

Index

Unit & Chapter	Name of the Chapter	Page No.
VI-01	Reproduction in Organisms	02 – 04
VI-02	Sexual Reproduction in Flowering Plants	05 – 12
VI-03	Human Reproduction	13 – 20
VI-04	Reproductive Health	21 – 25
VII-05	Principles of Inheritance and Variation	26 – 37
VII-06	Molecular Basis of Inheritance	38 – 53
VII-07	Evolution	54 – 61
VIII-08	Human Health and Disease	62 – 72
VIII-09	Strategies for Enhancement in Food Production	73 – 80
VIII-10	Microbes in Human Welfare	81 – 84
IX-11	Biotechnology : Principles and Processes	85 – 90
IX-12	Biotechnology and its Applications	91 – 94
X-13	Organisms and Populations	95 – 102
X-14	Ecosystem	103 – 109
X-15	Biodiversity and Conservation	110 – 114
X-16	Environmental Issues	115 – 124

Mr. P. K. Singh (PGT Bio) prshhtmsngh@gmail.com **KV Muzaffarpur (F/S)**

CHAPTER-01

REPRODUCTION IN ORGANISMS

Reproduction:

Defined as a biological process in which an organism gives rise to young ones (offspring) similar to itself.

Asexual reproduction:

- Offsprings produced by single parents.
- Does not involve gamete formation.
- Offsprings are *genetically identical* to their parents.

Methods of asexual reproduction:

- Cell division; eg. Protista and Monera.
- Binary fission; e.g. *Amoeba*, *Paramecium*.
- Budding: e.g. yeast.
- Asexual reproductive structures:
 - **Zoospores:** aquatic fungi, eg. *Chlamydomonas*.
 - **Conidia:** *Penicillium*.
 - **Bud:** *Hydra*
 - **Gemmules:** *sponges*.
- Vegetative propagation units in plant: (Vegetative propagules)
 - Runner, rhizome, sucker, tuber, offset, bulb.

Sexual reproduction:

- Involvement of single or two individual.
- Involves the production of male and female gametes (haploid)
- Gametes are fused to form a diploid zygote (2n).
- Zygote develops into new organism.
- The offsprings are not genetically identical with their parents.

Features of sexual reproduction:

- Period between birth and sexual maturity is called **juvenile phase**. It is known as **vegetative phase** in plant.
- Bamboo species flower only once in their life time generally after 50-100 yr.
- *Strobilanthus kunthiana* (neelakranji) flowers once in 12 years.
- **Oestrus cycle:** cyclical changes during reproduction in non-primate mammal like cows, sheep, rats, deers, dogs, tiger etc.
- **Menstrual cycle:** cyclical changes during reproduction in primate mammals like monkeys, ape, and humans.
- **Seasonal breeders:** reproductive cycle takes place in favourable seasons as in wild animals.

- **Continuous breeders:** reproductively active throughout their reproductive phase.

Pre-fertilization events:

- Process of gamete formation is called **gametogenesis**.
- If two gametes are similar in appearance, they are called **homogametes (isogametes)**.
- If the two gametes produced are morphologically different, they are called **heterogametes**.
- Male gamete is called **antherozoids** or **sperm** and the female gamete is called **ovum** or **egg**.

Sexuality in organism:

- Plant having both male and female sex organ called **homothallic** or **monoecious**.
- Plants having only one sex organ is called **heterothallic** or **dioecious**.
- In flowering plants, the unisexual male flower is **staminate**, i.e. bearing stamens, while the female is **pistillate** or bearing pistils.
- Animal having one type of reproductive system, called **unisexual**.
- Animal having both male and female reproductive system, called **hermaphrodite** or **bisexual**.

Cell division during gamete formation:

- Gametes in all heterogametic species two types namely **male** and **female**.
- Gametes are always haploid irrespective of parent's ploidy.
- A haploid parent produces gametes by **mitotic** division.
- Diploid parent produces gametes by **meiotic** division.
- In diploid organisms specialized cells called **meiocytes** (gamete mother cell) undergo meiosis to produce haploid gametes.

Gamete transfer:

- Male and female gamete must be physically brought together to facilitate fusion called fertilization.
- In most cases male gametes are motile, female gametes are non-motile.
- In case of few fungi and algae, both male and female gametes are motile.
- In most cases water is the medium for gamete transfer.
- Male gametes are produced in several thousand times the number of female gametes produced to compensate the loss during transfer.

Fertilization:

- Fusion of male and female gamete is called **fertilization or syngamy**.
- The female gamete undergoes development to form new organism without fertilization. This phenomenon is called **parthenogenesis**.
- Gametic fusion takes place outside the body i.e. water is called **external fertilization**.

- There must be synchrony of gamete release, large number of gametes released to enhance the chance of fertilization.
- Enable the individual to produce large number of offsprings.
- A major disadvantage is that the offsprings are extremely vulnerable to predators.
- Fertilization takes place inside the body is called **internal fertilization**.

Zygote:

- Formation of zygote after fertilization is universal in all sexually reproducing organisms.
- Zygote is formed usually **in water** in case of **external fertilization**.
- Zygote is formed **inside the body** of the organism in **internal fertilization**.
- Zygote of fungi and algae develops a thick wall that is resistant to desiccation and damage.
- In Organism with haplontic life cycle, the zygote undergoes meiosis to produce haploid spores.

Embryogenesis:

- Development of zygote into an embryo is called **embryogenesis**.
- Zygote undergoes **cell division** (mitosis) and cell **differentiation**.
- Oviparous animal which lays eggs and development takes place inside egg.
- Viviparous animal gives birth to the young. The development takes place inside the body of the female.
- In plants:
 - **Zygote** developed into **embryo**.
 - **Ovule** developed into **seed**
 - **Integument of the ovule** developed into **seed coat**.
 - **Ovary** developed into **fruit**.

Ovary wall developed into **pericarp**.

CHAPTER-02

SEXUAL REPRODUCTION IN FLOWERING PLANTS

Pre fertilization: structure and events:

- Hormonal and structural changes in plants leads to development of flower,
- **Androecium** consists of a whorl of **stamens** represents male sex organ.
- **Gynoecium** represents the female reproductive organ.

Stamen, Microsporangium and Pollen grain:

- Typical **stamen** consists of two parts, long and slender stalk called **filament** and terminal bilobed structure called **anther**.
- Atypical angiosperm anther is **bilobed**.
- Each lobe have two theca i.e. **ditheous**.
- Each anther contains four **microsporangia** located at the corners, two in each lobe.
- Microsporangia become **pollen sacs** and are packed with pollen grains.

Structure of microsporangium:

- Each **microsporangium** surrounded by four wall layers
 - **Epidermis**
 - **Endothecium**
 - **Middle layer.**
 - **Tapetum.**
- The innermost layer is **tapetum** which is multinucleated, with dense cytoplasm; it nourishes the developing pollen grain.
- The centers of each microsporangium contain homogenous cells called **sporogenous tissues**.

Microsporogenesis:

- The process of formation of microspores from pollen mother cell through meiosis is called **microsporogenesis**.
- The sporogenous tissue of microsporangium differentiated into **microspore mother cell** or **pollen mother cell**.
- Each microspore mother cell undergoes meiosis and gives rise to haploid **microspore tetrad**.
- On dehydration microspore tetrad dissociated to form four **microspores**.
- Each microspore developed into a **pollen grain**.

Pollen grain:

- Pollen grain represents the **male gametophytes**.
- It is spherical and measuring about 25-50 micrometer in diameter.
- It is covered by **two layers**.

- The hard outer layer called the **exine** is made up of **sporopollenin**, which is one of the most resistant organic materials known. It can withstand high temperature and strong acids and alkali. No enzyme can degrade sporopollenin is so far known.
- The exine has prominent apertures called **germ pore** where sporopollenin is absent.
- The inner wall of pollen grain is called **intine**. It is thin and continuous layer made of **cellulose** and **pectin**.
- On maturity the pollen grain contains two cells, the **vegetative cell** and **generative cell**.
- The vegetative cell is bigger, has abundant food reserve and a large irregularly shaped nucleus.
- The generative cell is small and floats in the cytoplasm of vegetative cell.
- In 60% of angiosperms, pollen grains are shed at this **2-celled stage**.
- In others the generative cell divides mitotically to form two male gametes before pollen grain are shed (**3-celled stage**).

Economic importance of pollen grain:

- Pollen grain may cause severe allergies and bronchial afflictions.
- It may cause chronic respiratory disorders – asthma, bronchitis, etc.
- Pollen grain of Parthenium or carrot grass causes pollen allergy.
- Pollen grains are rich in nutrients hence used as pollen tablets for food supplements.
- Pollen consumptions increase performance of athletes and race horses.
- After shedding the viability depends on temperature and humidity.
- In wheat and rice the pollen grain lose viability within 30 min. of their release.
- In Rosaceae, Leguminosae and Solanaceae they remain viable for months.
- Pollen grain can be preserved for years in liquid nitrogen (-196 °C).

The Pistil, Megasporangium (ovule) and Embryo Sac:

- The **Gynoecium** represents the female reproductive part of the flower.
- The Gynoecium may contain single pistil (**monocarpellary**) or may have more than one pistil (**multicarpellary**).
- Fused pistils are called **syncarpous** and free pistils are called **apocarpous**.
- Each pistil has three parts the **stigma**, **style** and **ovary**.
- Inside the ovary is the **ovarian cavity (locule)**.
- The **placenta** located inside the ovarian cavity.
- **Megasporangia** or **ovules** arise from the placenta.
- The number of ovule inside the ovary may be **single** or **many**.

The Megasporangium (Ovule):

- Ovule is a small structure attached to the placenta of locule with a stalk called **funicle**.
- The body of the ovule fused with the funicle in the region called **hilum**.

- **Hilum** is the junction between the funicle and ovule.
- Each ovule has one or two protective envelopes called **integuments**.
- Integument covered the ovule except an opening at the top called **micropyle**.
- Opposite of the micropylar end, is the **chalaza**, representing the basal part of the ovule?

Megasporogenesis:

- The process of formation of **megaspores** from the **megaspore mothercell** is called **Megasporogenesis**.
- In the centre of the ovule there is a mass of tissue called **nucellus**.
- Cells of nucellus have abundant reserve food materials.
- One cell of the nucellus towards micropylar end differentiated into **megaspore mother cell (MMC)**.
- It is a large **diploid cell**, dense **cytoplasm** with **prominent nucleus**.
- The MMC undergo **meiotic** division resulting four haploid **megaspores**.

Female gametophyte:

- Out of four megaspores, one megaspore is functional and other three degenerates.
- The functional **megaspore** developed into the **female gametophyte**.
- Female gametophyte is known as the **embryo sac**.
- Development of embryo sac from a single megaspore is called as **monosporic** type of embryo sac.
- The nucleus of the functional megaspore divided by **mitotic division** to form **two nuclei** which move to the opposite pole, **2-nucleated embryo sac**.
- Two successive mitotic division leads to formation of **4-nucleate** and later **8-nucleate** stages of the embryo sac.
- All mitotic divisions are free nuclear type; **karyokinesis** is not followed by **cytokinesis**.
- Six of the eight nuclei are surrounded by cell walls and organized into cells.
- Three cells are grouped together at the micropylar end, constitute the **egg apparatus**.
- The egg apparatus, in turn consists of two **synergids** and one **egg cell**.
- Synergids have special **filiform apparatus**, which play an important role in guiding the entry of pollen tube into the synergids.
- Three cells arranged towards chalazal end are called **antipodal** cells.
- The large **central cell** has two **polar nuclei**.
- A typical angiosperm embryo sac at maturity is **8-nucleated** and **7-celled**.

Pollination:

- Transfer of pollen grains from the anther to the stigma of a pistil is termed as pollination.
- Both male and female gametes are non-motile.

Kinds of pollination:

Autogamy:

- Pollination within same flower.
- In open and exposed anthers and stigma autogamy is rare.
- *Viola*, *Oxalis* and *Commelina* produce two types of flowers:
 - **Chasmogamous**: exposed anther and stigma
 - **Cleistogamous**: closed anther and stigma.
- Cleistogamous flower is invariably **autogamous** and **assured seed** set even in the absence of the pollinator.

Geitonogamy:

- Pollination between two flowers of the same plant.
- Pollination by pollinating agent.
- Genetically similar to the autogamy.

Xenogamy:

- Transfer of pollen grains from the anther to the stigma of different plant.
- It is commonly called as cross-pollination.
- It brings genetically different types of pollen grains to the stigma.

Agents of pollination:

- Plant use two **abiotic agent** i.e. wind and water for pollination.
- One **biotic agent** for pollination such as animals.
- Majority of plant use biotic agent for pollination.
- Few plant use abiotic pollinating agent.

Anemophily:

- Pollinating agent is wind.
- Plants produces enormous amount of pollen when compared to the number of ovules available for pollination to compensate the uncertainties of pollination.
- Flowers with well exposed stamens.
- Large feathery stigma to trap air-borne pollen grains.
- Most wind pollinated flower contains single ovule in one ovary and numerous flower packed into an inflorescence e.g. corn cob.
- Pollen grains are light and non-sticky.

Hydrophily:

- Pollination by abiotic agent like water.
- This type of pollination is very rare, about 30 genera, mostly monocot.
- *Vallisneria*, *Hydrilla* and *Zostera* are the common example for Hydrophily.
- All aquatic plants are not Hydrophily.
- Pollen grains released into the surface of water and carried to the stigma by air current as in *Vallisneria*.
- In **sea grass** the flowers remained **submerged**.

- Pollen grains are **long, ribbon** like and carried passively inside the water
- Pollen grains are protected from wetting by **mucilaginous covering**.

Pollination by biotic agent:

- Majority of flowering plants use a range of animals as pollinating agents.
- Among the animal, insect particularly bees are the dominant biotic agents for pollination.
- Insect pollinating flowers are very large, colorful, fragrant and rich in nectar.
- Small flowers present in cluster to make them conspicuous.
- Flower pollinated by flies and beetles secrete foul odours.
- Nectar and pollen grains are the usual floral rewards for insects.
- In some species floral rewards are in providing safe places to lay eggs: e.g. *Amorphophallus*.
- A species of **moth** and *Yucca* plant cannot complete their life cycle without each other. The moth deposits its eggs in the locule of the ovary and the flower in turn get pollinated by the moth.
- Many insects may consume pollen or nectar without bring about pollination. Such floral visitors are referred as **pollen/nectar robbers**.

Outbreeding Devices:

- Majority of the flowering plants produce hermaphrodite flower and undergo **autogamy**.
- Continuous **autogamy** or **self-pollination** results in **inbreeding depression**.
- Flowering plants have developed many devices to avoid self-pollination and to encourage cross-pollination. Such devices are called **Outbreeding devices**.
 - **Pollen released and stigma receptivity is not synchronized.**
 - **Spatial separation of anthers and stigmas**
 - **Anther and stigma are placed at different positions.**
 - **Self-incompatibility.**
 - **Production of unisexual flowers.**

Pollen pistil Interaction:

- All the events – from pollen deposition on the stigma until pollen tubes enter the ovule – are together referred as **pollen-pistil interaction**.
- Pollination does not guarantee the transfer of the right type of pollen grain to the right type of stigma.
- The pistil has the ability to recognize the pollen whether it is compatible or incompatible.
- If it is right type the stigma allow the pollen to germinate.
- If it is wrong type the stigma rejects the pollen, preventing germination.
- The ability of the pistil to recognize the pollen by continuous dialogue mediated by chemical like **Boron, Inositol and sucrose level**.
- Following compatible pollination, the pollen grain produce pollen tube through one of the germ pore.

- Content of the pollen grain move into the pollen tube.
- Pollen tube grows through the tissues of the stigma and style and reaches the ovary.
- If the pollen grain is in 2-celled stage the generative cell divides and forms two male gametes inside the pollen tube.
- If the pollen grain is in 3- cell stage the pollen tube carry two male gametes from the beginning.
- Pollen tube enters into the ovule through micropyle and then into the embryo sac through synergids guided by filiform apparatus.

Artificial hybridization:

- One of the major approaches of crop improvement programme.
- Only desired pollen grain used for pollination.
- Stigma is protected from contamination (from unwanted pollen grain).
- Removal of anthers from the flower bud before the anther dehisces is called **emasculatation**.
- Emasculated flowers covered by bag generally made up of butter paper, to prevent contamination of its stigma with unwanted pollen. This step is called **bagging**.
- If the female flower is unisexual there is no need of emasculatation.

Double fertilization:

- After entering one of the synergids, the pollen tube releases two male gametes into the cytoplasm of the synergids.
- **Syngamy:** one of the male gamete fused with egg cell, to form a diploid **zygote**.
- Two polar nuclei of central cell fused to form a diploid **secondary nucleus**.
- **Triple fusion:** The second male gamete fused with the secondary nucleus to form a triploid **primary endosperm nucleus**.
- Since two type of fusion, syngamy and triple fusion take place in the embryo sac the phenomenon is termed as **double fertilization**.
- The central cell after triple fusion becomes **primary endosperm cell** and developed into the **endosperm**.
- The zygote developed into an embryo.

POST- FERTILIZATION : STRUCTURE AND EVENTS :

Events of endosperm and embryo development, maturation of ovule into seed and ovary into fruit, are collectively termed as **post-fertilization events**.

Endosperm:

- Development of endosperm takes place before the embryo development.
- Primary endosperm cell divides repeatedly to form a triploid endosperm.
- Cells are filled with reserve food material and are used for the nutrition of the developing embryo.

- PEN undergoes successive nuclear division to give rise to free nuclei. This is called free-nuclear endosperm.
- Subsequently cell wall formation takes place and become cellular endosperm.
- The coconut water is free nuclear endosperm and the white kernel is the cellular endosperm.
- Endosperm may be consumed completely during embryo developed or it may be consumed during germination of seed.

Embryo:

- Zygote formed and placed at the micropylar end of the embryo sac.
- Zygote starts its development only after some amount of endosperm formed.
- Embryo development takes place in following stages:
 - **Proembryo**
 - **Globular stage**
 - **Heart shaped**
 - **Matured embryo.**

Dicot embryo:

- A typical dicotyledonous **embryo** consists of an **embryonal axis** and two **cotyledons**.
- Embryonal axis above the cotyledon is the **epicotyls**.
- Terminal part of the epicotyls is the **plumule** (gives rise to the shoot).
- Embryonal axis below the cotyledon is the **hypocotyl**.
- The terminal part of the hypocotyl is called the **radicle (root tip)**.
- The root tip is covered by the **root cap**.

Monocot embryo:

- Possesses only one cotyledon
- In grass family the **cotyledon** is called **scutellum**.
- Scutellum situated towards one side of the embryonal axis.
- Radicle and the root cap enclosed by a sheath called **coleorhiza**.
- The portion of the embryonal axis above level of attachment of scutellum is called **epicotyls**.
- Epicotyl has the shoot apex or plumule enclosed by hollow foliar structure called **coleoptile**.
- Seed is the final product of the sexual reproduction.
- Seed consists of seed coat, cotyledon and an embryo axis.
- Cotyledon stores the reserve food material for development and germination.
- Matured seed without endosperm called **non-albuminous**. (Ground nut)
- A part of the endosperm retained in matured seed is **Albuminous**.
- Remnants of nucellus in the matured seed is called **perisperm**. E.g. black peeper, beet.
- The wall of the ovary develops into the wall of fruit called **pericarp**.
- Fruit developed from the ovary is called **true fruit**.

- In apple, strawberry, cashew, the thalamus contributes in the fruit formation is called **false fruit**.
- Fruit developed without fertilization is called **Parthenocarpic fruits**.

APOMIXIS AND POLYEMBRYONY:

- Apomixis is very common in Asteraceae and grasses.
- Seeds are produced without fertilization.
- Apomixis is a type of asexual reproduction which mimics the sexual reproduction.
- Diploid egg cell is formed without meiosis and develops into seed without fertilization.
- In *Citrus* and *Mango* the nucellar cells starts dividing, protrude into the embryo sac and develop into embryo.
- Ovule having more than one embryo is termed as **polyembryony**.
- Hybrid plants are developed by apomixis to maintain the genetic identity.

CHAPTER-03

HUMAN REPRODUCTION

THE MALE REPRODUCTIVE SYSTEM:

- Located in the pelvis region.
- Male reproductive system includes
 - **A pair of testes.**
 - **Accessory ducts.**
 - **Accessory glands.**
 - **External genitalia**

Testes:

- Located outside the abdominal cavity within a pouch called **scrotum**.
- Scrotum provides **low temperature** required for spermatogenesis.
- Each testis is about 4 to 5 cm length and 2 to 3 cm width.
- Each testis has about 250 compartments called **testicular lobules**.
- Each lobule contains one to three seminiferous tubules.
- Seminiferous tubules lined by **male germ cells** and **Sertoli cells**.
- Male germ cell undergoes **meiosis** and produce **sperm**.
- Sertoli cells provide **nutrition** to the germ cell and the sperm.
- In between the seminiferous tubule there is **interstitial cell** or **Leydig cell**.
- Leydig cells produce testicular hormones called **androgen (testosterone)**.

Accessory ducts:

- Includes **rete testis, vasa efferentia, epididymis** and **vas deferens**.
- Seminiferous tubules open into vasa efferentia through rete testis.
- The vasa efferentia leaves the testis and open into epididymis.
- The epididymis leads to vas deferens that ascends to the abdomen through inguinal canal and loops over the urinary bladder.
- Vas deferens receives a duct from seminal vesicle and opens into the urethra as the **ejaculatory duct**.
- Urethra originates from the urinary bladder and extends through the penis to its external opening called **urethral meatus**.

Accessory glands:

- Includes
 - **Paired seminal vesicle**
 - **A prostate gland**
 - **Paired bulbourethral gland.**
- Secretion of these glands constitutes the **seminal plasma**.
- Seminal plasma rich in fructose, calcium, and certain enzyme.
- Secretion of bulbo-urethral glands helps in lubrication of penis.

External genitalia:

- **Penis** is the external genitalia.
- It is made of special tissue that helps in erection of the penis to facilitate **insemination**.
- The enlarged end of penis is called **glans penis**.
- Glans penis is covered by a loose fold of skin called **foreskin**.

THE FEMALE REPRODUCTIVE SYSTEM

- Located in the pelvic region of the female.
- The female reproductive system includes:
 - **A pair of ovaries**
 - **A pair of oviduct.**
 - **Uterus**
 - **Cervix**
 - **Vagina**
 - **External genitalia.**
 - **A pair of mammary gland.**
 - The ovarian stroma has two zones :
 1. A peripheral cortex.
 2. An inner medulla.

Oviduct:

- Oviducts, uterus and vagina constitute the female accessory ducts.
- Each fallopian tube is about 10-12 cm long and extends from the periphery of each ovary to the uterus.
- Close to the ovary the oviduct has a funnel shaped structure called **infundibulum?**
- The edges of the infundibulum possess finger-like projections called **fimbriae**, which helps in collection of the ovum after ovulation.
- The infundibulum leads to a wider part of the oviduct called **ampulla**.
The last part of the oviduct is called **isthmus** which joined to uterus.

Uterus:

- It is single and is called **womb**.
- It is inverted pear shaped.
- Attached the pelvic wall by ligaments.
- The uterus opens into vagina through a narrow cervix.
- The lumen of cervix is called cervical canal.
- Cervical canal along with vagina form the birth canal.
- The wall of the uterus has three layers of tissues
 - **Perimetrium:** external thin membranous.
 - **Myometrium:** middle thick layer of smooth muscles
 - **Endometrium:** inner glandular layer.
- Endometrium undergoes cyclical changes during menstrual cycle.
- Myometrium exhibits strong contraction during delivery of the baby.

External genitalia:

- It includes following structure:
 - **Mons Pubis:** cushion of fatty covered by skin and pubic hair.
 - **Labia majora:** fleshy folds of tissue which extends down from the mons pubis and surrounds the vaginal opening.
 - **Labia minora:** are paired folds of tissue under the labia majora.
 - **Hymen:** the opening of vagina is often covered partially by a membrane called hymen.
 - **Clitoris:** a tiny finger-like structure lies at the upper junction of two labia minora above the urethral opening.

Mammary glands:

- Mammary gland consists of glandular tissue and fat.
- Glandular tissue of each breast divided into 15-20 **mammary lobes**.
- Mammary lobes contain cluster of cells called **alveoli**.
- The cells of alveoli secrete milk, stored in the lumen of alveoli.
- The alveoli open into **mammary tubules**.
- The tubules of each lobe join to form a **mammary duct**.
- Several mammary ducts join to form a wider **mammary ampulla**.
- Mammary ampulla connected to **lactiferous duct**, through which milk is sucked out.

GAMETOGENESIS: (formation of gametes)

Spermatogenesis:

- Formation of sperm from the germ cell in the testes is spermatogenesis.
- The process begins at puberty.
- Spermatogonia present in the lining of seminiferous tubules undergo mitotic division to increase their number.
- Each spermatogonium is **diploid (2n)** which contain **46 chromosomes**.
- Innermost layer of spermatogonial becomes larger called **primary spermatocyte**.
- Primary spermatocyte undergoes meiosis-I to form two equal haploid (n) **secondary spermatocytes (n)**.
- Each secondary spermatocyte undergoes meiosis-II to form two equal, haploid **spermatids**.
- Each primary spermatocyte produces four spermatids.
- Spermatids transformed into **spermatozoa (sperms)** by the process called **spermiogenesis**.
- The sperm head embedded in the Sertoli cell.
- Release of sperm from the seminiferous tubule is called **spermiation**.

Hormonal control of spermatogenesis:

- This process is initiated at puberty due to secretion of gonadotrophins releasing hormone (GnRH).

- GnRH secreted from hypothalamus and stimulates anterior pituitary to secrete two gonadotrophins.
 - Luteinizing hormone (LH) and
 - Follicle stimulating Hormone (FSH)
- LH acts on Leydig cells and stimulates synthesis of androgens.
- Androgen stimulates spermatogenesis.
- FSH acts on Sertoli cells and stimulates spermatogenesis in other ways.

Structure of sperm:

- Ultrastructure of sperm consists of a **head**, **neck**, a **middle piece** and a **tail**.
 - Whole body of sperm surrounded by plasma membrane.
 - The sperm head contains an elongated haploid nucleus.
 - Above the nucleus a cap-like structure is present called **acrosome**.
 - The acrosome contains **enzymes** which help in fertilization of ovum.
 - The middle piece contains **mitochondria**, which provide energy for movement of tail that facilitates sperm motility.
 - Human male ejaculates **200-300 million sperms** during coitus.
 - 60 percent must have normal shape and size and 40 percent of them must show vigorous motility.
 - Sperm released from seminiferous tubules enters into accessory ducts.
 - On their way fluids from seminal vesicle and prostate gland are added which collectively are called as **Semen**.
- The function of male accessory ducts and glands are maintained by testicular hormone androgen.

Oogenesis:

- Formation of a mature female gamete or ovum is called **oogenesis**.
- Oogenesis starts during embryonic stage, 25th week of the fetal age.
- Germinal epithelium of ovary divides mitotically to produce millions of gamete mother cells or **oogonia**.
- No oogonia are formed or added after birth.
- Oogonia enter into meiosis-I and proceed up to diakinesis of Prophase-I and get suspended, at this stage called **primary Oocytes**.
- Each primary oocyte is surrounded by layers of granulosa cells and then called **primary follicle**.
- At puberty only **60,000 to 80,000** primary oocytes are left in each ovary.
- After puberty primary follicles get surrounded by more layers of granulosa cells and a new theca to form **secondary follicles**.
- The secondary follicle transforms into **tertiary follicle**, characterized by a fluid-filled cavity called **antrum**.
- The theca layers are organized into an inner theca interna and outer theca externa.
- During the growth of primary follicle into tertiary follicle during puberty, the primary oocyte restarts its first meiotic division and completes it within tertiary follicle resulting **two unequal haploid cells**.

- Large haploid cell is called **secondary oocyte**.
- A tiny cell called **first polar body**.
- The secondary oocyte retains bulk of the nutrient rich cytoplasm of primary oocyte.
- The tertiary follicle having secondary oocyte further changes into **Graafian follicle**.
- The **secondary oocyte** surrounded by a new membrane, **zona pellucida**.
- The secondary oocyte undergoes second meiotic division continued upto **metaphase-II** and get suspended until entry of sperm.
- At this stage Graafian follicle releases secondary oocyte from the ovary by the process called **ovulation**.
- On entry of a sperm into the secondary oocytes stimulates it to complete meiosis-II and there is formation of a haploid **ovum** and a **second polar body (n)**.

Menstrual cycle:

- Reproductive cycle of female primates is called **menstrual cycle**.
- The first menstruation begins at puberty is called **Menarche**.
- Menstrual cycle repeated at an average interval of 28/29 days.
- One ovum is released in the middle of each menstrual cycle.

Menstrual cycle has following phases:

Menstrual phase:

- 1st phase of menstrual cycle.
- Menstrual flow occurs.
- Lasts for 3-5 days.
- Breakdown of endometrial lining and blood vessel.
- Mucus and blood comes out through vagina.
- It occurs only when ovum released but no fertilization.
- Lack of menstruation is the indication of pregnancy.

Follicular phase:

- Menstrual phase followed by follicular phase.
- Primary follicle becomes Graafian follicle.
- Regeneration and proliferation of uterine endometrium.
- LH and FSH level increases gradually in follicular phase.
- Level of estrogen increases as it is secreted from growing follicle.
- It lasts for 5-13 days.

Ovulatory phase:

- FSH and LH attain peak level in this period (14th day).
- This is called **LH surge**, which induces rupture of Graafian follicle and release of ovum from the ovary called **ovulation**.

Luteal phase:

- Remaining part of Graafian follicle transformed into **corpus luteum**.
- Corpus luteum produces large amount of **progesterone**.
- Progesterone maintains the uterine endometrium, and prepares it for implantation.
- Thickness of uterine endometrium increase in many folds, due to proliferation.
- If there is fertilization, corpus luteum grows further and pregnancy continued, menstrual cycle stopped.
- In the absence of fertilization corpus luteum degenerates.
- Disintegration of endometrium leading to menstruation.
- Menstrual cycle ceases around 50 years of age, called **menopause**.

FERTILIZATION AND IMPLANTATION:

- During copulation (coitus) semen is released by the penis into the vagina is called **insemination**.
- The motile sperm swim rapidly, pass through cervix, uterus and finally reach the junction of isthmus and ampulla (**ampullary-isthmic junction**).
- The ovum released from the ovary also transported to ampullary isthmic junction where fertilization takes place.
- Fertilization only takes place if both sperm and ovum reach ampullary – isthmic junction simultaneously.
- The process of fusion of a sperm and ovum is called **fertilization**.
- Acrosome of sperm secretes enzymes helps in penetration into the ovum.
- Once a sperm comes contact with the zona pellucida of ovum and induces the changes in the membrane that blocks the entry of additional sperms.
- That ensures **monospermy** and prevents **polyspermy**.
- Only one sperm fertilize with one ovum.
- Entry of sperm into the ovum induces the ovum to complete its second meiotic division of secondary oocyte.
- Meiosis-II is also unequal cytokinesis resulting production of one large **ovum (ootid)** and one small **second polar body**.
- Haploid nucleus of sperm fused with the haploid nucleus of ovum to form a **diploid zygote**.

Sex determination:

- Sex of a baby has been decided during fertilization and in the zygote.
- Sex is determined by the sex-chromosomes present in gametes.
- Human female contain two XX chromosomes.
- Human male contain XY chromosomes.
- All the female gametes produced with only 'X' chromosome.
- Sperms produced by male, 50% with 'X' and 50 % with 'Y' chromosome.
- After fertilization zygote either carries XX or XY chromosomes.

- Zygote with XX chromosomes develop into female and with XY chromosome develops into male.

Cleavage:

- Repeated mitotic division of the zygote without growth resulting a multicellular ball like embryo is called **cleavage**.
- Cleavage starts soon after fertilization.
- Daughter cells produced during cleavage are called **blastomeres**.
- The product of cleavage is called **Morula**, which is 8 to 16 celled.
- The morula continues to divide and grow and transformed into **blastocyst**.
- The blastomeres in blastocyst arranged into an outer layer called **trophoblast** and an inner mass of cells attached to trophoblast called **inner cell mass**.
- Trophoblast cells attached to the endometrium helps development of placenta.
- Inner cell mass gets differentiated into the embryo.
- After attachment the uterine cells divide rapidly and cover the blastocyst.
- Blastocyst completely embedded in the uterine endometrium. This is called **implantation**.

Pregnancy and embryonic development:

- After implantation, finger like projections appears on the trophoblast called **chorionic villi**.
- Chorionic villi surrounded by uterine tissue and maternal blood.
- Temporary association between the fetal tissue (chorionic villi) and maternal tissue (uterine endometrium) is called **placenta**.

Function of placenta:

- The embryo connected to the placenta by umbilical cord, which transports substances to and from the embryo.
- Facilitate transport of oxygen and nutrient from mother to embryo.
- Removes CO₂ and waste material from the embryo.
- Acts as endocrine gland and produces several hormones like:
 - Human chorionic gonadotrophins (hCG)
 - Human placental lactogen (hPL)
 - Estrogen.
 - Progesterone
 - Relaxin produced from the ovary in the later stage of pregnancy.

Embryonic development:

- After implantation the inner cell mass of blastocyst differentiated into an outer layer called **ectoderm** and an inner layer called **endoderm**.
- **Mesoderm** differentiated in-between ectoderm and endoderm.
- The inner cell mass thus called **stem cells**, having potency to produce all types of cell, tissues and organs by differentiation.

Organogenesis:

- Formation of different organs in the embryo is called **organogenesis**.
- Human pregnancy lasts for **9 months**.
- After one month of pregnancy **heart** is formed in the embryo.
- By the end of 2nd month the foetus develops **limbs and digits**.
- By the end of 12 weeks (**first trimester**) most of organ system is formed (limbs and external genitalia are well developed).
- First movement of foetus and appearance of hairs observed in 5th month.
- By the end of 24th week (**2nd trimesters**) the body is covered with fine hairs, eye-lids separate, and eyelashes are formed.
- By the end of 9 months the foetus is fully developed and is ready for delivery.

PARTURATION AND LACTATION:

- The period of pregnancy is called gestation period. (9 months).
- Ejection or expulsion or delivery of foetus is called **parturition**.
- Parturition is due to vigorous contraction of **uterine Myometrium**.
- The signal of parturition is originated from the fully developed foetus and the placenta which induces mild contraction of uterus called **fetal ejection reflex**.
- Fetal ejection reflex triggers the release of **Oxytocin** from pituitary.
- Oxytocin induces stronger contraction of uterine endometrium.
- Stimulatory reflex continues stronger contraction leads to expulsion.
- After delivery the placenta is also expelled out of the uterus.

Lactation:

- The mammary gland of the female more differentiated during pregnancy,
- Mammary gland starts producing milk towards the end of the pregnancy.
- Process of milk production in mammary gland is called **lactation**.
- Milk produced during initial days of lactation is called **colostrum**.
- Colostrum contains several **antibodies** which provide immunity to the new born baby.

CHAPTER-04

REPRODUCTIVE HEALTH

REPRODUCTIVE HEALTH – PROBLEM AND STRATERIES:

- The programme “family planning” initiated in 1951.
- Reproductive and child health care (ACH)
- Sexually transmitted diseases (STD).
- **Amniocentesis:** A fetal sex determination test based on the chromosomal pattern in the amniotic fluid surrounding the developing embryo.
- ‘Saheli’ an oral contraceptive for female, developed by CDRI.

POPULATION EXPLOSION AND BIRTH CONTROL:

- Increased health facilities, better living conditions are the cause of population explosion.
- Out of 6 billion world population 1 billion are Indians.
- Rapid decline in death rate, **maternal mortality rate (MMR)** and **infant mortality rate (IMR)** are major cause of population growth.
- Indian population growth rate is around 1.7 percent.

Characteristics of ideal contraceptive:

- User friendly.
- Easily available.
- Effective
- Nor or least side – effects.
- No way interferes with sexual drive.

BIRTH CONTROL METHODS:

Natural methods: work on the principle of avoiding chances of ovum and sperms meeting.

Periodic abstinence:

- Avoid or abstain from coitus form day 10 to 17 of the menstrual cycle when ovulation could be expected.
- Chance of fertilization is very high in this period.
- It is called fertile period.

Withdrawal or coitus interruption:

- The male partner withdraws his penis from the vagina just before ejaculation, so as to avoid insemination into the vagina.

Lactational amenorrhea:

- No menstruation during lactation period.
- Chance of fertilization is nil.
- It is effective upto six month.

Barrier methods:

- **Principle of working:** prevents physical meeting of sperm and ovum.
- Such methods available both for male and female.

Condoms:

- Barriers made of thin rubber/latex sheath.
- Used to cover the penis in male or vagina and cervix in the female.
- Used just before coitus so that semen not entered into the female reproductive tract.
- Male and female condoms are disposable.
- Prevents AIDS and STDs.

Diaphragm, cervical caps and vaults:

- Barriers made of rubber.
- Inserted into the female reproductive tract to **cover the cervix**.
- Prevents conception by blocking the entry of sperm through cervix.
- They are reusable.

Intra Uterine Devices:

- These devices are only used by female.
- Inserted by doctor or by expert nurses in the uterus through vagina.
- Non-medicated IUDs e.g. Lippes loop.
- Copper releasing IUDs (CuT, Cu7, Multiload 375)
- Hormone releasing IUDs (Progestasert, LNG-20)

Principle of working:

- Increase phagocytosis of sperm within the uterus.
- Cu ion released suppresses sperm motility and fertilizing capacity of sperm.
- Hormone releasing IUDs make the uterus unsuitable for implantation and the cervix hostile to the sperm.

Oral contraceptives:

- This methods used by female only.
- Used in the form of tablets hence popularly called **pills**.
- Pills contain progestogens or progestogen-estrogen combination.
- Pills have to be taken daily for a period of 21 days.
- Started within first five days of menstruation.
- Pills are very effective with lesser side effect.
- **Saheli** - a **non-steroidal** preparation used as oral contraceptive pills.

Principle of working:

- Inhibit ovulation.
- Inhibit implantation.
- Alter the quality of cervical mucus to prevent/retard entry of sperms.

Injections or implants:

- Progestogens alone or in combination with estrogen used as injections or implants under the skin by female.
- Mode of action is similar as in pills
- It is very effective for long periods.

Emergency contraceptives:

- These methods are used within 72 hours of coitus, rape or casual unprotected intercourse.
- Administration of progestogens or progestogen-estrogen combination.
- Use of IUDs.

Surgical methods:

- It is also called as sterilization method.
 - Advised to both male and female partner.
 - **Permanent or terminal method** to prevent pregnancy.
 - Sterilization process in male is called '**vasectomy**,
 - Sterilization process in female is called '**Tubectomy**'
 - In vasectomy, a small part of the vas deferens is removed or tied up.
 - In Tubectomy a small part of the fallopian tube is removed.
- Reversibility is very poor.

MEDICAL TERMINATION OF PREGNANCY:

- Intentional or voluntary termination of pregnancy before full term is called **medical termination of pregnancy (MTP)** or **induced abortion**.
- MTP has significant role in decreasing population.
- It accounts for 1/5th of the total number of conceived pregnancies.
- Legal restriction only to reduce female foeticide.
- This method is safe within 1st trimester.

SEXUALLY TRANSMITTED DISEASES:

- Diseases or infections which are transmitted through sexual intercourse.
- Also known as Venereal diseases (VD) or reproductive tract infections (RTIs)
- **Gonorrhoea, Syphilis, Genital herpes, chlamydia, genital warts, trichomoniasis, hepatitis-B and HIV** are some common STDs.
- Except **hepatitis-B, genital herpes and HIV infections**, others are curable.

Symptoms:

- Itching, fluid discharge, slight pain, swelling in the genital region.
- STDs remain asymptomatic in female and remain undetected for long.
- In the later stage it may lead to **Pelvic inflammatory diseases (PID)**, abortion, still births, ectopic pregnancy, infertility or even cancer in RT.

Preventions:

- Avoid sex with unknown partners/ multiple partners.
- Always use condoms during coitus.
- In case of doubt, consult with a qualified doctor for early detection.
- Get complete treatment if diagnosed with disease.

INFERTILITY:

- The couple unable to produce children inspite of unprotected sex.
- The reason of infertility may be:-
 - **physical,**
 - **congenital,**
 - **diseases,**
 - **drugs,**
 - **immunological** or
 - Even **psychological.**
- Problems of infertility may be in male or female.
- Infertility clinic can diagnose and correct the cause of infertility.
- In case there no corrections are possible, some special technologies used to have children called **assisted reproductive technologies. (ART)**

Assisted reproductive technologies:

(a) In vitro fertilization:

- Fertilization outside the body in the laboratory.
- Condition created in laboratory similar to the body.

(b) Embryo transfer:

- Popularly known as **test tube baby** programme.
- Ova from the wife/donor and sperm from the husband/donor are collected and induced to form zygote under simulated conditions in the laboratory.
- The zygote or early embryos (with upto 8 blastomeres) could be transferred into the fallopian tube.
- **ZIFT- Zygote intra fallopian transfer.**
- **IUT- Intra Uterine transfer** (embryo with more than 8 blastomeres).
- Further development taken place within the female body.
- Embryo formed by **in-vivo fertilization** can also be transfer to assist those female who cannot conceive.

(c) Gamete intra fallopian transfer- GIFT

- Transfer of ovum collected from the donor into the fallopian tube of another female who cannot produce it.
- Such female can provide suitable environment for fertilization and development.

(d) Intra cytoplasmic sperm injection (ICSI):

- The sperm is directly injected into the ovum.
- After in vitro fertilization either ZIFT or embryo transfer technique is followed.

(e) Artificial insemination (AI)

- Semen is collected either from the husband or donor is artificially introduced into vagina or into the uterus (**IUI-intra uterine insemination**) of the female.
- Such technology is useful in cases either the male partner unable to inseminate the female or very low sperm counts in the ejaculates.

Abbreviation:

IUCD: Intra Uterine Contraceptive Device

RCH: Reproductive and Child Health care

STD: Sexually Transmitted Disease

HIV: human Immuno deficiency virus.

AIDS: Acquired immuno deficiency syndrome

CDRI: Central Drug Research Institute

MMR: Maternal Mortality Rate

IMR: Infant mortality rate.

MTP: Medical Termination of Pregnancy

VD: Venereal Disease

RTI: Reproductive Tract Infection

PID: Pelvic Inflammatory Disease

ART: Assisted Reproductive Technologies

IVF: In Vitro Fertilisation

ZIFT: Zygote Intra Fallopian Transfer

AI: Artificial insemination

IUI: Interna uterine insemination.

ET: Embryo transfer.

IUT: intra uterine transfer.

ICSI: Intra Cytoplasmic Sperm Injection

CHAPTER-05

PRINCIPLES OF INHERITANCE AND VARIATION

PRINCIPLES OF INHERITANCE AND VARIATION

- **Genetics:** deals with the inheritance, as well as the variation of characters from parents to offsprings.
- **Inheritance:** is the process by which characters are passed on from parent to progeny.
- **Variation:** is the degree by which progeny differ from their parents.

MENDEL'S LAWS OF INHERITANCE:

- Gregor Mendel. Conducted hybridization experiments on garden peas for seven years (1856 – 1863) and proposed laws of inheritance.
- Mendel conducted artificial pollination/cross pollination experiments using several true-breeding pea lines.
- A true breeding line is one that, having undergone continuous self-pollination for several generations.
- Mendel selected 14 true-breeding peas' plant varieties, as pair's which were similar except for one character with contrasting traits.
 - **True breed selected by Mendel**
 - **Stem height- Tall / dwarf**
 - **Flower color- Violet/white**
 - **Flower position – Axial / terminal**
 - **Pod shape- Inflated / beaded or constricted**
 - **Pod color- Green / yellow**
 - **Seed color- Yellow/ green**
 - **Seed shape – round / wrinkled**

INHERITANCE OF ONE GENE:

- Mendel crossed tall and dwarf pea plants to study the inheritance of one gene.
- He collected the seeds produced as a result of this cross and grew them to generate plants of the first hybrid generation. This generation is called **filial progeny** or the **F₁**.
- Mendel observed that all the F₁ progeny plants were tall, like one of its parents; none were dwarf.
- He made similar observations for the other pairs of traits – he found that the F₁ always resembled either one of the parents, and that the trait of the other parent was not seen in them.
- Mendel then self – pollinated the tall F₁ plants and to his surprise found that in the F₂ generation some of the offsprings were 'dwarf; the character that was not seen in the F₁ generation was now expressed.
- The proportion of plants that were dwarf was 1/4th of the F₂ plants while 3/4th of the F₂ plants were tall.

- The tall and dwarf traits were identical to their parental type and did not show any **blending**, that is all the offsprings were either tall or dwarf, none were of in between height.
- Similar results were obtained with the other traits that he studied: only one of the parental traits was expressed in the F_1 generation while at the F_2 stage both the traits were expressed in the proportion of 3:1.
- The contrasting traits did not show any blending at either F_1 or F_2 stage.

Mendel's proposition:

- Mendel proposed that something was being stably passed down, unchanged, from parent to offspring through the gametes, over successive generations. He called these things as '**factors**'.
- Now a day we call them as **genes**.
- Gene is therefore are the **units of inheritance**.
- Genes which codes of a pair of contrasting traits are known as **alleles** *i.e.* they are slightly different forms of the same gene.

Alphabets used:

- Capital letters used for the trait expressed at the F_1 stage.
- Small alphabet for the other trait.
- '**T**' is used for Tall and '**t**' is used for dwarf.
- '**T**' and '**t**' are alleles of each other.
- Hence in plants the pair of alleles for height would be **TT**, **Tt** or **tt**.
- In a true breeding tall or dwarf pea variety the allelic pair of genes for height are **identical** or **homozygous**, **TT** and **tt** respectively.
- **TT** and **tt** are called the **genotype**.
- **Tt** plant is **heterozygous** for genes controlling one character (height).
- Descriptive terms **tall** and **dwarf** are the **phenotype**.

Test cross:

- When F_1 hybrid is crossed back with the recessive parent, it is known as test cross.
- It is used to know the genotype of the given plant/animal.

Law of Dominance:

- Characters are controlled by discrete units called factors.
- Factors occur in pairs.
- In a dissimilar pair of factors one member of the pair dominates (dominant) the other (recessive).

Law of Segregation:

- The alleles do not show any blending and that both the characters are recovered as such in the F_2 generation though one of these is not seen at the F_1 stage.

- The parents contain two alleles during gamete formation; the factors or alleles of a pair segregate or separate from each other such that a gamete receives only one of the two factors.
- Homozygous parent produces all gametes that are similar *i.e.* contain same type of allele.
- Heterozygous parents' produces two kinds of gametes each having one allele with equal proportion.

Know the terms

Terms	Meaning	Example
Locus	Address/ location of a gene in a chromosome	T,A,b,d etc
Allele	Allelomorphs= alternative form of a gene	T and t OR A and a etc
Homozygous	Both alleles of a gene at a locus similar	AA or aa
Heterozygous	Both alleles of a gene at a locus dissimilar	Aa or Tt etc
Homozygous Dominant	Both alleles of a gene at a locus similar & dominant	AA
Homozygous recessive	Both alleles of a gene at a locus similar & recessive	aa

Incomplete dominance:

- When a cross between two pure breed is done for one contrasting character, the F₁ hybrid phenotype dose not resemble either of the two parents and was in between the two, called **incomplete dominance**.
- Inheritance of flower color in the **dog flower (snapdragon or Antirrhinum sp.)** is a good example of incomplete dominance.
- F₂ generation phenotypic ratio is 1:2:1 instead of 3:1 as Mendelian monohybrid cross.
- Genotypic ratio of F₂ generation is 1:2:1.

Co – dominance:

- F₁ resembled either of the two parents (**complete dominance**).
- F₁ offspring was in-between of two parents (**incomplete dominance**).
- F₁ generation resembles both parents side by side is called (**co-dominance**).
- Best example of co-dominance is the ABO blood grouping in human.
- ABO blood group is controlled by the **gene I**.
- The plasma membrane of the RBC has sugar polymers (antigen) that protrude from its surface and the kind of sugar is controlled by the **gene-I**.
- The gene I has three alleles **I^A, I^B** and **i**.

- The alleles I^A and I^B produce a slightly different form of sugar while allele i doesn't produce any sugar.
- Each person possesses any two of the three I gene alleles.
- I^A and I^B are completely dominant over i .
- When I^A , and I^B present together they both express their own types of sugar; this because of co-dominance. Hence red blood cells have both A and B type sugars.

Co Dominance and multiple allelism

Blood group	Possible genotype
A	$I^A I^A$ OR $I^A i$
B	$I^B I^B$ OR $I^B i$
AB	$I^A I^B$
O	ii

Crosses of blood group (CO DOMINANCE)

Blood group	Possible genotype	Possible phenotype
A X A	$I^A I^A \times I^A I^A$	A
	$I^A I^A \times I^A i$	A
	$I^A i \times I^A i$	A; O
B X B	$I^B I^B \times I^B I^B$	B
	$I^B I^B \times I^B i$	B
	$I^B i \times I^B i$	B; O
AB X AB	$I^A I^B \times I^A I^B$	AB; A; B
O X O	$ii \times ii$	O

POSSIBLE BLOOD GROUP OF PROGENY WITH RESPECT TO THE BLOOD GROUP OF PARENTS

Parent	Progeny			
	A	B	AB	O
A X A	+	-	-	+
A X O	+	-	-	+
A X B	+	+	+	+
B X B	-	+	-	+
B X O	-	+	-	+
AB X A	+	+	+	-
AB X B	+	+	+	-
AB X O	+	+	-	-
AB X AB	+	+	+	-
O X O	-	-	-	+
KEY	+ = POSSIBLE		- = NOT POSSIBLE	

Multiple Alleles:

- Example of ABO blood grouping produces a good example of multiple alleles.
- There are more than two i.e. three allele, governing the same character.

A single gene product may produce more than one effect:

- Starch synthesis in pea seeds is controlled by one gene.
- It has two alleles **B** and **b**.
- Starch is synthesized effectively by **BB** homozygote and therefore, large starch grains are produced.
- The '**bb**' homozygous has less efficiency hence produce smaller grains.
- After maturation of the seeds, **BB seeds are round** and the **bb seeds are wrinkle**.
- **Heterozygous (Bb)** produce round seed and so B seems to be dominant allele, but the starch grains produced are of intermediate size.
- If starch grain size is considered as the phenotype, then from this angle the alleles show incomplete dominance.

INHERITANCE OF TWO GENES:

Law of independent Assortment:

- When two characters (dihybrid) are combined in a hybrid, segregation of one pair of traits is independent of the other pair of traits.

CHROMOSOMAL THEORY OF INHERITANCE:

Why Mendel's theory was remained unrecognized?

- **Firstly** communication was not easy in those days and his work could not be widely publicized.
- **Secondly** his concept of genes (or factors, in Mendel's word) as stable and discrete units that controlled the expression of traits and of the pair of alleles which did not 'blend' with each other, was not accepted by his contemporaries as an explanation for the apparently continuous variation seen in nature.
- **Thirdly** Mendel's approach of using mathematics to explain biological phenomena was totally new and unacceptable to many of the biologists of his time.
- Finally he could not provide any physical proof for the existence of factors.

Rediscovery of Mendel's result:

- 1990 three scientists (**de Vries, Correns and von Tschermak**) independently rediscovered Mendel's result on the inheritance of character.

Chromosomal theory of inheritance:

- Proposed by **Walter Sutton** and **Theodore Boveri** in 1902.
- They work out the chromosome movement during meiosis.
- The behavior of chromosomes was parallel to the behavior of genes and used chromosome movement to explain Mendel's laws.

- Sutton united the knowledge of chromosomal segregation with Mendelian principles and called it the **chromosomal theory of inheritance**.
 - Chromosome and genes are present in pairs in diploid cells.
 - Homologous chromosomes separate during gamete formation (meiosis)
 - Fertilization restores the chromosome number to diploid condition.
 - The chromosomal theory of inheritance claims that, it is the chromosomes that segregate and assort independently.

Experimental verification of chromosomal theory:

- Experimental verification of chromosomal theory of inheritance by **Thomas Hunt Morgan** and his colleagues.
- Morgan worked with tiny fruit flies, *Drosophila melanogaster*.

Why Drosophila?

- Suitable for genetic studies.
- Grown on simple synthetic medium in the laboratory.
- They complete their life cycle in about two weeks.
- A single mating could produce a large number of progeny flies.
- Clear differentiation of male and female flies
- Have many types of hereditary variations that can be seen with low power microscopes.

Linkage and Recombination:

- Morgan hybridized **yellow bodied, white eyed** females to **brown-bodied, red eyed** male and intercrossed their F1 progeny.
- He observed that the two genes did not segregate independently of each other and the F2 ratio deviated very significantly from 9:3:3:1 ratio (expected when the two genes are independent).
- When two genes in a dihybrid cross were situated on the same chromosome, the proportion of parental gene combinations was much higher than the non-parental type.
- Morgan attributed this due to the physical association or linkage of the two genes and coined the term **linkage**.
- **Linage:** physical association of genes on a chromosome.
- **Recombination:** the generation of **non-parental** gene combinations.
- Morgan found that even when genes were grouped on the same chromosome, some genes were very **tightly linked** (showed very low recombination) while others were **loosely linked** (showed higher recombination).
- The genes white and yellow were very tightly linked and showed 1.3 percent recombination.
- The genes white and miniature wing showed 37.2 percent recombination, hence loosely linked.

- Alfred Sturtevant used the frequency of recombination between gene pairs on the same chromosome as a measure of the distance between genes and ‘**mapped**’ their position on the chromosome.

POLYGENIC INHERITANCE:

- Human have no distinct tall or short instead a whole range of possible heights.
- Such traits are generally controlled by three or more genes and are thus called **polygenic trait**.
- Besides the involvement of multiple genes polygenic inheritance also takes into account the influence of environment.
- Human skin color is another classic example of polygenic inheritance.
- In a polygenic trait the phenotype reflects the contribution of each allele i.e. the effect of each allele is additive.
- Assume that three genes A, B, C control the skin colour in human.
- Dominant forms A, B; AND C responsible for dark skin colour and the recessive forms a, b, c for light color of the skin.
- Genotype with dominant alleles (AABBCC) will have darkest skin color.
- Genotype with recessive alleles (aabbcc) will have lightest skin colour.
- Other combinations always with intermediate colour.

PLEIOTROPY:

- A single gene can exhibit multiple phenotypic expression, such gene is called **pleiotropic gene**.
- The mechanism of pleiotropy in most cases is the effect of a gene on metabolic pathways which contributes towards different phenotypes.
- **Phenylketonuria** a disease in human is an example of pleiotropy.
- This disease is caused due to **mutation** in the gene that code for the enzyme **phenyl alanine hydroxylase**.
- **Phenotypic expression characterized by:-**
 - Mental retardation
 - Reduction in hairs.
 - Reduction in skin pigmentation.

SEX DETERMINATION:

- **Henking (1891)** traced specific nuclear structure during spermatogenesis of some insects.
- **50 %** of the sperm received these specific structures, whereas 50% sperm did not receive it.
- Henking gave a name to this structure as the **X-body**.
- X-body of Henking was later on named as X-chromosome.

Sex-determination of grass hopper:

- Sex-determination in grasshopper is **XX-XO type**.
- All egg bears one ‘X’ chromosome along with autosomes.

- Some sperms (50%) bear's one 'X' chromosome and 50% do not.
- Egg fertilized with sperm (with 'X' chromosome) became female (22+XX).
- Egg fertilized with sperm (without 'X' chromosome) became male (22 + XO).

Sex determination in insects and mammals (XX-XY type):

- Both male and female has same number of chromosomes.
- Female have autosomes and a pair of X chromosomes. (AA+ XX)
- Male have autosomes and one **large 'X' chromosome** and one very small '**Y-chromosomes**. (AA+XY)
- This is called **male heterogammy** and **female homogammy**.

Sex determination in birds:

- Female birds have two different sex chromosomes designated as **Z** and **W**.
- Male birds have two similar sex chromosomes and called **ZZ**.
- Such type of sex determination is called **female heterogammy** and **male homogammy**.

Sex determination in Honey bee:

- Sex determination in honey bee based on the number of sets of chromosomes an individual receives.
- An offspring formed from the fertilization of a sperm and an egg developed into either queen (female) or worker (female).
- An unfertilized egg develops as a male (drone), by means of parthenogenesis.
- The male have half the number of chromosome than that of female.
- The female are diploid having 32 chromosomes and males are haploid i.e. having 16 numbers of chromosomes.
- This is called **haplodiploid sex determination system**.
- Male produce **sperms by mitosis**, they do not have father and thus cannot have sons, but have **grandsons**.

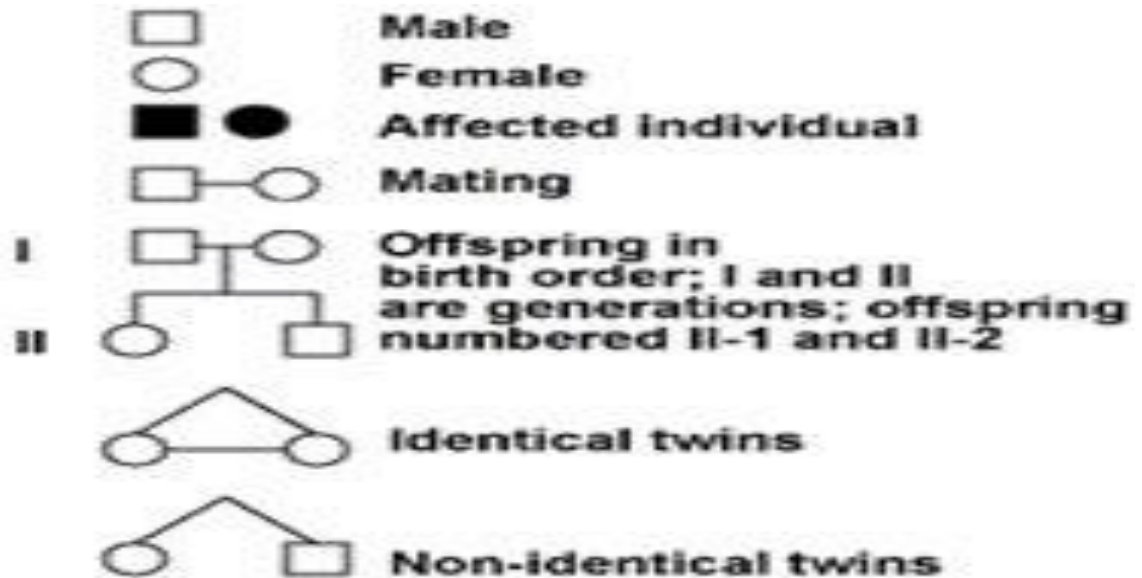
MUTATION:

- Mutation is a phenomenon which results in alteration of DNA sequences and consequently results in changes in the genotype and phenotype of an organism.
- In addition to recombination, mutation is another phenomenon that leads to variation in DNA.
- Loss (deletion) or gain (insertion/duplication) of a segment of DNA results in alteration in chromosomes.
- Since genes are located on the chromosome, alteration in chromosomes results in abnormalities or aberration.
- Chromosomal aberrations are commonly observed in cancerous cells.
- Mutations also arise due to change in a single base pair of DNA. This is known as point mutation. E.g. sickle cell anemia.
- Deletion and insertions of base pairs of DNA causes frame shift mutations.

GENETIC DISORDERS:

Pedigree Analysis:

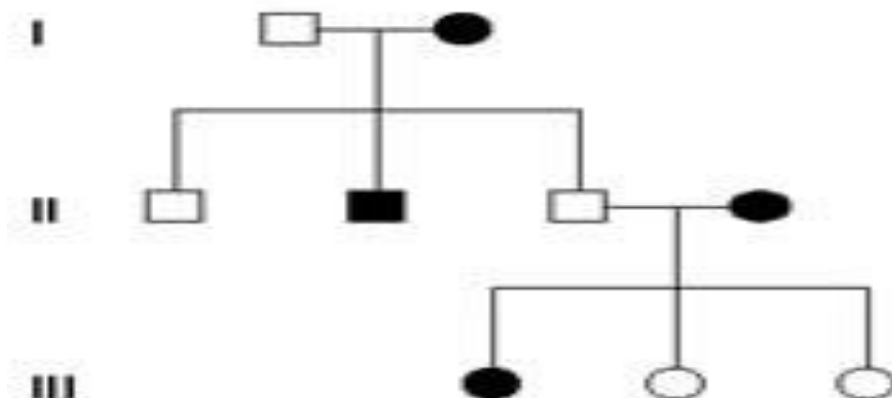
- Analysis of traits in several of generations of a family is called the **pedigree analysis**.
- In the pedigree analysis the inheritance of a particular trait is represented in the family tree over generations.



Autosomal Dominant: Affected individuals have at least one affected parent

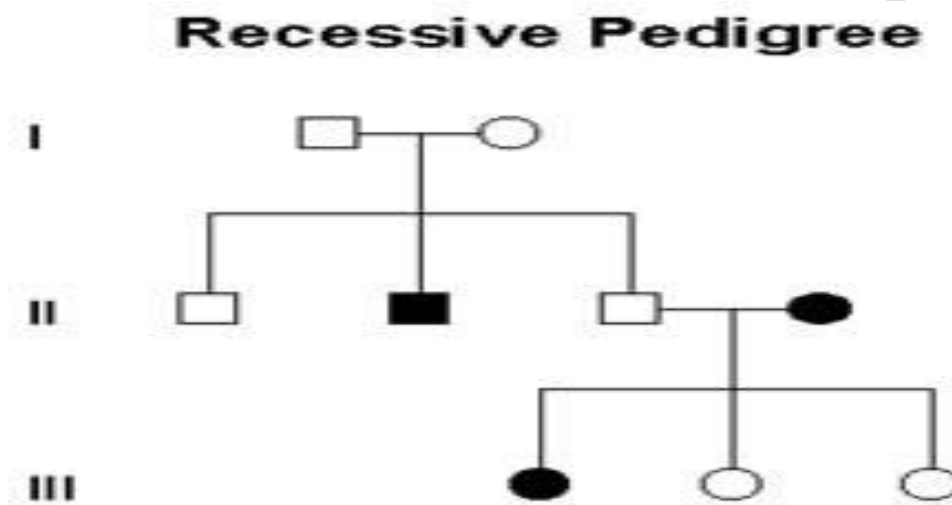
- The phenotype generally appears every generation
- Two unaffected parents only have unaffected offspring
- Traits are controlled by dominant genes
- Both males and females are equally affected
- Traits do not skip generations
- e.g. polydactyly, tongue rolling ability etc

Dominant Pedigree



Autosomal recessive:

- Unaffected parents can have affected offspring
- Traits controlled by recessive genes and
- Appear only when homozygous
- Both male and female equally affected
- Traits may skip generations
- 3:1 ratio between normal and affected.
- Appearance of affected children from normal parents (heterozygous)
- All children of affected parents are also affected.
- **e.g.- Albinism, sickle cell anaemia etc.**



Mendelian Disorder:

- Genetic disorders grouped into two categories –
 - Mendelian disorder
 - Chromosomal disorder
- Mendelian disorders are mainly determined by alteration or mutation in the single gene.
- Obey the principle of Mendelian inheritance during transmission from one generation to other.
- Can be expressed in pedigree analysis.
- **Example - Haemophilia, colorblindness, Cystic fibrosis, Sickle cell anemia, Phenylketonuria, Thalasemia etc.**

Hemophilia:

In this disease a single protein that is a part of the cascade of proteins involved in the clotting of blood is affected. Due to this in an affected individual a simple cut will result in non-stop bleeding.

- Sex linked recessive disease.
- The diseases transmitted from unaffected carrier female to some of the male progeny.

- Female becoming hemophilic is extremely rare because mother of such a female at least carrier and the father should be hemophilic.
- Affected transmits the disease only to the son not to the daughter.
- Daughter can receive the disease from both mother and father.

Sickle cell anaemia:

- The defect is caused due to substitution of Glutamic acid (Glu) by Valine (Val) at the sixth position of the beta globin chain of the haemoglobin molecule.
- Substitution of amino acid takes place due to the single base substitution at the sixth codon of the beta globin gene from GAG to GUG.
- The mutant haemoglobin molecule undergoes polymerization under low oxygen tension causing the change in the shape of the RBC from biconcave disc to elongated sickle like structure.
- This is an autosomes linked recessive trait.
- Transmitted from parents to the offspring when both the parents are carrier for the gene (heterozygous).
- This disease is controlled by single pair of allele, Hb^A , and Hb^S .
- There are three possible genotypes ($Hb^A Hb^A$, $Hb^A Hb^S$, and $Hb^S Hb^S$).
- Only homozygous individuals for Hb^S ($Hb^S Hb^S$) show the diseased phenotype.
- Heterozygous ($Hb^A Hb^S$) individuals appear apparently unaffected but they are carrier of the disease as there is 50 percent probability of transmission of the mutant gene to the progeny.

Phenylketonuria:

- Autosomal recessive trait.
- Inborn error of metabolism.
- The affected individual lack one enzyme called phenyl alanine hydroxylase that converts the amino acid phenyl alanine to tyrosine.
- In the absence of the enzyme phenyl alanine accumulated and converted into phenylpyruvic acid and other derivatives.
- Accumulation of these results in mental retardation.
- These derivatives excreted through kidney.

Chromosomal disorders:

- Caused due to absence or excess or abnormal arrangement of one or more chromosome.
- Failure of segregation of chromatids during cell division cycle results in the gain or loss of chromosome(s), called **Aneuploidy**.
- Failure of cytokinesis after telophase stage of cell division results in an increase in a whole set of chromosome in an organism and this phenomenon is called **polyploidy**.

Trisomy: additional copy of a chromosome may be included in an individual ($2n+1$).

Monosomy: an individual may lack one of any one pair of chromosomes ($2n-1$)

Down syndrome:

- Caused due to presence of an additional copy of the chromosome number 21 (**trisomy of 21**).
- This disorder was first described by **Langdon Down (1866)**.
 - Short stature with small round head.
 - Furrowed tongue
 - Partially opened mouth
 - Palm is broad with characteristic palm crease.
 - Physical, psychomotor and mental development is retarded.

Klinefelter's syndrome:

- Caused due to the presence of an additional copy of X-chromosome resulting into a karyotype of 47, ($44+XXY$).
 - Overall masculine development.
 - Also develop feminine character (development of breast i.e. Gynaecomastia)
 - Individuals are sterile.

Turner's syndrome:

- Caused due to the absence of one of the X-chromosomes i.e. $45 (44 + XO)$.
- Such females are sterile as ovaries are rudimentary.
- Lack of other secondary sexual characters.

CHAPTER-06

MOLECULAR BASIS OF INHERITANCE

THE DNA:

- DNA is a long polymer of deoxyribonucleotides.
- The length of the DNA depends on, number of nucleotide pair present in it.
- Characteristics of the organism depend on the length of the DNA.
- Bacteriophage ϕ 174 has 5386 nucleotides.
- Bacteriophage lambda has 48502 base pairs.
- Escherichia coli have 4.6×10^6 base pairs.
- Human genome (haploid) is 3.3×10^9 bp.

Structure of polynucleotide chain:

- A nucleotide has three component:-
 - A nitrogen base
 - A pentose sugar (ribose in RNA and deoxyribose in DNA)
 - A phosphoric acid.
- There are two types of nitrogen bases:
 - **Purines** (Adenine and Guanine)
 - **Pyrimidines** (Cytosine, Uracil and Thymine)
- Adenine, Guanine and Cytosine is common in RNA and DNA.
- Uracil is present in RNA and Thymine is present in DNA in place of Uracil.
- Pentose sugar is **ribose** in RNA and **Deoxyribose** in DNA.
- A nitrogen base attached to the pentose sugar at C¹ of pentose sugar by **N-glycosidic** linkage to form a nucleoside.
- According to the nature of pentose sugar, two types of nucleosides are formed **ribonucleoside** and **deoxyribonucleotides**.
- **Ribonucleosides** are:
 - Adenosine
 - Guanosine
 - Cytidine
 - Uridine
- **Deoxyribonucleosides** are:
 - Deoxyadenosine
 - Deoxyguanosine
 - Deoxycytidine
 - Deoxythymidine.
- Phosphoric acid attached to the **5' OH** of a nucleoside by **Phosphodiester linkage** a corresponding nucleotide is formed. (Ribonucleotide or deoxyribonucleotides depending on the sugar unit).
- Two nucleotides are joined by 3'-5' Phosphodiester linkage to form dinucleotide.
- More than two nucleotides joined to form polynucleotide chain.

- Polynucleotide chain has a free phosphate moiety at 5' end of sugar, is referred to as 5' end
- In the other end of the polymer with 3'-OH group called 3' end.
- The backbone of the polynucleotide chain is sugar and phosphate.
- Nitrogen bases linked to the sugar moiety project from the backbone.
- In RNA every nucleotide has an additional –OH group at 2' of ribose.
- In RNA Uracil is found in place of thymine.
- 5-methyl uracil is the other name of thymine.

History of DNA:

- DNA is an acidic substance in the nucleus was first identified by Friedrich Meischer in 1869. He named it as 'Nuclein'
- 1953 **double helix** structure of DNA was given by James Watson and Francis Crick, based on X-ray defraction data produced Maurice Wilkins and Rosalind Franklin.
- Hallmark of their proposition was base pairing between two strands of polynucleotide chains. This was based on observation of **Erwin Chargaff**.
- Chargaff's observation was that for a double stranded DNA, the ratio between Adenine and Thymine, and Guanine and Cytosine are constant and equal one.

Salient features of Double helix structure of DNA:

- Made of two polynucleotide chains.
- Sugar and phosphate forms the **backbone** and bases projected to inside.
- Two chains have **anti-parallel** polarity.
- Two strands are held together by **hydrogen bond** present in between bases.
- Adenine of one strand pairs with Thymine of another strand by two hydrogen bonds and vice versa.
- Guanine of one strand pairs with Cytosine of another strand by three hydrogen bonds and vice versa.
- A purine comes opposite to a pyrimidine. This generates approximately uniform distance between the two strands of the helix.
- The two chains are coiled in a right – handed fashion.
- The pitch of the helix is 3.4 nm or 34 Å⁰
- There are roughly 10 bp in turn.
- The distance between the bp in a helix is 0.34nm or 3.4 Å⁰.
- The plane of one base pair stacks over the other in double helix.
- H-bond confers stability of the helical structure of the DNA.
- Central dogma of flow of genetic information: DNA → RNA → Protein.

Packaging of DNA Helix:

- Distance between two conjugative base pairs is 0.34nm, the length of the DNA in a typical mammalian cell will be 6.6×10^9 bp x 0.34×10^{-9} /bp, it comes about 2.2 meters.

- The length of DNA is more than the dimension of a typical nucleus (10-6m), how is such a long polymer packaged in a cell?

Packaging in prokaryotes:

- They do not have definite nucleus.
- The DNA is not scattered throughout the cell.
- DNA is held together with some proteins in a region is called 'nucleoid'.
- The DNA in nucleoid is organized in large loops held be proteins.

Packaging in Eukaryotes:

- In eukaryotes the packaging is more complex.
- There is a set of positively charged, basic protein called **Histones**.
- Histones are positively charged due to rich in basic amino acids like **Lysine** and **arginine**.
- Histones are organized to form a unit of eight molecules called **histone octamer**.
- Negatively charged DNA wrapped around positively charged histone octamer to form a structure called **nucleosome**.
- A typical nucleosome contains **200 bp** of DNA helix.
- Nucleosome constitutes the repeating unit of a structure in nucleus called **chromatin**, thread like stained bodies seen in the nucleus.
- The nucleosomes are seen as '**beads-on-string**' structure when viewed under electron microscope.
- The chromatin is packaged to form **chromatin fibers** that are further coiled and condensed at metaphase stage to form **chromosome**.
- Packaging at higher level required additional set of proteins called **Non-histone Chromosomal (NHC) proteins**.
- In a typical nucleus some loosely coiled regions of chromatin (light stained) is called **euchromatin**.
- The chromatin that more densely packed and stains dark are called **Heterochromatin**.
- Euchromatin is **transcriptionally active** chromatin and heterochromatin is inactive.

THE SEARCH OF GENETIC MATERIAL:

Transforming principle:

- Given by **Frederick Griffith** in 1928.
- His experiment based on *Streptococcus pneumoniae* (caused pneumonia).
- There is change in physical form of bacteria.
- There are two colonies of bacteria:
 - Smooth shiny colonies called **S strain**.
 - Rough colonies called **R strain**.
- S-strain bacteria have a **mucous (polysaccharide) coat**.
- R-strain does not have mucous coat.
- S-strain is virulent and caused pneumonia in mice and died when infected.

- R-strain is non-virulent and dose caused pneumonia in mice when infected.
- Heat killed S-Strain is non-virulent and dose not causes pneumonia.
- The heat killed S-Strain mixed with live R-Strain injected into mice; the mice developed pneumonia and died.
- He recovered live S-Strain bacteria form the dead mice.

Conclusion of experiment:

- R – Strain bacteria had somehow been **transformed** by the heat killed S-Strain bacteria.
- Some ‘**transforming principle**’, transferred from heat killed S-Strain bacteria, had enabled the R-Strain to synthesize smooth polysaccharide coat and become virulent (S Strain).
- The transformation of R-Strain to S-Strain is due to transfer of **Genetic material**.
- However the biochemical nature of genetic material was not defined from his experiment.

Biochemical characterization of transforming principle:

- Biochemical nature of transforming principle was discovered by **Oswald Avery, Colin Macleod and Maclyn McCarty**. (1933-44)
- Prior to their work genetic material was thought to be protein.
- They worked to determine the biochemical nature of the ‘transforming principle’ of Griffith’s experiment.
- They purified biomolecules (proteins, DNA and RNA) from the heat killed S cells to see which one could transform live R cells to S cells.
- Heat killed S-Strain + protease + Live R-Strain → transformation.
- Heat killed S-Strain + RNase + Live R-Strain → transformation.
- Heat killed S-Strain + DNase + Live R-Strain → transformation.

Conclusion of the experiments:

- Protein of heat killed S-Strain is not the genetic material
- RNA of heat killed S-Strain is not the genetic material.
- DNA of heat killed S-Strain is the genetic material, because DNA digested with DNase mixed with R-strain unable to transform R-Strain to S-Strain.

The Genetic Material is DNA:

- ‘DNA is the genetic material’ is proved by **Alfred Hershey and Martha Chase** (1952).
- They worked on the virus that infects bacteria called **bacteriophage**.
- During normal infection the bacteriophage first attaches the bacteria cell wall and then inserts its genetic material into the bacterial cell.
- The viral genetic material became integral part of the bacterial genome and subsequently manufactures more virus particle using host machinery.
- Hershey and Chase worked to discover whether it was protein or DNA from the viruses that entered the bacteria.

Experiment (Blenders experiment) :

- They grew some viruses on a medium having **radioactive phosphorus** and some others on medium having **radioactive sulfur**.
- Viruses grown in **radioactive Phosphorus** have **radioactive DNA** but not radioactive protein because Phosphorus present in DNA not in protein.
- Viruses grown in **radioactive sulfur** have **radioactive protein** not radioactive DNA because sulfur present in protein but not in DNA.
- **Infection:** radioactive phages were allowed to attach to *E.coli* bacteria; the phages transfer the genetic material to the bacteria.
- **Blending:** the viral coats were separated from the bacteria surface by agitating them in a blender.
- **Centrifugation:** The virus particles were separated from the bacteria by spinning them in a centrifuge machine.

Observation:

- Bacteria infected with viruses that had radioactive DNA were radioactive and no radioactivity in the supernatant.
- Bacteria infected with viruses that had radioactive protein were not radioactive, but radioactivity found in the supernatant.

Conclusion of Experiment:

- **DNA is the infecting agent that made the bacteria radioactive hence DNA is the genetic material not the protein.**

PROPERTIES OF GENETIC MATERIAL (DNA VERSUS RNA):

Criteria for genetic material:

- It should be able to generate its replica (replication)
- It should be chemically and structurally stable.
- It should provide the scope for slow changes (mutation) that required for evolution.
- It should be able to express itself in the form of 'Mendelian Character'.
- Protein does not fulfill the criteria hence it is not the genetic material.
- RNA and DNA fulfill the criteria.

RNA is unstable:

- 2'-OH group present at every nucleotide (ribose sugar) in RNA is a reactive group and makes RNA liable and easily degradable.
- RNA is also now known as **catalyst**, hence reactive.
- RNA is unstable and mutates faster. Consequently the viruses having RNA genome and having shorter life span mutate and evolve faster.

DNA is more stable:

- Stability as one of the properties of genetic material was very evident in Griffith's 'transforming principle' itself that heat, which killed the bacteria at least did not destroy some of the properties of genetic material.

- Two strands being complementary if separated by heating come together, when appropriate conditions are provided.
- Presence of Thymine in place of uracil confers additional stability to DNA
- DNA is chemically less reactive and structurally more stable when compared to RNA.
- Therefore, among the two nucleic acids the DNA is a better genetic material.

Better genetic material (DNA or RNA)

- Presence of thymine at the place of uracil confers more stability to DNA.
- Both DNA and RNA are able to mutate.
- In fact RNA being unstable mutate at a faster rate.
- RNA can directly code for the synthesis of proteins, hence easily express.
- DNA however depends on RNA for protein synthesis.
- The protein synthesis machinery has evolved around RNA.
- Both RNA and DNA can functions as genetic material, but DNA being more stable is preferred for storage of genetic information.
- For the transmission of genetic information RNA is better.

RNA WORLD:

- RNA is the first genetic material.
- Essential life processes evolved around RNA.
- RNA used to act as a genetic material as well as catalyst.
- But RNA being catalyst was reactive and hence unstable.
- Hence DNA has evolved from RNA with chemical modifications that make it more stable.
- DNA being double stranded and having complementary strand further resists changes by evolving a process of repair.

REPLICATION: THE PROCESS:

- **Watson and Crick** proposed a scheme for replication of DNA.
- The Original statement that “It has not escaped our notice that the specific pairing we have postulated immediately suggests a possible copying mechanism for the genetic material” (Watson and Crick, 1953).
- The scheme suggested that the two strands would separate and act as template for the synthesis of new complementary strands.
- New DNA molecule must have one parental strand and one new strand.
- This scheme of replication is called **Semi-conservative** type of replication.

Experimental Proof of semiconservative nature of replication:

- It is now proved experimentally that replication is semiconservative type.
- It was first shown in *Escherichia coli* and subsequently in higher organism.
- **Mathew Messelson** and **Franklin Stahl** performed the following experiment in 1958.

STEPS OF THE EXPERIMENTS:

- They grew *E.coli* in $^{15}\text{NH}_4\text{Cl}$ medium for many generations. (^{15}N is heavy nitrogen not radioactive element)
- The result was that ^{15}N was incorporated into newly synthesized DNA and other nitrogen containing compound as well.
- This heavy DNA molecule could be distinguished from normal DNA by centrifugation in a cesium chloride (CsCl) density gradient.
- Then they transferred the *E.coli* into a medium with normal $^{14}\text{NH}_4\text{Cl}$ and let them grow (*E.coli* divides in 20 minutes).
- They took samples at definite time intervals as the cells multiplied, and extracted the DNA that remained as double-stranded helices.
- Various samples were separated independently on **CsCl gradients** to measure the densities of DNA.
- The DNA that was extracted from the culture one generation after the transfer from ^{15}N to ^{14}N medium had a **hybrid** or **intermediate density**.
- DNA extracted from the culture after another generation (after 40 minutes) was composed of equal amount of this hybrid DNA and of 'light DNA'.

Experiment by Taylor and colleagues:

- Used **radioactive thymidine** to detect distribution of newly synthesized DNA in the chromosomes.
- They performed the experiment on *Vicia faba* (faba beans) in 1958.
- They proved the semiconservative nature of DNA replication in eukaryotes.

Replication Machinery and Enzymes:

- In all living cells such as *E.coli* replication requires a set of enzymes.
- *E.coli* completes the replication of its DNA in within **38 min**.
- The average rate of polymerization has to be approx. **2000 bp per sec**.
- The polymerization process must be accurate; any mistake during replication would result into **mutation**.
- **Deoxyribonucleoside triphosphates** (dATP, dGTP, dCTP, dTTP) serve **dual purposes**.
 - Provide **energy** for polymerization.
 - Acts as **substrates** for polymerization.
- The replication process occurs within a small opening of the DNA helix called **replication fork**.
 - The region where, replication fork formed is called **origin of replication**.
 - The replication fork is formed by an enzyme called **helicase**.
 - Two separated strand is called **template strands**.
 - Main enzyme is DNA-dependent **DNA polymerase**, since it uses a DNA template to catalyze the polymerization of deoxyribonucleotides.
 - DNA polymerase catalyses polymerization only in one direction i.e. **5'→3'**.
 - On one strand (template with 3'→5' polarity) the replication is continuous hence called **leading strand**.

- In another strand (template with 5'→3' polarity) the polymerization takes place in the form of short fragment called **Okazaki fragment**.
- The short fragments are joined by **DNA ligase**, hence called **lagging strand**.
- In eukaryotes replication takes place in **S-phase** of cell cycle.
- A failure of cytokinesis after replication results into **polyploidy**.

TRANSCRIPTION:

- 'The process of copying genetic information from one strand of the DNA into RNA is termed as transcription'.

Transcription vs. Replication:

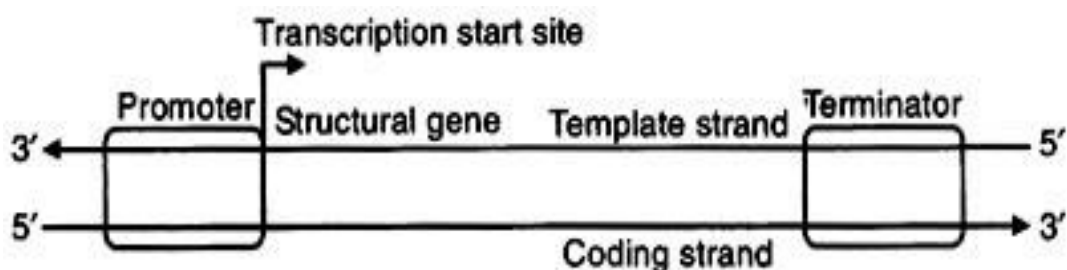
- Principle of **complementarity** governs the process of transcription except Adenosine of DNA forms base pair with the **Uracil** instead of thymine. During replication Adenine pairs with **thymine** instead of uracil.
- During replication once started the **whole DNA** is duplicated, whereas transcription takes place only **a segment of DNA**.
- In replication **both strand** acts as template, where as in transcription only **one strand** is acts as template to synthesize RNA.
- In replication **DNA copied** from a DNA, where as in transcription **RNA copied** from the DNA.

Why both strands of DNA not copied during transcription:

- If both strand of DNA acts as template, they would translated into two RNA of different sequences and in turn if they code for proteins, the sequence of amino acids in the protein would be different. Hence one segment of DNA would be coding for two different proteins.
- The two RNA molecules if produced from simultaneously would be complementary to each other, hence will form double stranded RNA. This would prevent RNA translation into protein.

Transcription unit:

- A transcription unit in DNA consists of three regions:
- A promoter
- The structural gene
- A terminator.
- **DNA dependent RNA polymerase** catalyses the polymerization in only one direction that is 5'→3'.



Structural gene:

- The DNA strand having polarity $3' \rightarrow 5'$ is called **template strand** for transcription.
- The other strand of DNA having polarity $5' \rightarrow 3'$ is called **coding strand**.
- The sequences of nitrogen base in the RNA transcribed from the template strand are same as the coding strand of DNA except having Thymine in place of Uracil.
- All the reference point defining a transcription unit is made with the **coding strand** only, not the template strand.

Promoter:

- **Promoter** and **Terminator** present on either side of structural gene.
- The promoter located towards 5' end (**upstream**) of the structural gene.
- It is a short sequence of DNA that provides binding site for RNA polymerase. (mostly TATA, Commonly called TATA box)
- Presence of the promoter defines the template and coding strands.
- If the position of promoter is changed with terminator the definition of coding and template strand will be reversed.

Terminator:

- The **terminator** located towards 3' end (**downstream**) of coding strand.
- It terminates the process of transcription.
- It is also a short segment of DNA which recognizes the termination factor. (ρ -factor)

Transcription unit and the gene:

- Gene is defined as the functional unit of inheritance.
- Genes are located on the DNA.
- The DNA sequence coding for tRNA and rRNA molecule also define a gene.
- **Cistron**: a segment of DNA (structural gene) coding for a polypeptide.
- **Monocistronic**: most of eukaryotic structural gene codes for single polypeptide.
- **Polycistronic**: Most prokaryotic structural gene code for more than one polypeptides.
- In eukaryotes the monocistronic structural genes have interrupted coding sequences, the genes are said to be **split gene**:
 - The coding sequences or expressed sequences are called **Exons**.
 - Exons are interrupted by **Introns**.
- Exons are said to be those sequences that appear in mature or processed mRNA.
- Introns never appear in mature or processed mRNA. They are spliced out.

Types of RNA:

- In prokaryotes there are three major types of RNAs: **mRNA** (messenger), **tRNA** (transfer), and **rRNA** (ribosomal).
- All three RNAs are required to synthesize protein in a cell.

- The mRNA provides the template and having genetic information in the form of genetic code.
- The tRNA brings the amino acids and read the genetic code of mRNA.
- The rRNA is the structural part of the ribosome and also as catalytic role during process of translation.

Process of transcription: prokaryotes.

- There is a single **DNA dependent RNA polymerase** that catalyses transcription or synthesis of all three types of RNAs in prokaryotes.
- The process of transcription completed in three steps:

Initiation:

- RNA polymerase binds to the specific site of DNA called **promoter**.
- Promoter of the DNA is recognized by **initiation factor or sigma (σ)**.
- RNA polymerase along with initiation factor binds to the promoter.

Elongation:

- RNA polymerase unzipped the DNA double helix and forms an open loop.
- It uses **ribonucleoside triphosphates** as substrate and polymerizes in a DNA template following the rule of complementarity.
- Only a short stretch of polymerized RNA remains binds with the enzyme.
- The process of polymerization continued till the enzyme reaches the terminator gene.

Termination:

- RNA polymerase recognizes the terminator gene by a **termination-factor** called **rho (ρ) factor**.
- The RNA polymerase separated from the DNA and also the transcribed RNA.

Additional complexities in eukaryotes:

- There are **three** different types of RNA polymerases in the nucleus:
 - **RNA polymerase I** transcribes **rRNA (28S, 18S and 5.8S)**
 - **RNA polymerase II** transcribes **heterogeneous nuclear RNA (hnRNA)**.
 - **RNA polymerase III** transcribes **tRNA, 5srRNA and snRNA**.
- **Post transcriptional processing** (occurs inside the nucleus).

(a) Splicing:

- The primary transcript (**hnRNA**) contain both **exons** and **introns** and required to be processed before translationally active (**mRNA**).
- The introns are removed and exons are joined in a defined order.
- This process is catalyzed by **SnRNP**, introns removed as **spliceosome**.

(b) Capping: an unusual nucleotide called **methyl guanosine triphosphate** is added to the 5' end of hnRNA.

(c) **Tailing:** Adenylate residues (200-300) are added at 3' end of hnRNA in a template independent manner.

The processed hnRNA is now called mRNA and transported out of the nucleus for translation.

GENETIC CODE:

Contribution to discovery:

- The process of replication and transcription based on complementarity.
- The process of translation is the transfer of genetic information from a polymer of nucleotides to a polymer of amino acids. There is no complementarity exist between nucleotides and amino acids.
- If there is change in the nucleic acid (genetic material) there is change in amino acids in proteins.
- There must be a genetic code that could direct the sequence of amino acids in proteins during translation.
- **George Gamow** proposed the code should be combination of bases, he suggested that in order to code for all the 20 amino acids, the code should be made up of three nucleotides.
- **Har Govind Khorana** enables instrumental synthesizing RNA molecules with desired combinations of bases (**homopolymer and co-polymers**).
- **Marshall Nirenberg's** cell – free system for protein synthesis finally helped the discovery of genetic code.
- **Severo Ochoa** enzyme (**polynucleotide phosphorylase**) was also helpful in polymerizing RNA with desired sequences in a template independent manner (enzymatic synthesis of RNA)

Salient features of genetic code:

- The **codon is triplet**. Three nitrogen base sequences constitute one codon.
- There are 64 codon (61 codes for amino acids and 3 codons are stop codon).
- One codon codes for only one amino acid, hence it is **unambiguous**.
- **Degeneracy:** some amino acids are coded by more than one codon.
- **Comma less:** the codon is read in mRNA in a continuous fashion. There is no punctuation.
- **Universal:** From bacteria to human UUU codes for phenyl alanine.
- **Initiation codon:** **AUG** is the first codon of all mRNA. And also it codes for methionine (met), hence has **dual function**.
- **Non-overlapping:** The genetic code reads linearly
- **Direction:** the code only read in **5' → 3'** direction.
- **Anticodon:** Each codon has a complementary anticodon on tRNA.
- **Non-sense codon:** UAA, GUA, and UAG do not code for amino acid and has no anticodon on the tRNA.

Mutation and Genetic code:

- Relationship between DNA and genes are best understood by mutation.

Point mutation:

- It occurs due to replacement nitrogen base within the gene.
- It only affects the change of particular amino acid.
- Best understood the cause of **sickle cell anemia**.

Frame shift mutation:

- It occurs due to insertion or deletion of one or more nitrogen bases in the gene.
- There is change in whole sequence of amino acid from the point of insertion or deletion.
- Best understood in **β -thalasemia**.

tRNA (the Adaptor molecule):

- The tRNA is called sRNA (soluble RNA).
- It acts as an adapter molecule.
- tRNA has an **anticodon loop** that base complementary to the codon.
- It has an **amino acid acceptor end** to which it binds with amino acid.
- Each tRNA bind with specific amino acid i.e 61 types of tRNA found.
- One specific tRNA with anticodon UAC called **initiator tRNA**.
- **There is no tRNA for stop codons. (UAA, UGA, UAG)**
- The secondary structure is like clover-leaf.
- The actual structure of tRNA is compact, looks like inverted 'L'.

TRANSLATION:

- It refers to polymerization of amino acids to form a polypeptide.
- The number and sequence of amino acids are defined by the sequence of bases in the mRNA.
- The amino acids are joined by **peptide bond**.
- Amino acids are activated in the presence of ATP and linked to their specific tRNA is called **charging of tRNA** or **aminoacylation of tRNA**.
- Ribosome is the cellular factory for protein synthesis.
- Ribosome consists of structural rRNA and 80 different proteins.
- In inactive state ribosome (70S) present in two subunits :
 - A large sub unit 50S.
 - A small sub unit 30S.

Initiation:

- The process of translation or protein synthesis begins with attachment of mRNA with small subunit of ribosome.
- The ribosome binds to the mRNA at the start codon (AUG).
- AUG is recognized by the initiator tRNA.

Elongation:

- Larger subunit attached with the initiation complex.
- Larger subunit has two site 'A' site and 'P' site.

- Initiator tRNA accommodated in 'P' site of large subunit, the subsequent amino-acyl-tRNA enters into the 'A' site.
- The subsequent tRNA selected according to the codon of the mRNA.
- Codon of mRNA and anticodon of tRNA are complementary to each other.
- Formation of peptide bond between two amino acids of 'P' and 'A' site, catalyzed by **ribozyme** (23S rRNA in bacteria)
- The ribosome moves from codon to codon along the mRNA called **translocation**.

Termination:

- Elongation continues until a stop codon arrives at 'P' site.
- There is no tRNA for stop codon.
- A **release factor** binds to the stop codon.
- Further shifting of ribosome leads to separation of polypeptide.
- An mRNA also has some additional sequences that are not translated called **untranslated regions (UTR)**.

REGULATION OF GENE EXPRESSION:

- Regulation of gene expression in eukaryotes takes place in different level:
- Transcriptional level (formation of primary transcript)
- Processing level (regulation of splicing)
- Transport of mRNA from nucleus to the cytoplasm.
- Translational level.
- In prokaryotes control of rate of transcriptional initiation is the predominant site for control of gene expression.
- The activity of RNA polymerase at the promoter is regulated by accessory proteins, which affects its ability to recognize the start site.
- The regulatory proteins can act both positively (activators) or negatively (repressor)
- The regulatory proteins interact with specific region of DNA called **operator**, which regulate the accessibility of RNA polymerase to promoter.

Lac operon:

- **Francois Jacob and Jacques Monod** first to describe a transcriptionally regulated system of gene expression.
- A polycistronic structural gene is regulated by common promoter and regulatory genes. Such regulation system is common in bacteria and is called **operon**.
- Lac operon consists of:-
 - One regulator gene (i-gene)
 - Three structural genes (z, y, and a)
 - Operator. (binding site of repressor protein)
 - Promoter (binding site of the RNA polymerase).
- The i-gene codes for **repressor** of the lac operon.
- The structural gene consists of three genes (z, y and a)

- 'z'-gene codes for **beta-galactosidase**, which hydrolyze lactose into Galactose and glucose.
- 'y' –gene codes for **permease**, which increases the permeability of bacterial cell to lactose.
- 'a'-gene codes for **transacetylase**.
- All three genes are required for the metabolism of lactose in bacteria.
- **Inducer:** lactose is the substrate for β - galactosidase and it regulates the switching on and off of the lac operon. Hence it is called inducer.
- In the absence of glucose, if lactose is added in the growth medium of the bacteria, the lactose is transported into the cell by **permease**.
- Very low level of expression of lac operon has to be present in the cell all the time; otherwise lactose cannot enter the cell.

Mechanism of regulation of lac operon:

- The **repressor protein** is synthesized from **i-gene** (all time constitutively)
- In the **absence of the inducer** i.e. lactose the active repressor binds to the operator and prevents RNA polymerase from transcribing the structural gene
- **In the presence** of the inducer such as **lactose** or **allolactose**, the repressor is inactivated by interaction with inducer.
- This allows RNA polymerase access to the promoter and transcription proceeds.
- The regulation of lac operon by **repressor** is referred to as **negative regulation**.

HUMAN GENOMIC PROJECT:

- Genetic make-up of an organism or an individual lies in the DNA sequences.
- Two individual differs in their DNA sequences at least in some places.
- Finding out the complete DNA sequence of human genome.
- Sequencing human genome was launched in 1990.

Goals of HGP:

- Identify all the approximately 20,000 – 25,000 genes in human DNA.
- Determine the sequence of all 3 billion chemical base pairs.
- Store this information in data bases.
- Improve tools for data analysis.
- Transfer related technologies to other sectors, such as industries.
- Address the ethical, legal, and social issues (ELSI) that may arise from the project.

Methodology:

- To identify all the genes that expressed as RNA referred as **Expressed Sequence Tags (ESTs)**.
- Simply sequencing the whole set of genome that contained all the coding and non-coding sequence, and later assigning different regions in the sequence with functions called **Sequence Annotation**.

- The commonly used hosts for sequencing were bacteria and yeast and vectors were called as **BAC** (bacterial artificial chromosome) and **YAC** (yeast artificial chromosome).

Salient features of Human Genome:

- The human genome contains **3164.7 million** nucleotide bases.
- The average gene consists of **3000 bases**.
- The largest known human gene being **dystrophin** at 2.4 million bases.
- The total number of gene is estimated at **30,000 million**.
- **99.9** percent nucleotide base sequences are same in all peoples.
- The function of 50% genes discovered is unknown.
- **Less than 2 percent** of the genome codes for **proteins**.
- **Repeated sequences** make up very large portion of human genome.
- **Chromosome I** has most genes (2968) and the **Y** has the fewest (231).
- It is identified about 1.4 million locations where single-base DNA differences (**SNPs – single nucleotide polymorphism**) occurs in humans.

DNA FINGER PRINTING:

- DNA finger printing is a very quick way to compare the DNA sequences of any two individuals.
- DNA fingerprinting involves **identifying differences in some specific regions in DNA** called **repetitive DNA**, because in these sequences, a small stretch of DNA is repeated many times.
- During centrifugation the bulk DNA forms major peak and the other **small peaks** are called **satellite DNA**.
- Depending on base composition (A:T rich or G:C rich), length of segment, and number of repetitive units, the satellite DNA classified into many types, such as mini –satellite and micro – satellite.
- These sequences dose not code for any proteins.
- These sequences show high degree of polymorphism and form basis of DNA fingerprinting.
- Polymorphism in DNA sequence is the basis of genetic mapping of human genome as well as of DNA fingerprinting.
- **Polymorphism** (variation at genetic level) arises due to mutations.
- If an inheritable mutation is observed in a population at high frequency it is referred as **DNA polymorphism**.

The process:

- DNA fingerprinting was initially developed by **Alec Jeffreys**.
- He used satellite DNA as the basis of DNA fingerprinting that shows very high degree of polymorphism. It was called as **Variable Number Tandem Repeats (VNTR)**.

- **Different steps of DNA fingerprinting are :**
 - Isolation of DNA.
 - Digestion of DNA by restriction endonucleases.
 - Separation of DNA fragments by gel electrophoresis.
 - Transferring (blotting) of separated DNA fragments to synthetic membranes, such as nitrocellulose or nylon.
 - Double stranded DNA made single stranded.
 - Hybridization using labeled VNTR probe.
 - Detection of hybridized DNA fragments by autoradiography.
- The VNTR belongs to a class of satellite DNA referred to as mini-satellite.
- The size of VNTR varies from 0.1 to 20 kb.
- After hybridization with VNTR probe the autoradiogram gives many bands of different sizes. These bands give a characteristic pattern for an individual DNA. It differs from individual to individual.
- The DNA from a single cell is enough to perform DNA fingerprinting.

Applications:

- Test of paternity.
- Identify the criminals.
- Population diversity determination.
- Determination of genetic diversity.

CHAPTER-07

EVOLUTION

ORIGIN OF LIFE:

- Stellar distances are measured in light years.
- The universe is very old – almost 20 billion years old.
- The **Big Bang** theory attempts to explain to us the origin of universe.

The Big Bang theory:

- A singular huge explosion unimaginable in physical term.
- The universe expanded and hence the temperature came down.
- Hydrogen and Helium formed sometime later.
- The gases condensed under gravitation and formed the galaxies of the present day universe.
- In the solar system of the Milky Way galaxy, earth was supposed to have been formed about 4.5 billion years back.

Condition of early earth:

- Earth formed 4.5 billion years back.
- There was no atmosphere on early earth.
- Water vapor, methane, carbon dioxide and ammonia released from molten mass covered the surface.
- The UV rays from the sun broke up water into Hydrogen and oxygen and lighter H_2 escaped.
- Oxygen combined with ammonia and methane to form water, CO_2 and others.
- The ozone layer was formed.
- As it cooled, the water vapor fell as rain, to fill all the depressions and form oceans.
- **Life appeared 500 Million years after the formation of earth.**

Origin of life:

- Early Greek thinkers thought units of life called **spores** were transferred to different planets including earth.
- ‘Panspermia’ is still a favorite idea for some astronomers.
- For a long time it was also believed that life came out of decaying and rotting matter like straw, mud etc. This was the **theory of spontaneous generation**.

Louis Pasteur experiment:

- Careful experimentation demonstrated that life comes only from pre-existing life.
- He showed that in pre-sterilized flasks, life did not come from killed yeast while in another flask open to air, new living organism arose from ‘killed yeast’.
- This disproved the theory of spontaneous generation.

Oparin – Haldane theory of origin of life:

- Oparin of Russia and Haldane of England proposed that the first form of life could have come from pre- existing non-living organic molecule (e.g. RNA, protein etc.).
- Formation of life was preceded by chemical evolution i.e. formation of diverse organic molecule from inorganic constituents.

Urey and Miller experiment:

- **The conditions on earth were –**
 - High temperature.
 - Volcanic storms.
 - Reducing atmosphere containing CH₄, NH₃ etc.
- In **1953, S. L. Miller** an American Scientist created similar conditions in a laboratory scale.
 - He created **electric discharge** in a closed flask to raise temperature upto 800^oC as it was in primitive earth.
 - Used CH₄, H₂, NH₃ and water vapor inside the flask.
 - He observed the formation of **amino acids**.

Acceptance of chemical evolution theory (evidences):

- Miller observed the synthesis of amino acids from simple inorganic chemicals in simulated condition in the laboratory.
- In similar experiments others observed, formation of sugars, nitrogen bases, pigment and fats.
- Analysis of meteorite content also revealed similar compounds indicating that similar processes are occurring elsewhere in space.

Theory of biogenesis:

- The first non-cellular forms of life could have originated 3 billion years back.
- They would have been giant molecules (RNA, proteins, Polysaccharides etc.).
- These capsules reproduced their molecules perhaps, named as **coaservates**.
- The first cellular form of life did not possibly originate till about 2000 million years ago.
- The first cellular forms of life were probably unicellular.
- All life forms were in water environment only.
- This theory of **biogenesis** from **non-living molecules** was accepted by majority.

EVOLUTION OF LIFE FORMS – A THEORY:

- Conventional religious literature tells us about the **theory of special creation**.
- **The theory of special creation has three connotations:-**
 - All the living organisms (species types) that we see today were created as such.
 - The diversity was always the same since creation and will be same in future.
 - Earth is about 4000 years old.

Challenge to special creation theory:

- Observation made during a sea voyage in a sail ship called H.M.S. Beagle round the world. Charles Darwin concluded that existing life forms share similarities to varying degrees not only among themselves but also with life forms that millions of years ago.
- Many such life forms exist anymore. There had been extinctions of different life forms in the years gone by just as new forms of life arose at different periods of history of earth.
- There has been gradual evolution of life forms.
- Any population has built in variation in characteristics.
- Those characteristics which enable some to survive better in natural conditions (climate, food, physical factors etc.) would outbreed others that are less-endowed to survive under such natural condition.
- Survival of the fittest. The fitness according to Darwin refers ultimately and only leaves more progeny than others.
- These, therefore, will survive more and hence are selected by nature. He called it as **natural selection**.
- **Alfred Wallace**, a naturalist who worked in **Malay Archipelago** had also come to similar conclusions around the same time.
- The geological history of earth closely correlates with the biological history of earth.

WHAT ARE EVIDENCES FOR EVOLUTION?

Paleontological evidence:

- Fossils are remained of hard parts of life-forms found in rocks.
- Different-aged rock sediments contain fossils of different life-forms who probably died during the formation of the particular sediment.
- They represent the extinct organisms (e.g. Dinosaurs).
- A study of fossils in different sedimentary layers indicates the geological period in which they existed.
- The study showed that life-forms varied over time and certain life forms are restricted to certain geological time-span.
- Hence new lives have arisen at different times in the history of earth.
- All this called **Paleontological evidence**.

Comparative anatomy and morphological evidence:

- Comparative anatomy and morphology shows similarities and differences among organisms of today and those that existed years ago.

Divergent evolution:

- Whale, bats, cheetah and human share similarities in the pattern of bones of forelimbs.

- These forelimbs perform different functions in these animals, they have similar anatomical structure – all of them have humerus, radius, ulna, carpals, metacarpals and phalanges in their forelimbs.
- Hence in these animals, the same structure developed along different directions due to adaptation to different needs.
- This is **divergent evolution** and these structures are **homologous**.
- Homology indicates common ancestry.
- Other examples of homologous organ are vertebrate hearts and brains.
- **Thorn of *Bougainvillea* and tendrils of *Cucurbita*** represent homology.

Convergent evolution:

- Wings of butterfly and of birds look alike.
- They are anatomically similar structure though they perform **similar function**.
- Hence **analogous** structures are a result of **convergent evolution**.
- **Eye of octopus and eye of mammals.**
- **Flippers of Penguins and Dolphins.**
- **Sweet potato (root modification) and potato (stem modification).**

Biochemical evidences:

- Similarities in proteins and genes performing a given function among diverse organisms give clues to common ancestry.

Embryological support for evolution:

- Proposed by **Ernst Haeckel** based upon observation of certain features during embryonic stage common to all vertebrates that are absent in adult.
- The embryos of all vertebrates including human develop a row of vestigial gill slits just behind the head but it is a functional organ only in fish and not found in any other adult vertebrates.
- This is **disproved** on careful study performed by **Karl Ernst von Baer**. He noted that embryos never pass through the adult stages of other animals.

Evolution by natural selection:

- Based on observation of moth population in England made in 1850.
- Before industrialization set in, it was observed that there were more white-winged moths on trees than dark-winged or melanised moths.
- After industrialization i.e. 1920 there were more dark-winged moths in the same area i.e. the proportion was reversed.

Evolution by anthropogenic action:

- Excess use of herbicides, pesticides etc. has only resulted in selection of resistant varieties in a much lesser time scale.
- This is also true for microbes against which we employ antibiotics or drugs against eukaryotic organisms/cell.

- Hence resistance organisms/cells are appearing in a time scale of months or years and not in centuries.
- These are the examples of evolution by anthropogenic action.
- Evolution is a stochastic process based on chance events in nature and chance mutation in the organisms.

WHAT IS ADAPTIVE RADIATION?

Darwin's Finches:

- In Galapagos Islands Darwin observed small black birds later called Darwin's Finches.
- He realized that there were many varieties of finches in the same island.
- All the varieties, he came across, evolved on the island itself.
- From the original seed-eating features, many other forms with altered beaks arose, enabling them to become insectivorous and vegetarian finches
- This process of evolution of different species in a given geographical area starting from a point and literally radiating to other areas of geography (habitats) is called **adaptive radiation**.

Australian marsupial:

- A number of marsupials each different from the other evolved from an ancestral stock. But all within the Australian island continent.
- When more than one adaptive radiation appeared to have occurred in an isolated geographical area (representing different habitats), one can call this **convergent evolution**.
- **Placental mammals** in Australia also exhibit adaptive radiation in evolving into varieties of such placental mammals each of which appears to be 'similar' to a corresponding **marsupial** (e.g.- placental wolf and Tasmanian wolf-marsupial).

BIOLOGICAL EVOLUTION:

- The essence of Darwinian Theory about evolution is natural selection.
- The rate of appearance of new forms is linked to the life cycle or the life span.
- There must be a genetic basis for getting selected and to evolve.
- Some organisms are better adapted to survive in an otherwise hostile environment.
- Adaptive ability is inherited.
- It has genetic basis.
- Fitness is the end result of the ability to adapt and get selected by nature.
- Branching descent and natural selection are the two key concepts of Darwinian Theory of Evolution.

Lamarck theory of evolution (theory of inheritance of acquired characters):

- French Naturalist Lamarck had said that evolution of life forms had occurred but driven by use and disuse of organs.

- He gave the example of Giraffes who in an attempt to forage leaves on tall trees had to adapt by elongation of their necks.
- They passed on this acquired character of elongated neck to succeeding generations.
- Giraffes, slowly over the years, came to acquire long necks.

MECHANISM OF EVOLUTION:

- In the first decade of twentieth century, **Hugo de Vries** based on his work on **evening primrose** brought forth the idea of **mutations**.
- Mutation is the large difference arising suddenly in a population.

How de Vries theory of mutation differs from Darwin's theory of natural selection?

- It is the mutation which causes evolution and not the minor variations that Darwin talked about.
- Mutations are random and directionless while Darwinian variations are small and directional.
- Evolution for Darwin was gradual while de Vries believed mutation caused speciation and hence called it **saltation** (single step large mutation).

HARDY – WEINBERG PRINCIPLE:

- In a given population one can find out the frequency of occurrence of alleles of a gene on a locus.
- This frequency is supposed to remain fixed and even remain the same through generations.
- Hardy-Weinberg principle stated it using algebraic equations.
- The principle states that allele frequencies in a population are stable and is constant from generation to generation.
- The gene pool (total genes and their alleles in a population) remains a constant. This is called **genetic equilibrium**:
- Sum total of all the allelic frequencies is 1.
- $(p + q)^2 = p^2 + 2pq + q^2 = 1$.
- When frequency measured, differs from expected values, the difference (direction) indicates the extent of evolutionary change.
- Disturbance in genetic equilibrium, or i.e. change of frequency of alleles in a population would then be interpreted as resulting in evolution.
- Five factors are known to affect Hardy-Weinberg equilibrium:
 - **Gene migration or gene flow.**
 - **Genetic drift.**
 - **Mutation.**
 - **Genetic recombination.**
 - **Natural selection.**
- **Gene migration:** When migrations of a section of population to another place occur, gene frequencies change in the original as well as in the new population.

New genes /alleles are added to the new population and these are lost from the old population.

- **Gene flow:** Gene migration occurs many time is termed as gene flow.
- **Genetic drift:** change in gene frequency takes place by chance.
- **Founder effect:** sometimes the change in allelic frequency is so different in the new sample of population that they became a different species. The original drifted population becomes **founder species** and the effect is called **founder effect**.

Operation of natural selection on different trait:

- Natural selection can lead to :
 - **Stabilization:** in which more individuals acquire mean character value.
 - **Directional changes** i.e. more individuals acquire value other than the mean character value.
 - **Disruption:** more individuals acquire peripheral character value at both ends of the distribution curve.

A BRIEF ACCOUNT OF EVOLUTION:

- About 2000 million years ago (mya) the first cellular forms of life appeared on earth.
- Some cellular form had the ability to release O₂.
- Slowly single cell organisms became multi-cellular life forms.
- By the time of 500 mya invertebrates were formed and active.
- Jawless fish probably evolved around 350 mya.
- Sea weeds and few plants existed probably around 320 mya.
- **Coelacanth** a lobe finned fish discovered in South Africa in 1938 evolved into first amphibians that lived on both land and water. These were **ancestors of modern day frogs and salamanders**.
- The amphibian evolved into reptiles.
- Reptiles' lays eggs which don not dry up in sun unlike those of amphibians.
- **Giant ferns** (pteridophytes) were present but they fell to form **coal deposits** slowly.
- Some of the reptiles went back into water to evolve into fish like reptiles probably 200 mya (*Ichthyosaurs*)
- The land reptiles were the dinosaurs.
- The biggest dinosaurs are *Tyrannosaurus rex* was about 20 feet in height and had huge fearsome dagger like teeth.
- About 65 mya the dinosaurs suddenly disappeared from the earth.
- Some of them evolved into birds.
- The first mammals were like shrews. Their fossils were small sized.
- Mammals were viviparous and protected their unborn young inside the mother's body.
- Due to **continental drift, pouched mammals** of Australia survived because of lack of competition from any other mammals.

ORIGIN AND EVOLUTION OF MAN:

- About 15 mya primates called *Dryopithecus* and *Ramapithecus* were existing.
- They were hairy and walked like gorillas and chimpanzees.
- *Ramapithecus* was more **man like** while *Dryopithecus* was more **ape-like**.
- Few fossils of man-like bones have been discovered in Ethiopia and Tanzania.
- Two mya *Australopithecines* probably lived in East African grasslands.
 - They hunted with stone weapons.
 - Essentially ate fruit.
- The first human-like being the hominid and was called *Homo habilis*.
 - Brain capacity between 650 – 800 cc.
 - They did not eat meat.
- Fossils discovered in Java in 1891 revealed the next stage i.e. *Homo erectus* about 1.5 mya.
 - Had large brain around 900 cc.
 - Probably ate meat.
- **Neanderthal man:**
 - Brain size 1400 cc
 - Lived in east and central Asia between 1,00,000-40,000 years back.
 - They used hides to protect their body.
 - Buried their dead.
- ***Homo sapiens:***
 - Arose in Africa and moved across continents and developed distinct races.
 - During ice age between 75,000-10,000 years ago modern *Homo sapiens* arose.
 - Pre historic cave art developed about 18,000 years ago.
 - Agriculture came around 10,000 years back and human settlement started.

CHAPTER-08

HUMAN HEALTH AND DISEASE

HUMAN HEALTH AND DISEASE :

- Improper functioning of one or more organs or systems of the body is adversely affected, gives rise to various signs and symptoms *i.e.* we have **disease**.
- Diseases which can easily transmit from one person to other by any means are called **infectious** or **communicable diseases**.
- Diseases which cannot be transmitted from one person to another are called **non-infectious** or **non-communicable diseases**.
- Disease causing organisms are said to be **pathogen**.

TYPHOID :

Pathogen: *Salmonella typhi* (bacterium).

Organs affected: small intestine, migrate to other organs through blood.

Method of transmission: contamination of food and water.

Symptoms:

- Sustained high fever (39°C to 40°C).
- Weakness, stomach pain, constipation, headache and loss of appetite.
- Intestinal perforation and death may occur.

Test: Typhoid fever could be confirmed by **Widal test**.

PNEUMONIA :

Pathogen: *Streptococcus pneumonia* and *Haemophilus influenzae*.

Organs affected: Alveoli of lungs, alveoli get filled with fluid.

Method of transmission:

- Inhaling the droplets/aerosols released by infected person.
- Sharing glasses and other utensils.

Symptoms:

- Fever, chills, cough and headache.
- In severe cases the lips and finger nails turn gray to bluish colour.

COMMON COLD :

Pathogen: Rhino viruses.

Organs affected: nose and respiratory passage

Method of transmission:

- Direct inhalation of droplets from infected person.
- Through contaminated objects like pen, books, cups, computer key board.

Symptoms:

- Nasal congestion and discharge, sore throat, hoarseness, cough.

MALARIA :

Pathogen: *Plasmodium* (*P. vivax*, *P. malariae*, *P. ovale*, *P. falciparum*).

Malignant malaria : caused by *P. falciparum* is fatal.

Organs affected: liver, RBC.

Method of transmission: by biting of female anopheles mosquito (vector).

Symptoms: high fever and chill, fever occurs on every alternate day, vomiting.

life cycle of malaria parasite:

- Life cycle of plasmodium starts with inoculation of **sporozoites** (infective stage) through the bite of infected female *Anopheles* mosquitoes.
- The parasite initially multiplied within the liver cells and then attack the red blood cells (RBCs) resulting in their rupture.
- There is release of a toxic substance called **hemozoin** from the ruptured RBCs which responsible for the **chill and high fever**.
- From the infected human the parasite enters into the body of Anopheles mosquito during biting and sucking blood.
- Further development takes place in the body of Anopheles mosquitoes.
- The female mosquito takes up **gametocytes** with the blood meal.
- Formation of gametes and fertilization takes place in the intestine of mosquito.
- The zygote develops further and forms thousands of sporozoites which migrated into the salivary gland of mosquito.
- When the mosquito bite another human sporozoites are injected.
- The malarial parasite requires two hosts – human and Anopheles, to complete their life cycle.

AMOEBIASIS (Amoebic dysentery) :

Pathogen: *Entamoeba histolytica* a protozoan parasite.

Organs affected: large intestine of man.

Method of transmission:

- House fly acts as mechanical carrier.
- Contamination water and food with faecal matter.

Symptoms:

- Constipation, abdominal pain and cramps.
- Stools with excess mucous and blood clots.

ASCARIASIS :

Pathogen: *Ascaris lumbricoids* (nematode).

Organs affected: intestine of man.

Method of transmission: Contaminated water, vegetables, fruits.

Symptoms:

- Internal bleeding, muscular pain, fever, anemia.
- Blockage of the intestinal passage.

FILARIASIS (ELEPHANTIASIS) :

Pathogen: Wuchereria (*W. bancrofti* and *W. Malayi*) is nematode parasite.

Organs affected: lymphatic vessels of the lower limbs, genital organs.

Methods of transmission: biting of infected female *Culex* mosquito.

Symptoms:

- Chronic inflammation of the organs where they live for many years.
- Abnormal swelling of lower limb, scrotum, penis.
- Hence the disease named as **elephantiasis** or **Filariasis**.

RING WORMS :

Pathogen: *Microsporum*, *Trichophyton* and *Epidermophyton* (fungi).

Organs affected: Skin, nails, folds of skin, groin.

Method of transmission:

- Acquired from the soil.
- Using towel, clothes or even comb of infected individuals.

Symptoms:

- Appearance of dry, scaly lesions in skin nails and scalp.
- Lesion accompanied with intense itching.
- **Heat** and **moisture** help these fungi to grow.

PREVENTION AND CONTROL OF INFECTIOUS DISEASES :

- Maintenance of personal and public hygiene is very important for prevention and control of many infectious diseases.
- **Personal hygiene** includes:
 - Consumption of clean drinking water, food vegetable fruits.
 - Keeping the body cleans.
- **Public hygiene** includes:
 - Proper disposal of waste and excreta
 - Periodic cleaning and disinfection of water reservoirs, pools, cesspools.
 - Standard practices of hygiene in public catering.
- In case of **air-borne diseases**, close contact with the infected persons or their belongings should be avoided.

For vector borne diseases :

- To control or eliminating the vectors and the breeding places.
- Avoiding stagnation of water in and around residential areas.
- Regular cleaning of household coolers.
- Use of mosquito nets.
- Introducing fishes like *Gambusia* in pond that feeds on mosquito larvae.
- Spraying of insecticides in ditches, drainage area and swamps.
- Window and doors must be fitted with wire mesh.
- All these precautions are use full for vector borne disease like dengue and Chickungunya, malaria and filarial etc.

Immunization:

- By massive immunization there is complete eradication of disease like smallpox.
- Diseases like polio, diphtheria, pneumonia, and tetanus have been controlled in large extent.

IMMUNITY:

- The overall ability of the host to fight the disease causing organism by immune system is called **immunity**.
- There are two types of immunity:
 - Innate Immunity.
 - Acquired Immunity.

Innate (non-specific) immunity:

- Called **inborn immunity**.
- Always available to protect out body.
- This is called the **first line of defense**.
- Consists of various barriers that prevent entry of foreign agents into the body.
- If enters they are quickly killed by some other components of this system.
- Different types of barriers are as follows:

Physical barriers:

- Skin is the main barrier which prevents entry of micro-organism.
- Mucous coating of the epithelium lining of respiratory, gastrointestinal and urinogenital tracts helps in trapping microbes.

Physiological barriers:

- **Acidity** of the stomach kills most ingested microbes.
- **Lysozyme** in tears, saliva, and snot kills bacteria by digesting bacterial wall.
- **Pyrogen** released by WBC raise body temperature to prevents growth of microbes in out body.
- **Interferon** induces antiviral state in non-infected cells.

Phagocytic barrier:

- **Polymorpho-nuclear leukocytes** (PMNL-neutrophils), **macrophages**, and **natural killer cells** in the blood and tissues kill pathogen by **phagocytosis**.

Inflammatory barrier:

- When there is injury to the tissue there is release of histamine and prostaglandins by the mast cells.
- Due to vasodilation there is leakage of vascular fluid containing serum proteins with antibacterial activity.
- Further there is influx of Phagocytic cells into the affected area.

Acquired (specific) immunity:

- It is also known as **adaptive immunity**.
- This immunity developed **after birth** when encountered with pathogen.
- It supplements the immunity provided by the innate immunity.
- Acquired immunity has following **unique features**:
 - **Specificity**: distinguish specific foreign molecules.

- **Diversity:** recognize vast variety of foreign molecules.
- **Discrimination between self and non-self:** it is able to recognize and respond to molecules that are foreign or non-self. It will not respond to our own cell or molecules.
- **Memory:** after responding to the foreign microbes and elimination, this immune system retains the memory of that encounter (**primary immuneresponse**). The second encounter with the same microbe evokes a heightened immune response. (**Secondary immune response**)
- Acquired immunity is carried out by two special types of lymphocytes:
 - **B-lymphocytes.**
 - **T-lymphocytes.**
- The B-lymphocytes produce a group of proteins in response to pathogen into the blood to fight with them called **antibody**.
- T-lymphocytes do not produce antibody but help B-cells to produce them.

Structure of antibody:

- Each antibody has four polypeptide chains.
- Two small chains called light chains.
- Two longer chains called heavy chains.
- Antibody represented as H₂L₂.
- Different classes of antibody produced in our body are IgA, IgM, IgD, IgE and IgG.

AMI Vs CMI :

- Immune response by the B-cells by production of antibody is called **Antibody mediated immune response** or **humoral immune response**.
- Immune response by T-cells is by activation of **cytotoxic killer** cells which detects and destroys the foreign cells and also cancerous cells called **cell mediated immune** response.
- Rejection of organs transplants are due to T-lymphocytes.
- Tissue matching, blood group matching are essential for organ transplantation.
- Even after tissue typing immune-suppressants is required before and after transplantation.

Active immunity:

- When the host is exposed to **antigens**, which may be in the form of living or dead microbes or other proteins, antibodies are produced in the host body.
- Active immunity is slow and takes time to give its full effective response.
- Injecting microbes deliberately during immunization or infection of microbes naturally induce **active immunity**.

Passive immunity:

- Readymade antibodies are directly given to protect the body against foreign agents.

- **Colostrums** of mother contain abundant **antibody (IgA)** to protect the child.
- Foetus receives some antibody (IgG) from mother during pregnancy.

Vaccination and Immunization:

- The principle of immunization or vaccination is based on the property of 'memory, of the immune system.
- In vaccination, a preparation of antigenic protein of pathogen or inactivated/weakened pathogen (vaccine) is introduced into the body.
- The antibodies produced in the body against vaccine (antigen) would neutralize the pathogenic agents during actual infection.
- The vaccines also generate memory B and T-cells that recognize the pathogen quickly on subsequent exposure.

Passive immunization:

- Preformed antibody or antitoxin injection for specific antigen.
- Injection of antivenin for snake bites to counter the snake venom.

Vaccine production:

- Recombinant DNA technology has allowed the production of antigenic polypeptide of pathogen in **bacteria and yeast**.
- Vaccine produced by this approach allows large scale production of antigen for immunization. E.g.- **hepatitis-B** produced from yeast.

Allergies:

- The exaggerated response of the immune system to certain antigens present in the environment is called **allergy**.
- The substance to which such immune response is produced is **allergen**.
- **IgE** is produced during allergic reactions.
- Common allergens are dust, pollen, animal dander etc.
- Common symptoms are sneezing, watery eyes, running nose etc.
- Allergy is due to release of **histamine** and **serotonin** from the **mast cells**.
- Drugs like anti-histamine, adrenalin and steroid quickly reduce symptoms of allergy.

Auto immunity:

- Memory based acquired immunity able to distinguish foreign molecules or cells (pathogen) from self-cells.
- Sometimes due to genetic and other unknown reasons the body attacks self-cells. This results in damage to the body cells and is called auto-immune disease. E.g. **Rheumatoid arthritis, Multiple sclerosis**.

Immune system in our body:

- The immune system consists of
 - **Lymphoid organs**

- **Lymphoid tissues**
- **T and B-cells.**
- **Antibodies.**
- Immune system recognizes the foreign antigens, responds to them and remembers them.
- The immune system also plays important role in:
 - Allergic reaction
 - Auto-immuno diseases and
 - Organ transplantation.
- **Primary lymphoid organs:** bone marrow and thymus where **production and maturation** of lymphocytes take place.
- **Secondary lymphoid organs:** spleen, tonsil, lymph node, Payer's patches of small intestine and appendix where **proliferation** and **differentiation** of lymphocyte take place.
- **Bone marrow** is the main lymphoid organ where all blood cell including lymphocytes are produced.
- **Thymus** is a bilobed organ located near the heart, beneath the breastbone.
- B-lymphocytes are produced and matured in bone marrow.
- T-lymphocytes are produced in bone marrow but matured in thymus.
- **The spleen**
 - Large bean shaped organ mainly contain **lymphocytes** and **phagocytes.**
 - Acts as a filter of the blood by trapping blood-borne micro-organisms.
 - Spleen is also serves as the large reservoir of **erythrocytes.**
- **Lymph node:**
 - Small solid structure located at different points along the lymphatic system.
 - Traps the micro-organisms or other foreign antigens.
 - Antigen trapped into the lymph node responsible for activation and differentiation of lymphocytes and cause immune response.
- **Mucosal associated lymphoid tissues (MALT):**
 - Located within the lining of major tract (respiratory, digestive and urogenital tracts)
 - It constitutes 50% of lymphoid tissues.

AIDS:

- Stands for **Acquired Immuno Deficiency Syndrome.**
- Deficiency of immune system that acquired during life time and not congenital disease.
- Syndrome means a group of symptoms.
- AIDS was first reported in 1981.
- AIDS is caused by HIV (Human Immuno deficiency Virus).
- HIV is **retrovirus**, having RNA as the genetic material.

Method of transmission:

- Sexual contact with infected persons.

- Transfusion of contaminated blood and blood products.
- Sharing infected needles as intravenous drug user.
- From infected mother to the foetus through placenta.

Life cycle of HIV:

- After getting into the body the HIV enters into **macrophages** or **T-helper** cells.
- The viral RNA genome replicated to form viral DNA with the enzyme called **reverse transcriptase**.
- The viral DNA gets incorporated into the host cell's DNA by an enzyme called **integrase** and directs the infected cells to produce virus particle.
- The macrophage continues to produce virus and acts as HIV factory.
- Virus released from macrophage attack T-helper cells.
- There is progressive reduction in the number of T-helper cells.
- Due to reduction of T-helper cells the person starts suffering from infections of other virus, fungi and even parasites like *Toxoplasma*.
- The patient becomes immuno deficient and more prone to other disease.

Diagnosis:

- ELISA (**Enzyme Linked Immuno Sorbent Assay**).

Prevention of AIDS:

- AIDS has no cure, prevention is the best option.
- Safe blood for transfusion
- Use of disposable needles
- Free distribution of condoms.
- Prevention of drug abuse
- Advocating safe sex and promoting regular checkup.

CANCER:

- Uncontrolled cell division leads to production of mass of cell called cancer.
- Cancerous cell lost the property of **contact inhibition**.
- Cancerous cell just continue to divide giving rise to masses of cell called **tumors**.
- **Benign tumors:**
 - Normally remain confined to their original location
 - Do not spread to other location.
 - Cause little damage.
- **Malignant tumors:**
 - Mass of proliferating cells called neoplastic or tumor cells.
 - These cells grow very rapidly.
 - Invade and damage surrounding tissues.
 - These cells actively divide and grow; they also starve the normal cells.
 - Cancerous cells escape from the site of origin and moves to distant place by blood, wherever they get lodged make the normal cell cancerous. This property is called **metastasis**.

Causes of cancer:

- Normal cells transformed into cancerous neoplastic cells by physical, chemical and biological agents. These agents are called **carcinogen**.
- **Physical agents:** ionizing radiation like X-rays, gamma rays non-ionizing radiations like UV-rays.
- **Chemical agents:** Tobacco smoke, sodium azide, Methyl ethane sulphonate.
- **Biological agents:**
 - Cancer causing viruses called **oncogenic viruses** have a gene called **viral oncogenes**, induce transformation of neoplastic cells.
 - **Cellular oncogenes** (C-oncogene) or **proto oncogenes** in normal cells when activated lead to oncogenic transformation of the normal cells.

Cancer detection and diagnosis:

- Biopsy and histopathological study of the tissues.
- Radiography like X-rays, CT (computerized tomography).
- MRI (magnetic resonance Imaging).
- Presence of antibodies against cancer-specific antigen.

Treatment of cancer:

- Surgery
- Radiation therapy
- Immunotherapy
- Chemotherapy
- Cryosurgery
- Laser therapy.
- **α -interferone** a response modifier used to detect the cancer.

DRUGS AND ALCOHOL ABUSE :

Opioid:

- The drugs which bind to specific opioid receptor present in **central nervous system** and **gastrointestinal tract**.
- **Heroin** commonly called **smack**, chemically **diacetylmorphine**.
- It is white, odourless, bitter crystalline compound.
- Obtained by **acetylation of morphine**.
- Extracted from latex of poppy plant *Papaver somniferum*.
- Generally taken by snorting and injection.
- Heroin is depressant and slows down body function.

Canabinoids:

- Group of chemicals that interact with the cannabinoid receptors of brain.
- Obtained from inflorescence of *Cannabis sativa*.
- Flower top, leaves and resin of cannabis plant are used in various combinations to produce **marijuana, hashish, charas and ganja**.
- Generally taken by inhalation and oral ingestion
- Effects on **cardiovascular system** of the body.

Cocaine:

- Coca alkaloid or cocaine is obtained from coca plant *Erythroxylum coca*.
- It interferes with transport of neurotransmitter **dopamine**.
- Cocaine is commonly called as **coke** or **crack** is usually snorted.
- Potent stimulating effect on central nervous system.
- Produces sense of **euphoria** and increased energy.
- Excessive dosage causes **hallucination**.
- Other plants with hallucinogenic properties are :
 - *Atropa belladonna*
 - *Datura*.
- Canabinoids are also being abused by some sportspersons.

Medicinal use of drugs:

- Barbiturates, amphetamines, benzodiazepines, lysergic acid diethyl amide (LSD) used as medicines to help patients cope with mental illnesses, depression and insomnia.
- Morphine is a very effective sedative and painkiller used for surgery patient
- Plant product with hallucinogenic property have used as folk-medicine, religious ceremonies and rituals.

Tobacco:

- It is smoked, chewed or used as a snuff.
- Tobacco contains **nicotine an alkaloid**.
- Nicotine stimulates Adrenal glands to raise blood pressure and increased heart rates.
- Smoking tobacco is associated with cancer of lung, urinary bladder, and throat, bronchitis, emphysema, coronary heart disease, gastric ulcer etc.
- Smoking increased Carbon monoxide content of blood which reduces oxygen carrying capacity of hemoglobin.
- Tobacco chewing is associated with cancer of oral cavity.

Adolescence and Drug/Alcohol Abuse:

- The period between 12-18 years of age may think of an adolescent period.
- Adolescent is a bridge linking childhood and adulthood.
- Curiosity, need for adventure and excitement, and experimentation, are the common cause of drug/alcohol abuse.

Addiction and dependence:

- Addiction is a psychological attachment to certain effects such as euphoria and a temporary feeling of well-being associated with drugs and alcohol.
- With repeated use of drugs the tolerance level of the receptors present in our body increases. Consequently the receptors respond only to higher doses of drugs or alcohol leading to greater intake and **addiction**.
- Use of drugs even once, can be a fore runner to addiction.

- Dependence is the tendency of the body to manifest a characteristic and unpleasant **withdrawal syndrome** if regular dose of drugs/alcohol is abruptly discontinued.
- Withdrawal syndrome characterized by anxiety, shakiness, nausea and sweating.

Effects of Drug / Alcohol Abuse:

- Immediate effects are reckless behavior, vandalism and violence.
- Excessive doses of drugs may lead to coma and death due to respiratory failure, heart failure or cerebral hemorrhage.
- Warning sign of drug and alcohol abuse among youth include:
 - Drop in academic performance,
 - Unexplained absence from school/college.
 - Lack of interest in personal hygiene
 - Withdrawal, isolation, depression fatigue, aggressive and rebellious behavior.
 - Deteriorating relationship with family and friends.
 - Loss of interest in hobbies.
 - Change in eating and sleeping habits.
 - Fluctuation in weight and appetite.
- Intravenous drug user more prone to acquire infections like AIDS and hepatitis.
- The chronic use of drugs and alcohol damages nervous system and cause of **liver cirrhosis**.
- Use of drug and alcohol during pregnancy affect the foetus.

Prevention and control:

- Avoid undue peer pressure.
- Education and counseling.
- Seeking help from parents and peers.
- Looking for danger signs.
- Seeking professional and medical help.

Abbreviations:

- PMNL : Polymorpho Nuclear Leukocytes
- CMI : Cell Mediated Immunity
- ELISA : Enzyme Linked Immuno Sorbent Assay
- HLA : Human Leukocyte Antigen
- MALT : Mucosal Associated Lymphoid Tissue
- SCID : Severe Combined Immuno Deficiency
- NACO : National AIDS Control Organization
- MRI : Magnetic Resonance Imaging
- CT : Computerized Tomography.

CHAPTER-09

STRATEGIES FOR ENHANCEMENT IN FOOD PRODUCTION

ANIMAL HUSBANDRY:

- The agricultural practice of breeding and raising livestock.
- Deals with care and breeding of livestock like buffaloes, cows, pigs, horses, cattle, sheep, camels, goats etc.
- Extended form includes poultry farming and fisheries.
- Fisheries include rearing, catching selling etc. of fish, molluscs (shell fish) and crustaceans (prawns, crabs etc.)

Dairy farm management:

- Dairying is the management of animals for milk and its products.
- Use of improved breed of cow such as **Jersey**.
- Well housed.
- Should have adequate water
- Maintained disease free
- Feeding should be scientific manner.
- Quantity and quality of fodder
- Stringent cleanliness and hygiene.
- Regular visit by a veterinary doctor would be mandatory.

Poultry farm management:

- Poultry is the class of domesticated fowl (birds) used for food and eggs.
- Selection of disease free and suitable breeds.
- The improved breed of poultry is **Leghorn**.
- Proper and safe farm conditions
- Proper feed and water
- Hygiene and health care.

Animal breeding:

- A group of animals related by descent and similar in most characters like general appearance, features size, configuration, etc, are said to be a **breed**.
- **Inbreeding:** crosses between same breed.
- **Outbreeding:** crosses between different breeds.

Inbreeding:

- Mating of more closely related individuals within the same breed for 4-6 generations.
- Superior male and female is identified and mated in pairs.
- Progeny obtained are evaluated and superior males and females among them are identified for further mating.

- **More milk per lactation** is the criteria for superior female for cow and buffalo. Superior male which gives rise to **superior progeny**.
- Inbreeding increases **homozygosity**.
- Inbreeding is necessary to create **pure line** in any animal.
- Inbreeding exposes harmful recessive genes that are eliminated by selection.
- Helpful in **accumulation of superior genes**.
- Continuous inbreeding reduces fertility and even productivity. This is called **inbreeding depression**.

Outbreeding:

- Out-breeding is the breeding of **unrelated animals**.

Out-crossing:

- Mating of animals within the same breed but having no common ancestor on either side of their pedigree upto 4-6 generations.
- Offspring of such mating is called out-cross.
- A single outcross often helps to overcome inbreeding depression.

Cross-breeding:

- Superior male of one breed are mated with superior female of another breed.
- It allows the desirable qualities of two different breeds to be combined.
- **Hisardale** is a new breed of sheep developed in Punjab by crossing **Bikaneri ewes** and **Marino rams**.

Interspecific hybridization:

- Male and female of two different species are mated.
- The progeny may combine desirable features of both the parents (e.g. - mule).

Artificial insemination:

- **Controlled breeding experiments** are carried out using **artificial insemination**.
- The semen is collected from the male and injected into the reproductive tract of the selected female by the breeder.
- The semen collected may be used immediately or can be frozen for later use. The semen can be transported in a frozen form to where the female is housed.

Multiple Ovulation Embryo Transfer Technology:

- It is used to improve chances of successful production of hybrids.
- Cow is administered hormones with **FSH-like activity**
- induce follicular maturation and **super ovulation**
- Production of **6-8 eggs** instead of one egg per cycle.
- The female is either mated with an elite bull or **artificially inseminated**.
- Non-surgical recovery of fertilized eggs at **8-32 cells** stages.
- Each one transferred to **surrogate mother**.
- The **genetic mother** is available for another round of super ovulation.
- This technology is used to increase **herd size** in a short time.

Bee-keeping:

- Bee-keeping is called apiculture.
- It includes maintenance of hives of honeybees for production of honey.
- Honey is a food of high nutritive values and also used as medicine.
- Honey bees also produce beeswax which has many uses in industry, like preparation of cosmetics and polishes of various kinds.
- Bee-keeping practiced in areas with sufficient bee pastures of some wild shrubs, fruit orchards and cultivated crops.
- *Apis indica* is the most common species used in apiculture.
- The following points are important **for successful bee-keeping**:
 - Knowledge of the nature and habits of bees.
 - Selection of suitable location for keeping the beehives.
 - Catching and hiving of swarms (group of bees)
 - Management of beehives during different seasons.
 - Handling and collection of honey and of beeswax.
 - Bees are the pollinators for many plants, hence keeping beehives in crop fields during flowering period, increases pollination and improves honey yield.

Fisheries:

- Fishery industry related to catching, processing or selling of fish, shellfish or other aquatic animals.
- Common **fresh water fish**: *Catla*, *Rohu* and common carp.
- Common **marine fishes**: Hilsa, Sardines, Mackerel and Pomfrets.
- Production of aquatic plants and animals, both freshwater and marine water is increased by **Pisciculture** and **aquaculture**.
- Increasing production of fish is called **Blue revolution**.

PLANT BREEDING:

- Plant breeding as a technology has helped increase yields to a large extent.
- **Green revolution** was not only responsible to meet the national requirement of food, but also helped us even to export it.
- Green revolution is due to plant breeding techniques which developed high yielding varieties of wheat, rice, maize etc.

What is plant breeding?

- Plant breeding is the purposeful manipulation of plant species in order to create desired plant types that are better suited for cultivation, give better yields and are disease resistant.
- Classical plant breeding involved crossing or hybridization of pure lines followed by artificial selection to produce plants with desirable traits of higher yield, nutrition and resistance to diseases.

Trait for which plant breeding done:

- Trait or characters that the breeders have tried to incorporate into the plants are as follows:
 - Increased crop yield
 - Improve quality
 - Increased tolerance to environmental stresses (salinity, extreme temperature, and drought).
 - Resistant to pathogens (viruses, fungi, and bacteria)
 - Increase tolerance to insect pest.

Steps in plant breeding techniques:

- **Collection of variability:**
 - Genetic variability is the root of any breeding programme.
 - Pre-existing genetic variability is available from wild relatives of crop.
 - Collection and preservation of all the different wild varieties, species and relatives of the cultivated species.
 - Evaluation for their characteristics.
 - The entire collection (of plants /seeds) having all the diverse alleles for all genes in a given crop is called **germplasm collection**.
- **Evaluation and selection of parents:**
 - The germplasm is evaluated so as to identify plants with desirable combination of characters.
 - The selected plants are multiplied and used in hybridization.
 - Pure line is created wherever desirable and possible.
- **Cross hybridization among the selected parents:**
 - Cross hybridization of two selected parent by emasculation and bagging, to produce hybrid of combined character of both parents.
 - For example high protein quality of one parent may need to be combined with disease resistance from another parent.
 - Usually one in few hundred to a thousand crosses offspring shows desirable combinations.
- **Selection and testing of superior recombinants:**
 - Selection is done from the progeny of hybrids produced by **cross hybridization**.
 - It requires careful scientific observations and evaluation of progeny.
 - Hybrid plants that are superior to both of the parents are selected.
 - These hybrids are self-pollinated for several generations till they reach a state of uniformity (homozygosity).
- **Testing, release and commercialization of new cultivars:**
 - Selected pure lines are evaluated for their yield and other agronomic traits of quality, disease resistance etc.
 - This evaluation is done in the research fields and recording their performance under ideal fertilizer, irrigation
 - Testing is done in the farmers 'fields' at least for three generations.
 - The material is compared with best available local crop cultivar.

Product (Wheat and Rice):

- Production of wheat and rice increased in many folds due to **semi-dwarf variety** during the period of 1960-2000.
- Nobel laureate **Norman E. Borlaug**, at international centre for wheat and Maize improvement in Mexico, developed semi-dwarf variety of wheat.
- In 1963 several varieties such *Sonalika* and *Kalyan Sona* high yielding variety was introduced in India.
- Semi-dwarf rice was derived from **IR-8** (developed at International Rice Research Institute (IRRI), Philippines) and **Taichung Native –I** (from Taiwan).
- **Jaya** and **Ratna**, semi dwarf rice variety developed in India.

Product (sugarcane):

- *Saccharum barberi* of north India with poor sugar content and yield crossed with *Saccharum officinarum* with thick stems and higher sugar content to produce sugar cane of high yield, thick stems and high sugar.

Plant breeding for Disease Resistance:

- A wide range of fungal, bacterial and viral pathogens affects the yield of cultivated crop species, they lessen the yield upto 20-30 % sometime total.
- Development of cultivars resistant to diseases is essential.
- These also reduce the dependence on the fungicide or insecticide.
- Pathogen causing different diseases in plants:
 - **Fungi:** brown rust of wheat, red rot of sugarcane, late blight of potato.
 - **Bacteria:** black rot of crucifer,
 - **Virus:** tobacco mosaic, turnip mosaic etc.

Method of breeding for disease resistant:

- Screening of germplasm for resistance sources.
- Hybridization of selected parent.
- Selection and evaluation of hybrids
- Testing and release of new varieties.

Mutation breeding:

- Genetic variability is created by induced mutation (by application of mutagen, chemical or physical).
- Screening and selection of the parent with desirable character used as a parental plant for breeding programme.
- In mung bean, resistance to yellow mosaic virus and powdery mildew were induced by mutation.
- Natural wild varieties of plant with disease resistant genes are available but low yield.
- These wild varieties are hybridized with high yield varieties to make them disease resistant and also high yielding variety.

- Resistance to yellow mosaic virus in bhindi (*Abelmoschus esculentus*) was transferred from a wild species and resulted a new variety of *A. esculentus* called **Parbhani kranti**.

Plant breeding for Developing Resistant to insect pest:

- Another major cause of large scale destruction of crop plants is the insect and pest infestation.
- Insect resistance in host crop is due to morphological, biochemical or physiological characteristics.

Characters that make the plant resistance to insect pest:

- **Hairy leaves** in several plants make them resistant to insect pest.
- **Solid stem** in wheat lead to non-preference by stem sawfly.
- **Smooth leaves** and **nectar-less** cotton variety do not attract **bollworms**.
- **High aspartic acid, low nitrogen** and sugar content in maize make them resistant to **stem borers**.
- Steps for developing insect pest resistant variety of crop are same as others.
- The resistant variety selected either form the wild variety of from other available cultivars.

Plant breeding for Improved Food quality:

- Around three billion people suffer from micronutrient, protein and vitamin deficiencies called **Hidden hunger**.
- Diets lacking essential micronutrients particularly iron, vitamin A, iodine or zinc-increase the risk of diseases; reduce life span, reduce mental ability.
- **Biofortification** – Breeding crops with higher levels of vitamins and minerals or higher protein and healthier fats is the most practical means to improve public health.
- **Objectives of biofortification is to improve :**
 - **Protein content and quality.**
 - **Oil content and quality**
 - **Vitamin content and**
 - **Micronutrient and mineral content.**
- Hybrid maize developed with twice the amount of amino acids **lysine** and **tryptophan**, compared with existing maize.
- Wheat variety Atlas 66, having high protein content has been used as donor for improving cultivated wheat.
- **Iron fortified** rice developed with five times more iron than existing variety.
- IARI New Delhi developed:
 - Vitamin-A enriched carrots, spinach pumpkin.
 - Vitamin-C enriched bitter gourd, bathua mustard tomato.
 - Iron and Calcium enriched spinach and bathua
 - Protein enriched beans- broad, lablab, French and garden peas.

SINGLE CELL PROTEIN (SCP):

- More than 25% of human population is suffering from hunger and malnutrition.
- One of the alternating sources of proteins for animal and human is **SCP**.
- Production of biomass (protein) in large scale using micro-organism and low cost raw material is called **single cell proteins**.
- Microbes like *Spirulina* grown on waste water from potato processing plants, straw, molasses, animal manure and even sewage, to produce large quantities of biomass with rich in protein, mineral, fats, carbohydrate and vitamins.
- It has been calculated that 250 kg cow produces 200gm of protein per day. In the same period 250gm of micro-organism like *Methylophilus methylotophus*, expected to produce 25 tons of protein.
- Another example is production of biomass like mushroom from straw.

TISSUE CULTURE:

- Potency or power or ability of a single cell/ explants to develop a whole plant is called **totipotency**.
- This property led the scientist able to develop whole plant from **explants** – any part of plant, cell grown in a test tube, under sterile condition in special nutrient medium.
- The nutrient medium provides a carbon source such as sucrose. Inorganic salts, vitamins amino acids and growth regulator like auxin, cytokinin.
- The method of production of thousands of plants through tissue culture is called **micro-propagation**.
- Plants grown by micro-propagation are genetically identical called **soma-clones**.

Application of tissue culture:

- Production of large number of plant from small tissue or single cell.
- Production of genetically identical plants (soma-clones)
- Recovery of healthy plants from diseased plants by **meristem** culture. Although the plant infected with virus, the meristem is free of virus.

Somatic hybridization:

- Isolation of single cells from the plants.
- Digestion of cell wall to get protoplast of different donor cells, by use of cellulase and pectinase.
- Two protoplasts of two different plants with desirable character are fused to form hybrid protoplast, either by using electric field or by PEG (polyethylene glycol).
- These hybrids are called **somatic hybrid** and the process called **somatic hybridization**. E.g. – production of pomato plant from potato and tomato.

Abbreviation:

- ET : Embryo Transfer
- IARI : Indian Agricultural Research Institute
- IRRI : International Rice Research Institute
- ICAR : Indian Council of Agriculture Research
- MOET : Multiple Ovulation Embryo Transfer
- NDRI : National Dairy Research Institute.

P. K. Singh

CHAPTER-10

MICROBES IN HUMAN WELFARE

MICROBES IN HUMAN WELFARE

- Microbes are diverse – protozoa, bacteria, fungi and microscopic plants viruses, viroids and also prions (proteinous infectious agents).
- Microbes like bacteria and fungi can be grown in nutrient media to form colonies and can be seen in naked eyes.
- Some microbes' causes diseases and some are useful for human being.

MICROBES IN HOUSEHOLD PRODUCTS :

Lactic acid Bacteria:

- Lactic acid Bacteria (LAB) grow in milk and convert it to curd.
- LAB produces acids that coagulate and partially digest milk proteins.
- A small amount of curd added to fresh milk as inoculums or starter.
- LAB improves nutritional quality of milk by increasing vitamin B₁₂
- LAB plays very important role in checking disease causing microbes.
- Dough, used to make *dosa* and *idli* is also fermented by bacteria.
- The puffed-up appearance of dough is due to the production of CO₂.
- Baker's yeast (*Saccharomyces cerevisiae*) is used to making bread.
- 'Toddy' a traditional drink is made by fermentation of sap from palms.
- Large holes in 'Swiss cheese' are due to production of large amount of CO₂ by a bacterium named *Propionibacterium sharmanii*.
- The '**Roquefort cheese**' is ripened by specific fungi, which gives specific flavor.

MICROBES IN INDUSTRIAL PRODUCTS :

- Microbes are used in industry to synthesize a number of products
- Beverages and antibiotics are some examples.
- Microbes are grown in very large vessels called **fomenters**.

Fermented Beverages:

- **Yeasts** are used for production of beverages like wine, beer, whisky, brandy or rum.
- *Saccharomyces cerevisiae* commonly called '**brewer's yeast**' used for fermenting malted cereals and fruit juices to produce **ethanol**.
- The type of raw material used for fermentation and the processing, different types of alcoholic drinks are produced.
- **Wine and beer** are produced **without distillation**.
- **Whisky, brandy** and **rum** are produced by **distillation** of the fermented broth.

Antibiotics:

- **Antibiotics** are the chemical substances which are produced by some microbes and can kill or retard the growth of other microbes.

- The first antibiotic discovered is the penicillin, from a mould (fungus) *Penicillium notatum*.
- Antibiotics have greatly improved our capacity to treat deadly diseases such as plague, whooping cough, Diphtheria and leprosy.

Chemicals, Enzymes and other Bioactive Molecules:

- *Aspegillus niger* (a fungus) produces **citric acid**.
- *Acetobacter aceti* (a bacterium) produce **acetic acid**.
- *Clostridium butylicum* (a bacterium) produce **butyric acid**.
- *Lactobacillus* (a bacterium) produces **lactic acid**.
- *Saccharomyces cerevisiae* (yeast) used for production of **ethanol**.
- **Lipases** are used in detergent produced by microbes.
- **Pectinase, proteases and cellulase**, make bottled fruit juices clearer.
- **Streptokinase** produced by **Streptococcus** used as a ‘**clot buster**’, for removing clots from the blood vessels.
- **Cyclosporin-A** produced by a fungus called *Trichoderma polysporum* used as **immunosuppressive agent** in organ transplantation.
- **Statins** produced by *Monascus purpureus* used as blood cholesterol lowering agents. It acts as competitive inhibitor for the enzyme responsible for synthesis of cholesterol.

MICROBES IN SEWAGE TREATMENT :

- The waste water generated in cities and town containing human excreta. This water (municipal water) is called sewage.
- Before disposal to the natural body sewage is treated in sewage treatment plants (STPs) to make it less polluting.
- Treatment is done by heterotrophic microbes naturally present in sewage.

Primary treatment:

- Involves the physical removal of particles – large and small from sewage through filtration and sedimentation.
- Initially **floating debris** is removed by **sequential filtration**.
- The **grit** (soil and small pebbles) are removed by **sedimentation**.
- All solids that settle form the **primary sludge**, and the supernatant forms the effluents.
- The effluents are from the primary settling tank taken for secondary treatment.

Secondary treatment or Biological treatment:

- The primary effluent is passed into large aeration tanks.
- This allows vigorous growth of useful aerobic microbes into **flocs**.
- The growth of microbes consumes the major part of the organic matter in the effluent. This significantly reduces the **BOD (biochemical oxygen demand)** of the effluent.

- BOD refers to the amount of oxygen required to oxidize total organic matter by bacteria, present in one liter of water.
- BOD is the measures of the organic matter present in the water.
- **Greater the BOD of the waste water more is its polluting potential.**
- Once the BOD of sewage reduced significantly, the effluent is then passed into the settling tank where the bacterial 'flocs' are allowed to sediment. This sediment is called **activated sludge**.
- Small part of activated sludge is pumped back to aeration tank to serve as the inoculums.
- The remaining sludge is pumped into **anaerobic sludge digester**.
- In the anaerobic sludge digester there is other kinds of bacteria which grow anaerobically, digest the bacteria and fungi in the sludge.
- During this digestion bacteria produce **biogas** (mixture of methane, hydrogen sulphide and carbon dioxide)
- The effluent from the secondary treatment plant is released into natural water body like rivers and streams.
- **Ganga Action Plan** and **Yamuna Action Plan** initiated by Ministry of Environment and Forest to save these major rivers of our country.
- It is proposed to build a large number of sewage treatment plants so that only treated sewage may be discharged into the rivers.

MICROBES IN PRODUCTION OF BIOGAS :

- **Biogas** is a mixture of gases (predominantly **methane**) produced by the microbial activity and is used as fuel.
- Certain bacteria grow anaerobically on cellulosic material, produce large amount of methane along with CO₂ and H₂S. These bacteria are collectively called **methanogens**. One common bacterium is *Methanobacterium*.
- These bacteria present in the rumen of cattle, plays essential role in nutrition of cattle by digesting cellulose. Hence the excreta (dung) used for the production of biogas.

MICROBES AS BIOCONTROL AGENT :

- Bio-control refers to the use of biological methods for controlling plant diseases and pests.
- Effect of use of chemical, insecticide and pesticide to control disease and pests:
 - These chemicals are toxic and extremely harmful to human beings and animals.
 - Polluting our environment (soil, ground water), fruits and vegetables.
 - Soil is polluted through use of weedicides to remove weeds.

Biological control of pest and disease:

- Use of bio-control measures will greatly reduce our dependence on toxic chemical and pesticides.
- The Ladybird and Dragonflies are used to get rid of aphids and mosquitoes.

- *Bacillus thuringiensis* (Bt) used to control butterfly caterpillars.
- Dried spores are mixed with water and sprayed onto vulnerable plants where these are eaten by the insect larvae.
- In the gut of the larvae, the toxin is released and the larvae get killed.
- **Trichoderma** a free living fungus used to control several plant pathogens.
- Baculoviruses are pathogen that attack insects and other arthropods
- The majority of **baculoviruses** used as biological control agents are in the genus *Nucleopolyhedrovirus*.
- These viruses are excellent candidates for species-specific, narrow spectrum insecticidal application.
- They have no negative impacts on plants, mammals, birds, fish etc.
- This is very use full in integrated pest managements (IPM).

MICROBES AS BIOFERTILIZERS :

- **Biofertilizers** are organisms that enrich the nutrient quality of the soil.
- Main biofertilizers are the bacteria, fungi and cyanobacteria.
- *Rhizobium* form root nodules in legumes and fix atmospheric nitrogen.
- *Azospirillum* and *Azotobacter* free living bacteria fix atmospheric nitrogen and thus increasing nitrogen content of the soil.
- **Mycorrhiza** – fungi symbiotically associated with root of plants.
- Many members of the genus *Glomus* form Mycorrhiza.
 - Provide **phosphorus** to the plants from the soil.
 - Make the plant resistant to **root-borne pathogen**.
 - Increase tolerance to **salinity and drought**.
- Cyanobacteria like *Anabaena*, *Nostoc* and *Oscillatoria* etc :
 - Fix atmospheric nitrogen.
 - add organic matter to the soil and
 - Increase soil fertility.

Abbreviation:

- DO : Dissolved Oxygen
- GAP : Ganga Action Plan
- KVIC : Khadi and Village Industries Commission
- TMV : Tobacco Mosaic Virus
- YAP : Yamuna Action Plan
- IPM : Integrated Pest Management.

CHAPTER-11

BIOTECHNOLOGY: PRINCIPLES AND PROCESSES

BIOTECHNOLOGY: PRINCIPLES AND PROCESSES

- Two core techniques that enabled birth of modern biotechnology:
 - **Genetic engineering:** Techniques to alter the chemistry of genetic material (DNA and RNA) to introduce into host organisms and thus change the phenotype of the host organism.
 - **Maintenance of sterile** (microbial contamination-free) ambient chemical engineering processes to enable growth of only the desired microbe/eukaryotic cell in large quantities.

Conceptual development of the principle of genetic engineering:

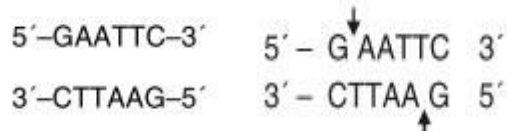
- Asexual reproduction preserves the genetic identity of species.
- Sexual reproduction creates variation and creates unique combinations of genetic makeup.
- Traditional hybridization procedures used in plant and animal breeding lead to inclusion of **undesirable genes** along with **desired genes**.
- The techniques of genetic engineering which includes creation of **recombinant DNA**, use of **gene cloning** and **gene transfer**, overcome this limitation and allows us to isolate and introduce only one or a set of desirable genes without introducing undesirable genes into target organism
- Three basic steps in genetically modifying an organism –
 - **Identification of DNA with desirable gene**
 - **Introduction of the identified DNA into the host.**
 - **Maintenance of introduced DNA in the host and transfer of the DNA to its progeny.**

TOOLS OF RECOMBINANT DNA TECHNOLOGY :

Restriction Enzymes:

- In the year 1963 two enzymes discovered from *Escherichia coli* which **restrict the growth of bacteriophage** in it.
 - One of these added **methyl groups** to DNA.
 - Other cut the phage DNA (**restriction endonuclease**).
- The first restriction endonuclease discovered is ***Hind II***.
- ***Hind II*** always cut DNA molecule at particular point by recognizing a specific sequence of six base pairs. This is called **recognition sequence** for ***Hind II***.
- Till date around 900 restriction enzymes isolated from 200 strains of bacteria each of which recognize different recognition sequences.
- Restriction enzyme belongs to **nucleases**.
- **There are two kind of nucleases:**
 - **Exonuclease**
 - **Endonuclease**

- Exonuclease removes nucleotides from the **free ends** of the DNA.
- Endonucleases make cuts at specific positions **within** the DNA.
- Each restriction endonuclease recognizes a specific **palindromic nucleotide sequences** in the DNA.
- Palindromes are the group of letters that read same both forward and backward. e.g.- MALAYALAM.
- The palindrome in DNA is a sequence of base pairs that reads same on the two strands when orientation of reading is kept same.



- The restriction enzyme cut the strand of DNA little away from the centre of the palindrome sites, but between the same two bases on the opposite strand. This leaves single stranded portions at the ends. There are overhanging stretches called **sticky ends** on each strand.
- This stickiness of the ends facilitates the action of the enzyme **DNA ligases**.
- The foreign DNA and the host DNA cut by the **same restriction endonuclease**, the resultant DNA fragments have the same kind of 'sticky-ends' and these can be joined together using DNA ligases.

Convention for naming restriction endonuclease:

- The first letter of the name comes from the **genus**.
- Second two letters come from the species of the prokaryotic cell from which the enzyme isolated
- The fourth letter is in capital form derived from the Strain of microbes.
- The Roman letter followed is the order of discovery
- Best example: **EcoRI** comes from *Escherichia coli* **RY 13**.

Separation and isolation of DNA fragments:

- The cutting of DNA by restriction endonucleases results in the fragments of DNA.
- These fragments are separated by a technique called **gel electrophoresis**.
- Since the DNA fragments are **negatively charged**, they can be separated by forcing them to move towards **anode** under an electric field through a **medium/matrix**.
- Most commonly used matrix is **agarose**, a natural polymer extracted from sea weed.
- DNA fragments separate according to their size through **sieving effect** provided by the agarose gel. Hence the smaller the fragment size, farther it moves.
- The separated fragments are visualized by staining them with **Ethidium bromide** followed by exposure to UV radiation.
- The separated bands of DNA are cut out from the agarose gel and extracted from the gel piece. This step is called **elution**.

Cloning vectors:

- The plasmid and bacteriophages have the ability to replicate within bacterial cells independent of the control of chromosomal DNA.
- Alien DNA linked with the vector multiply its number equal to the copy number of the plasmid or bacteriophage.

Features of cloning vector :

Origin of replication:

- This is the sequence where the replication starts called *ori* gene.
- The alien DNA linked with vector also replicates.
- Controls the copy number of the linked DNA.

Selectable marker:

- It is required to identify **recombinant** from the **non-recombinant**.
- Helps in identifying and eliminating non-transformants and selectively permitting the growth of the transformants.
- Transformation is a procedure through which a piece of foreign DNA is introduced in a host bacterium.
- Normally, the gene coding resistance to antibiotics such as ampicilin. Tetracycline, chloramphenicol or kanamycins etc are considered as useful selectable markers for *E. coli*.
- The normal *E. coli* cells do not carry resistance against any of antibiotics.

Cloning sites:

- In order to link the alien DNA, the vector needs to have very few, preferably single, **recognition sites (palindromic site)** for the commonly used restriction endonuclease.
- Commonly used vector is pBR322 for *E. coli*.
- The ligation of foreign DNA is carried out at a restriction site present in one of the two **antibiotic resistance** genes.
- If a foreign DNA ligated or inserted at the **Bam HI** site of tetracycline resistance gene in the vector pBR322, the recombinant plasmid will lose tetracycline resistance (insertional inactivation).
- The **recombinant** can be identified from the **non-recombinant** in following steps:
 - All are grown in **ampicilin** medium.
 - One replica of above plate grown in ampicilin medium (control).
 - Other replica grown in the medium containing both **tetracycline and ampicilin**.
 - The colonies grows in plate-I but failed to grow in plate-II are identified as **recombinants**.

Alternative selectable marker:

- In *E. coli*, a plasmid called **PUK-18** is used as selectable marker which is better than pBR322.
- The foreign DNA is introduced within the coding sequence of an enzyme β -galactosidase which convert X-Gal (chromatogenic substrate) into Galactose and 5-bromo-4-chloroindigo (blue color).
- The non-recombinant produce enzyme and give blue colored colonies.
- The recombinant unable to produce **β -galactosidase** and does not produce blue colored colonies after addition of chromatogenic substrate i.e. X-Gal.
- This inactivation of insertion of foreign DNA called **insertional inactivation**.

Vectors for cloning genes in plants and animals:

- *Agrobacterium tumefaciens*, a pathogenic bacterium of several dicot plants.
- This bacterium contains a plasmid called **Ti-plasmid** (tumor inducing).
- In natural condition the *A. tumefaciens* transfer the **T-DNA** into the plant which transform normal plant cells into a **tumor** and direct these tumor cells to produce the chemical required by the pathogen.
- **Retroviruses** in animals have the ability to transform normal cells into **cancerous** cells.
- The dis-armed retroviruses are being used to transfer gene into animals.
- In Ti-plasmid the T-DNA is replaced by the gene of interest, still *A. tumefaciens* able to transfer the gene into the plant without causing tumor in plants.

Competent Host (for transformation with recombinant DNA):

- DNA is a hydrophilic molecule; it cannot pass through cell membranes.
- In order to force bacteria to take-up the plasmid, the bacterial cells must first be made 'competent' to take up DNA.
- The bacterial cell is treated with divalent cations such as calcium, which increases the efficiency of DNA up take by the bacteria.
- Recombinant DNA and the bacterial cells are incubated in ice, followed by placing them briefly at 42°C (heat shock) and then putting them back in ice.
- By **microinjection** the recombinant DNA directly injected into the nucleus of the animal cell.
- Plant cells are bombarded with high velocity micro-particles of gold or tungsten coated with DNA in a method known as **biolistics** or **gene gun**.
- The disarmed pathogen vectors which when allowed infecting the cell transfer the recombinant DNA into the host.

PROCESS OF RECOMBINANT TECHNOLOGY:

- Isolation of DNA.
- Fragmentation of DNA by **restriction endonuclease**.
- Isolation of desired DNA fragment by **gel electrophoresis**.
- Ligation of DNA fragment with a vector by **DNA ligase**
- Transferring the recombinant DNA into the host.

- Culturing the host cells in a medium at large scale in a **bioreactor**.
- Extraction of desired product by **downstream processing**.

Isolation of the Genetic material (DNA):

- Bacterial cell wall digested by **Lysozyme**.
- Plant cell wall is digested by **cellulase** and **pectinase**.
- Fungal cell wall is digested by **chitinase**.
- RNA of the cellular content is digested by **ribonuclease**.
- Proteins are removed by **Proteases**.
- Purified DNA ultimately precipitated out after addition of **chilled ethanol**.
- The precipitated DNA is separated and removed by **spooling**.

Amplification of Gene of Interest using PCR:

- PCR stands for **Polymerase chain reaction**:
- Multiple copy of gene of interest can be synthesized in vitro.
- PCR includes following steps:

Denaturation:

- Double stranded DNA made single stranded.
- It is done by heating the DNA at 94°C.
- Each single stranded DNA is called Template strand.

Annealing:

- Two sets of **primer** (small oligonucleotide chain that are complementary to the DNA at 3' end of the DNA template) added to the medium.
- This is done at around 50°C.

Extension:

- Deoxyribonucleotides triphosphates are added in the medium.
- **Taq polymerase** catalyses the polymerization reaction using nucleotides extending from the primer towards 5' end of the template.
- Taq polymerase is a thermo-stable polymerase isolated from a bacterium called ***Thermus aquaticus***.
- It catalyses polymerization reaction at 74°C.

Obtaining the Foreign Gene product or Recombinant product:

- The protein encoding gene is expressed in a **heterogeneous host** is called a **recombinant protein**.
- The host is cultured in a continuous culture system provided in bioreactor.
- A bioreactor provides optimum growth conditions (temperature, pH, substrate, salts, vitamins, oxygen).
- Bioreactor convert the raw materials into specific product, specific enzyme.

Downstream processing:

- After biosynthesis inside the bioreactor, the product has to be subjected through a series of processes before it is ready for marketing.
- The process includes separation and purification which are collectively referred as downstream processing.
- The product has to be formulated in suitable preservatives.
- Such formulation has to undergo through clinical trials as in case of drugs.

P. K. Singh

CHAPTER-12

BIOTECHNOLOGY AND ITS APPLICATIONS

The critical areas of biotechnology are:

- Providing the best catalyst in the form of improved organism usually a microbe or pure enzyme.
- Creating optimal condition through engineering for a catalyst to act.
- Downstream processing technologies to purify the protein/organic compound.

BIOTECHNOLOGICAL APPLICATIONS IN AGRICULTURE:

- Plants, bacteria, fungi and animals whose genes have been altered by manipulation are called **Genetically Modified Organisms (GMO)**.
- **Advantages of Genetic Modification in plants.**
 - Made crops more tolerant to abiotic stresses (cold, drought, salt, heat).
 - Reduce reliance on chemical pesticides (pest resistant crop)
 - Helped to reduce post-harvest losses.
 - Increased efficiency of mineral usage by plants.
 - Enhanced nutritional values of food e.g. vitamin A enriched rice.

Bt-Cotton:

- Some strains of *Bacillus thuringiensis* produce proteins that kill certain insects such as **lepidopterans** (tobacco budworm, armyworm), **coleopterans** (beetles) and **dipterans** (flies, mosquitoes).
- *B. thuringiensis* forms protein crystals during a particular phase of their growth. These crystals contain a toxic **insecticidal protein**.
- These proteins are present in inactive **protoxin** form, but become active toxin in the alkaline pH of insect gut.
- The activated toxin binds to the surface of midgut epithelial cells and creates pores that cause cell swelling and lysis and eventually cause death of insect.
- Specific Bt toxin genes were isolated from *B. thuringiensis* and genetically transferred to several plants such as cotton.
- **Crystal proteins** are produced by a gene called **cry** in *B. thuringiensis*.
- The protein coded by genes **cryIAc** and **cryIIAb** control the cotton **bollworms**.
- The protein coded by gene **cryIAb** controls **corn borer**.

Pest resistant plants:

- Several **nematodes** parasitize a wide variety of plants and animals including human beings.
- A nematode *Meloidegryne incognitia* infects the root of **tobacco plants** and causes a great reduction in yield.
- Strategy based on **RNA interference (RNAi)** prevents this infestation.
- Process by which double-stranded RNA (dsRNA) directs sequence-specific degradation of mRNA.

Steps of RNA interference:

- Double stranded RNA is produced endogenously or exogenously.
- Using Agrobacterium vectors, nematode specific genes were introduced into the host plant (tobacco plant).
- Introduction of DNA produces both sense and antisense RNA in the host.
- These two RNA's being complementary to each other formed a double stranded (dsRNA) that initiated RNAi.
- The dsRNA injected into the host plant from outside called exogenous dsRNA.
- The dsRNAs are cleaved into 21-23 nucleotide segments (**small interfering RNAs** or **siRNAs**) by an enzyme called **Dicer**.
- siRNAs are incorporated into **RNA-induced silencing complex (RISC)**.
- Guided by base complementarity of the siRNA, the RISC targets mRNA for degradation.
- The consequence was that the parasite could not survive in a transgenic host.

BIOTECHNOLOGICAL APPLICATIONS IN MEDICINE:

- Biotechnology enables **mass production** of safe and more effective therapeutic drugs.
- Recombinant therapeutics does not induce **unwanted immunological responses** as is common in case of similar products isolated from non-human sources.
- At present, around 30 recombinant therapeutics are approved for human use.

Genetically Engineered Insulin:

- Taking insulin at regular interval of time is required for adult-onset diabetes.
- Previously the source of insulin was the slaughtered cattle and pigs.
- This insulin caused allergy in some patients.
- Insulin made of two short polypeptide chains : chain-A and chain-B that are linked together by disulphide linkage.
- Insulin synthesized in pancreas as pro-hormone which is a single polypeptide with an extra stretch called **C-peptide**.
- C-peptide is removed during matured insulin.
- In 1983, Eli Lilly an American company prepared two DNA sequences corresponding to A and B, chains of human insulin and introduced them in plasmids of *E. coli* to produce insulin chains.
- Chain-A and chain-B produced separately, extracted and combined by creating disulfide bonds to form mature human insulin.

Gene therapy:

- Gene therapy is an attempt to cure hereditary or genetic diseases.
- Genes are inserted into a person's cells and tissue to treat the disease.
- The first clinical gene therapy was given in 1990 to a 4 years old girl with adenosine deaminase (ADA) deficiency.
- This enzyme is required for breakdown of **deoxyadenosine** into uric acids.

- In the absence of ADA toxic deoxyadenosine is accumulated and destroy the infection fighting immune cells called T-cells and B-cells.
- This disorder is caused due to the deletion of the gene for adenosine deaminase in chromosome 20.

Treatment for ADA deficiency disease:

- Treated by bone marrow transplantation.
- Enzyme replacement therapy, involving repeated injections of the ADA enzyme
- Lymphocytes from the blood of the patient are grown in a culture. A functional ADA cDNA is then introduced into these lymphocytes and returned into the body.
- The patient required periodic infusion of genetically engineered lymphocytes because these cells are not immortal.
- Functional ADA cDNA introduced into cells at early embryonic stages, could be the permanent cure.

Molecular diagnosis:

- Early detection of disease is not possible by conventional methods (serum and urine analysis).

Molecular diagnosis techniques:

- Recombinant DNA technology.
- Polymerase chain reaction (PCR).
- Enzyme linked Immuno-sorbent Assay (ELISA).
- Very low concentration of a bacteria or virus can be amplified and detected by PCR.
- It used to detect genetic disorders.
- PCR is use full to mutation in genes in suspected cancerous patient:
 - A single stranded DNA or RNA tagged with radioactive molecule (probe) is allowed to hybridize to its complementary DNA in a clone of cells followed by detection using autoradiography.
 - The clone having mutated gene unable make complementary bonding of probe, hence not appears in photographic film.

TRANSGENIC ANIMALS:

- Animals that have an **alien DNA** which able to express in it is called **transgenic animals**.

Reasons for creation of transgenic animals:

- **Normal physiology and development:**
 - Transgenic animals are specifically designed to allow study of:
 - How the genes are regulated.
 - How the gene affects normal functioning of body.
 - How it affects growth and development. E.g.- Insulin like growth factor.

- The animals made transgenic to know the biological effect and result.
- **Study of disease:**
 - Transgenic animals are designed to understand how genes contribute to the development of disease like cancers, cystic fibrosis, rheumatoid arthritis and Alzheimer's.
- **Biological products:**
 - Transgenic animals are used to produce biological product of human interest:
 - **α -1-antitrypsin** used to treat emphysema.
 - Proteins for treatment for PKU and cystic fibrosis.
 - Transgenic cow Rosie, produce human protein enriched milk (2.4 gm/lit. **human α -lactalbumin**).
- **Vaccine safety:**
 - Transgenic mice are being developed and use in testing the safety of vaccines before they are used for humans.
 - Polio vaccine is tested in mice.
- **Chemical safety testing:**
 - This is also known as toxicity/safety testing.
 - Transgenic animals are made to know the effect of toxic chemicals.

ETHICAL ISSUES:

- GEAC (Genetic Engineering Approval Committee) set up by Indian Govt. which will make decisions regarding validity of GM research and safety of introducing GM-organisms for public services.
- A **patent** is the right granted by a government to an inventor to prevent others from commercial use of his invention.
- Patents granted for biological entities and for products derived from them; these patents are called **bio-patents**.
- 27 documented varieties of Basmati are grown in India.
- **Biopiracy** is the term used to refer to the use/exploit or patent, of biological resources by multinational companies and other organizations without proper authorization from the countries and people concerned without compensatory payment.

CHAPTER-13

ORGANISMS AND POPULATIONS

ORGANISM AND ITS ENVIRONMENT :

- Rotation of sun and the tilt of its axis cause annual variations in the intensity and duration of temperature, resulting distinct seasons.
- These variations along with annual variations in precipitation, forms major biomes, such as **desert**, **rain forest** and **tundra**.
- Temperature, water, light and soil are the key elements that lead to so much variation in the physical and chemical conditions of habitats.
- Both **abiotic** (physic-chemical) and **biotic** components (pathogen, parasites, predators, competitions) characterize the habitat of an organism.

Major abiotic factors :

Temperature:

- Temperature decreases progressively from equator towards the pole and high altitudes to $>50^{\circ}\text{C}$ in tropical deserts in summer.
- Thermal springs and deep-sea hydrothermal vents are unique with $>100^{\circ}\text{C}$.
- Temperature affects the kinetics of enzymes, BMR and other physiological actions.
- **Eurythermals**: organism which can tolerate wide range of temperatures.
- **Stenothermal**: organism which can tolerate narrow range of temperatures.

Water:

- Water is also important factor that influence the life of organism.
- The productivity and distribution of plants is also depends on water.
- The salinity varies in aquatic environment:
 - 5% in inland waters (fresh water).
 - 30-35 in sea water.
 - More than 100% in hyper saline lagoons.
- **Euryhaline**: organism which can tolerate wide range of salinity.
- **Stenohaline**: organism which can tolerate narrow range of salinity.

Light:

- Plant produce food by photosynthesis, which only possible in presence of light. Hence it very important for autotrophs.
- Plant species (herbs and shrubs) adapted for photosynthesize under canopy.
- Sunlight is required for photoperiodic response like flowering.
- Animals use diurnal and seasonal variations in light intensity and photoperiod as cues for timing their foraging, reproductive and migration.

Soil:

- Properties of soil vary according to the climate, the weathering process.

- Soil composition, grain size and aggregation determine the percolation and water holding capacity of the soil.
- These characteristic along with pH, mineral composition and topography determine to a large extent the vegetation in any area.
- The sediment-characteristic often determine the type of benthic animal in aquatic environment.

Response to Abiotic Factors :

- **Homeostasis** – the process by which the organism maintain a constant internal environment in respect to changing external environment.

How does organism cope with the changing environment?

Regulate:

- Some organisms are able to maintain homeostasis physiological (sometimes behavioral also) means which ensures constant body temperature, constant osmotic concentration.
- All birds and mammals and few lower invertebrates are capable of such regulation i.e. thermoregulation and osmoregulation.
- Success of mammals is due to thermoregulation.
- We maintain a constant body temperature of 37°C.
- When outside temperature is high we sweat profusely and evaporative cooling take place to bring body temperature down.
- In winter, due to low temperature outside our body temperature falls below 37°C, we start to shiver, to generate heat to raise body temperature.

Conform:

- Majority (99%) of animals and plants cannot maintain a constant internal environment; their body temperature varies according to ambient temperature.
- In aquatic animals the osmotic concentration of body fluid varies with ambient water osmotic concentration.
- All the above animals and plants are simply called as **conformer**.

Why the conformer not evolved to became regulators?

- Thermoregulation is energetically expensive for many animals.
- Small animal like shrews and humming birds cannot afford so much energy for thermoregulation.
- Heat loss or heat gain is a function of surface area.
- Small animals have larger surface area relative to their volume, they tend to lose body heat very fast when it is cold outside; then has to expend much energy to generate body heat through metabolism.
- This is why very small animals are rarely found in Polar Regions.
- **Alternative response for stressful conditions is localized or remains for short duration.**

Migrate:

- The organism moved away temporarily from the stressful habitat to a more hospitable area and return when stressful condition is over.
- Bird migrate form the colder region to warmer region.

Suspend:

- Thick walled spores are formed in microbes to overcome unfavourable stressful external environment. Spores germinate in favourable condition.
- In higher plants seeds and other vegetative reproductive structures are means to tide over the stress. They reduce their metabolic activity and going into a state of 'dormancy'.
- **Hibernation:** during winter animals like bears escape in time.
- **Aestivation:** animals like snail and fish avoid summer related problem like heat and desiccation.
- **Diapauses:** many zooplanktons undergo a stage of suspended development in unfavourable conditions.

ADAPTATION :

- **Adaptation:** is any attribute of the organism (morphological, physiological, and behavioral) that enables the organism to survive and reproduce in its habitat.

Adaptation of animal in desert:

- Kangaroo rat meets their water requirement from **oxidation of fat**.
- Excrete very **concentrate urine** to conserve water.

Adaptation of plant in desert (xerophytes):

- **Thick cuticle** on their leaf surfaces.
- **Sunken stomata**, both to reduce transpiration.
- Have special photosynthetic pathway (**CAM**), stomata closed during day time and remained open during night.
- Opuntia has no leaf- they are reduced to **spines**.
- Photosynthesis takes place in **flat green stems**.

Adaptation of animal in cold climate:

- **Allen's Rule:** mammals from colder climates generally have shorter ears and limbs to minimize heat loss.
- Seals of polar aquatic seas have a thick layer of fat called **blubber** below their skin that acts as insulator and reduces loss of body heat.

Adaptation in high altitude:

- A person moves to high altitude (>3,500 meter), develop **altitude sickness**.
- Symptoms developed are nausea, fatigue and heart palpitations.
- This is due to low atmospheric pressure of high altitudes; the body does not get enough oxygen.

How the bodies solve the problem?

- The body compensates low oxygen availability by increasing red blood cell production.
- The body compensates decreasing binding capacity of hemoglobin with oxygen by increasing rate of breathing.

Behavioral adaptation:

- **Desert lizards are conformer hence they cope with the stressful environment by behavioral adaptations:**
 - They bask in the sun and absorb heat when their body temperature drops below the comfort zone in winter.
 - Move to shade when the ambient temperature starts increasing.
 - Some species burrowing into the soil to hide and escape from the above-ground heat.

POPULATION :

Population attributes:

- **Population:** a group of individual living in a well-defined geographical area, share or compete for similar resources, potentially interbreed.
- Birth rate and death rate refers to per capita births and deaths respectively.
- Another attribute is **sex ratio**. The ratio between male female in a population.
- If the age distribution is plotted for a population the resulting structure is called age pyramid.
- The shape of the pyramids reflects the growth status of the population like **growing, stable** or **declining**.
- The population size is more technically called as **population density**.

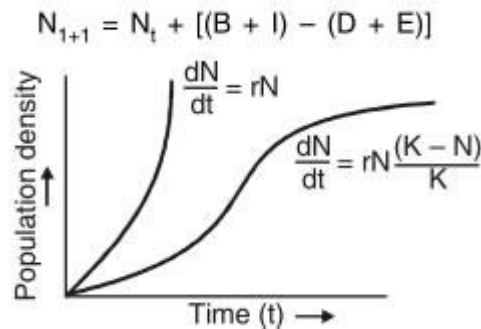
Methods for measurement of population density:

- Counting the number
- Percent cover
- Biomass.
- **Pug marks** and **fecal pellets** for tiger census.

Population growth:

- The size of the population changes depending on food availability, predation pressure and reduce weather.
- Population size fluctuated due to changes in four basic processes, two of which (Natality and immigration) contribute an increase in population density and two (mortality and emigration) to a decrease.
- **Natality:** number of birth in given period in the population.
- **Mortality:** number of deaths in the population in a given period of time.
- **Immigration:** is the number of individuals of same species that have come into the habitat from elsewhere during a given period of time.

- **Emigration:** number of individuals of the population who left the habitat and gone elsewhere during a given time period.
- If 'N' is the population density at time 't' then its density at time (t+1) is :



Where, B = the number of births.

I = the number of immigrants.

D = the number of deaths.

E = the number of Emigrants.

N = Population Density.

r = Intrinsic rate of natural increase.

t = Time period.

K = Carrying capacity (The maximum population size that an environment can sustain).

Exponential growth:

- The Exponential growth equation is $N_t = N_0 e^{rt}$
- N_t = Population density after time 't'.
- N_0 = Population density at time zero.
- r = intrinsic rate of natural increase.
- e = the base of natural logarithms (2.71828).

Exponential growth ('J' shape curve is obtained):

- When resources are not limiting the growth.
- Any species growth exponentially under unlimited resources conditions can reach enormous population densities in a short time.
- Growth is not so realistic.

Logistic growth model:

- Verhulst-Pearl Logistic Growth is described by the following equations :

$$dN/dt = rN (K-N) / N$$

Where, N = Population density at time 't'.

r = Intrinsic rate of natural increase.

K = Carrying capacity.

Logistic Growth (Sigmoid curve is obtained):

- When responses are limiting the Growth.
- Resources for growth for most animal populations are finite and become limiting.
- The logistic growth model is a more realistic one.

POPULATION INTERACTIONS :

Predation:

- Organism of higher trophic level (predator) feeds on organism of lower trophic level (prey) is called the predation.
- Even the herbivores are not very different from predator.
- Predator acts as a passage for transfer of energy across trophic level.
- Predators keep prey populations under control.
- Exotic species have no natural predator hence they grow very rapidly (**prickly pear cactus** introduced in Australia created problem).
- Predators also help in maintaining species diversity in a community, by reducing the intensity of competition among competing prey species (**Pisaster** starfish field experiment).

Defense developed by prey against predators:

Animals:

- Insects and frogs are cryptically coloured (**camouflaged**) to avoid being detected by the predator.
- Some are poisonous and therefore avoided by the predators.
- **Monarch butterfly** is highly distasteful to its predator (bird) due to presence of special chemical in its body. The chemical acquired by feeding a poisonous weed during caterpillar stage.

Plants:

- **Thorns** in Acacia, Cactus are morphological means of defense.
- Many plants produce and store some chemical which make the herbivore sick if eaten, inhibit feeding, digestion disrupt reproduction, even kill the predators.
- **Calotropis** produces poisonous **cardiac glycosides** against herbivores.
- Nicotine, caffeine, quinine, strychnine, opium etc. are produced by plant actually as defenses against the grazers and browsers.

Competition:

- Interspecific competition is a potent force in organic evolution.
- Competition generally occurs when closely related species compete for the same resources that are limiting but this not entirely true :
- **Firstly:** totally unrelated species could also compete for the same resources.
 - American lakes visiting flamingoes and resident fishes have their common food, zooplanktons.
- **Secondly:** resources need not be limiting for competition to occur.
 - **Abingdon tortoise** in Galapagos Islands became extinct within a decade after **goats** were introduced on the island due to greater browsing ability.
- **Competitive release:** A species whose distribution is restricted to a small geographical area because of the presence of a competitively superior species, is

found to expand its distributional range dramatically when the competing species is experimentally removed.

- **Connell's** elegant field experiment showed that superior barnacle *Balanus* dominates the intertidal area and excludes the smaller barnacle *Chathamalus* from that zone.
- Gause's '**competitive Exclusion Principle**': two closely related species competing for the same resources cannot co-exist indefinitely and the competitively inferior will be eliminated eventually.
- **Resource partitioning**: If two species compete for the same resource, they could avoid competition by choosing, for instance, different times for feeding or different foraging pattern.
 - **MacArthur** showed five closely related species of warblers living on the same tree were able to avoid competition and co-exist due to behavioral differences in their foraging activities.

Parasitism:

- Parasitic mode of life ensures free lodging and meals.
- Some parasites are host-specific (one parasite has a single host) in such a way that both host and parasite tend to co-evolve.

Parasitic adaptation

- Loss of **unnecessary sense organs**.
- Presence of **adhesive organs** or **suckers** to cling on to the host.
- Loss of **digestive system**.
- High reproductive capacity.
- Parasites having one or more intermediate host or vectors to facilitate parasitisation of its primary host.
- Liver fluke has two intermediate hosts (snail and a fish) to complete its live cycle.

Effects on the host:

- Parasite always harms the host.
- They reduce the survival, growth and reproduction of the host.
- Reduce its population density.
- They make the host more vulnerable to the predators, by making it physically weak.
- **Ectoparasite**: feeds on the external surface of the host.
 - Lice on human.
 - Ticks on dog.
 - Marine fish infested with copepods.
 - *Cuscuta* (a parasitic plant) grow on hedge plants.
- **Endoparasites**: are those that live inside the host body at different sites.
 - Life cycle is more complex.
 - Morphological and anatomical features are greatly simplified.
 - Highly developed reproductive system.

- **Brood parasitism:**

- Special type of parasitism found in birds.
- The parasitic birds lay its eggs in the nest of its host and let the host incubate them.
- The egg of the host is very similar with the egg of the host.
- Cuckoo lays eggs in the nest of the crow.

Commensalism: This is the interaction in which one species benefits and the other is neither benefited nor harmed.

- Orchids growing as an *epiphyte* on a mango branch.
- Clown fish living among tentacles of sea anemone.
- Barnacles on back of whales.
- Cattle Egret and grazing cattle.

Mutualism: Interaction between two living organism, both are equally benefited, no one is harmed.

- **Lichen:** a mycobiont and a phycobiont.
- **Mycorrhiza:** relationship between fungi and root of higher plant.
- Pollinating insects and flowering plants.
- **Fig trees** and its pollinating agent **wasp**.

Sexual deceit:

- Mediterranean orchid **Ophrys** employs '**sexual deceit**'.
- Petal of the flower resembles the female bee.
- The male bee attracted to what it perceives as a female, 'pseudo-copulates' with the flower but does not get any benefits.

CHAPTER-14

ECOSYSTEMS

ECOSYSTEMS :

- The interaction between the living organism and the non-living environment is called **ecosystem**.

ECOSYSTEM – STRUCTURE AND FUNCTION :

- Interaction of biotic and abiotic components results in a physical structure that is characteristic of each type of ecosystem.
- Identification and description of plant and animal species of an ecosystem gives its **species composition**.
- Vertical distribution of different species occupying different levels is called **stratification**.
- The components of the ecosystem are seen to function as a unit:
 - Productivity.
 - Decomposition.
 - Energy flow and
 - Nutrient cycle.
- **Description of pond as an ecosystem:**
 - The abiotic components include all dissolved inorganic and organic substances and the rich soil deposit at the bottom of the pond.
 - The solar input, cycle of temperature, day length, regulates the rate of function of the entire pond.
 - The **producer** (autotrophic) includes phytoplankton, some algae and the floating, submerged and marginal plants found in edge of pond.
 - The **consumers** are represented by zooplankton, free swimming and bottom dwelling animals.
 - The decomposers are the fungi, bacteria especially abundant at the bottom of the pond.
- **Basic events (in terms of function) in an ecosystem:**
 - Conversion of inorganic into organic material (photosynthesis) by producers.
 - Consumption of the autotrophs by heterotrophs.
 - Decomposition and mineralization of the dead organic matter to release them back for reuse by the autotrophs
 - There is unidirectional flow of energy towards the higher trophic levels and its dissipation and loss as heat to the environment.

PRODUCTIVITY :

- **Primary productivity:**
 - The amount of biomass or organic matter produced per unit area over a time period by plants during photosynthesis.

- It is expressed in terms of weight (g-2) or energy (kcal m-2)
- The rate of biomass production is called **productivity**.
- **Gross primary productivity: (GPP)** is the rate of production of organic matter during photosynthesis.
- **Net primary productivity:**
 - A considerable amount of energy is utilized by plants in respiration.
 - Gross primary productivity minus respiration losses (R) is the net primary productivity.
 - $GPP - R = NPP$.
- Net primary productivity is the available biomass for the consumption to heterotrophs (herbivore and decomposers).
- **Secondary productivity:** is defined as the rate of formation of new organic matter by the consumer.

DECOMPOSITION :

- Earthworm is said to be '**friends**' of farmer :
 - Breakdown the complex organic matter.
 - Loosening of the soil helps in aeration and entry of root.
- The decomposers break down complex organic matter into inorganic substances like carbon dioxide, water and nutrients, called **decomposition**.
- Dead plant remains such as leaves, bark, flowers and dead remains of animals, including fecal matter, constitute the **detritus**.
- The process of decomposition completed in following steps:
 - **Fragmentation** : Break down of detritus into smaller particles by detritivore (earthworm).
 - **Leaching**: Water soluble inorganic nutrients go down into the soil horizon and get precipitated as unavailable salts.
 - **Catabolism** : Bacterial and fungal enzymes degrade detritus into simple inorganic substances.
 - **Humification**: Accumulation of dark coloured amorphous substances called humus.
 - **Importance of humus:**
 - Highly resistance to microbial action.
 - Undergo decomposition at an extremely slow rate.
 - Being colloidal in nature, it serves as reservoir for nutrients.
 - **Mineralization**: The humus is further degraded by some microbes and release of inorganic nutrients occur.

Factor affects rate of decomposition:

- Decomposition is largely an oxygen-requiring process.
- Detritus rich in chitin and lignin has slow rate of decomposition.
- Detritus rich in nitrogen and water-soluble substance like sugar has faster decomposition.
- **Temperature** and **soil moisture** are most important climatic factor that regulate decomposition.

- **Warm** and **moist** environment favours decomposition.
- **Low temperature, dryness** and **anaerobiosis** inhibit decomposition.

ENERGY FLOW IN ECOSYSTEM:

- Except for deep sea hydrothermal ecosystem, sun is the only source of energy for all ecosystems on earth.
- Less than 50% of incident solar radiation is **photosynthetically active radiations (PAR)**.
- Plants capture 2-10 % of PAR and used in photosynthesis.
- All organisms depend on the producers, either directly or indirectly.
- Energy flow in the ecosystem is **unidirectional** *i.e.* energy transferred from producer to consumers.
- Energy transfer is not absolute, spontaneous; unless energy is degraded it can't be transfer. When energy transferred from one trophic level to another, lot of energy lost in the form of heat to the environment.
- Only 10% of energy transferred from one trophic level to other.

Food chain:

- **Grazing food chain:** it extends from producers through herbivore to carnivore.
- **Detritus food chain:** Begins with dead organic matter (**detritus**) and pass through detritus feeding organism in soil to organisms feeding on detritus-feeders.
- In aquatic ecosystem **GFC** is the major conduit for energy flow.
- In terrestrial ecosystems a much larger fraction of energy flows through the **detritus food chain** than through GFC
- Different food chains are naturally interconnected e.g.- a specific herbivore of one food chain may serve as food of carnivores of other food chains. Such interconnected matrix of food chains is called **food web**.
- **Trophic level:** A group of organism irrespective of their size having same source of energy or similar food habit constitute a **trophic level**.
- **Standing crop:** each trophic level has a certain mass of living material at a particular time called as the standing crop.
- The standing crop is measured as the mass of living organisms (**biomass**) or the **number** in a unit area.
- The number of trophic levels in a food chain is restricted by 10 % flow of energy, less amount of energy available to the last trophic level.

ECOLOGICAL PYRAMID:

- The base of the pyramid is broad and it narrows down at the apex. The similar shape is obtained when food or energy relationship between organisms at different trophic level.
- The relationship can be expressed in terms of number, energy or biomass.
- The base of the pyramid represented by producer and apex is the top consumer; other trophic levels are in between.

- In most ecosystems, all the pyramids, of number, of energy and biomass are upright.
- The pyramid of **number** in a tree ecosystem is **inverted**.
- The pyramid of **biomass** in sea also **inverted** because the biomass of fishes is far exceeding that of phytoplankton.
- Pyramid of **energy** is **always upright**, can never be inverted, because when energy flows from a particular trophic level to the next, some energy is always lost as heat at each step.

Limitations of ecological pyramids:

- It does not take into account the same species belonging to two or more trophic levels.
- It assumes a simple food chain; it never exists in nature.
- It does not accommodate food web.
- Saprophytes are not given place in ecological pyramids.

ECOLOGICAL SUCCESSION:

- The gradual and fairly predictable change in the species composition of a given area is called **ecological succession**.
- Composition and structure of the community constantly change in response to changing environmental condition.
- This change is orderly and sequential, parallel with the changes in the physical environment.
- All the changes lead finally to a community that is in near equilibrium with the environment and that is called **climax community**.
- During succession some species colonize an area and their populations become more numerous, whereas populations of other species decline and even disappear.
- The entire sequences of communities that successively change in a given area are called **sere**.
- The individual transitional communities are termed as **seral stages**.
- In the successive seral stages there is a change in the diversity of species of organisms, increase in number of species and total biomass.
- **Primary succession:** Succession that starts where no living organisms are there. These could be areas where no living organism ever existed may be a bare rock or new water body.
- **Secondary succession:** Succession that starts in areas that somehow, lost all the living organisms that existed there.
- Primary succession occurs in :
 - newly cooled lava,
 - bare rock,
 - Newly created pond or reservoir.
- Secondary succession begins in areas where natural biotic communities have been destroyed such as :
 - In abandoned farm lands.

- Burned or cut forest,
- land that have been flooded
- Since some soil or sediment is present, secondary succession is faster than primary succession.

Succession in plants:

- Based on the nature of habitat – whether it is water or it is on very dry areas- succession of plants is called **hydrarch** or **xerarch**.
- **Hydrarch succession** takes place in water areas and the successional series progress from **hydric** to **mesic** condition.
- **Xerarch succession** takes place in dry areas and the series progress from **xeric** to **mesic** conditions.
- Both hydrarch and xerarch successions lead to medium water conditions (mesic) *i.e.* neither too dry (xeric) nor too wet (hydric).

Xerarch succession (Succession in bare rock) :

- The species that invades bare area are called **pioneer species**.
- In primary succession on bare rock the pioneer species is the **lichen**.
- Lichen secretes acid to dissolve rock, helping in weathering and soil formation.
- The little soil, leads to growth of **bryophytes (mosses)**.
- The mosses speed up the process of soil accumulation by trapping wind-blown particles.
- Lichen moss carpet provides suitable substratum for the germination of seeds of **herbaceous plants**.
- Gradually more soil is accumulated and herbaceous species make way for the invasion of **shrubs** followed by **trees**.
- The **climax community** is generally dominated by **trees**.

Hydrarch (succession in aquatic environment) :

- In primary succession in water, the **pioneer species** are **phytoplankton**.
- Zooplanktons.
- Sub merged plant stage. (rooted hydrophytes)
- Sub merged and free-floating plant stage.
- Reed-swamp stage.
- Marsh-meadow stage.
- Shrub stage
- Trees
- The climax again would be the forest
- All the succession whether taking place in water or on land, proceeds to a similar climax community *i.e.* the mesic.

NUTRIENT CYCLING:

- Organism needs constant supply of nutrients to grow, reproduce and regulate various body functions.

- **Standing state** : The amount nutrients such as carbon, nitrogen, phosphorus, calcium etc. present in soil at any given time.
- **Nutrient cycling** : The movement of nutrient elements through the various component of an ecosystem is called nutrient cycling.
- Another name of nutrient cycling is **biogeochemical cycle**.
- Nutrient cycles are of two types :
 - **Gaseous cycle**
 - **Sedimentary cycle**.
- The reservoir for gaseous type of nutrient cycle (nitrogen, carbon) exists in the atmosphere.
- The reservoir for sedimentary cycle (sulphur, phosphorus) is Earth's crust.
- Environmental factors like soil, moisture, pH temperature regulate the rate of release of nutrient into the atmosphere.
- The function of the reservoir is to meet the deficit which occurs due to imbalance in the rate of influx and efflux.

Ecosystem Carbon cycle :

- Carbon constitutes **49 percent** of dry weight of organism.
- Out of total global carbon :
 - 71% carbon found dissolved in ocean.
 - About 1% in the atmosphere.
- 4×10^{13} kg of carbon is fixed in the biosphere by photosynthesis annually.
- Large amount of carbon returned to the atmosphere as CO_2 through respiration of producers and consumers.
- Decomposers also return CO_2 to reservoir during decomposition process.
- Some amount of Carbon is lost to sediments and removed from circulation.
- Burning wood, forest fire, combustion of organic matter, fossil fuel, volcanic activities are additional sources for releasing CO_2 to atmosphere.

Influence of human activity on Carbon cycling :

- Rapid deforestation.
- Massive burning of fossil fuel for energy and transport
- Increased the rate of release of CO_2 into the atmosphere.

Ecosystem Phosphorus cycle :

- Phosphorus is a major constituent of biological membranes, nucleic acids and cellular energy transfer system(ATP).
- Animals need phosphorus to make shell, bones and teeth.
- Reservoir pool of phosphorus is the rock which contain phosphorus in the form of **phosphates**.
- During weathering of rock small amount of phosphates dissolved in soil solution and are absorbed by the roots of the plants.
- Herbivore and other animals obtain organic form of phosphorus from plants.

- The waste product and dead organisms are decomposed by phosphate-solubilising bacteria releasing phosphorus.

How phosphorus cycle differs from carbon cycle?

- There is no respiratory release of phosphorus into atmosphere.
- Atmospheric inputs of phosphorus through rainfall are much smaller.
- Gaseous exchange of phosphorus between organism and environment are negligible.

ECOSYSTEM SERVICES :

- The products of ecosystem processes are named as **ecosystem services**.
- Healthy forest ecosystems purify air and water.
- Mitigate droughts and flood.
- Cycle nutrients.
- Generates fertile soil.
- Provide wildlife habitat.
- Maintain biodiversity.
- Pollinate crops.
- Provide storage site for carbon
- Provides aesthetic, cultural and spiritual values.

Abbreviation:

- PAR : Photosynthetically Active Radiation
- GAP : Gross Primary Productivity
- NPP : Net Primary Productivity
- DFC : Detritus Food Chain
- GFC : Grazing Food chain

CHAPTER-15

BIODIVERSITY AND CONSERVATION

Biodiversity:

- The term biodiversity refers to the totality of genes, species, and ecosystems of a region.
- Types of biodiversity described by **Edward Wilson** :
 - **Genetic diversity:** A single species might show high diversity at the genetic level over its distributional range.
 - Medicinal plant *Rauwolfia vomitoria* of Himalayan range produces active chemical **reserpine** shows genetic variation.
 - India has more than 50000 different strain of rice.
 - 1000 varieties of mango.
 - **Species diversity:** different species of a single animal like frog.
 - **Ecological diversity:** diversity in the ecosystem level like desert, rain forest, mangroves, coral reef, wetlands, estuaries etc.

How many species are there on Earth and How many in India?

- According to **IUCN** (2004), 1.5 million of plants and animals are in our biosphere.
- **Robert May** places global species diversity at about 7 millions.
- More than 70 percent of all the species recorded are animals.
- All plants constitute about 22 percent.
- Among animals insects constitute 70 percent.
- India has only 2.4% of the world's land area, its share of global species diversity is impressive 8.1%.
- India is considered one of the mega diversity countries of the world.

Pattern of Biodiversity :

Latitudinal gradients:

- Species diversity decreases as we move away from the equator towards the pole.
- Tropic (23.5° N to 23.5° S) harbors more species than temperate and pole
- The largely tropical Amazonian rain forest in South America has the greatest biodiversity on earth:
 - 40,000 species of plants.
 - 3000 species of fishes.
 - 1300 of birds.
 - 427 amphibians
 - 378 reptiles
 - More than 1,25,000 invertebrates.

Why tropical rain forest has greater biodiversity:

- Unlike temperate regions subjected to frequent glaciations in the past, tropical latitudes have remained relatively undisturbed for millions of years. Thus, this had taken a long evolutionary time for species diversification.
- **Tropical environments:** Unlike temperate ones, these are less seasonal, relatively more constant and predictable, promotes niche specialization and lead to greater species diversity.
- There is more solar energy available in the tropics, which contribute to higher productivity.

Species area relationship:

- Alexander Von Humboldt observed within a region species richness increased with increasing explored area but only up to a limit.
- The relation between species richness and area for a wide variety of taxa turns out to be a rectangular hyperbola.
- On a logarithmic scale the relationship is a straight line describe by the equation **$\text{Log } S = \text{log } C + Z \text{ log } A$**
Where, S= species richness
A = Area
Z = slope of the line (regression coefficient)
C = Y-intercept.
- It has been noted that regardless of the taxonomic group or region the slope of the regression line is amazingly similar. However, for a very large area like the entire continent the slope of the line is steeper.

Importance of species diversity to the Ecosystem:

- Community with more species generally tends to be more stable than those with less species.
- A stable community should not show too much variation in productivity from year to year, it must be resistant or resilient to occasional disturbances (natural or man-made).
- Stable community must be resistant to invasion by alien species.
- **David Tillman's long-term field experiment finds that :**
 - Plots with more species showed less year to year variation in biomass
 - Increased diversity contributed to higher productivity.
- **The rivet popper hypothesis :**
 - In an airplane (ecosystem) all parts are joined together by thousands of rivets (species).
 - If every passenger starts popping a rivet to take home (species extinct), it may not affect flight safety initially but as more and more rivets are removed the plane becomes dangerously weak.
 - Furthermore, which rivet is removed may also be critical.

- Loss of rivets on the wings (key species) is obviously a more serious threat to flight safety than loss of a few rivets on the seats or windows inside the plane.

Loss of Biodiversity:

- The IUCN Red List (2004) documents the extinction of 784 species.
- Recent extinction includes :
 - Dodo (Mauritius).
 - Quake (Africa)
 - Thylacine (Australia)
 - Stiller's cow (Russia)
 - Three subspecies of tiger (Bali, Java, Caspian).
- Since the origin and diversification of life on earth there were **five episodes of mass extinction** of species.
- The sixth mass Extinctions in progress now.

How the 'sixth Extinction' is different from the previous five extinctions?

- The current extinction rate is 100 to 1000 times faster.
- All others are pre-human period, this one is anthropogenic.

Effect of biodiversity loss:

- Decline in plant production.
- Lowered resistance to environmental perturbations such as drought.
- Increased variability in certain ecosystem processes such as plant productivity, water use and pest & disease cycle.

Causes of biodiversity loss:

- The present loss is all due to human activity (anthropogenic)
- There are four major causes "**The Evil Quartet**" are as follows :

➤ **Habitat loss and fragmentation:**

- Most important cause driving animals and plants to extinct.
- The tropical rain forest reduced to 6 % from 14 % of earth land surface.
- The Amazonian rain forest is called as '**lungs of the planet** 'is being cut cleared for cultivating soya beans.
- Degradation of many habitat by pollution is also threatens the loss of diversity.
- Large areas are broken into figments also the cause of diversity loss.

➤ **Over-exploitation:**

- When 'need' turns to 'greed' it leads to over-exploitation of natural resources.
- Many species extinctions in the last 500 years (Stiller's cow, passenger pigeons) were due to over-exploitation.
- Many marine fish populations around the world are over harvested.

➤ **Alien species invasion:**

- The alien species became invasive and cause decline or extinction of indigenous species.
- **Nile perch** introduced into Lake Victoria in east Africa led to extinction of 200 species of **cichlid fish** in the lake.
- **Parthenium** (carrot grass), **Lantana** and water hyacinth (**Eichornia**) posed a threat to indigenous species.
- African cat fish **Clarias gariepinus** for aquaculture purposes is posing a threat to indigenous catfishes in our rivers.

➤ **Co-extinction:**

- When a species becomes extinct, the plant and animal species associated with it and obligatory way also become extinct.
- Extinction of **Host species** leads to extinction of the **parasite** also.
- Co-evolved **plant-pollinator** mutualism where extinction of one invariably lead to the extinction of the other.

BIODIVERSITY CONSERVATION :

Why should we conserve Biodiversity?

- Reason for conservation biodiversity is grouped into three categories :
 - **Narrowly utilitarian.**
 - **Broadly utilitarian**
 - **Ethical**

Narrowly utilitarian:

- Human derive countless direct economic benefits from nature.
- Food (cereals, pulses, fruits), firewood, fiber, construction material.
- Industrial products (tannins, lubricants, dyes, resins, perfumes)
- Products of medicinal importance.
- **Bio-prospecting** : Exploring molecular genetics and species-level diversity for products of economic importance.

Broadly Utilitarian:

- Amazonian forest along produce 20% of oxygen during photosynthesis.
- **Pollinator layer** : Bees, bumblebees, birds and bat that pollinate the plant without which seed cannot be produced by plants.
- Aesthetic pleasure we get from the biodiversity.

How do we conserve biodiversity?

In situ conservation:

- When we conserve and protect the whole ecosystem, its biodiversity at all level is protected – we save the entire forest to save the tiger. This approach is called **in situ (on site) conservation.**

- **Biodiversity hot spot** : Regions with very high levels of species richness and high degree of **endemism** (species confined to that region and not found anywhere else).
- Hot spot in biodiversity is also regions of accelerated habitat loss.
- Out of 34 hot spot in the world, three hot spot located in India :
 - Western Ghats and Sri Lanka.
 - Indo-Burma.
 - Himalaya.
- Other protected area under in situ conservations are :
 - 14 biosphere reserve
 - 90 national park
 - 448 wild life sanctuary
- **Sacred groves** : Tract of forest were set aside, and all the trees and wildlife within were venerated and given total protection.

Ex situ conservation:

- Threatened animals and plants are taken out from their natural habitat and placed in special setting where they can be protected and given special care.
- Zoological Park.
- Botanical garden
- Wildlife safari.
- Conservation of gamete by **cryopreservation**.
- Genetic strains are preserved in **seed bank**.

Convention on Biodiversity:

- **“The earth Summit”** held in Rio de Janeiro in 1992 called upon all nations to take appropriate measures for conservation of biodiversity and sustainable utilization of its benefits.
- **World Summit** on Sustainable development held in 2002 in Johannesburg (South Africa), 190 countries pledged their commitment to achieve by 2010 a significant reduction in the current rate of biodiversity loss at global, regional and local level.

CHAPTER-16

ENVIRONMENTAL ISSUES

Pollution: is any undesirable change in physical, chemical or biological characteristics of air, land, water or soil.

Pollutant: Any solid, liquid or gas released into the environment in such a huge quantity that makes our environment unhealthy is called a pollutant.

Environment (protection) Act, 1986 to protect and improve the quality of our environment (air, water and soil)

AIR POLLUTION AND ITS CONTROL :

Effect of air pollution:

- Cause injury to all living organisms.
- Reduce growth and yield of crops.
- Cause premature death of plants.
- Affects the respiratory system of human beings.
- Particulate size 2.5 micrometers or less are responsible for breathing and respiratory symptoms like irritation, inflammation and damage to the lungs and premature death.

Pollution caused by thermal power plant:

- Sources of **particulate matter:** thermal power plant, smelters
- These plants release particulate matter and gaseous air pollutant.
- A harmless gas released by these plants is **Nitrogen** and **Oxygen**.

Prevention of air pollution (ways to remove particulate matter):

- **Electrostatic precipitator :**
 - Widely used to remove particulate matter in the exhaust from a thermal power plant.
 - Electrode wires that are maintained at several thousand volts, which produce a corona that releases electrons.
 - Electron binds with particulate matter giving them a net negative charge.
 - Positively charged collecting plates attract the charged dust particle.
- **Scrubber:**
 - Removes gases like **sulphur dioxide**.
 - The exhaust is passed through a spray of water or lime.
- **Methods to reduce vehicular pollution:**
 - Use of lead free petrol or diesel can reduce vehicular pollution.
 - **Catalytic converter :**

- Having expensive metals namely platinum, palladium and rhodium as the catalyst.
- These metals reduce emission of poisonous gases.
- The **unburnt hydrocarbons** are converted into **CO₂ and H₂O**.
- **Carbon monoxide** and **nitric oxide** are changed to **carbon dioxide** and **nitrogen gas** respectively.
- Motor vehicle equipped with catalytic converter should use **unleaded petrol** because lead in the petrol **inactivates** the **catalyst**.

Controlling Vehicular pollution :

- **A case study of Delhi : Use of CNG (compressed natural gas).**

Advantages of CNG :

- CNG burns most efficiently.
- Very little remain unburnt.
- Cannot be siphoned
- Cannot be adulterated like petrol or diesel.
- CNG is cheaper than petrol and diesel.

Problem of use of CNG:

- Difficulty in laying down pipelines to deliver CNG.
- Non-assurance of uninterrupted supply.

Other parallel steps taken in Delhi:

- Phasing out old vehicles.
- Use of unleaded petrol.
- Use of low sulphur petrol and diesel.
- Use of catalytic converter in vehicle.
- Application of strict pollution level norms for vehicle.

- **New auto fuel policy to cut down vehicular pollution.**

- Steadily reducing the sulphur and aromatic content in petrol and diesel fuels.
- **Euro-II norms :**
 - Sulphur reduced to 350 ppm in diesel.
 - Sulphur reduced to 150 ppm in petrol
 - Aromatic hydrocarbon to be reduced to 42%.
 - Up gradation of vehicle engines.

- **Due to above steps taken by Delhi Govt. there is substantial fall in CO₂ and SO₂ level between 1997 and 2005.**

NOISE POLLUTION :

- Undesirable high level of sound is called **noise pollution**.

Harm full effect of noise pollution:

- Psychological and physiological disorder in humans.

- High sound level, 150dB or more may damage ear drums.
- Noise causes sleeplessness
- Increased heart rate.
- Altered breathing pattern.

Prevention of Noise Pollution:

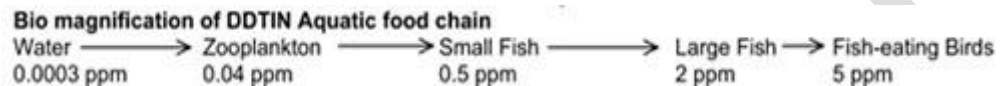
- Use of sound absorbent materials or by muffling noise in industries
- Demarcation of horn free zones around hospitals and schools.
- Permissible sound levels of crackers,
- Timings after which Loudspeakers cannot be played .

WATER POLLUTION AND ITS CONTROL :

Domestic sewage and industrial effluents:

- A mere 0.1 percent impurities make domestic sewage unfit for human use
- Sewage contains dissolve salts like nitrates, phosphates, other nutrients, toxic metal ions and organic compounds.
- The amount of organic matter in water is estimated by **BOD**.
- **Biochemical oxygen demand:** the amount of Oxygen required oxidizing all organic matter present in one liter of water.
- **Changes take place on discharge of sewage into the river.**
 - Micro-organism involved in biodegradation of organic matter in the receiving water body consume a lot of oxygen, hence there is sharp decline in dissolved oxygen downstream from the point of discharge.
 - Due to low DO there is mortality of fish and other aquatic animals.
- Presence of large amount of nutrients in water also causes excessive growth of **Planktonic** (free floating) algae, called **algal bloom**.
 - Algal bloom imparts distinct color to water bodies.
 - Deterioration of water quality and fish mortality.
 - Some bloom-forming algae are extremely toxic to human and animals.
- **The world's most problematic aquatic weed is water hyacinth (*Eichhornia crassipes*) called 'Terror of Bengal'.**
 - Introduced to India for their lovely flowers.
 - Excessive growth causes blocks in waterways.
 - They grow abundantly in eutrophic water bodies.
 - Causes imbalance in ecosystem and dynamics of water body.
- **Sewage associated with diseases:**
 - Sewage from home and hospital contain pathogenic microbes.
 - Discharge of such sewage without proper treatment causes diseases like dysentery, typhoid, jaundice, cholera etc.
- **Toxic heavy metals (defined as elements with density > 5g/cm³) released from:**
 - Petroleum industry.
 - Paper manufacturing.
 - Metal extraction and processing.

- Chemical manufacturing industries.
- **Bio-magnifications** : Increase in concentration of the toxicant at successive trophic level is called biological magnification or bio-magnifications.
 - Toxic substance accumulated by an organism cannot be metabolized or excreted.
 - The accumulated toxic passed to the next trophic level.
 - This phenomenon is well known for **mercury** and **DDT**.
- **Bio magnification of DDT in Aquatic food chain.**



- **Eutrophication:** The process of nutrient enrichment of water and consequent loss of species diversity is referred to as **Eutrophication**.
- **Natural Eutrophication:**
 - Streams draining into the lake increase nutrients like nitrogen and phosphorus.
 - Increase in nutrient encourages growth of aquatic organisms.
 - Over centuries, as silt and organic debris pileup the lake grows shallower and warmer.
 - Warm-water organisms dominate over that thrive in a cold environment.
 - Marsh plants take root in the shallows and begin to fill in the original lake basin.
 - Eventually the lake gives way to large masses of floating plants (bog), finally converting into land.
- **Cultural or Accelerated Eutrophication:**
 - Pollutants from man's activities like effluents from the industries and homes can radically accelerate the aging process. This phenomenon is called **Cultural** or **Accelerated Eutrophication**.

Causes:

- Sewage and agricultural and industrial wastes.
- Prime contaminants are nitrates and phosphates.

Effects:

- Unsightly scum and unpleasant odors.
- Robbing the dissolved oxygen from water.
- Pollutant inflow kills the fish.
- Decomposition of dead fish causes further depletion of DO.
- Finally a lake can literally choke to death.

Thermal pollution:

Cause:

- Heated (thermal) waste waters flowing out of electricity-generating units. e.g.- Thermal power plants.

Effects:

- Thermal wastewater eliminates or reduces the number of organism sensitive to high temperature.
- Enhance the growth of plants and fish in extremely cold areas but only after causing damage to the indigenous flora and fauna.

A case study of integrated waste water treatment:

- Wastewater including sewage can be treated in an integrated manner, by utilizing a mix of artificial and natural process.
- It has been done in town of Arcata, in the northern coast of California.
- **The treatment is done in two stages :**
 - The conventional sedimentation, filtering and chlorine treatment are given.
 - The biologist developed a series of six connected marshes over 60 hectares of marshland.
 - Appropriate plants, algae, fungi and bacteria were seeded into this area, which neutralize, absorb and assimilate the pollutant
 - The water flows through the marshes, it get purified naturally.
- The marshes also constitute a sanctuary, with high level of biodiversity in the form of fishes, animals and birds that now reside there.
- A citizens group called **Friends of the Arcata Marsh (FOAM)** is responsible for the upkeep and safeguarding of this project.

Ecological sanitation:

- Ecological sanitation is a sustainable system for handling human excreta, using dry composting toilets.
- This is a practical, hygienic, efficient and cost-effective solution to human waste disposal.
- With this composting method human excreta can be recycled into a resource (as natural fertilizer).
- 'EcoSan' toilets are being used in Kerala and Srilanka.

SOLID WASTES:

- **Solid wastes** refer to everything that goes out in trash.
- **Municipal solid wastes** are wastes from homes, offices, stores, schools, hospitals etc. It comprises paper, food wastes, plastics, glass, metals rubber, leathers, textiles etc.
- Open dump of these wastes serve as the breeding ground for rats and flies.
- **Sanitary landfills** were adopted as substitute for open-burning dumps.

Sanitary landfills:

- Wastes are dumped in a depression or trench after compaction and covered with dirt every day.

Disadvantages:

- Shortage of space for huge garbages.
- Danger of seepage of chemicals, polluting the ground water resources.

Solution to solid wastes:

- All solid wastes are categorized into three types :
 - **Bio-degradable.**
 - **Recyclable.**
 - **Non-biodegradable.**
- All the garbage generated is sorted first.
- Recyclable material to be separated and send for recycles.
- Biodegradable wastes can be put into deep pits in the ground and be left for natural breakdown.
- Only Non-biodegradable wastes are left and required to be disposed.

Prevention:

- The need to reduce our garbage generation should be a prime goal.
- Reduction in use of plastics and use of eco-friendly packaging.
- Carrying cloth or other natural fiber carry bags when we go shopping.
- Refusing polythene bags.

A Case study of Remedy for Plastic wastes (Ahmad Khan) :

- **Polyblend**, a fine powder of recycled modified plastic, was developed by his company.
- Polyblend is mixed with bitumen that is used to lay roads.
- It increases the water repelling property of bitumen, and helped to increase road life by a factor of three.
- The raw material used for polyblend is plastic film waste.

Hospital wastes:

- Hospitals generate hazardous wastes that contain disinfectants and other harmful chemicals, and also pathogenic organisms.
- The use of **incinerators** is crucial to disposal of hospital wastes.

Electronic wastes (e-wastes):

- Irreparable computers and other electronic goods are known as **electronic wastes (e-wastes)**.
- E- Wastes are buried in landfills or incinerated.
- Metals like copper, iron, silicon, nickel and gold are recovered during recycling process of e-wastes.
- Manual recycling process exposes workers to toxic substances present in e-wastes.
- Recycling is the only solution for the treatment of e-wastes.

AGRO-CHEMICAL AND THEIR EFFECTS :

- Use of inorganic fertilizers and pesticides has increased manifold for enhancing crop production.
- Pesticides, herbicides, fungicides etc. are being increasingly used.

- These are toxic to non-target organisms that are important components of the soil ecosystem.
- Increasing amounts of artificial fertilizers causes eutrophication.

A Case study of organic farming (Ramesh Chandra, Dagar of Sonipat) :

- Integrated organic farming is a cyclical, zero waste procedure, where waste products from one process are cycled in as nutrients for other processes.
- Maximum utilization of resource and increase the efficiency of production.
- He includes bee-keeping, dairy management, water harvesting, composting and agriculture in a chain of processes, which support each other and allow an extremely economical and sustainable venture.

Advantages:

- There is no need of use of chemical fertilizers for crops
- Cattle excreta are used as manure.
- Crop waste used to create compost, which can be used as a natural fertilizer or can be used to generate natural gas for energy need.

RADIOACTIVE WASTES :

- Nuclear energy was hailed as a non-polluting way for generating electricity.
- Later on it was realized that it has two very **serious inherent problem** :
 - Accidental leakage, as occurred in Three Mile Island and Chernobyl.
 - Safe disposal of radioactive wastes.
- Radiation from radioactive waste causes mutation at very high rate.
- High dose of nuclear radiation is lethal, but lower doses create genetic disorders and also cause cancer.

Disposal of nuclear wastes:

- Storage of nuclear waste after sufficient pre-treatment should be done in suitably shielded containers buried within the rocks about 500m deep below the earth's surface.

GREEN HOUSE EFFECT AND GLOBAL WARMING :

- The term "Greenhouse effect" has been derived from a phenomenon that occurs in greenhouse.
- In a greenhouse, the glass panel lets the light in but does not allow heat to escape. Therefore, the greenhouse warms up very much like inside a car that has been parked in the sun for a few hours.
- The greenhouse effect is a naturally occurring phenomenon that is responsible for heating of Earth's surface and atmosphere.
- Without greenhouse effect the average temperature at surface of earth would have been a chilly -18°C rather than the present average of 15°C .

- Clouds and gases reflect about one-fourth of the incoming solar radiation and absorb some of it but half of incoming solar radiation falls on Earth's surface heating it while a small portion is reflected back.
- Earth's surface re-emits heat in the form of infrared radiation but some part of this does not escape into space because of atmospheric gases (e.g.- carbon dioxide, methane etc.).
- The molecule of these gases radiate heat energy and a major part of which again comes to Earth's surface, thus heating it up once again.
- Carbon dioxide and methane are commonly called as greenhouse gases because they are responsible for greenhouse effect.
- Increase in the level of greenhouse gases has led to considerable heating of Earth leading to **global warming** or **enhanced greenhouse effect**.
- During the past century, the temperature of Earth has increased by 0.6° C.

Effect of global warming:

- Deleterious changes in the environment and resulting in odd climatic changes (e.g.- El Nino effect).
- Increased melting of polar ice caps as well as of other places like the Himalayan snow caps.
- Rise in sea level that can submerge many coastal areas.

Control of global warming:

- Reduce use of fossil fuel.
- Improving efficiency of energy usage.
- Reducing deforestation.
- Promoting afforestation programme.
- Slowing down growth of human population.
- International initiative to be taken to reduce emission of greenhouse gases.

OZONE DEPLETION IN THE STRATOSPHERE :

- **'Bad' ozone** formed in the lower atmosphere (troposphere) that harms plants and animals.
- There is **'good' ozone** also. This ozone is found in the upper part of the atmosphere called **stratosphere** and it acts as a shield absorbing ultraviolet radiation from the sun.
- The thickness of ozone layer is measured in terms of **Dobson units (DU)**.
- Ozone (O₃) gas is continuously formed by the action of UV rays on molecular oxygen and also degraded into molecular oxygen in the stratosphere.
- There should be proper balance of formation and degradation of ozone.

Ozone depletion:

- Balance of ozone in stratosphere is disrupted due to enhancement of ozone degradation by **chlorofluorocarbons (CFCs)**.
- CFCs find wide use as refrigerants.

- CFCs discharged in the lower part of atmosphere move upward and reach stratosphere.
- In stratosphere, UV rays acts on CFCs and release **active Cl-atoms**.
- Cl-atom degrades ozone releasing molecular oxygen.
- Cl-atom acts as catalysts and not consumed during reaction.
- Whatever CFCs are added to the stratosphere, they have permanent and continuing affects one Ozone levels.
- The depletion is marked particularly over the **Antarctic region**. This has resulted in formation of a large area of thinned ozone layer commonly called as the **ozone hole**.

Effects of UV rays:

- UV radiations shorter than UV-B are almost completely absorbed by Earth's atmosphere, if the ozone layer is intact.
- DNA and proteins of living organisms are damaged by UV rays as they potentially absorb it.
- The high energy of UV rays breaks the chemical bond in these molecules.
- UV-B damages DNA and mutation may occur.
- It causes ageing of skin.
- Damage skin cells and causes skin cancers.
- In human eye, cornea absorb UV-B radiation and high dose of UV-B causes inflammation of cornea called **snow-blindness, cataract etc**.
- Such exposes may damage cornea.

Prevention:

- **Montreal Protocol** was signed at Montreal (Canada) in 1987 to control emission of ozone depleting substances.
- Many efforts are being made to reduce emission of ozone depleting substances.

DEGRADATION BY IMPROPER RESOURCE UTILIZATION AND MAINTENANCE :

Soil erosion :

- The removal of top fertile layer due to human activities.
- **Reasons :**
 - Over cultivation
 - Unrestricted grazing
 - Deforestation
 - Poor irrigation practices

Water logging and soil salinity :

- Irrigation with proper drainage, leads to water lodging in the soil.
- Draws salt to the surface of the soil.
- The salt starts collecting at the roots of the plants.
- The salt damages the roots and crop productions.

Deforestation:

- Conversion of forested areas to non-forested one.

How deforestation does occurs :

- Slash and burn agriculture/jhum cultivation.
- Farmers cut down the trees of the forest and burn the plant remains.
- Ash is used as fertilizer and land is used for farming or cattle grazing
- Later, Land is left uncultivated for several years for replenishment of minerals

Effects of deforestation :

- Leads to global warming due to excess carbon-dioxide
- Loss of biodiversity
- Damage to hydrological cycle
- Leads to soil erosion
- Desertification of land

Reforestation :

- Restoring forest that was existing earlier E.g. Observing Van-Mahotsav
- It also occurs naturally.
- Aforestation develops a forest in a new area where no such forest existed in that area.

A case study of people's participation in forest conservation :

- A king of Jodhpur wanted to arrange wood for his new palace in 1731.
- Few people of Bishnoi community hugged the trees and asked to cut them first rather than cutting trees.
- 365 persons lost their lives in this act.
- A small temple is now present there in remembrance of this act.
- Amrita Devi Bishnoi's Wild Life Protection Award is instituted for individuals of rural areas who take keen interest in protecting wild life.
- Chipko movement.
- It was started by local women of Garhwali; they hugged the trees to protect them from the axes of contractors.
- Joint forest management (JFM).
- Strategy Government of India in 1980.
- Local communities worked with the government to save the forest.
- Communities get forest products for encouragement.

Abbreviation:

- CPCB : Central Pollution Control Board
- BOD : Biological Oxygen Demand
- CNG : Compressed Natural Gas
- FOAM : Friends of Arcata Marsh
- JFM : Joint Forest Management.