hatched for an egg and becomes a larva, it lives in water and respires through gills. When it becomes an adult frog it respires by lungs and skin. Frog's skin has a network of blood capillaries so that gas exchange may occur there.

**Birds**

Birds have a very efficient system of respiration. Birds respire through lungs. They also have air sacs in their in their body and bones, which also helps in capturing air and hence efficiency of gas exchange is improved.

**Respiratory organs**

Land animals have their respiratory organs named lungs. In aquatic animals, gills are the organs for gas exchange.

**Topic No 92**

**Respiratory system in humans**

Respiration in human beings occurs by lungs and associated air passageways. Human respiratory system consists of:

Air passageways in humans

- Nostril and nasal cavity
- Pharynx
- Larynx
- Trachea
- Bronchi
- Bronchioles

Lungs in humans

- Pleural cavity
- Body of lungs
- Alveoli – the smallest units

**Structure and functions of various parts of air passageways**

**Nose and nasal cavity**

Nose is the part for inhalation. Nose and nasal cavity are lined with ciliated epithelium and its surface is covered with mucus. The ciliated and moist surface traps dirt and other particles. The air, which enters inside is filtered, moist and warm.

**Pharynx**

It is a small muscular passage, which is lined with mucous membrane. Air moves down to larynx through pharynx.

**Larynx: the voice box**

Larynx is a complex cartilaginous structure, through which air moves down to trachea. Its opening is ciliated and covered with mucous. Mucus membrane is stretched across into thin edged fibrous bands called vocal cords. Vocal cords help in voice production.

**Trachea (windpipe)**

Trachea is a long tube stretched from larynx to lungs. It has a tubular structure, and it lies ventral to the esphagous. It has C-shaped cartilaginous rings which are supporting structure and prevent tracheal collapse. Trachea extends through the chest cavity or thorax. In thorax, it divides into 2 tubes called bronchi one entering each lung.

**Bronchi, bronchioles and alveoli**

Trachea divides to form bronchi in thorax region. Bronchi enter the lungs and divide further to make bronchioles (when they attain a diameter of 1mm or less). Bronchi have same cartilaginous rings but these become irregularly distributed plates when reaches at the end of bronchioles.

**Alveoli**

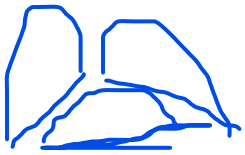
Brochioles keep dividing further and enter deep into the lungs to give rise to air sacs. Air sac is the functional unit of lungs. Air sacs consist of several microscopic structures called alveoli. Alveoli and air sacs are covered by a network of capillaries. This network is the site of gas exchange.

**Topic No 93**

**Structure of lungs**

There is a pair of lungs, one is right and other is left. These are present in chest cavity. Lungs are protected by ribs and pleural cavity (double membrane fluid filled cavity). Lungs are spongy in structure because of the presence of millions of alveoli. Below lungs

a muscular floor of the chest is present, which is called diaphragm.



**Breathing**

Breathing is the process of inhalation and exhalation. Lungs are spongy, cannot pull or push air themselves. Breathing movements occurs with the help of pressure and movements of diaphragm. Diaphragm is dome-like structure present as the floor of chest cavity. When it is contracted, it becomes less dome-like, which results in expansion of chest and inhalation of air. When it is relaxed then it becomes more dome-like and pressure in the chest is decreased, which results in exhalation of air. When muscles between the ribs contract, the rib cage is elevated; and when relax, ribs settle down. This movement also helps in the process of breathing.

**Exchange of gases in alveoli**

Gases are exchanged against a pressure difference in the blood and lungs (alveoli). Blood capillaries are distributed around alveoli in very thin layers. Blood cells and alveolar air are very close for exchange of gases. Gases are exchanged at the alveoli. Hemoglobin (Hb) in blood cells help in exchange of oxygen against the concentration difference.

$Hb + O_2$ $HbO_2$ (reversible if pressure of gases in air is changed)

**Transport of $CO_2$**

Carbon dioxide is more soluble and dissolves in tissue fluids. From here it enters into blood capillaries. Carbon dioxide is transported in blood as:

- 20 % as carboxyhemoglobin
- 5% by few plasma proteins
- About 70 % as bicarbonate ions combined with sodium.
- $CO_2 + H_2O$ –Carbonic anhydrase- $H_2CO_3$

- $H_2CO_3$ $H^+ + HCO_3^-$

**Topic No 94**

**Factors affecting gas exchange**

- Carbon dioxide

  If CO2 increases, then O2 tension decreases. The result is that capacity of hemoglobin for O2 is reduced.

- Temperature

  Rise in temperature decreases the oxygen carrying capacity of hemoglobin.

- pH

  If pH declines, then oxygen carrying capacity declines.

- H+ ions bind to hemoglobin to decrease oxygen carrying capacity.

  Lung capacity and effect of exercise on breathing rate

In humans, fully inflated lungs have a capacity of 5 liters. Normally, at rest or sleep exchange is half a liter. During exercise, it may increase up to 3.5 liters. It means that there is a 1.5 L residual volume that cannot be expelled. Inhalation per minute is 15-20 times; during exercise it may be up to 30 times to fulfill the needs of exercising muscles.

**Topic No 95**

**EXCRETORY SYSTEM**

**Introduction and need of excretion**

Living organisms have to metabolize which consist of anabolism and catabolism. Anabolism consists of all reactions for formation of compounds. Catabolism is break

down of various compounds including glucose, proteins and lipids for the formation of energy.

Catabolism of carbohydrates, proteins and lipids

carbohydrates + oxygen $CO_2 + H_2O$ + energy

proteins + oxygen $CO_2 + H_2O + NH_3$ + energy

lipids + oxygen $CO_2 + H_2O$ + energy

**Metabolic wastes**

We can see in the reactions above that metabolic waste products are carbon dioxide, water and ammonia. These are waste and surplus materials. These have to be removed. Ammonia is highly toxic for the tissues. Water and CO2 are dangerous for the body if these are present in excess. Rise in CO2 may result in decrease in pH in blood which is dangerous. Ammonia is highly toxic and has to be converted into less toxic form. But those forms also need to be removed. Changes in water concentration in tissue fluids also create problems. Higher concentration may cause dropsy, which is accumulation of water in tissues. Decrease in water concentration may lead to dehydration. For all of these reasons an excretory system is required.

**Excretion in Vertebrates**

**Major waste products in vertebrates**

- Carbon dioxide
- Mineral salts
- Urea
- Creatinine
- Uric acid
- Excess water

**Excretion in animals**

- 
  - 
    - Unicellular organisms
    - Small animals
    - Large animals
    - Adaptations to habitat

      water - fresh and brackish e.g. marine

      desert

      terrestrial

**topic No 96**

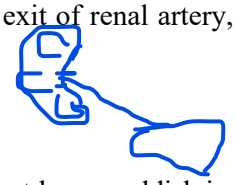
**Excretory Organs**

- 
  - 
    - Skin acts as excretory structure
    - Salt glands are present in some marine or brackish water inhabiting organisms
    - Intestine sometimes act as excretory organ
    - Kidney
  - Major organ for excretion in vertebrates
  - Help also in osmoregulation

Structure of kidney is different in organisms living in fresh and marine waters. Kidneys in freshwater organisms is modified to produce dilute urine while kidneys of marine or brackish water inhabiting animals are designed to produce concentrated urine.

**External and internal structure of kidney**

Kidney is a bean shaped structure. A pair of kidneys is present in abdominal cavity attached to dorsal body wall. The concave part of kidney lies towards vertebral column. There is a depression present towards the vertebral column which is called hilus. The concave part provides space for entrance and exit of renal artery, vein and nerves. A thin tube ureter arises out of the concave part of kidney.



**Renal cortex and renal medulla**

If we look at a section of a kidney, then the outermost layer, reddish in color is called cortex. After cortex the second layer consists of structure like pyramids occur. This region is called medulla. A funnel shaped cavity is present towards inside that receives urine called renal pelvis.

**Nephron**

Nephron is the functional unit of kidney. Kidneys have many small structures present in

cortex and medulla called nephrons. Function of the nephron is to produce urine.

Nephron consists of following structures:

- Glomerulus, which is a capillary network.
- Bowman's capsule (Glumerular capsule) is the capsule like structure in which the glumerulus is present.
- Convoluted tubule
- Loop of Henle

Collecting duct

**Topic No 97**

**Functioning of the kidney: Urine formation**

Function of the kidney is to produce urine. Kidney produces urine by following

mechanism.

**Pressure Filtration**

- Pressure filtration in the renal corpuscle occurs due to blood pressure in renal artery. Molecules other than RBCs and plasma are filtered, i.e., these enter into the renal capsule.
- It is called Bowman's filtrate
- It consists of salts, glucose, urea and uric acid

**Reabsorption**

The filtrate moves down the tubular system. Next step is called reabsorption. Reabsorption means that those compounds which are useful for the body return back to the blood and the remaining part of filtrate in the tubules makes urine consist of water and nitrogenous wastes. Reabsorption occurs through the network of capillaries surrounding the tubules. Reabsorption is selective process. Glucose, water and salts are reabsorbed.

In the last step, only nitrogenous wastes remain inside the urine. This urine now goes down to pelvis and uretes. Through ureters it enters the urinary bladder for temporary storage. When bladder is full, there is a reflex for urination through urethra. Urination is not under conscious control in infants. In adults, however, this is under the conscious control through a sphincter present on the junction of urinary bladder and urethera.

**Topic No 98**

**Osmoregulation**

Kidneys help in osmoregulation. This also helps in the maintenance of blood pressure. In case of increased blood pressure dilute urine is produced to temporarily reduce the quantity of water. In case of decreased blood pressure concentrated urine is produced and water is reabsorbed to compensate for fluid loss like in case of too much perspiration. Thirst is also a reflex in response to decreased quantity of water in blood.

**Topic No 99**

**Ecology**

Ecology is the study of interactions among organisms and their environment.

**Environment**

The physical surroundings of an organism including air, water, soil etc. Organisms have

to interact with environment. Environment provides organisms everything required to live. Environment is always changing, may be supportive or hostile. Organisms living in an environment are "best fit" for that particular environment.

Environment consist of biotic and abiotic factors

- Biotic factors

  Animals, plants, fungi, algae, bacteria

- Abiotic factors

  Air, water, soil

What is Ecology?

- The study of interactions between organisms with each other and their environment.
  - 
  - Biosphere and environment.

    Surface of Earth's crust

    Water bodies on Earth's surface

- Atmosphere that surrounds Earth

**Topic No 100**

**Levels of Organization in an Ecosystem**

- 
  - 
    - Species
    - Population
    - Community
    - Ecosystem

Then comes:

- 
  - 
    - Biome
    - Biosphere

**Ecosystem**

Ecosystem is the area where organisms live in interaction with non-living factors.

**Organisms and their environment**

Organisms live in specific locations or areas suitable for them. Habitat is the term used for physical location of organism. Another term is niche. Niche is defined as ecological role of organisms. Organisms interact with their surroundings; their interaction is termed as niche.

**Biotic and Abiotic Components**

- Living / Biotic Components of an Environment

  Animals, plants; Interactions between organisms.

- Non-living / Abiotic Components of an Environment:

  Air, water; Interactions between organisms and their environment.

**Energy Flow in Ecology**

Main source of energy in an ecosystem is "Sunlight". Autotrophs (producers) are the organisms which utilize the energy stored in inorganic compounds and use sunlight to make their food. These organisms carry out "photosynthesis" and use this energy to convert "carbon dioxide and water into oxygen and glucose". The other type of autotrophs makes carbohydrates using chemical energy e.g., many bacteria. These are called chemoautotroph. Heterotrophs (consumers) are the organisms that cannot make their own food and depends upon organic source.

- 
  - Herbivores (the organisms that eat plants, i.e., producers; also called primary consumers)
  - Carnivores (the organisms that eat herbivores or other carnivore animals)
  - Omnivores (the organisms that eat upon plants and animals both)

**Pyramids in Ecology**

- 
  - 
    - Decomposers (these are the organisms which break down the dead organic matter)
    - Consumers (the organisms which feed upon producers or other consumers)
    - Producers (the organisms which makes their own food by either using sunlight or inorganic chemical)

**Topic No 101**

**Factors that Affect Environment**

- Air currents
- Temperature
- Soil
- Water
- Light

**Human interference**

**Industrial effluents**

Industrial growth results in the production of more and more effluents. Industries produce effluents which may be solid or liquid. These may contain heavy metals, toxic chemicals.

These are disastrous for the biodiversity. These should be treated well. The solution is treatment plants for removal of dangerous materials.

**Deforestation**

Forests are cut down for wood; wood may be used in buildings, furniture etc. Forests are also cut for making towns. This result in destruction of habitats of organisms which may leads to extinction of many organisms. As organisms are related, results may be increase or decrease of number of other organisms, e.g., if sparrows are decreased then insect pests may increase and destroy crops in that area.

**Urban spread**

As populations are increasing there is an increasing need of towns and cities. Urban spread requires the cutting of forests or other type of habitat destruction.

**Construction of dams, waterways**

Dams and water ways etc are constructed on water bodies. These may result in habitat destruction of fish and other aquatic animals. Loss of biodiversity results in instability of ecosystem.

**Agriculture**

Agriculture also affect environment. In grasslands keeping too many animals may result into loss of grass and destruction of ecosystem. In crop growing fields fertilizer use and growing exhaustive crops (the crops that uses much of soil's resources) may result in damage to the ecosystem.

**Natural factors**

Natural disasters like floods, earthquakes etc. may lead towards total or partial habitat destruction. Both biotic and abiotic factors are affected. The whole structure of ecosystem may change.

**Topic No 102**

**Ecology II – Ecosystem**

**Ecosystems - An Introduction**

Ecosystem is an area where the organisms are living in an area in interactions with each other and their physical surroundings. There are many types of ecosystems, mainly divided into two categories:

- Aquatic ecosystems (ecosystem in water)
- Terrestrial ecosystems (ecosystem on land)

**Basic components of an ecosystem; and their roles / significance**

**Ecosystem consists of:**

- Abiotic factors

Air, water, soil

Temperature, pH, humidity

- Biotic factors

Animals, plants, protists, bacteria

**Biotic components**

- Producers

Plants, algae

- Consumers

Animals

- Decomposers

Fungi, bacteria

**Topic No 103**

**Pyramids in Ecosystems**

Ecological relationships are also shown in the form of pyramids in ecology. These are

called ecological pyramids or trophic levels. On the base come producers, then consumers and then decomposers.

**Producers**

The living organisms which make their own food are called producers. Examples include plants, bacteria and algae. They carry out photosynthesis, carbon dioxide and sunlight to water and oxygen. They synthesize carbohydrate by a process called Calvin's cycle. Entry point of energy in the ecosystem is producers.

**Consumers**

These are the organisms that uses organic carbon source. These use carbohydrates produced by plants. Types include primary, secondary and tertiary consumers. Consumers include herbivores, carnivores and omnivores.

**Decomposers**

These organisms feed upon dead organic matter. These include fungi and bacteria.

**Abiotic components of Ecosystem**

**Water, Light, Air and Soil**

Water is a limiting factor in an ecosystem. It is required by living organisms.

Organisms have to adapt according to the availability of water.

• Sun light is the primary source of energy, e.g. zones of a lake.

• Air manages temperature and other things.

• Soil supports life

**Topic No 104**

**ECOSYSTEMS – EXAMPLES**

Climate and weather affect the ecosystem. There are various ecosystems in the world and in Pakistan according to the climate of the area.

**Aquatic Ecosystems**

Water is a liquid medium to support life. Aquatic ecosystems may be of fresh water or marine; a lake or a sea.

**Water has some properties**

- It changes its temperature slowly, more appropriate for life.
- It absorbs considerable amount of energy (sunlight) but at depths its level
- decreases for photosynthesis.
- Nutrients are concentrated at bottom.
- Water is abundantly available in this ecosystem.

**Fresh water lakes**

- 
    - 
        - Lakes vary in nutrient, physical conditions and depths.
        - Life also varies according to the conditions.
        - Lakes have three zones:

Littoral – shallow water, photosynthesis occur, most diverse, anchored plants, submerged plants, phyto and zooplankton and fishes.

Limnetic – upper layer of deep water, have good light penetration, photosynthesis occur, cynobacteria – protozoans – crustaceans and fishes.

Profundal – bottom layer of deep part, almost no light for photosynthesis, high nutrients on bottom, bacteria – decomposers.

**Human interference**

Wastes comes from different areas and results in eutrophication. Excessive growth of cyanobacteria create a scum on the surface; result is that plants and animals die. Decomposer bacteria further decompose result in more organic matter. Ultimately habitat destruction is the result.

**Terrestrial Ecosystems**

Plants and animals adapt to the changes from water to land habitat. Supporting tissue developed for the purpose; animals have skeleton and plants have vascular bundles to support these organisms on land. Water conservation is another adaptation to conserve water.

**Types of Terrestrial Ecosystems**

Divided into four main types:

- Forest ecosystems

    - Tropical rain forests
    - Temperate deciduous forests
    - Coniferous alpine and boreal forests

- Grassland ecosystems
- Desert ecosystems
- Tundra ecosystems

**Topic No 105**

**Major ecosystems in Pakistan**

- 
    - 
        - Pakistan has a variety of climate and seasons.
        - Have these main ecosystems:
    - Temperate deciduous forests – Shogran and Neelum valley
    - Coniferous alpine and boreal forests – Kaghan, Malam Jabba, Dir and Chillas
    - Grassland ecosystems – Gilgit, Kashmir, Waziristan, Chitral and Kallat
    - Desert ecosystem – Mianwali, Bahawal Nagaar, Bahawalpur and more, Thal, Thar and Cholistan
    - Tundra ecosystem – mountains of Karakoram and Hindukash

Temperate deciduous forest

- 
    - 
        - Present in Southeast Asia, India, North America, China, Japan etc.
        - Have moderate temperature – 4 – 30$^{o}$C
        - Rain fall 750 to 1500 mm
        - Plants – Taxus, Pinus, Berberis, ferns, grasses, herbacious; shed their leaves in dry season
        - Animals – Rehesus monkeys, leopard, black bears
        - Soil - very fertile and rich in nutrients
        - Humans hunt animals and cut wood

**Coniferous alpine and boreal forest**

- 
    - 
        - Eurasia, North America, Canada
        - Low temperature – freezing to 10C
        - Snow cover is present
        - Harsh climate – less suitable for life
        - Highly adapted species lives here, Marco polo sheep, bisons, wolf, black bear
        - Plants – Pinus species grow; have long waxy needle like leaves to survive cold
        - Human disturbs less because less accessible

**Grassland ecosystem**

- 
    - 
        - Pakistan, Eurasian countries, North America
        - Two types – Prairies, Savana
        - Rain fall 250-750 mm, water is a crucial factor
        - Plants are grasses – tall and short, legumes, herbs, mosses, lichens
        - Animals are reptiles, amphibians, mammals
        - Decomposers are fungi, bacteria
        - Human impact – agriculture and livestock

**Desert ecosystem**

- 
    - 
        - Rain fall – 25-50 mm
        - Perennial plants, cacti, succulent leaves and stems
        - Animals adapt to little water, kangroo rats, reptiles, birds
        - Human impact – desertification

**Tundra ecosystem**

- 
    - 
        - Very cold, snowy
        - Small perennial flowers
        - Mosquitoes grow well, birds feed upon these (geese, ducks)
        - White bears, foxes, snow owls are present
        - Human interference can produce long lasting effects because plants grow slowly but this interference is low due to harsh weather

## Topic No 106

Evolution is the branch of biology which deals with the study of change in the heritable characteristics of biological populations over successive generations. These characteristics are the expressions of gene, which are passed on from parents to offsprings during reproduction. Variations come from evolution and cause biodiversity. The history of evolution can be divided into 5 stages, classical, medieval, pre-darwinian, darwininan revolution and modern synthesis. In the classical era it was believed that all living things were related and that they had changed over time. One type of organism could descend from another type. Lucretus is one of the dominant philosophers of the classical era. In medieval period the concepts evolved from classical ideas, and it was believed that life forms were fixed. This concept is called fixed natural possibilities and states that all the life forms were fixed since the beginning of the time and would remain the same till the end. Aristotle added the concept that new types of living things could come to be.

Pre-darwinian period began in 17th century when philosopher and scientist began to explain natural phenomena in terms of physical laws. John Ray introduced the term 'species'. In 1735 Linnaeus explicitly recognized the hierarchical nature of species relationships and introduced the five-kingdom classification system. Buffon suggested that species could degenerate into different organisms. Erasmus Darwin proposed that all warm-blooded animals could have descended from a single microorganism. In 1809 Jean-Baptiste Lamarck presented very first evolutionary scheme. He proposed the theory of changes/mutations over the time. It was a break from the concept of fixed creation, but his theory faced strong resistance.

Darwinian revolution is the next era in which Malthus's (1798) concepts about population growth and resultant struggle for resources. Seeking his inspiration from Malthus, Charles Darwin proposed the groundbreaking Theory of natural selection in 1859. Darwin's ideas were applied to humans by Huxley using paleontology and comparative anatomy. Gregor Mendel proposed the laws of genetics. Weismann for the first time made the important distinction between germ cells and somatic cells. He demonstrated that heredity passes through the germ line only. De Vries proposed that hereditable material is in the nucleus and transferred to cytoplasm of a cell.

The modern synthesis is the current age of evolution it connects natural selection and population genetics based on Mendelian inheritance, into a unified theory that applied generally to any branch of biology. Watson and Crick presented the structure of DNA.

The study of evolution requires evidence based on biogeography, paleontology, comparative anatomy, and molecular biology.

In this lesson the students will learn about

- Evolution
- Eras of evolution

## Topic No 107

Based on the observations and the collected data, Darwin proposed his theory of natural selection in the form of 'Origin of Species' in 1859. This theory has four major postulates and each one describes the driving force behind the evolution.

Competition; all organisms have a far greater reproductive potential than is ever realized. Female sea star releases about 1 million eggs each season, if all of them are fertilized and developed to reproductive adults by following year than it could result in a significant pressure on the resources on the habitat and can affect the sustainability of the ecosystem. The limited resources result in intraspecific and interspecific competitions between the individuals.

In this lesson the students will learn about

- Competition as the driving force behind evolution

## Topic No 108

Individuals belonging to same population exhibit variations as the two individuals can't be same. Individuals vary in traits directly related to their ability to survive and reproduce. Inherited variations arise by random mutations, which can be ether beneficial or disadvantageous. These mutations could also be neither harmful nor helpful for the survival of the individual. Variations can be passed on to offspring. Individuals with beneficial or helpful variations can outperform the individuals lacking that variation. This results in increase of the population of the advantageous individuals.

Mendel suggested that genes are responsible for traits, and these are inherited to offspring by transfer of genes. The phenomena of independent assortment and crossing over bring variations. Mendel's work confirmed that second postulate of the Darwin's theory.

In this lesson the students will learn about

- Variations as the driving force behind evolution

**Topic No 109**

Evolution as a result of competition and variation, leads to the individuals which are more competitive and capable of surviving at better scale in their habitat and niche. These individuals evolve in response limited resources and competition (both inter- & intra- specific). These individuals have higher rates of population growth and would increase their population. Reproduction provides variations and the traits which promote successful reproduction are said to be adaptive. This is the third postulate of the Darwin's theory, the survival of the fittest. Only those individuals/species will survive which have advantageous traits thus making them nature's favorite in evolution.

The last postulate of the theory of natural selection is the transfer of adaptive traits to next generation. Once an individual acquired any beneficial trait via reproduction, it will not only help the individual to survive but will also be transferred to the next generation. On the other hand, individuals with maladaptive traits are less competitive and are less likely to reproduce. With each generation, the population of individuals with maladaptive traits decrease and is eliminated eventually. This process of selection of individuals/traits is called natural selection. This selection is triggered by multiple factors affecting the environment (such as change in climate, food/water shortage, other stresses etc.), and will lead to extinction of ill-adapted species.

Alfred Russel Wallace is considered as the co-discoverer of the theory of natural selection. He collected data and specimens from Amazon and Malay Archipelago. Wallace independently proposed that populations out pressure on the resources of the habitat (competition) and variations arise in response to environmental changes. His theory has certain differences with the Darwin's natural selection. Wallace argued that each new variation arising on new generations is the result of adaptive radiations and are not necessarily caused by the changes in the environment. However, Darwin did not insist on finding adaptive significance for every modification.

In this lesson the students will learn about

- Survival of the fittest as the driving force behind evolution
- Natural selection as the driving force behind evolution
- Alfred Russel Wallace's theory of evolution

**Topic No 110**

**FOOD CHAIN**

**Ecosystem**

An area where the organisms are living in interactions with each other and their physical

surroundings is called an ecosystem. There are many types of ecosystems, mainly divided

into two categories:

- Aquatic ecosystems (water ecosystems)
- Terrestrial ecosystems (land ecosystems)

**Ecosystem**

Ecosystem consist of biotic (living) and abiotic (non-living) components.

- Biotic factors:

Producers, Consumers, decomposers

Plants, animals, fungi / bacteria

Biotic factors are in relationship with each other in an ecosystem. These also interact with

abiotic factors present in their surroundings like air, water, sunlight.

**Trophic levels**

Trophic levels or feeding levels in an ecosystem define roles of the groups of organisms.

- Producers

Plants, algae

Consumers

Animals

- Decomposers

Fungi, bacteria

**Topic No 111**

**Autotrophs – Producers**

These are the organisms that make their own food using sunlight as an energy source. These convert carbon dioxide and water into carbohydrates. Producers are the only source for all organic food in the planet Earth; this is the entry point of energy. These provide food to all other life forms. Examples include plants, algae.

**Chemotrophs**

These are the organisms, which uses inorganic chemicals as energy source and makes carbohydrates. These lives in deep oceans, where there is no light source. Examples are some bacteria and deep-sea worms.

**Heterotrophs – Consumers**

Organisms that do not make their own food and get food from other organisms are called consumers. They get energy by eating either plants or other animals. Their source of food hence is organic compounds produced by producers. Examples are small and large animals.

**Types of Consumers**

Consumers are divided into three classes:

- Primary consumers – herbivores (plant eaters)

e.g., goats, cows

- Secondary consumers – primary carnivores (meat eaters that eat herbivores)

e.g., frogs, snakes

- Tertiary consumers – secondary carnivores (meat eaters that eat other meat eaters)

e.g., eagle, hawk

**Decomposers**

These are the organisms that eat dead organic matter. These feed upon animals and plants – their fallen parts or dead organisms. These recycle food in the ecosystem, convert non available organic matter into available matter for producers.

**Topic No 112**

**Food chains**

**What is a food chain?**

Food chain is a one-way flow of energy in an ecosystem.

Producers to consumers to decomposers

In ecosystem different food chains exist. This is a relationship of eating and being eaten.

**Phytoplankton □ Fish Herbivore □ Fish Carnivore □ Fish Eater Bird**

**Example of a food chain from land (grassland) ecosystem**

Grassland ecosystem has following characteristics:

- Rain falls 250-750 mm, water is a crucial factor
- Plants are grasses – tall and short, legumes, herbs, mosses, lichens
- Animals are reptiles, amphibians, mammals
- Decomposers are fungi, bacteria

Sunlight □ Grass □ Primary Consumer (Rat/Mole) □ Secondary Consumer (Snake) □ Tertiary Consumer (Hawk)

**Example of a food chain from aquatic ecosystem**

- Aquatic ecosystem is marked by:

Water, which is a liquid medium to support life.

May be fresh water or marine, a lake or a sea.

- Light, temperature penetration is important for biodiversity

**A Simple Food Chain in a Lake**

Phytoplankton  Crustaceans  Fishes  Fishes

**Arctic Ecosystem**

Algae  Shrimp  Arctic cod (fish)  Seal  Polar Bear

**Food web**

What is a food web?

- 
  - 
    - A network of eating and being eaten.
    - Feeding relationships between organisms are not as simple as food chains.
    - There is a diversity of organisms, and one organism may be eaten by more than one other organisms.

Example is grass is eaten by cows, buffaloes

**Topic No 113**

**Example of a food web from land (grassland) ecosystem**

- Grassland ecosystem have small plants, reptiles, amphibians and mammals.
- Grass is eaten by most of the herbivores.
- Herbivores are eaten by more than one carnivores.

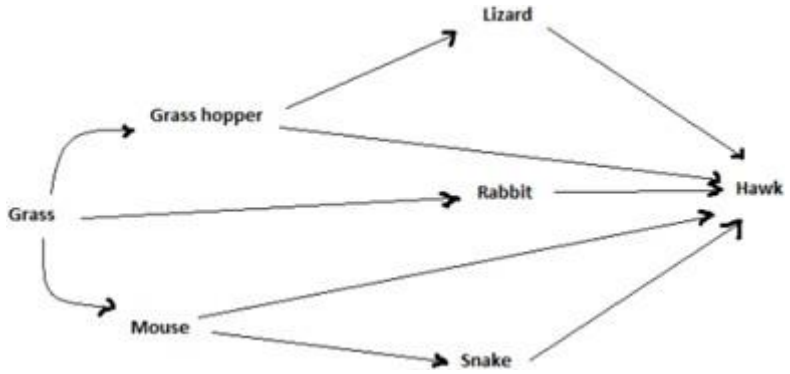



*Figure 113.1 A food web of*

**Example of a food web from aquatic ecosystem**

Aquatic ecosystem has phytoplanktons, zooplanktons, plants, animals and decomposers.

Food webs may be simple or complex starting from producers.

**Desert Ecosystem**

- Dry and hot environment.
- Few specific plants.
- Well adapted animals.

**Forest Ecosystem**

- Dense vegetation, animal diversity is high
- Simple to complex food webs

**Topic No 114**

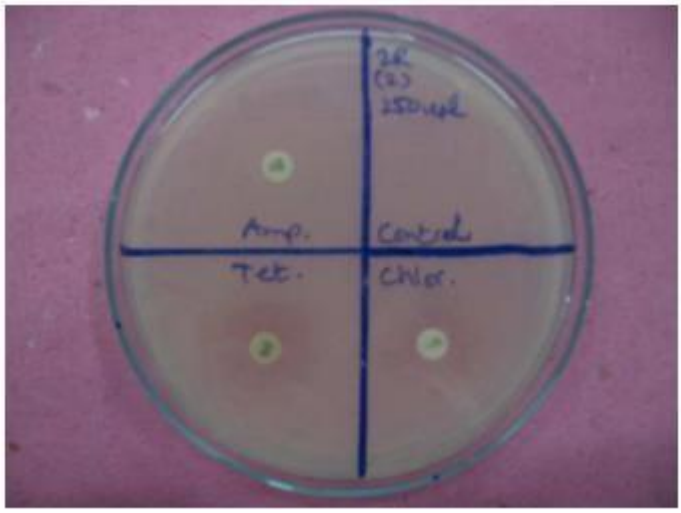
~~**Topic No 102**~~

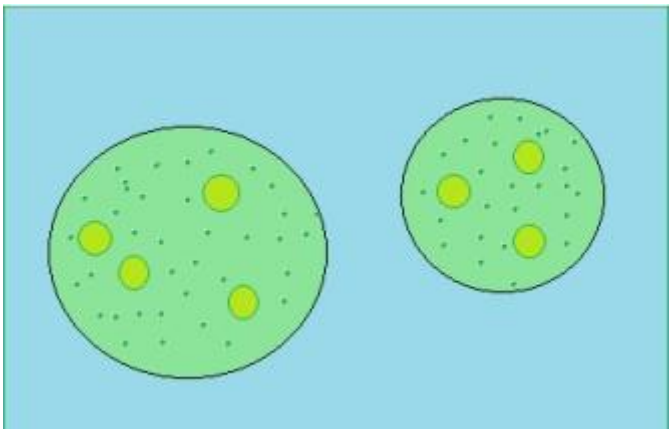
**ALGAE AND FUNGI**

Algae are:

The organisms' range in size from microscopic unicellular to large sea weeds, demonstrate following characteristics:

- Major producers of oxygen
- Provide food for animals in various food chains
- Source of chemicals with pharmaceutical value
- Utilize carbon dioxide to reduce the greenhouse effect
- Source of valuable materials including biofuels, food and agar (part of media for microbial growth)
- Thrived for 1.5 billion years, are an integral part of the global ecosystem



**Distinguishing characteristics of algae, with examples**

- Photosynthetic, autotrophic
- Eukaryotic, have cell wall consist of cellulose
- Classified by their:

  - energy storage products
  - cell walls
  - colour (due to types and abundance of pigments their plastids)
  - cellular organization
  - Reproduce both asexually and sexually
  - Asexually by mitosis
  - Sexually when there is environmental stress
  - Plus and minus gametes fuse to form zygospore which give rise to new organism e.g. Chlamydomonas
  - Some reproduce by conjugation like spirogyra

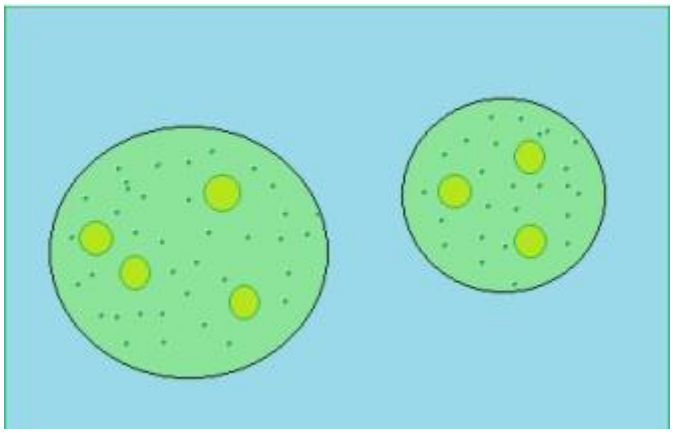


**Figure 114.1 Volvox is algae consists of rounded colonies**

**Topic No 115**

**Types of Algae based upon the pigments**

- Green algae (chlorophyll, soluble in organic solvents only, such as alcohol), 7000 species
- Red algae (have "phycobilins" water soluble pigment) 4000 species, cosmeics, gelatin etc
- Brown algae 1500 species, weeds
- Golden brown algae (formation of petroleum products)
- Carotenoids are also algal pigments soluble in organic solvents
- Euglenoids, both plant and animal like characteristics

**Algae could be distinguished by cellular organization:**

- Unicellular algae: species occur as single, unattached cells that may or may not be motile. For example, Chlamydomonas, Euglena.

- Filamentous algal species occur as chains of cells attached end to end. These filaments may be few to many cells long and may be unbranched or branched in various patterns. For example, Spirogyra.

- Colonial algae occur as groups of cells attached to each other in a nonfilamentous manner. For example, a colony may include several cells adhering to each other as a sphere, flat sheet, or other three-dimensional shape. For example, Volvox.

**Distribution and Significance of algae**

Algae are widely distributed in various areas of the world in small to large water bodies. These are present in small ponds to huge oceans. Green algae are found almost everywhere. Red algae and brown algae are mostly marine organisms. Euglenoids are freshwater inhabitants. When algae grow a lot covering large area of water it is called algal blooms.

**Important producers of the planet Earth**

Algae are producers in many food chains particularly in aquatic ecosystems. These convert sunlight into energy and carbon dioxide into carbohydrates. These help in reducing the Greenhouse effect by absorbing carbon dioxide.

**Algae as human diet alternatives**

Agar is used in the place of gelatin to make gels in many foods. Weeds are part of the salads and other materials for making various kinds of salads and desserts. Some are used in making ice creams.

**Production of Agar**

These are also used in production of Agar which is material extracted from sea weeds and help in solidifying bacterial growth media.

- Agar is very important in scientific research
- Used to culture microorganisms on solid surface
- A plentiful and cheap source for growing microorganisms
- Used in almost all microbiology research laboratories, for example in clinical laboratory

**Algal Biofuels**

Algae can produce huge biomass. This biomass could be used to convert into fuels like ethanol. Algae are potential source of biofuels.

**Topic No 116**

**Topic No 109**

**The Fungi**

Fungi are the heterotrophs and major decomposers which feed upon decaying organic matter. These range in size from microscopic to large ones. These include heterotrophs, symbionts and parasites.

**Distinguishing characteristics of fungi, with examples**

- Unicellular to multicellular
- Eukaryotic
- Body is made of long filaments of hyphae (singular: hypha) which form a mycelium (collection of hyphae)
- Have a cell wall, made up of chitin
- Reproduce both asexually and sexually

  - Spores
  - Mating of hyphae

**Some important groups of Fungi**

Phylum Zygomycota = the Bread Molds

e.g. Rhizopus – black bread mold

Phylum Ascomycota = the Sac Fungi

e.g. Yeast

Phylum Basidiomycota = the Club Fungi

e.g. Mushrooms, puffballs, bracket fungi, rusts, smuts

Phylum Deuteromycota = the Fungi Imperfecti

e.g. Penicillium

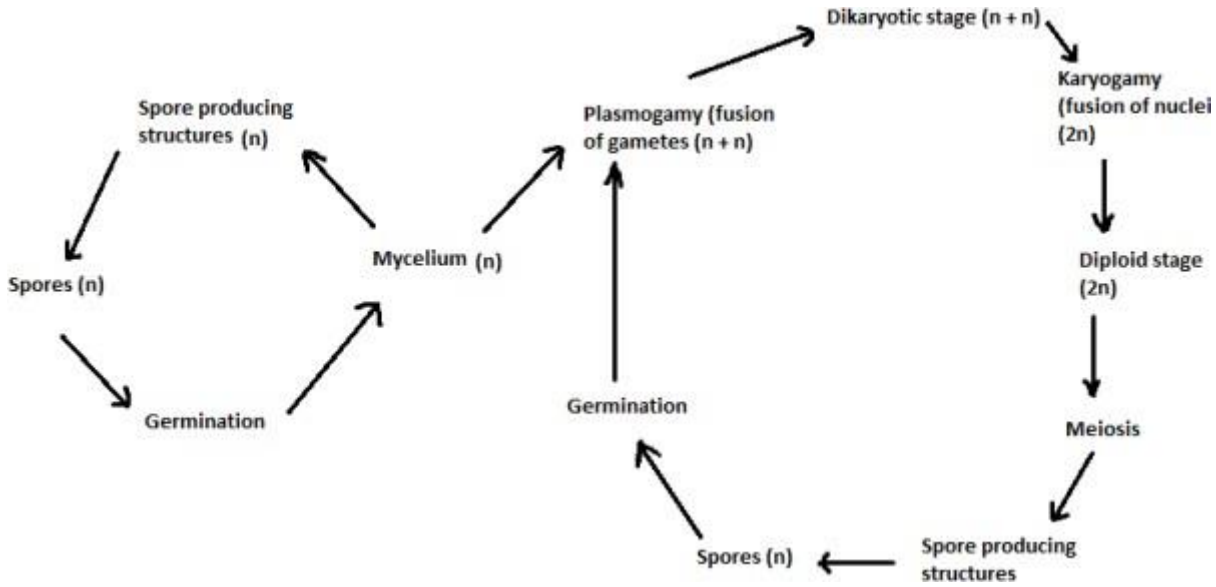



**Figure 116.1 Life cycle of fungi**

**Topic No 117**

**Habitats and Significance of fungi**

Fungi grow in almost every habitat imaginable, as long as there is some type of organic matter present, and the environment is not too extreme. It is a diverse group; number of described species is somewhere between 69,000 to 100,000 (estimated 1.5 million species total). It is present in soil, water, or on foods. These may also live on animal or plant bodies. Mycorrhizae are the fungi associated with the roots of vascular plants. Lichens are the association between fungi and algae.

**Lichens**

Lichens consist of association between algae and fungi. It is a symbiotic association. These are usually the first inhabitants of a new ecosystem.

**Fungi are important reducers / decomposers**

- Feed on dead organic matter
- Convert it to less toxic form
- Convert into useable form for plant's use

**Food source and food spoilage**

Many fungi are source of food and many other spoils foods.

- Mushrooms
  - Various types
  - Poisonous to edible
  - Used as food, protein source
- Some fungi cause spoilage of foods
  - Bread molds

**Sources of medicines**

Some fungi are sources for medicines.

- Truffles
  - Used in medicines
  - Grow inside soil
- Penicillium
  - Source of penicillin
- Other drugs

**Pathogenic fungi causing diseases in plants and humans**

Many fungi are pathogenic and cause infections in animals, plants and human beings. These may cause skin infections, eye infections and others. Parasites can harm human beings and plant infections can damage crops.

**Yeast**

Yeast is a very important organism for biotechnology and food industry. It is used to make various biotechnological products. It is eukaryotic and better in making proteins in comparison to prokaryotes. It is also used in wastewater treatments because it absorbs heavy metals and converts other contaminants into less toxic form.

**Topic No 118**

**ANIMALS AND THEIR IMPORTANCE**

**Distinguishing Characteristics of Animals, with examples**

- Consist of eukaryotic cells
- Multicellular organisms, from simplest sponges to complex animals like mammals
- Highly adapted to their environments
- Widely distributed, present in all types of aquatics to terrestrial ecosystems even in air, e.g., the birds
- Important parts of food chains as consumers of different levels
- Heterotrophic, rely on organic carbon sources
- Independently living or parasitic
- Reproduce asexually or sexually
- Very important for human beings and the ecosystems

**Diversity of animals**

These are widely distributed and are present in almost all habitats. There are two majors groups including invertebrates and vertebrates. Animals are wild if these are living in natural habitat, or these are termed as domestic if these are kept for some benefit or as pets. Animals are kept in captivity or controlled habitats if there is a need in zoological gardens and safari parks.

**Topic No 119**

**Phyla of Invertebrates with examples**

**Porifera**

These are simple animals with one opening as mouth and a body cavity. These are aquatic organisms.

- e.g. Sponges
- Two layered, no specific organ, porous body
- Highly specialized cells

**Coelenterates**

The animals with 3 body layers. These have stinging cells which have a poison to paralyze or kill the prey.

- e.g. Jelly fishes which are marine
- Have simple organs
- Have stings, mouths, tentacles

**The worms**

These are of different kinds including round worms, flat worms and earth worms.

- Regeneration: These have ability to regenerate their lost part.
- Parasites, decomposers
- Grow very long

**Arthropods, widely distributed**

These are one of the most diverse groups of animals. These organisms have an exoskeleton.

- Four major groups

  - Arachnids (spiders)
  - Crustaceans
  - Insects
  - Millipede, centipede

- Have sense organs, are segmented, have exoskeleton

**Molluska**

These organisms are specific to have shells around their soft bodies.

- Soft bodies, hard shells
- Aquatic mostly
- Snails, lobsters
- Some of these make pearls

**Echinoderms**

These are the animals with spiny skins hence called echinoderms (echino-spiny).

- Exclusively marine
- Have a water vascular system for the movement
- Examples are star fish, sea cucumber, brittle star

**Importance of Invertebrates**

- Invertebrates are very important part of lot of food chains hence important for stability of ecosystems

- For human use:

  - Sponges are used widely in sound proofing, washing
  - Worms are important parasites of domestic animals and human beings
  - Insects are pests of many crops, many useful insects like honeybee, lac insect
  - Lobsters makes pearls, cultured for pearls

**These are components of Food Webs**

- Component of aquatic food webs
- Components of terrestrial food webs
- Maintain ecosystem stability, if one becomes extinct then the whole food chain or web may become disturbed.

**Role in Soil Fertility**

- Some invertebrates are decomposers, e.g., Earthworm
- They feed upon dead organic matter and increase soil fertility
- When animals die their bodies become part of soil and decomposed to add nutrients to soil

**Parasitic Invertebrates**

These could harm the humans and also harm our domestic animals and crops. There are many treatments available for these.

- Ticks, mites, lice are ectoparasites
- Worms are endoparasites
- Leeches are ectoparasits

**Topic No 120**

**Major Classes of Vertebrates and their Importance**

Vertebrates are the organisms with a vertebral column precisely with 3 main characteristics at any stage of their life including notochord, nerve cord and pharayngeal slits.

Vertebrates are divided into:

- Fishes
- Amphibians
- Reptiles
- Birds
- Mammals

**Fishes**

- Aquatic animals, occupies fresh water and marine habitats, widely distributed, bony or cartilaginous
- Have gills for gas exchange, 5 chambered single circuit heart
- Have streamlined and slimy body, ectotherms
- Have fins (appendages) and tail
- Cultured in ponds and caught from natural habitats, are important food source of proteins
- Cultured for ornamental purposes
- Examples are Rohu, Carps, sharks, electric rays

**Amphibians**

- Are transition between aquatic and land animals

- Gills, lungs and cutaneous respiration
- 5 chambered double circuit heart, mixing of blood
- Some part of the life is aquatic and some part terrestrial
- Produce large number of eggs, no parental care
- Ectotherms
- Examples are frogs, toads

**Reptiles**

- Of diverse types, widely distributed, aquatic and marine
- Highly adapted for hot dry habitats such as deserts
- Terrestrial animals, ectotherms
- Scaly skin, four chambered double circuit heart
- Lungs for gas exchange
- e.g. lizards, chameleon, turtles, tortoises, crocodiles, snakes, gavials



**Birds**

- Arial mode of life, terrestrial life, fore arms modified into wings
- Endotherms, 4 chambered heart
- Lungs and air sacs for respiration
- Lays fewer eggs, provide parental care
- Bones light weight called hollow bones
- Grain eaters to omnivores
- e.g. parrots, pigeons, peacock, pelican, king fisher

**Mammals**

- Widely distributed, most complex, endotherms
- 4 chambered heart, lungs for gas exchange
- Highly developed brains
- Young ones are born mostly, mother feed them with milk, provide parental care
- Have hairs on their body, provide insulation
- Thick skin with skin sensory receptors
- Heterotrophs, herbivores to omnivores

**Importance of Vertebrates**

These are beneficial

- Important parts of food chain and webs
- Beauty and diversity of nature
- Provides protein foods to human beings, eggs, milk, meat, honey
- Furs and feathers are used for various purposes
- By products of fish industry
- Pearls, clam culture
- Lac insect and silk worm
- Snake venom for medicines

**These are harmful**

- Poisonous animals, e.g. frogs, snakes,
- Parasitic animals, e.g. worms, mites, ticks
- Insect pests of crops

- Other animal pests e.g. rats
- Termites damage woody structures of buildings

**Topic No 122**

## REPRODUCTIVE SYSTEM

### Introduction of reproduction

Reproduction is one of the basic characteristics of life. It is a biological process by which the organisms produce their young ones which are similar to their parents. This ability also allows organisms to adapt itself to the changing environment.

### Types of Reproduction

There are two types of reproduction, asexual and sexual. In asexual reproduction, offspring are produced by simple cell division. No gametes are involved, and offspring have same characteristics as their parents. In sexual reproduction, two organisms are involved in reproduction and offspring have characteristics which are combination of their parents and some new. Gametes are involved in this kind of reproduction. Sexual reproduction has advantage of increase in genetic variability. Gametes are formed by a process called meiosis in which alterations in genetic material takes place. As two organisms are involved, mixing of their characteristics takes place that increase more genetic variability. Genetic variability leads towards more adaptability to the environment.

**Topic No 123**

### Types of asexual reproduction

Asexual reproduction involves no gametes. There are many methods of asexual reproduction in organisms including:

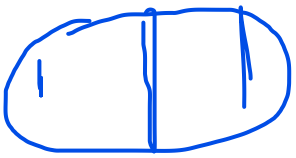
- Binary fission
- Multiple fission
- Budding
- Regeneration
- Vegetative propagation in plants

### Binary fission

This is a type of asexual reproduction in which one unicellular organism divides into two by simple division. In bacteria, for example, binary fission takes place:

1. Replication of single chromosome takes place.

2. Daughter chromosomes move towards opposite poles.



3. Cell membrane invaginates and meets in the center.

4. Cell wall follows the cell membrane and cell divides into two.

Binary fission also takes place in unicellular organisms like amoeba and paramecium. In these organisms:

1. nucleus elongates and divide into two

2. Cytoplasm also divides and cell divides into two

**Multiple fission**

This is a process in which a single cell divides into many cells. For example, amoeba makes a cyst in unfavorable conditions. Its nucleus divides into many nuclei and every part is surrounded by some cytoplasm. When favorable conditions come, the cyst split into many cells, each one a new amoeba.

**Budding**

In this process an outgrowth (called bud) appears on the surface of an animal and may separate later on to develop into a new organism. Examples are yeast, hydra.

**Regeneration**

Regeneration is the formation of lost part of the body. Examples are starfish and planaria. If their one body part is lost then they have ability to grow it again. In star fish an arm is lost then the arm grows again.

**Vegetative propagation in plants**

It is a process that involves the vegetative propagation of some plant part such as stem or leaves. Examples are runners, suckers. Runners like grasses and strawberry and suckers like banana and mint.

**Topic No 124**

**Sexual reproduction in Plants**

Flowering plants reproduce by sexual reproduction. Flowers have male and female parts

called stamen and carpel. Fertilization occurs with the help of a process called

pollination, i.e., transfer of pollen grains from anthers of stamen to carpel and then ovary.

Pollination is of two types: self-pollination and cross pollination.

Formation of seed and fruit

- Seed is formed from the zygote. It consists of embryo and its food.
- When the seed are formed then the wall of ovary becomes fleshy or scaly and forms fruit wall.
- The floral parts (sepals, petals etc.) fall off.
- The ripened ovary is called fruit.

**Topic No 125**

**Sexual reproduction in Animals**

In animals, it occurs through fertilization of gametes (sperms and ova). Gametes are produced by meiosis which is the reduction division. These have half a number of chromosomes. Females and males are different in only one pair of chromosomes. Males have a chromosome different called a Y chromosome in one pair. So, females have an XX pair while males have XY.

**Example of Frogs**

- Frogs carry out sexual reproduction.
- Female frog lays a large amount of ova in water and male release sperms on these.
- Sperms and ova fertilize to produce zygote.
- Zygote the divide by mitosis to develop a larva which lives in water for a specific period of time and then by a process called morphogenesis develops into adult frog.

**Reproductive system in humans**

Humans reproduce sexually by fertilization of gametes called sperms and ova. Gametes are produced by specialized organs called testes and ovaries. Gametes have a haploid number of chromosomes, i.e., 23 chromosomes. 22 of these pairs are alike but 23rd pair is different in males and females. Females have X chromosomes in their gamete while males have an X or Y. Gender of the child is hence determined by the sperm, X or Y. Female always contribute an X in gamete. If male gamete has a Y then a male child will be produced. If sperm have an X then female child will be produced.

**Basic components of male reproductive system**

Human male reproductive system consists of a pair of gonads called testes, and a system

of accessory tubes. (testis: sing.; testes: plural). Testes are paired which produces sperms. Testes lie outside the body in the sac called scrotum. From testes, sperms are transferred

to main duct called vas deferns. Vas deferns makes a complex system of ducts called epididymis. From epididymis, sperms pass through the urinogenital tract and discharged.

**Function of testes**

Function of testes is to produce sperms and the male hormone testosterone. Testis consists of a system of tubules called seminiferous tubules.

**Process of Spermatogenesis**

1. In the seminiferous tubules, germinal epithelium produces the cells called spermatogonia.

2. Spermatogonia divide into primary spermatocytes (2N).

3. Primary spermatocytes undergo meiosis to produce secondary spermatocytes (N).

4. Secondary spermatocytes divide further to make 4 haploid cells called spermatids.

5. Spermatids are converted to sperms by morphological changes

Testis also has some large cells in seminiferous tubules called sertoli cells. These cells

provide support and nourishment to the developing sperms.

**Basic components of female reproductive system**

Female reproductive system consists of a pair of ovaries and an associated system of tubes. A pair of ovaries lies inside the female body cavity which produces ova. Ovaries leads to the tubes called oviducts also called fallopian tubes. Oviduct opens into uterus. Ovum is fertilized in the oviducts then it passes to the uterus where it is implanted. A placenta is established between the uterine and fetal tissues for the exchange of materials. Uterus is connected to the vagina through cervix.

**Function of ovaries**

Function of ovaries is to produce ova and female hormones estrogen and progesterone. Germ cells in ovary produce many cells called oogonia.

**Process of Oogenesis**

1. Oogonia produce primary oocyte (2N) by mitosis.

2. The primary oocyte divides by meiosis into secondary oocyte (N) and polar body.

3. The secondary oocyte divides to form one ovum and a polar body.

4. The first polar body also divides to form 2 polar bodies.

5. Hence from secondary oocyte, one ovum and three polar bodies are produced. Usually, ovary releases one ovum at a time, this is called ovulation. Sometimes it releases more which may result into fraternal (non-identical) twins.

**Topic No 126**

**APPLICATIONS OF BIOLOGY**

**Biology – Useful for Humankind and World**

- Demands and problems are increasing with increase in population.
- Biologists investigate phenomena of nature and problem and find ways to resolve these.
- They introduced techniques and technologies to explore all biological resources to cope demands and resolve problems.

**Biotechnology**

- Biotechnology is the use and manipulation of biological processes, organisms and systems to manufacture products to improve the quality of human life.
- The term biotechnology was first used in 1970's.
- Some important applications

  - Genetic Engineering
  - Fermentation and Microorganisms

**Genetic Engineering**

- It is a process in which a useful gene is taken from one organism and inserted into another organism. The resulting organism is called transgenic.
- This process is carried out with the help of vectors and enzymes.
- Most common vector is bacterial plasmid.
- Most common enzymes are restriction endonucleases or restriction enzymes.

**Topic No 127**

**Transgenic bacteria**

- E. coli – most commons microorganisms in biotechnology because these have many strains (types) which are non-pathogenic, i.e., these cannot produce disease.
- GRAS (Generally regarded as Safe) Organisms are those which are known for their non-pathogenecity.
- Chemical industry: Acids and alcohols are produced using bacteria.
- Pharmaceutical industry

  - Formation of insulin by E. coli for diabetic patients

- Formation of human growth hormone for dwarf children
- Formation of vaccines

**Transgenic Bacteria**

Transgenic bacteria are used in many industries, for example:

- Food industry

- Yogurt, Cheese, Sour creams are prepared using microorganisms
- Breads b use of yeast
- Beverages, drinks by yeasts or some bacteria

- Treatment of waste materials is also carried out using different form of bacteria or even yeast. The microorganisms are used to treat:

- Solid wastes
- Wastewater
- Oil spills
- Nuclear wastes

- Bacteria and algae are also helpful in biofuels production (biogas, biodiesel).

**Transgenic plants**

These plants are produced to develop certain specific properties in these, for example, resistance to viruses. Some examples are following:

- Virus resistant crops (cotton)
- Insect resistant corn
- Genetically modified banana
- Pesticide resistant crops
- FOOD SAFETY AND SECURITY: This is very important in modifying food crops particularly that if we are inserting a gene of resistance or else, the plant should not develop any harmful characteristic. For this purpose, before selling any transgenic seed or other product to market, it is tested for safety. This is called biosafety.

**Transgenic animals**

- Transgenic mice as model of human disease

- Obese rats, diabetic mice

- Genetically modified organisms to increase milk yield
- Genetically modified farm animals to produce antibodies
- FOOD SAFETY AND SECURITY: This is very important in modifying food producing animals particularly that if we are inserting a gene of interest the animal should not develop any harmful characteristic. For this purpose, before selling any transgenic product to market, it is tested for safety. This is called biosafety.

**Fermentation Pathways**

Microorganisms' carryout fermentation, i.e., utilization of anaerobic pathway for production of energy. They produce many useful products for us during these pathways. These products are commercially produced by growing such organism in mass culture in very big vessels called fermenters or bioreactors.

Following products are obtained by this process:

- Biofuels

- Acids and alcohols
- Yogurt, cheese, sour creams
- Bread
- Beer, wine

**Topic No 128**

**Applications of biology in culturing Animal**

**Animals are traditionally cultured for their products**

- Meat from fish, chicken, goats, cows
- Milk from cows, buffaloes
- Eggs from hens, ducks
- Wool from sheep
- Leather industry uses skins
- Honey from Honeybee
- Silk from silkworm

**Common types of animal cultures**

- Aquaculture
- Ornamental fish
- Seri culture (Silkworm)
- Dairy and Poultry
- Apiculture (honeybee)
- Biological pest control

We discuss these one by one briefly.

**Aquaculture**

Culturing animals in water is called aquaculture for example fish farming.

- Fish farming, Prawn culture
- Pearl oyster culture
- Other aquacultures (clams, lobsters etc.)
- Byproducts of fishing industry include many cosmetics and some animal diets.

Biology helps in improvement of fish varieties and also extraction of various products

from these, for example:

- Selection of fishes in a fishpond (herbivores, carnivores)
- Feed for fishes which may increase their production
- Diseases and their treatment: biology help in finding causes of diseases and their treatments.

**Aquaculture for Ornamental Fish**

Ornamental fishes are the beautiful usually small fishes kept in small aquaria in houses or other places like hotels. Biology helps in:

- Feed: which is good for their maintenance
- Diseases: how can we handle diseases
- Requirements for healthy culture in small to large aquaria

**Sericulture**

Culture of silk moth for making silk on mulberry leaves is called sericulture. It is a popular home industry in many parts of the world

Biology helps in improvement of this culture:

- Process of making silk
- Diseases in moth or larvae can result in huge loss
- Biology helps in making new varieties, e.g., wingless variety, varieties which completes 5-6 cycles in a year

**Topic No 129**

**Dairy and poultry**

Biology plays very important roles in dairy and poultry industries. Biology helps in many ways for example:

- Improved breeding techniques and cross breeds (more vigorous and resistant).
- Improved quality and quantity yield of meat, milk, wool and eggs.
- Diagnosis and cure for diseases in farm animals.
- Biological knowledge is also helping in providing balanced food for best yield.

**Apiculture (honeybee)**

Keeping the honeybee for honey production is called Apiculture (Bee keeping).

Biologists explore the phenomena behind Honey production, like knowledge of biology

tells us that:

- Honeybee is a social insect and lives in large colonies
- Queen bee, drones and workers

Biology helps in understanding how this system works and can help in production of

better varieties and improved system for culturing these animals to increase yield of

honey.

Biological pest control

A large number of organisms damage the useful plants or animals and can cause huge

losses. These are called pests, e.g., many insects damage crops.

There are few ways to control pests:

- Primary or cultural control
  - Crop rotation rather than monoculture, i.e., different crops are grown at different seasons.
- Chemical control
  - Various chemicals are used to kill insects but insects may develop resistance and chemicals are dangerous for health

- Biological control means that the pests are controlled or killed by living organisms.

  - This is the most promising method of pest control
  - The natural enemies and predators of the pests are used to kill the pests
  - Lady beetles for citrus fruits
  - Bacillus thuringienesis for cotton

Selection of biological control agent needs care. These should be ecologically safe. For

example if we introduce a sparrow for eating an insect and that sparrow also start eating some plants. This will be even more harmful for crops and environment.

**Advantages of biological pest control**

- They do not pollute the environment.
- They do not disturb the natural ecosystem.
- A more permanent control because pest usually do not develop resistance

**Applications of Biology in agriculture and plant growth**

**Plants are in many ways useful**

- Parts of ecosystems, producers
- Food crops: crops, which makes our food like wheat, rice, vegetables.
- Many useful products like perfumes, medicines, furniture
- Beauty of the nature, ornamental plants

**Biology helps in:**

- Understanding the life cycles and methods of spread
- Better varieties by cross breeding or genetic engineering
- Disease mechanisms and control
- Appropriate soil, fertilizers and water availability

**Topic No 130**

**Science, Ethics & Values**

Science: Latin term “Scio”

Observation and theoretical form

Observation and experiment

**Science:**

- Investigation of the universe by a set of methodologies
- Progress made by scientific methods
- Stepwise, not a single activity, not a value free

**Ethics:**

- Associated with science
- Issues arises from scientific research
- Scientists are trying to do so

**Values:**

- Science has entered into our daily lives
- Proper resource allocation
- Reflects what society at the time deems to be valuable.

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