STUDY MATERIAL FOR BOTANY (Class 12th)

State Institute of Education & Trainings, Kashmir

STUDY MATERIAL BASED ON JK BOSE SYLLABUS

CLASS: 12TH

Subject : BOTANY

PATRON & MENTOR

Mr. Muhammad Younis Malik

DIRECTOR
SCHOOL EDUCATION KASHMIR

CHIEF COORDINATOR

Ms. Suriya Banoo

JT. DIRECTOR (TRGS) /
PRINCIPAL, SIE KASHMIR

CO-ORDINATOR

Mr. Manzoor Ahmad Wani

RESEARCH OFFICER SIE KASHMIR

CONTRIBUTORS

Dr. Mudasir Ahmad Shah

LECTURER GOVT. BOYS HR. SEC. SCHOOL SOPORE

Dr. Riyaz Ahmad Mir

LECTURER GOVT. GIRLS HR. SEC. SCHOOL KUPWARA

Preface

There is a unanimous agreement that science learners engage in a process of knowledge integration where they make sense of diverse information including their own experiences, classroom instruction, and related ideas rather than absorb information from courses.

In order to eliminate the disparity in learning opportunities of students that exists due to regional and economical reasons, worthy Director School Education, Kashmir Mr. Mohammad Younis Malik envisioned a project to provide a uniform, concise and relevant study material to the student community of senior secondary classes. The need for generating study material was also felt due to fast changing global educational scenario as well as because of distinctive local requirements. Although ample material is available both in the form of books and as online sources, the information is scattered and enormous, therefore difficult for a teenager to process it according to his needs. A good proportion of students cannot afford to buy these voluminous books because of their expensive costs. The current project was therefore envisaged in order to present a filtered, relevant and all encompassing study material that will cater to the needs of students even residing in the remotest parts of the region. Keeping it available free of cost in the form of an e-book that too in a printable format is another uniqueness of the material. It is not only the students but the subjects teachers too will find it beneficial for their classroom teaching. We hope that the students will grab the opportunity and use this one stop study material in best possible way.

As per the guidelines of National Curriculum Framework (NCF 2005), it is emphasized that knowledge should be connected with the outside world. Such knowledge will nurture thinking process, generate curiosity and develop creativity among the pupils. The contributing authors have tried to follow NCF by connecting the knowledge of Botany with the locally available information. Unlike information available elsewhere, most of the examples of plants and the photographs cited in the material are native and local names (kashmiri names) have been added to plants so that the students can understand better and construct knowledge in their own capacity.

We are thankful to Ms. Suriya Banoo, Joint Director/Principal SIE Srinagar for providing us an opportunity to be part of the material development team. The efforts of Scientific Officer, Mr. Manzoor Ahmad who coordinated this joint venture are commendable. He was very keen for the timely completion of the project. We would also like to thank Dr. Pervez Maqbool Lone and Dr. Syed Sabhi Andrabi for their valuable suggestions.

Mudasir Ahmad Shah
Riyaz Ahmad Mir

MODEL PAPER BIOLOGY

The Jammu and Kashmir State Board of School Education
Academic Division, New Campus, Bemina Srinagar

Model Paper

BIOLOGY (12th)

Section A (Zoology)

Max. Marks: 35 **Time 1 :30 hrs**

Instruction:

SECTION A

Long Answer Type Questions:

1. Reproduction generally involves two parents. Does it occur uniparently. How?

OR

Assisted reproductive techniques bring hope to infertile couples. Explain How.

5Marks

2. Describe Darwins theory of Natural Selection.

OR

Sex is generally determined by chromosomes of an organism .Does environment also determines sex. Illustrate with examples. **5 Marks**

SECTION B

Short Answer Type Questions:

3. What changes occur in the uterus of a human female if the ovum does not receive a sperm? **3 Marks**

4. What phenotypic symptoms are produced in an individual due to trisomy of 21st chromosome? **3 Marks**

5. Match the entries under section A with the choice given under B and C. **3Marks**

	A	B	C
a.	*Blood and mucus with* faeces abdominal pain	*W. bancrofti*	*Anopheles*
b.	Periodic fever followed by sweating, shivering	*E. histolytica*	worm
c.	Excessive swelling due to blockade of lymph vessels feet and legs	Plasmodium	protozoa

6. How Genetic engineering helps in producing pest resistant crops? **3 Marks**

SECTION C

Short Answer Type Questions:

7. Draw a labeled sketch of human sperm? **2 Marks**

8. Mention two differences between analogous and homologous organs. **2 Marks**

9. Mention two applications of cloning. **2 Marks**

10. How does DNA fingerprinting helps in detecting a criminal. **2 Marks**

SECTION D

Objective Type Questions:

11. Explain ZIFT. **1 Mark**

12. Write the scientific name of Silk moth. **1 Mark**

13. Name the antibody present in Colostrum. **1 Mark**

14. Human genome is said to have approximately ----------base pairs. **1Mark**

15. Give one example of point mutation. **1 Mark**

Section B (Botany)

Max. Marks: 35 **Time 1:30 hrs**

SECTION A

Long Answer Type Questions:

1. How does dominance differ from incomplete dominance? Illustrate with suitable example.

OR

Hershey and Chase conducted an experiment on T_2 Phage using radioisotopes. **5 Marks**

What were there observations and conclusions?

2. Define an ecosystem. Write an account of energy flow in an ecosystem.

OR

Major pollutants of air are CO_2, SO_2 and NO_2. What are their sources and harmful effects on human beings and plants? **5 Marks**

SECTION B

Short Answer Type Questions:

3. Vegetative propagation does not lead to variations. Why? **3 Marks**

4. What are GM crops? Give two disadvantages of GM crops? **3Marks**

5. Biofertilizers are better than Chemical fertilizers. Justify? **3 Marks**

6. What is ecological succession? Distinguish between primary and secondary succession. **3 Marks**

SECTION C

Short Answer Type Questions:

7. Why cross pollination cannot occur in cleistogamous flowers. **2 Marks**

8. What do you mean by degeneracy of genetic code? **2Marks**

9. How many phosphorus atoms are there in a DNA double helix having 10,000base pairs? **2 Marks**

10. An orchid plant grows on the branch of a mango tree. Give the interaction between the two. **2 Marks**

SECTION D

Objective Type Questions:

11. Define apomixis? **1 Marks**

12. How many cells are there in a typical angiospermic embryo sac? **1 Marks**

13. Define totepotency? **1Marks**

14. Which type of ecological interaction is represented in lichens? **1Marks**

15. What are schiophytes? **1Marks**

REPRODUCTION IN FLOWERING PLANTS

Reproduction: One of the essential characteristics of living organism is that they reproduce i.e. produce their own kind. Reproduction is a biological process by which an organism produces new individuals (offspring) similar to itself. Reproduction ensures that a species survives over long periods of time, even though the individual members may die. Different Plants multiply in a variety of ways, however these methods of reproduction in flowering plants can broadly be grouped into two: *Asexual* and *Sexual*.

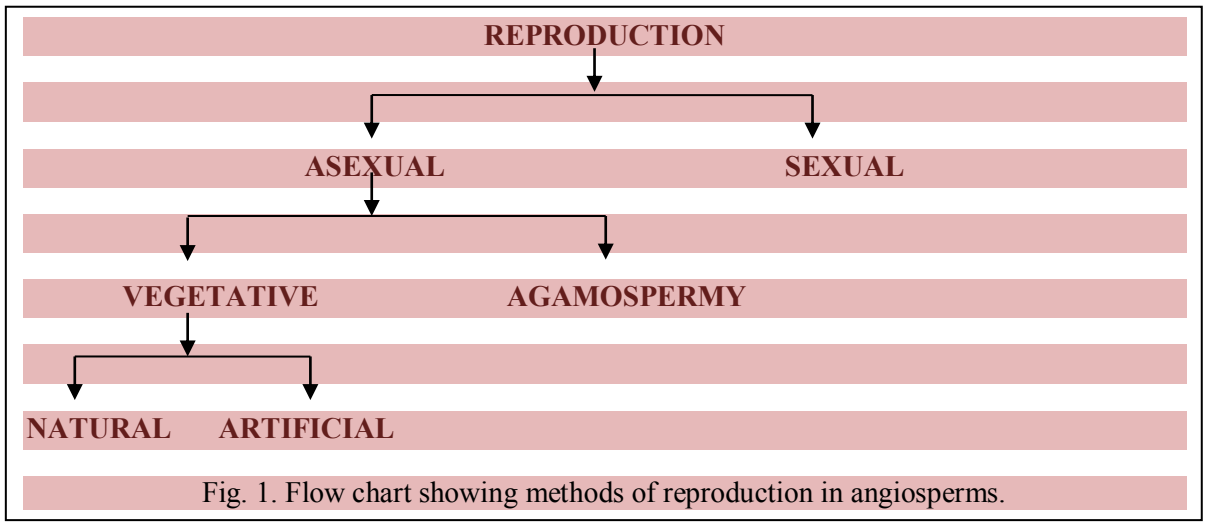


Fig. 1. Flow chart showing methods of reproduction in angiosperms.

ASEXUAL REPRODUCTION: That type of reproduction in which new individuals are produced without meiosis and fertilization is called Asexual reproduction. Asexual reproduction leads to the production of genetically identical individuals (clones) from a single parent. Angiosperms multiply asexually by two methods

1. **Vegetative reproduction**: A type of asexual reproduction in which new plants are produced from vegetative parts (roots, stems, leaves and buds) without producing embryos and seeds.
2. **Apomixis**: Although any reproduction that does not involve mixing of gametes is apomixis, but specifically it is that reproduction in which seeds are formed without meiosis and fertilization. It is also termed as agamospermy.

VEGETATIVE REPRODUCTION: In vegetative reproduction (also called Vegetative Propagation) a plant multiplies by using its vegetative parts. Although vegetative reproduction takes place by various methods like budding (Yeasts), Fragmentation

(Filamentous Algae; Bryophytes), Spore formation (Ferns; Fungi) etc., however in flowering plants vegetative propagation happens differently involving vegetative parts viz. roots, stems, leaves and shoot buds. Vegetative reproduction in higher plants can be divided into two types:

A. Natural Vegetative Propagation
B. Artificial Vegetative Propagation

A. Natural Modes of Vegetative Propagation: Several plants reproduce naturally using various vegetative parts.

1. Vegetative propagation by roots: Many plants form tuberous roots by storing food in them. These roots possess vegetative buds that develop leafy shoots and later separate thereby giving rise to new plants. Sweet potato (*Ipomoea batatus*) and Garden Dahlia (*Dahlia pinnata*) are common examples of plants that multiply by their roots.

2. Vegetative propagation by stems: Several flowering plants use stems for their multiplication. Such stems can be categorised into underground, sub-aerial and aerial stems.

a). Underground Stems: Many plants use their underground stem for multiplication. Such modified stems include:

Suckers: They are branches arising from the underground part of the erect shoot. They develop their own adventitious roots and later detach to form separate plants. Mints (*Mentha sp*; **Pudna**) and Chrysanthemums (*Chrysanthemum sp.*) that are commonly found in Kashmir reproduce by suckers.

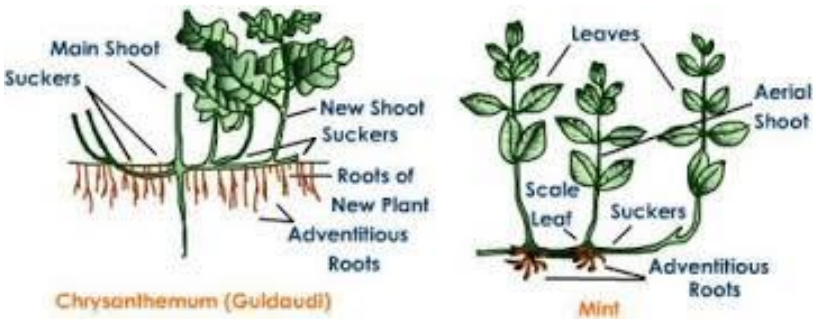


Figure 2: Sucker of Chrysanthemum and Mint. Source: Toppr.com

Rhizome: It is a horizontal underground stem that develops lateral shoots and adventitious roots at intervals (nodes). Plants that reproduce with the help of rhizome include Ginger (*Zingiber officinale;* **Adrakh**) and Turmeric (*Curcuma longa*; **Lidder**). Our local example for rhizome is Lotus (*Nelumbo nucifera*; **Nadur**).

Figure 3: Rhizome and flower of Lotus. Source: picuki.com; Shutterstock

Corm: It is a modified, short, upright swollen underground stem that bears many buds. Corms serve as perennating structures and organs of multiplication. Gladiolus (*Gladiolus dalenii*) and Saffron (*Crocus sativus*; **Zafran**) are common examples of corm.

Figure 4: Corm of Saffron. Source: pintrest.com

Bulb: A bulb is a rounded underground organ in which a highly reduced stem is surrounded by fleshy leaves. Onions (*Allium cepa;***Ganda**), garlic (*Allium sativum*; **Ruhan**) and Lillies (*Lillium sp.*) reproduce with the help of bulbs.

Tuber: These are swollen tips of modified underground stem branches. Each tuber possess several buds (eyes). Potato (*Solanum tuberosum*; **Oalva**) is a good example of tuber.

b). Sub-Aerial Stems (Creeping stems): Many plants are weak and creep on the surface of soil. They reproduce by forming stem branches which detach forming new plants. Creeping reproductive stems include:

Runners: These are creeping stems that produce adventitious roots at various nodes. Later they detach from the main plant and develop into independent plants. Lawn grass (*Cynodon dactylon*; **Dramun**) and creeping wood sorrel (*Oxalis corniculata*; **Tchok-tchin**) reproduce by runners.

Stolons: These are arched runner like structures that form roots and shoots on touching the ground. The arched parts later dry and decompose forming several new plants from a single parent plant. Stolons are commonly found in Strawberries (*Fragaria nubicola*; **Istabar**).

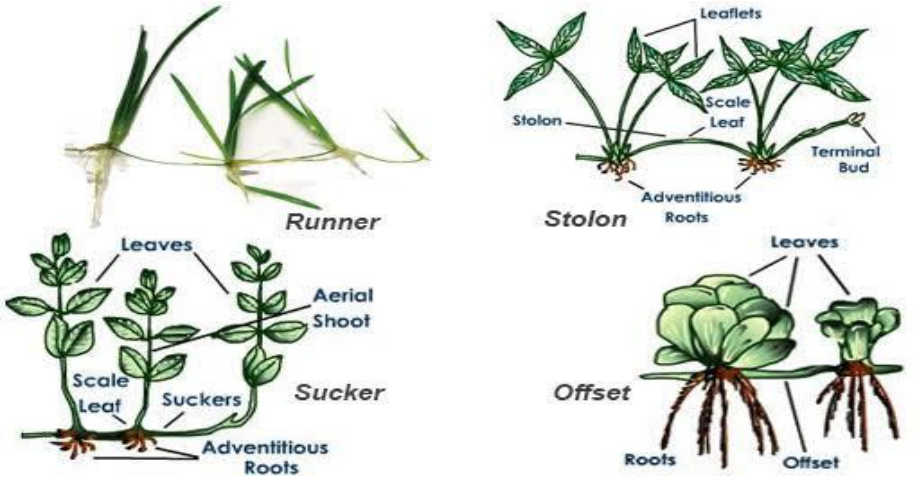


Figure 5: Runner, Stolon, Sucker and Offset. Source: Toppr.com

Offsets: A very short horizontal branch arising from the base of stem that develops adventitious roots and a tuft of leaves at the end. It is a one internode long runner of some aquatic plants. Common examples include Water Lettuce (*Pistia sp.*) and Water hyacinth (*Eichhornia crassipes*).

c). Aerial Stems: In many plants stem is segmented and these segments get detached to form new plants. In cacti each fragment (a Phylloclade) develops into a new plant after separating from parent plant. Prickly pear (*Opuntia ficus-indica*) is a common example.

Figure 6:*Opuntia sp*. Source: lucidcentral.org

Fig. 7: *Bryophyllum sp.* Source: Pedia.com

3. Vegetative propagation by Leaves: A few plants possess adventitious buds on their leaves (epiphyllous buds). Such leaf buds form new plants when the leaf is shed on ground. Miracle plant (*Bryophyllum pinnatum*) reproduces with the help of epiphyllous buds.

Fig. 8: Bulbils of Agave. Source: marriedtoplants.com

4. Bulbils: These are bulb-like fleshy buds that directly develop into fresh plants once they get detached from the parent plant. Century plant (*Agave americana*), *Dioscorea sp.* and some species of *Oxalis* reproduce via bulbils.

B. Artificial Methods of Vegetative Propagation: Horticulturists and plant growers have developed various methods for propagating crops using vegetative parts. Such methods of plant multiplication which require human intervention are termed as artificial methods. Artificial vegetative propagation is done using cutting, layering and grafting.

a) **Cutting**: In this method, parts of plant mainly stem or leaf with buds is cut off and planted in soil. These parts (cuttings) later develop adventitious roots and form new plants. Plants that are propagated by stem cuttings include willow (*Salix alba*, **Vir**;), Poplar (*Populus alba*; **Frast**), Rose (*Rosa sp.,***Gulab**) and Grape vine (*Vitis vinifera*; **Dachh**). *Begonia sp.* and some species of *Bryophyllum* are multiplied by leaf cuttings.

b) **Layering**: Here the branches of plant are induced to develop roots before they are cut from the parent plant. Depending on the habit of the plant, layering is of two types – mound layering and air layering.

1. ***Mound layering***: In mound layering (simple layering), lower soft stem branches are first defoliated, injured and then bent downwards. The portion with injury is covered with soil, however the growing tip is kept exposed. The injured portion under soil develops adventitious roots and this rooted branch is later separated from the parent plant. It is done in Jasmine (*Jasminium humile*; **Yasmin**), some varieties of Grapes (*Vitis vinifera*; **Dachh**) and Blackberry (*Rubus fruticosus*).
2. ***Air layering* (*Gootee***): It is a method of layering employed in propagating plants with hard rigid branches. Here girdling is done in aerial branches and the injured portion is covered with grafting clay (Soil and compost mixture) and then wrapped with polythene sheet. It develops roots around the injured portions and then the branch with roots is cut and planted in the soil. This method is employed in Pomegranate (*Punica granatum*; ***Daen***), Orange (*Citrus* x *sinensis*; ***sangtar***) and Litchi (*Litchi chinensis*).

c) **Grafting**: It is a technique by which two plants are vegetatively joined into one. Here an organic union or fusion of parts of two plants of the same or allied species is ensured. The plant part with root system (from which branches are removed) is called Stock or Root Stock and the portion that is grafted on the root stock is known as Scion. The union is made in such a manner that the xylem, cambium and the phloem

of the two parts coincide with each other. This method of propagation is employed in Apple (*Malus domestica*), Mango (*Mangifera indica*), Pear (*Pyrus communis*), etc. grafting is done through a variety of methods

i) **Tongue or Whip Grafting**: In this method, the stock and the scion (of the same diameter) are given oblique cuts of about two to three inches. The scion and stock are then notched for grip. The two parts are then fitted into each other tightly and moist grafting clay is applied around the joint followed by bandaging. Graft union occurs within a few days.

ii) **Wedge Grafting**: Here again the stock and the scion are almost of the same diameter. In this method a vertical cut is made through the diameter on the cut end of stock. The lower end of scion is shaped into a wedge like structure. The wedged end of scion is then fitted into the vertical notch of stock. Later moist grafting clay and cloth or polythene is wrapped on the joint.

iii) **Crown grafting**: In this case, several scions with wedge shaped ends are inserted into the peripheral slits of a stock in such a manner that it appears like a crown. Crown grafting is possible only in mature trees. Since the diameter of stock is larger compared to scion, it accommodates many scions simultaneously. The joints are later sealed with grafting clay and tied with cloth and polythene for some days.

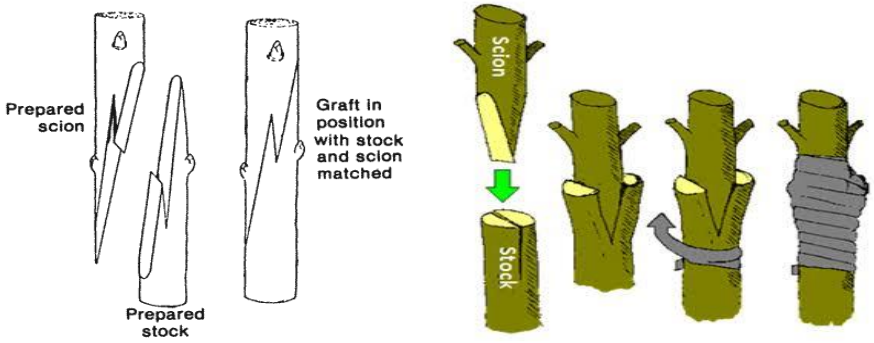


Fig. 9: Types of grafting: Tongue; Wedge; Crown and bud grafting . Source: rfcarchives.org; Galveston.agrilife.org; maaelu.postimees; gardening.stackexchange.com;

iv) **Bud grafting**: This method utilized an active bud instead of a stem branch as scion. A bud along with a portion of bark (acting as scion) is inserted under a T-shaped or H-shaped incision in the bark. The joint is sealed with bandage but the bud portion is kept exposed. After two to three weeks the bud resumes growth and forms a shoot. Later the stock above the level of graft union is removed.

MICROPROPAGATION: It is the artificial process of multiplying plants vegetatively through tissue culture or cell culture techniques. In this method a tissue or cell from parent plant is excised (now called explants) and grown on a predefined nutrient medium under aseptic conditions of a laboratory. This tissue divides profusely forming an undifferentiated mass of cells known as callus. The callus differentiates into small plantlets with the help of growth regulators. These plantlets are separated from each other and grown separately. They are finally shifted from lab to land where they grow into full plants. Such a method wherein small portions of plant in the form tissues or cells are used for propagation is known as micropropagation. This method of propagation is expensive and only employed when other artificial methods are either non-feasible or fail to give results.

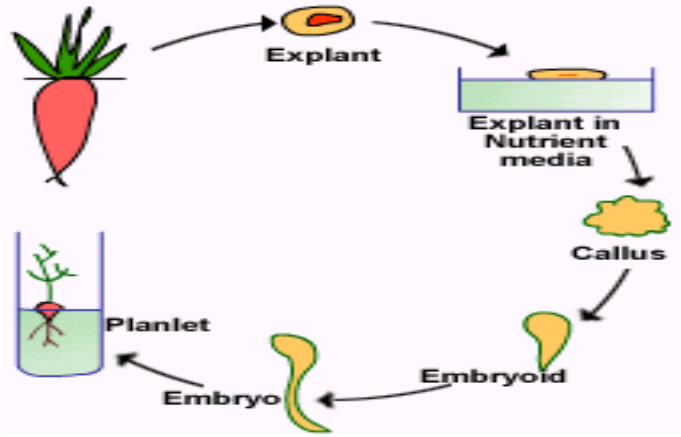


Fig. 10. Methodology of Micropagation. Source: toppr.com

Advantages of vegetative reproduction:

1. Vegetative propagation helps in propagation of those plants which have lost the power to produce seeds. Banana, Oranges, Grapes, Rose, Dahlia etc.
2. Such plants which produce only a small quantity of seeds are also propagated vegetatively.
3. It is a rapid, easier and cheaper method of multiplying those plants that have prolonged dormancy or poor viability.
4. All the plants produced by this method are clones to each other. They retain the characters of parent plant completely. Good quality is retained and conserved in vegetative reproduction.

5. Vegetative propagation by tissue culture produces disease free plants.

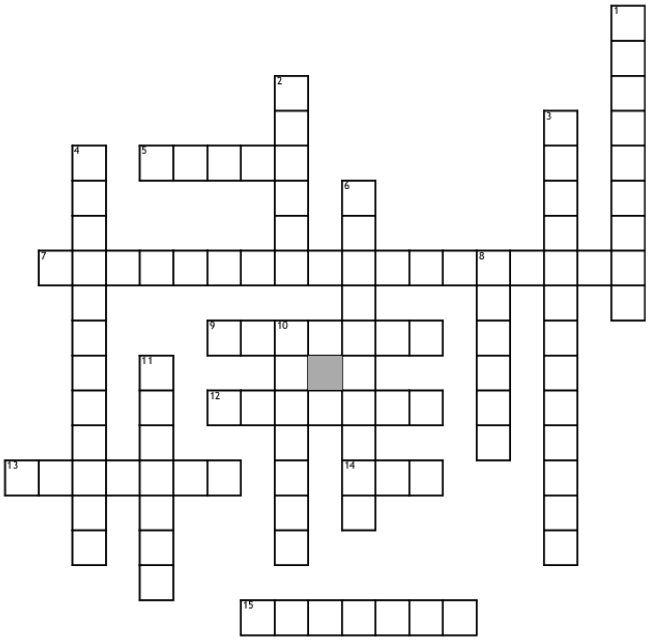
Disadvantages of vegetative reproduction:

1. Good quality cannot be introduced nor can bad quality be eliminated in plants grown by vegetative method.
2. Plants produced through this method have least adaptability to changing environment due to lack of variability.
3. Subsequent generations show a general fall in vigour and vitality.
4. Diseases contracted by parent plant propagates in all daughter plants.

SOLVE THE CROSSWORD PUZZLE

Name: _____ Date: _____ Period: _____

Asexual Reproduction



Across

5. Male gamete
7. Offspring produced by union
9. A small outgrowth of a parent organism develops into an offspring
12. Single set of each chromosome in a cell or cell nucleus
13. Cell division which reduces chromosomes
14. Female gamete
15. Cell division which divides chromosomes

Down

1. Young of living organisms
2. Sex cell
3. Combining the two gametes creating a new organism
4. Asexual reproductive process in which there is regrow the of lost or destroyed parts or organs
6. Substance or chemical that can cause cancer
8. Cells of body part become abnormal
10. Two sets of each chromosome
11. Parent organism splits into two

SEXUAL REPRODUCTION IN FLOWERING PLANTS

SEXUAL REPRODUCTION: The type of reproduction that results from the fusion of opposite gametes is known as sexual reproduction. The male and female gametes are produced inside anthers and ovules respectively by the process of meiosis. This fusion results in the formation of zygote which develops into embryo.

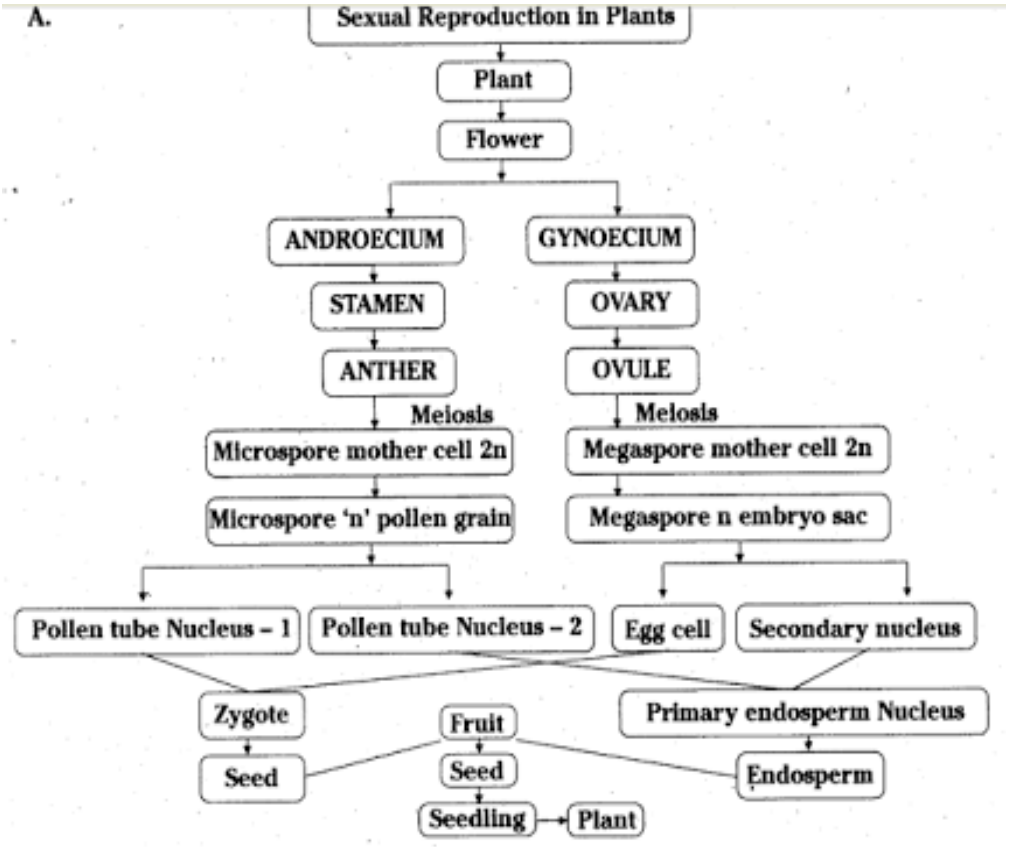


Fig. 11. Flow chart for sexual reproduction in flowering plants. Source: ask.learncbse.in

The Flower: The whole process of sexual reproduction takes place inside a specialized organ known as Flower. Flowers are objects of aesthetic, ornamental, social, religious and cultural value. A flower is a modified shoot formed from a floral bud. A typical flower consists of a

pedicel (stalk), receptacle, Calyx (sepals), Corolla (petals), Androecium (stamens) and Gynoecium (Pistil/Carpels). A stamen consists of a filament and anther while a pistil is made up of stigma, style and ovary.

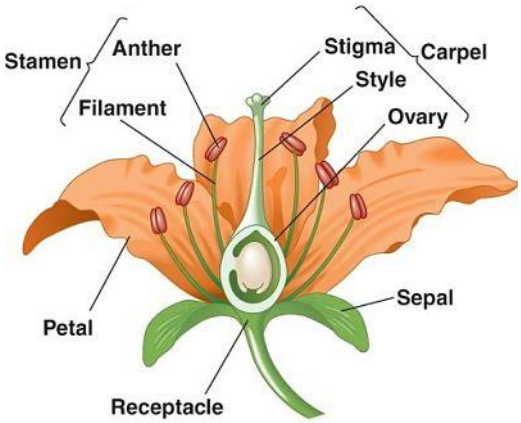


Diagram 12: Structure of flower. Source: askiitians.com

Structure of Anther: A typical angiosperm anther is bilobed with each lobe having two thecae, i.e., they are dithecous. Often a longitudinal groove runs lengthwise separating the theca. The bilobed nature of an anther is very distinct in the transverse section of the anther. The anther

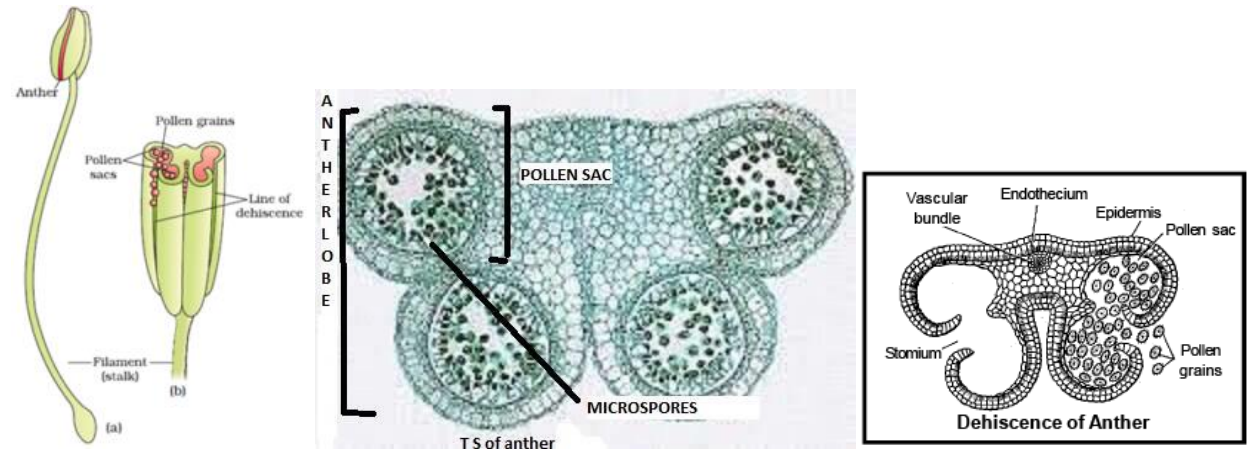


Fig. 13. Structure of anther; T.S of anther and dehiscence of anther. Source: Meritnation.com

is a four-sided (tetragonal) structure consisting of four microsporangia located at the corners, two in each lobe.

Structure of microsporangium: A typical microsporangium in a transverse section appears near circular in outline. It is surrounded by four concentric wall layers which include the epidermis, endothecium, middle layers (1-3) and the tapetum. The outer three wall layers perform the function of protection and help in dehiscence of anther to release the pollen. The innermost wall layer called tapetum nourishes the developing pollen grains. Tapetum also participates in the formation of pollen cell wall. Cells of the tapetum possess dense cytoplasm and generally have more than one nucleus.

Development of pollen sacs or Microsporangia: Development of microsporangia begins with the differentiation of ARCHESPORIAL cells in the hypodermis, at the four corners of a

young anther. The archesporial cells divide by periclinal divisions to give rise to outer Primary Parietal Layer and an inner Primary Sporogenous Layer. The cells of the primary parietal layer divide by successive divisions to form concentric layers of the pollen sac wall. The wall layers from periphery to center are:

i) A single layer of Epidermis
ii) A single layer of endothecium which helps in dehiscence of anther.
iii) One to three cell layers called Middle Layers which are ephemeral (short lived).
iv) Single layer of Tapetum that provides nourishment to sporogenous tissue and helps in formation of pollen wall.

Microsporogenesis: The cells of Sporogenous tissue inside the developing anther may divide in various planes and finally separate from each other to function as Microsporocytes or MICROSPORE MOTHER CELLS. Some of these MMCs degenerate and provide nourishment to growing microspores. Each MMC divide by meiosis to give rise to a four microspores (Microspore tetrad). The microspores within a tetrad later separate from each other to form Pollen grains. However compound pollen grains (unseparated pollen grains of a tetrad) are found in *Typha* and *Drosera* while in Orchids an entire anther lobe remains united as a unit called Pollinium.

Dehiscence of Anther: Inside the mature pollen sacs, the pollen grains dry up. The sterile partition wall between the two pollen sacs of an anther lobe gets destroyed forming a single chamber. Thus a mature anther has only two chambers one in each anther lobe. The cells of the endothecium contract from their outer thin walls so that anther lobe forms longitudinal slits. This results in the rupture of anther lobe walls in the region of stomium thereby dispersing the pollen grains.

Structure of mature pollen grain: Pollen grains are haploid structures. They are generally spherical measuring about 25-50 micrometers in diameter. Each pollen grain has a prominent two-layered wall, the outer exine and inner intine. Exine is hard and made up of sporopollenin which is one of the most resistant organic materials known. It can withstand high temperatures and strong acids and alkali. No enzyme that degrades sporopollenin is so far known. Pollen grain exine has prominent apertures called germ pores where sporopollenin is absent. Pollen grains are well preserved as fossils because of the presence of sporopollenin. The exine exhibits a fascinating array of patterns and designs. The inner wall of the pollen grain is called the intine. It is a thin and continuous layer made up of cellulose and pectin.

The cytoplasm of pollen grain is surrounded by a plasma membrane. At maturity, each pollen grain consists of two cells, a vegetative cell and a generative cell. In most angiosperms pollen grains are shed at two celled stage.

Development of Male gametophyte: The development of male gametophyte is uniform in all flowering plants. Microspore or immature pollen grain is the first cell of this gametophyte. The development of male gametophyte can be divided into two parts:

i) **Pre-pollination development**: After separation from the tetrad, the size of the nucleus increases and is displaced to peripheral position due to formation of a large central vacuole. Microspore nucleus then divides mitotically to produce two unequal cells, the large Vegetative cell (tube cell) and a small Generative cell. The generative cell acquires ellipisoidal, lenticular or horseshoe shape and later comes to lie free inside the cytoplasm of Vegetative cell. Generative cell also has a limited number of organelles and in contrast vegetative cell is packed with organelles and food material. In majority of angiosperms, pollen grains are shed at this two celled stage.

ii) **Post-pollination development**: The released pollen grain reach stigma by the process of pollination. The pollen grain absorbs water and swells. It releases wall held recognition factors. After mutual recognition, the vegetative cell enlarges and comes out through one of the apertures in the form of pollen tube which grows through the style. Gradually the tube nucleus migrates into pollen tube. This is followed by migration of generative cell into pollen tube. The generative cell divides here to form two small male gametes. The tube nucleus later degenerates.

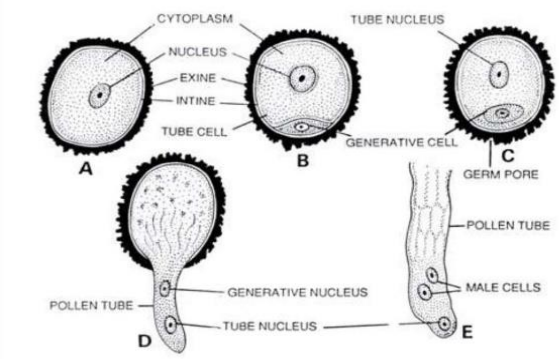


Fig. 14. Development of male gametophyte. Source: Slideshare.net

The Gynoecium (Pistil or carpel): A typical pistil or carpel consists of three parts, a swollen basal part called ovary, a stalk called style and a terminal receptive portion called stigma. Inside the swollen ovary are present ovules or Megasporangia.

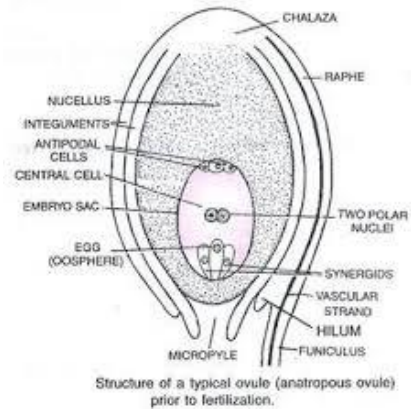


Fig. 15. Structure of ovule. Source: capebiology

Megasporangium or Ovule: A typical angiospermic ovule is a small structure attached to the placenta by means of a stalk called funicle or funiculus. The body of the ovule fuses with funicle in the region called hilum. Thus, hilum represents the junction between ovule and funicle. Each ovule has one or two protective envelopes called integuments. Integuments encircle the whole body of ovule except at the tip where a small pore called the micropyle is present. Opposite to the micropylar end, is the chalaza, representing the basal part of the ovule. Inside the integuments is a mass of cells with abundant reserve food and called the nucellus. Located in the nucellus is the embryo sac or female gametophyte. An ovule generally has a single embryo sac formed from a megaspore.

Development of Female Gametophyte: Female gametophyte or embryosac development takes place in two steps.

Megasporogenesis: The process of formation of haploid megaspores from the diploid megaspore mother cell by the process of meiosis is called megasporogenesis. Within the ovule a single cell in the micropylar region of the nucellus differentiates as archesporium. This archesporial cell may directly function as megaspore mother cell or divides into outer parietal cell and inner sporogenous cell. The sporogenous cell enlarges and acts as megaspore mother cell. The MMC undergoes meiotic division and results in the production of a linear row of four megaspores. In a majority of flowering plants, only one of the four megaspores (lying on the chalazal side) is functional while the other three degenerate. The functional megaspore develops into the female gametophyte (embryo sac). This method of embryo sac formation from a single megaspore out of four is termed **monosporic development**. However bisporic embryosacs (embryosac is formed from two megaspores) and tetrasporic embryosacs (all the four megaspores participate in embryosac formation) do exist.

Development of female gametophyte (embryosac) from the megaspore: The nucleus of the functional megaspore divides mitotically to form two nuclei which move to the opposite poles due to the appearance of a large central vacuole, forming the binucleate embryo sac. Two more sequential mitotic nuclear divisions at each pole result in the formation of the 4-nucleate and later the 8-nucleate stages of the embryo sac. From both poles, one nucleus each moves towards the centre. At this 8-nucleate stage, cell walls are laid down leading to the organisation of the typical female gametophyte or embryo sac. Three nuclei at the micropylar end form the Egg Apparatus (one egg cell and two synergids) and three nuclei of the chalazal end form three antipodal cells. The remaining two nuclei called polar nuclei form part of a large binucleate cell, the central cell. Thus a typical female gametophyte or embryo sac of angiosperms is seven celled and eight nucleated structure.

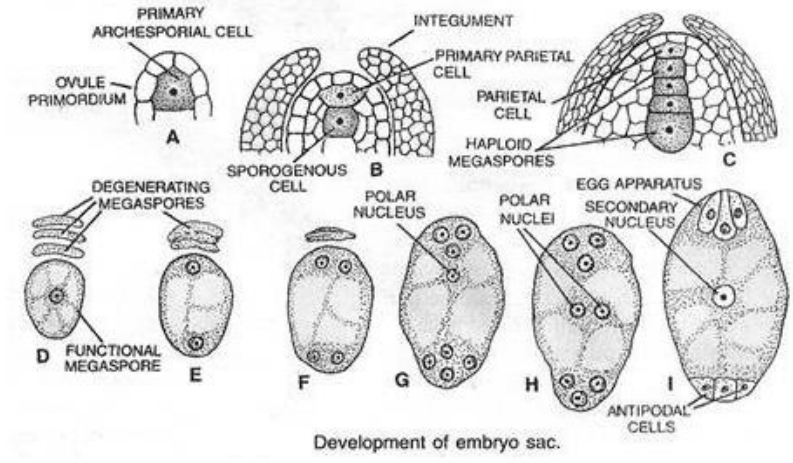


Fig. 16. Development of female gametophyte. Source: meritnation.com

POLLINATION: Transfer of pollen grains from a mature anther to the receptive stigma of a pistil is termed pollination. Pollination is mainly of two types based on the source of pollen. They are Self pollination and Cross pollination.

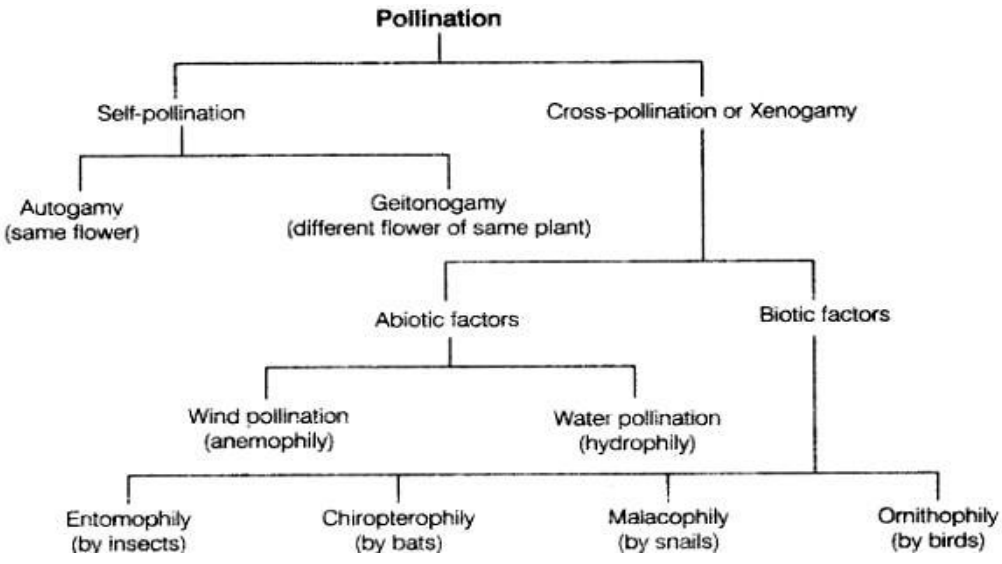


Fig.17. Pollination types. Source:cbse.net

Self pollination: Pollination that involves the transfer of pollen grains from anther to the stigma of same flower or different flower present on the same plant. Self pollination can be divided into two types:

i) **Autogamy**: Transfer of pollen grains from anther to the stigma of same flower. Autogamy requires synchrony in pollen release and stigma receptivity and also, the anthers and the stigma should lie close to each other so that self-pollination can occur. This type of pollination is seen in *Oryza sativa* (Rice), *Pisum sativum* (garden peas). Auto gamy is favoured by the following inbreeding devices

ii) **Geitonogamy**: Transfer of pollen grains from anther of one flower to the stigma of another flower present on the same plant. This is seen is *Lagenaria cicenaria* (Bottle gourd; **Kashir Aal**), *Cucurbita pepo* (Pumpkin; **Mashed Aal**), *Cucumis sativus* (Cucumber; **Laer**), *Salix sp* (willows; **Vir**).

Inbreeding devices or Contrivances to Self Pollination: Conditions that favour self pollination include:

a) **Cleistogamy**: Here the plant bears bisexual flowers that always remain closed thereby ensuring self pollination. *Viola odorata* (Banafsha; **Nunposh**), *Viola tricolor* (Pansy), *Oxalis sp* (Wood sorrel; **Chock-chin**) and *Commilina* are examples of cleistogamous flowers.

b) **Homogamy**: In this case the bisexual flowers open but the anther and stigma mature at the same time. Examples include *Helianthus annus* (Sunflower; **Guli aftab**) and *Mirabilis jalapa* (Marvel of Peru; 4' O clock plant).

c) **Bud Pollination**: Pollination occurs before the bisexual flowers open. This is seen in grass family including *Oryza sativa* (Rice) and *Triticum aestivum* (Wheat). *Pisum sativum* (pea plant) also shows bud pollination.

Cross pollination (Allogamy or Xenogamy): In this case, pollen grains are transferred from anther of a flower to the stigma of another flower of a different plant belonging to the same species.

Pollination types:
Autogamy: One Flower; One Plant
Geitonogamy: Two Flowers; One Plant
Xenogamy: Two Flowers; Two Plants

Agencies of pollination: The transfer of pollen grains from anther to stigma is carried out either by a non-living entity or a living organism. Thus plants use abiotic and biotic agents for achieving pollination. Abiotic agents include Wind and water while biotic agents are the animals.

Pollination through abiotic agents: This includes wind (Anemophily) and water (Hydrophily). Only 20% of total pollination takes place through abiotic agents, of which 98% is carried out by wind and only 2% by water.

i) **Anemophily**: Pollination that takes place by the agency of wind is called anemophily. This is the most common abiotic agency of pollination. It is found in *Plantago lanceolata* (Plantain; **Gulla**), *Zea mays* (maize), *Salix sp* (willows) and most conifers. The main features of such flowers include:

a) Flowers are inconspicuous and without colourful petals
b) They produce large quantities of light, non-sticky and sometimes winged pollen grains.

Fig. 18. Anemophilous flower (*Plantago sp*). flickr.com

c) Have exposed stamens and feathery stigma
d) Nectar and scent is absent.

ii) **Hydrophily**: When pollination is carried out by the agency of water, the phenomenon is called hydrophily. This is found in a few aquatic plants like *Valliseneria* (Tape grass), *Hydrilla* (Water thyme) and *Zostera* (Common eelgrass). Main features of hydrophilous flowers are:

a) Flowers are small and inconspicuous.
b) Perianth and other floral parts are unwettable.
c) Nectar and odour are absent.
d) Pollen grains are light and unwettable due to presence of mucliage.
e) Stigma is long, sticky but unwettable.

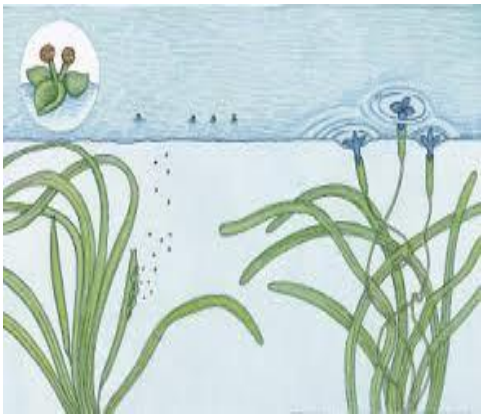


Fig. 19. Hydrophily in Vallisneria sp.. Source; semanticscholar

Pollination through biotic agents: Pollination through biotic agents accounts for almost 80% of total pollination. Animals that take part in pollination include insects, birds, bats and sometimes mammals.

i) **Entomophily**: Pollination that is carried out by insects is termed as entomophily. Bees (Honey bees, Bumble bees), beetles, butterflies, moths and even ants act as pollinators. Entomophily is seen in a large majority of plants including Magnolias, orchids, *Salvia sp*, *Ficus sp*, *Yucca sp*, *Rafflesia sp* (largest flower), *Amorphophallus sp* (Tallest flower), etc. Major features of entomophilous flowers include:

a) The flowers are large, brightly coloured and often scented. If small, they are aggregated.
b) Nectaries containing nutritious nectar are also a common feature of such flowers.
c) The pollen grains are thick-walled and sticky to enhance adhesion to insect bodies.
d) Blooming and maturation of corresponding insect pollinator usually coincide with each other.
e) Pollen grains are edible particularly in bee pollinated ones.

Fig. 20. Entomophily and Ornithophily Source: pacifichorticulture.org; coastalwildscape

ii) **Ornithophily**: When the agency of pollination is birds, the type of pollination is called ornithophily. Hummingbirds, sunbirds, honeyeaters and flowerpeckers are common bird pollinators. Bird pollinated species include *Callistemon sp* (Bottlebrush), *Campsis radicans* (trumpet vine), *Hibiscus rosa-sinensis* (China rose) and *Aquilegia sp* (Columbine). Flowers adapted to bird pollination have the following features:

a) Flowers are large, bright coloured and tubular (funnel-shaped).
b) Nectar is present in abundance.
c) They are odourless.
d) Remain open during the day (diurnal).

iii) **Chiropterophily**: Pollination through the agency of bats is called chiropterophily. Examples of chiropterophilous plants include *Kigelia sp* (Sausage tree) and *Adansonia sp* (Baobab tree). Adaptations found in bat pollinated flowers include:

a) They bloom during the night.
b) They are large, wide mouthed and pale coloured.
c) They emit a strong scent.
d) Floral parts are sometimes edible.

Outbreeding devices or Contrivances to cross pollination: Conditions that favour cross pollination include:

a) **Dichogamy**: When androecium and gynoecium of a flower mature at different times, it is called dichogamy. This is adaptation to avoid self pollination. In some species the androecium matures first (protandry) and the gynoecium matures only when the flower has shed all the pollen (*Gloriosa sp*., *Aquilegia sp*.). In others, (walnut, maize) the gynoecium matures before maturation of androecium (protogyny).
b) **Dioecism**: When male and female flowers are present of two separate plants (unisexual plants), the phenomenon is called dioecism. Dioecious plants are strictly cross pollinating varieties. Common examples are Willows, Poplars and Papaya (*Carica papaya)*.
c) **Herkogamy**: Flowers possess some mechanical barrier on their stigmatic surfaces that discourages self pollination. e.g. *Calotropis sp*.
d) **Self incompatibility**: In this case the pollen grains are unable to germinate on the receptive stigma of the same flower. There are definite genetic reasons for self incompatibility. e.g. *Brassica sp*.
e) **Male sterility**: Some varieties of plants produce non-functional pollen grains inside anthers. e.g. some varieties of maize.
f) **Heterostyly**: When the lengths of styles and stamens vary in two varieties of the same species, the phenomenon is called heterostyly. The difference in stylar length helps the plant to avoid self pollination. This is seen in *Primula* sp and *Linum* sp. (linseed; **Alish**).

Difference between Self pollination and Cross pollination.

Self Pollination	Cross Pollination
1. Transfer of pollen grains from anther to stigma of same flower or different flower but of the same plant.	1. Pollen grains are transferred from anther of one flower to stigma of another flower of a separate plant.
2. Anther and stigma of a flower mature at the same time (Homogamy).	2. Anther and stigma of a flower mature at different times (Dichogamy).
3. External agents are not required for pollination.	3. External agents are always required for pollination.
4. It is economical as there is no need for making nectar and huge amount of pollen.	4. Expensive process as large quantities of pollen and/or nectar is produced.
5. Self pollination results in formation of homozygous progenies (pure lines).	5. In cross pollination, heterozygous progenies (Hybrids) are produced.
6. Characters are preserved. Useful ones are conserved but useless ones are not eliminated.	6. Characters are not preserved. Useful characters can be lost but useless traits can be eliminated.
7. No new variations are introduced. Thus offspring are unable to adapt to changed environment.	7. Here variations are introduced which help plants to adapt to the changed environmental conditions.
8. T.his pollination can take place even when flowers are closed (Cleistogamy).	8. Flowers must open to ensure pollination (Chasmogamy).
9. No new varieties or species are formed by self pollination	9. New varieties and species are produced through cross pollination.

FERTILIZATION: Fusion of opposite gametes to form a diploid zygote is called fertilization. In angiosperms the male gamete is found inside pollen grain produced inside anther, while the female gamete (egg cell) is present inside the embryosac of ovule located in

the ovary of gynoecium. The male gamete after pollination is carried to the egg by pollen tube. This type of fertilization is known as **Siphonogamy**.

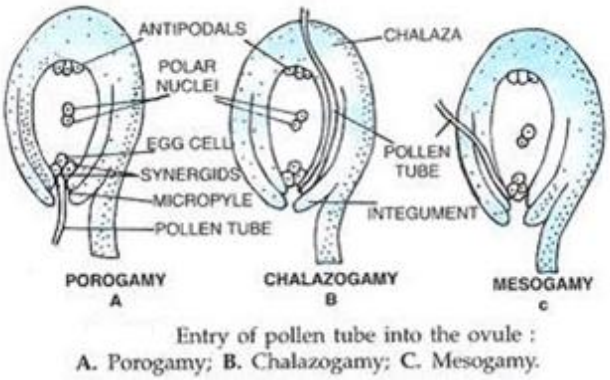
Steps to Fertilization: The various steps in fertilization include:

Pollen tube growth: To reach the ovule, the compatible pollen after falling on the stigma absorbs water and swell. The pollen grain germinates leading to the emergence of a pollen tube through one of the germ pores. The contents of the pollen grain move into the pollen tube. The vegetative cell materials along with its nucleus and the small undivided generative cell also move into the pollen tube. The generative cell later divides inside the pollen tube to form two small male gametes. The tube secretes exogenous enzymes and grows through the tissues of the stigma and style and reaches the ovary.

Entry of Pollen tube into the ovule: On reaching the ovary, the pollen tube enters the ovule by any one of the following methods:

a) **Porogamy**: When the pollen tube enters into an ovule through the micropyle, it is called porogamy. Porogamy is the most method of fertilization in angiosperms. E.g. *Lillium sp*. (Lily).

b) **Chalazogamy**: The entry of pollen tube into ovule through the chalazal region is called chalazogamy. This is found in *Juglans sp*. (walnut) and *Betula sp*. (Birches).



Entry of pollen tube into the ovule :
A. Porogamy; B. Chalazogamy; C. Mesogamy.

Fig. 21. Methods of pollen entry into ovule.
Source: yourarticlelibrary.com

c) **Mesogamy**: When the pollen tube enters an ovule laterally through the integuments at any place other than micropylar and chalazal region, it is known as mesogamy. It is the rarest mode of pollen entry. This is seen in *Cucurbita sp* (Pumpkin) and *Populus sp* (Poplar).

After entering into the ovule, the growth of pollen tube is directed by synergids as the later secrete some chemicals substances. The tip of the pollen tube enters into one of the synergids via the filiform apparatus. Inside the synergid, pollen tube enlarges and ruptures, releasing its contents that include two male gametes and vegetative nucleus inside the synergid. The tube

nucleus degenerates immediately after entering the synergid. The synergid ruptures and the two male gametes are set free.

DOUBLE FERTILIZATION: In angiosperms the fertilization process is a double event unlike other plant groups. Double fertilization was first reported by Russian embryologist Sergius Nawaschin (1898) in *Lillium martagon* (Lily). The same was later also independently discovered by Leon Guignard (1899). Among the two male gametes released by pollen tube, one fuses with the egg cell to form a diploid zygote. This process is called **Syngamy** or Generative fertilization. This diploid zygote is the first cell of embryo. The other male gamete fuses with the secondary nucleus to form a triploid Primary Endosperm Cell. Secondary nucleus is the nucleus formed by fusion of two polar nuclei prior to their fusion with male gamete. This process in which a cell with two nuclei fuses with a male gamete is called **triple fusion** or vegetative fertilization. This twin event of Syngamy and triple fusion is called Double fertilization. Thus the two gametes of the male gametophyte fuse with the two cells of the female gametophyte giving rise to zygote and primary endosperm cell simultaneously.

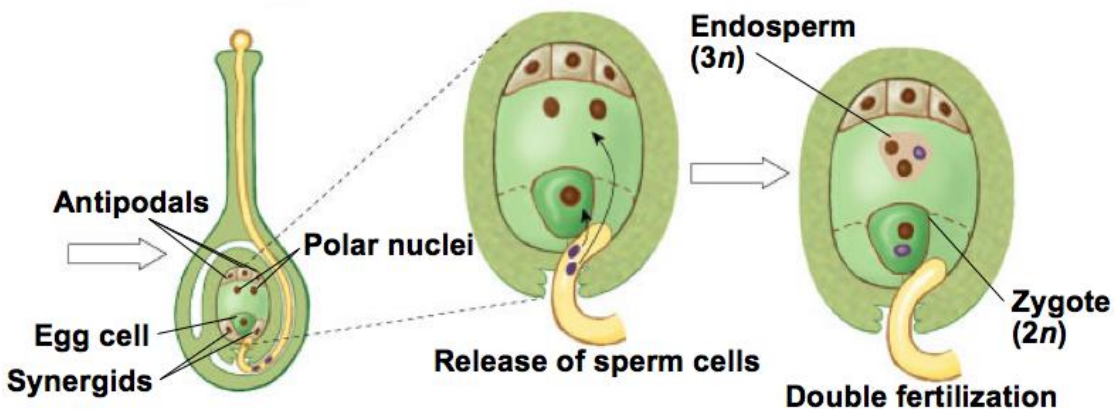


Fig. 22. Double fertilization. Source: toppr.com

Post fertilization Events: Soon after the event of double fertilization, a number of changes take place in the flower. The petals, stamens and style fall off. The main post fertilization events are marked by:

i) The triploid primary endosperm cell divides mitotically and develops into endosperm. Endosperm is the nutritive tissue that gives nourishment to growing embryo during its early stages of growth and development.

ii) The diploid zygote divides and develops into an embryo. Embryo is the miniature plant present in the seed.

iii) The ovule finally matures into seed.

iv) The ovary grows and develops into a fruit.

ENDOSPERM: Endosperm is the nutritive tissue formed from the triploid primary endosperm cell of embryosac within the ovule. It provides essential food material to embryo during growth and early stages of seedling formation. In angiosperms the endosperm is a triploid structure but in gymnosperms it is haploid tissue.

In many dicotyledons, the food reserve present in the endosperm is completely consumed for the development of the embryo and therefore endosperm is absent in the seed. Such seeds are called **non-endospermic** or **exalbuminous** seeds. The reserve food for future growth is kept in the massive cotyledons (Pea, Beans etc). However in cereals like Maize, wheat and rice, the endosperm is enlarged and retains the food material within the mature seeds. Such seeds are therefore called **endospermic** or **albuminous** seeds.

Types of endosperms: Based on the patterns of division and development, angisopermic endosperms can be divided into three types namely Nuclear, Cellular and Helobial endosperms.

i) **Nuclear Endosperm**: In this case, the triploid nucleus of the Primary endosperm cell undergoes repeated free nuclear divisions without the formation of cell walls. It results in the formation of a large number of free nuclei within the large central cell (Primary endosperm cell). A big central vacuole develops in the embryosac which pushes these nuclei towards the periphery. Thus all the nuclei lie in the peripheral cytoplasm between the cell wall and the vacuolar membrane. Later cell wall formation takes place by cleavage of peripheral cytoplasm in a centripetal manner. This results in conversion of whole endosperm into a cellular tissue. Nuclear endosperm is the most common type of endosperm and is seen in *Phoenix dactylifera* (Date palm), *Areca catechu* (Areca nut) and *Cocus nucifera*

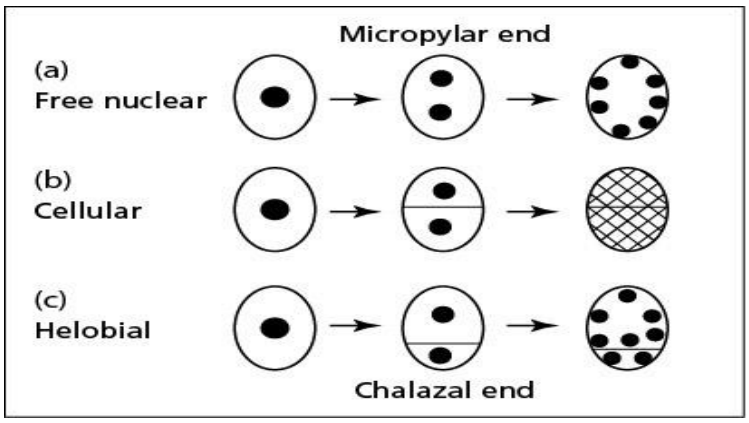


Fig. 23. Types of endosperms. Source: plantphys.net

(coconut). In coconut however the endosperm forms cellular tissue in the outer part but remains free nuclear in central part (liquid).

ii) **Cellular Endosperm**: Here the first nuclear division of primary endosperm cell is immediately followed by the formation of cell wall either longitudinally or transversely. The same process continues after every nuclear division thereby resulting in a mass of cells without any free nuclear stage. Such endosperms in which each nuclear division is followed by cellularization are called cellular endosperm. This is found in *Datura stramonium* (Jimsonweed; **Datur**), *Impatiens roylei* (Balsam) and *Petunia sp*.

iii) **Helobial endosperm**: In this case, the first nuclear division is immediately followed by cytokinesis resulting in the formation of two unequal cells. The small cell towards the chalazal end does not divide further and functions as haustorium. The nucleus of the larger micropylar cell divides repeatedly several times and results in the formation of numerous free nuclei within a single compartment. Later cellularization takes place centripetally to form a large helobial endosperm. This type of endosperm is seen in monocotyledons like *Asphodelus tenuifolius* (Onion weed).

EMBRYOGENY IN ANGIOSPERMS: Development of embryo from the zygote is called embryogeny.

Embryogeny in Dicotyledons: The best cited example of dicot embryogeny is of *Capsella bursa-pastoris* (Shepherd's purse; vern. **Kraalmund**). Here the development begins with the division of enlarged zygote into two unequal cells. The larger basal cell (on micropylar end of embryosac) is called Suspensor cell and the smaller terminal cell (towards chalazal end) is called Embryonal cell.

The basal suspensor cell now divides by a transverse division and the terminal embryonal cell divides by a longitudinal division resulting in the formation of an inverted T-shaped tetrad. Among the two cells of suspensor cell, the cell towards the micropyle enlarges to form a vesicular basal cell. The other cell divides by several transverse divisions to form a linear 7 – 8 celled Suspensor.

The two embryonal cells divide by longitudinal division that is perpendicular to its previous division to form a four celled Quadrant stage. These four cells further divide by transverse

division to form octant stage (two tiers of 4 cells). All the cells of octant stage divide by periclinal divisions to form eight outer protodermal cells and eight inner cells.

The 8- protodermal cells divide anticlinally while as the 8-inner cells divide in various planes. At this stage the embryo proper is almost spherical showing radial symmetry and is called globular Proembryo. Derivatives of protodermal cells form a single layered embryonic epidermis. From the derivatives of inner cells towards the distal end, two ridges arise and transform the globular proembryo into heart-shaped embryo. These ridges later form two cotyledons. Few cells at the base of the two cotyledons differentiate to form a dome shaped plumule.

The terminal cell of the suspensor that touches the proembryo towards the proximal end is called Hypophysis. The hypophysis divides both transversely and longitudinally and forms root apical meristem which consists of cortical initials of radicle, root protoderm and root cap.

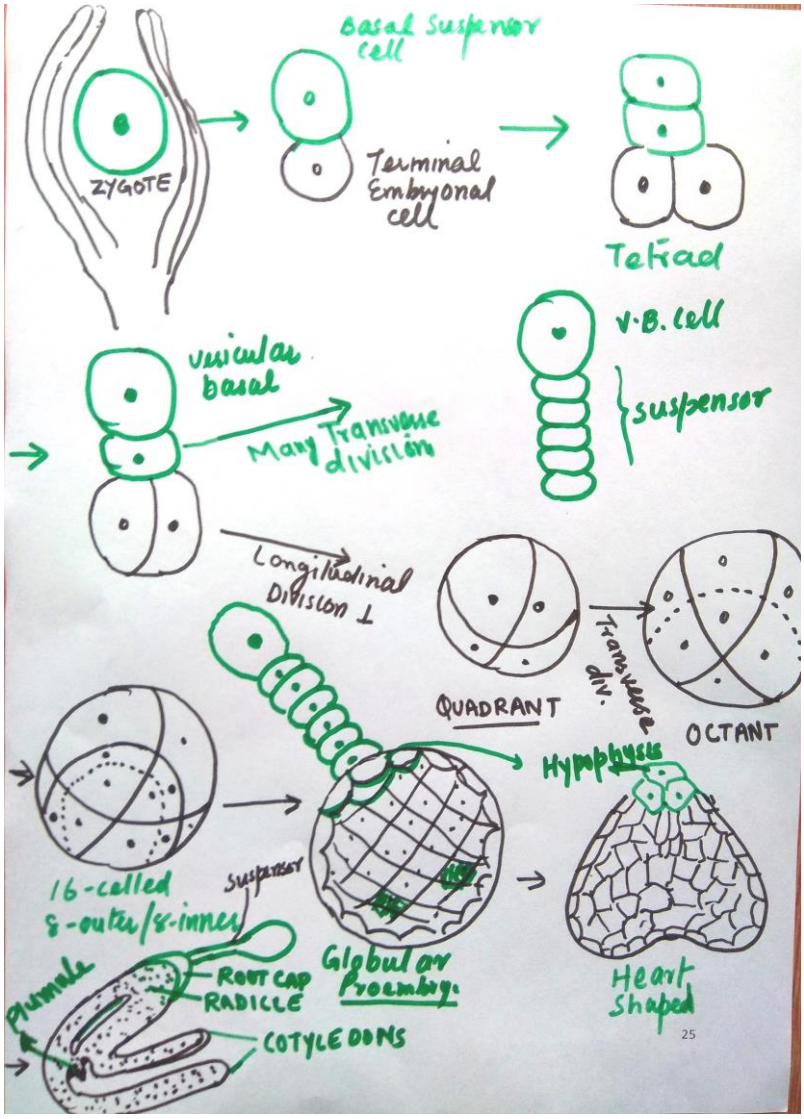


Fig. 24. Embryogeny in dicotyledons.

Thus a dicotyledonous embryo consists of an embryonal axis and two cotyledons. The portion of embryonal axis above the level of cotyledons is the epicotyl, which terminates with the plumule or stem tip. The cylindrical portion below the level of cotyledons is hypocotyl that terminates at its lower end in the radicle or root tip. The root tip is covered with a root cap.

APOMIXIS: The term was coined by Hans Winkler in 1908. Etymologically '*apo'* means without and '*mixis'* means mixing. According to Winkler (1908, 1934), the substitution for sexual reproduction of an asexual process, which does not involve any nuclear fusion is termed as apomixis. The term is in contrast to Amphimixis (reproduction involving fusion of opposite haploid gametes).

There are two main categories of apomixis - vegetative reproduction and Agamospermy. Vegetative reproduction has already been discussed in detail.

Agamospermy:

In this category of apomixis the plants have retained seed as the agent of propagation but the embryo is formed by some process in which normal meiosis and syngamy have been eliminated. This phenomenon is known as agamospermy.

There are three different types of agamospermy:

1. **Adventive embryony:** In this type of agamospermy, **embryos** arise directly from the diploid sporophytic cells of nucellus or integuments of an ovule. Thus embryos are formed but without meiosis nor syngamy. Adventives embryos are usually formed in addition to normal sexual embryos. Adventives embryony is common in *Citrus sp., Euphorbia dulcis, Capparis frondosa and Mangifera indica.*

2. Recurrent Apomixis: Formation of embryosac from diploid cell. it has the following types:

i) **Diplospory**: In this type of agamospermy, a diploid **embryosac** is formed from a megaspore mother cell, without a regular meiotic division. Thus all the seven cells of embryosac are diploid. It is divided into two types:

a) ***Diploid Parthenogenesis***: When the diploid egg cell of the unreduced embryosac forms an embryo, it is termed as Diploid parthenogenesis.

   Diploid Apogamy: The embryo develops from any diploid cell of unreduced embryosac except egg cell. Diploid embryo can develop from a diploid synergid or a diploid antipodal.

ii) **Apospory**: This phenomenon was reported in angiosperms for the first time by Rosenberg (1907). Here a somatic cell in the nucellus directly forms an unreduced **embryo sac**, and the diploid egg parthenogenetically develops into embryo. This is called ***Somatic apospory***. Somatic apospory has been reported in *Hieracium excelens, H. flagellare* and *H. aurantiacum*

Sometimes the archesporial cell in the nucellus directly forms a diploid embryosac without meiosis which has been termed as ***Generative apospory***. This has been found in *Eupatorium glandulosum* and *Parthenium argentatum*.

3.Non-recurrent apomixis: The embryosac is haploid and any haploid cell of this embryosac directly forms a haploid embryo. Embryosac is formed by usual meiotic divisions of the megaspore mother cell. When the embryo arises from the haploid egg cell of the embryosac, it is called ***haploid parthenogenesis.***

If any cell of the haploid embryosac other than the cell forms the embryo, it is called ***haploid apogamy***. Such haploid plants are generally sterile and do not reproduce sexually any more. Common examples of species that show non-recurent apomixis include *Solanum nigrum* (**Kambai**)*, Lilium, Bergenia* (**Zakhmi Hayat)***,** *and Nicotiana tabacum* (**Tamokh**).

EVALUATION

Question 1: Why is reproduction essential for organisms?

Answer: Reproduction ensures the continuance of various species on the Earth. In the absence of reproduction, the species will not be able to exist for a long time and may soon get extinct.

Question 2: Which is a better mode of reproduction sexual or asexual? Why?

Answer Sexual reproduction is a better mode of reproduction. It allows the formation of new variants by the combination of the genes from two different individuals of opposite sex. It involves the fusion of the male and the female gamete to produce variants, which are not identical to their parents and to themselves. This variation allows the individual to adapt to constantly changing and challenging environments. Also, it leads to the evolution of better suited organisms which ensures greater survival of a species. On the contrary, asexual reproduction allows very little or no variation at all. As a result, the individuals produced are exact copies of their parents and themselves.

Question 3: Why is the offspring formed by asexual reproduction referred to as clone?

Answer A clone is a group of morphologically and genetically identical individuals. In the process of asexual reproduction, only one parent is involved and there is no fusion of the male and the female gamete. As a result, the offspring so produced are morphologically and genetically similar to their parents and are thus, called clones.

Question 4: Offspring formed due to sexual reproduction have better chances of survival. Why? Is this statement always true?

Answer Sexual reproduction involves the fusion of the male and the female gamete. This fusion allows the formation of new variants by the combination of the DNA from two (usually) different members of the species. The variations allow the individuals to adapt under varied environmental conditions for better chances of survival. However, it is not always necessary that the offspring produced due to sexual reproduction has better chances of survival. Under some circumstances, asexual reproduction is more advantageous for certain organisms. For example, some individuals who do not move from one place to another and are well settled in their environment. Also, asexual reproduction is a fast and a quick mode of

reproduction which does not consume much time and energy as compared to sexual reproduction.

Question 5: How does the progeny formed from asexual reproduction differ from those formed by sexual reproduction?

Answer: Vegetative propagation is a process in which new plants are obtained without the production of seeds or spores. It involves the propagation of plants through certain vegetative parts such as the rhizome, sucker, tuber, bulb, etc. It does not involve the fusion of the male and the female gamete and requires only one parent.Hence, vegetative reproduction is considered as a type of asexual reproduction.

Question 6: What is vegetative propagation? Give two suitable examples.
Answer Vegetative propagation is a mode of asexual reproduction in which new plants are obtained from the vegetative parts of plants. It does not involve the production of seeds or spores for the propagation of new plants. Vegetative parts of plants such as runners, rhizomes, suckers, tubers, etc. can be used as propagules for raising new plants. Examples of vegetative reproduction are:

1.***Eyes of potato***: The surface of a potato has several buds called eyes. Each of these buds when buried in soil develops into a new plant, which is identical to the parent plant.

2. ***Leaf buds of Bryophyllum***: The leaves of Bryophyllum plants bear several adventitious buds on their margins. These leaf buds have the ability to grow and develop into tiny plants when the leaves get detached from the plant and come in contact with moist soil.

Question 7: Define (a) Juvenile phase (b) Reproductive phase and (c) Senescent phase.
Answer (a) Juvenile phase: It is the period of growth in an individual organism after its birth and before it reaches reproductive maturity.

(b) Reproductive phase: It is the period when an individual organism reproduces sexually.

(c) Senescent phase: It is the period when an organism grows old and loses the ability to reproduce.

Question 8: Higher organisms have resorted to sexual reproduction in spite of its complexity. Why?

Answer Although sexual reproduction involves more time and energy, higher organisms have resorted to sexual reproduction in spite of its complexity. This is because this mode of

reproduction helps introduce new variations in progenies through the combination of the DNA from two (usually) different individuals. These variations allow the individual to cope with various environmental conditions and thus, make the organism better suited for the environment. Variations also lead to the evolution of better organisms and therefore, provide better chances of survival. On the other hand, asexual reproduction does not provide genetic differences in the individuals produced.

Question 9: Explain why meiosis and gametogenesis are always interlinked?
Answer Meiosis is a process of reductional division in which the amount of genetic material is reduced. Gametogenesis is the process of the formation of gametes. Gametes produced by organisms are haploids (containing only one set of chromosomes), while the body of an organism is diploid. Therefore, for producing haploid gametes (gametogenesis), the germ cells of an organism undergo meiosis. During the process, the meiocytes of an organism undergo two successive nuclear and cell divisions with a single cycle of DNA replication to form the haploid gametes.

Question 10: Identify each part in a flowering plant and write whether it is haploid (n) or diploid
(2n).

(a)	Ovary	_____
(b)	Anther	_____
(c)	Egg	_____
(d)	Pollen	_____
(e)	Male-gamete	_____
(f)	Zygote	_____

Question 11: Define external fertilization. Mention its disadvantages.
Answer External fertilization is the process in which the fusion of the male and the female gamete takes place outside the female body in an external medium, generally water. Fish, frog, starfish are some organisms that exhibit external fertilization. Disadvantages of external fertilization: In external fertilization, eggs have less chances of fertilization. This can lead to the wastage of a large number of eggs produced during the process. Further, there is an absence of proper parental care to the offspring, which results in a low rate of survival in the progenies.

Question 12: Describe the post-fertilization changes in a flower.
Answer Fertilization is the process of the fusion of the male and the female gamete to form a diploid zygote. After fertilization, the zygote divides several times to form an embryo. The fertilized ovule forms a seed. The seed contains an embryo, enclosed in a protective covering, called the seed coat. As the seed grows further, other floral parts wither and fall off. This leads to the growth of the ovary, which enlarges and ripens to become a fruit with a thick wall called the pericarp.

Question 13: What is a bisexual flower? Collect five bisexual flowers from your neighborhood and with the help of your teacher find out their common and scientific names.
Question 14: Examine a few flowers of any cucurbit plant and try to identify the staminate and pistillate flowers. Do you know any other plant that bears unisexual flowers?
Answer: Cucurbit plant bears unisexual flowers as these flowers have either the stamen or the pistil. The staminate flowers bear bright, yellow coloured petals along with stamens that represent the male reproductive structure. On the other hand, the pistillate flowers bear only the pistil that represents the female reproductive structure. Other examples of plants that bear unisexual flowers are corn, papaya, and cucumber.

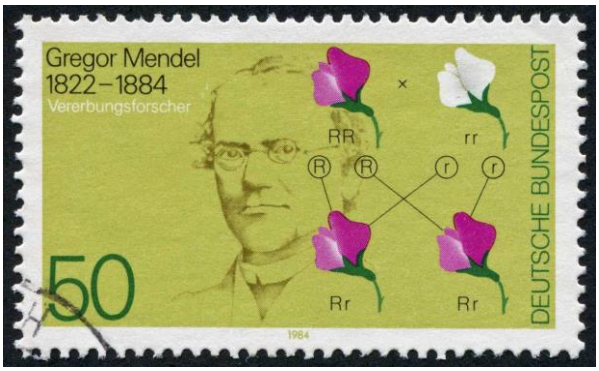
UNIT II - GENETICS

Common terms used in Genetics (Vocabulary of Genetics):

1. *Genetics*: The science of heredity and variation. The study of functions and behaviour of genes.
2. *Heredity*: The transmission of characters from parents to their offspring, generation after generation.
3. *Variation*: The degree with which the offspring differ from their parents.
4. *Gene*: A functional segment of DNA.
5. *Allele*: One of the alternative forms of the same gene responsible for determining contrasting characteristics. The alternative forms of a gene are alleles to each other. Alleles control a particular character and are located on the same locus of a homologous pair.
6. *Locus*: Position of an allele within a DNA molecule/ chromosome.
7. *Diploid*: A cell or an organism with two sets of chromosomes in each nucleus. 2n.
8. *Haploid*: Opposite of diploid. A cell or organism with only one set of chromosomes in the nucleus. 1n or n.
9. *Homozygous*: The diploid condition in which the alleles at a given locus are identical. Both the alleles are either dominant or recessive(XX, xx,TT, tt)
10. *Heterozygous*: the diploid condition in which the alleles at a given locus are different i.e. one dominant and the other recessive. (Xx, Tt)
11. *Phenotype*: The observable characteristics of an individual.
12. *Genotype*: The genetic constitution of an organism with respect to the alleles under consideration.
13. *Dominant*: The allele which influences the appearance of the phenotype even in the presence of an alternative allele. (T in Tt)
14. *Recessive*: The allele whose appearance remains hidden in the phenotype in the presence of an alternative allele. (t in Tt). The allele which influences the appearance of the phenotype only in the presence of another identical allele.

15. *F_1 generation* (First Filial generation): The generation produced by crossing homozygous parental stock.
16. *F_2 generation* (Second Filial Generation): The generation produced by crossing two F_1 organisms. In monoecious plants, it is also the generation obtained after self pollination of F_1 individuals.
17. *Emasculation*: Removal of stamens of a flower before the maturation of anthers.

Father of Genetics: Gregor Johann Mendel (1822 – 1884) is considered the father of genetics for his pioneering work in the field. He developed the principles of heredity by studying the heredity and variation patterns of seven pairs of characters in *Pisum sativum* (pea) plants.



Mendel was a monk at an Augustinian monastery at Brunn in Austria (presently Brno in Czech Republic). He later went to University of Vienna in 1951 where he studied natural history and mathematics for two years. At Vienna, Mendel had become interested in plant hybridization and in particular, the different forms in which hybrid offspring appear and the statistical relationships between them. Mendel began his scientific investigations on inheritance in Pea plants in 1856 at his monastery in Brunn.

Mendel's Experiments:

Gregor Johann Mendel, conducted hybridisation experiments on garden peas (*Pisum sativum*) for seven years (1856-1863) and proposed the laws of inheritance in living organisms. Mendel investigated seven characters in the garden pea plant viz. stem length, flower colour, flower position, seed shape, seed colour, pod shape and pod colour. These were manifested as two opposing or contrasting traits (tall or dwarf plants, yellow or green seeds etc). He then conducted artificial cross pollination experiments using several true-breeding pea lines. He was the first person to employ statistical analysis and mathematical logic to problems in biology. His experiments had a large sampling size, which gave greater credibility to the data that he collected. Also, the confirmation of his inferences from experiments on successive generations of his test plants proved that his results pointed to general rules of inheritance

rather than being unsubstantiated assumptions and ideas. Thus Mendel set up a basic framework of rules governing inheritance.

Seven pairs of contrasting characters selected by Mendel:

S.No	Character	Dominant	Recessive
1.	Length of stem (height)	Tall	Dwarf
2.	Flower colour	Voilet/Purple	White
3.	Flower position	Axial	Terminal
4.	Seed Shape	Round	Wrinkled
5.	Seed colour	Yellow	Green
6.	Pod shape	Inflated	Constricted
7.	Pod colour	Green	Yellow

INHERITANCE OF ONE GENE: MONOHYBRID CROSS

A hybridisation experiment that is aimed to study the inheritance pattern of a single pair of contrasting characters is called Monohybrid cross. In one such experiment, Mendel selected true breeding tall and dwarf pea plants. Emasculation was done in the flowers of one plant type (either tall or dwarf) and they acted as female. The other pea variety which served as pollen donor would act as male. Then he cross pollinated (cross hybridised) them by depositing the pollen from male flowers on the stigma of female flower (emasculated flowers). He collected the seeds and grew them as first hybrid generation. This he called *First Filial generation* or *F_1 Generation*. All the plants in the F_1 generation were tall resembling only one parental type. The absence of dwarf plants in F_1 puzzled Mendel. He then experimented with other traits but the result was same. Even the reciprocal crosses did not make any change in the F_1 phenotype. The F_1 generation always resembled only one of the parents.

The tall F_1 generation was allowed to self pollinate and the plants he generated from the seeds was termed as *second filial generation* or *F_2 generation*. In F_2 generation, Mendel was surprised to observe that *25* percent of the plants were dwarf, the character that was absent in F_1. The tall and dwarf traits were identical to their parental type and did not show any

blending. He obtained similar results with other traits i.e. only one of the parental traits was expressed in the F_1 generation while at the F_2 stage both the traits were expressed but in the ratio of **3:1**. The contrasting characters did not show any intermediate type at F_1 or F_2 level.

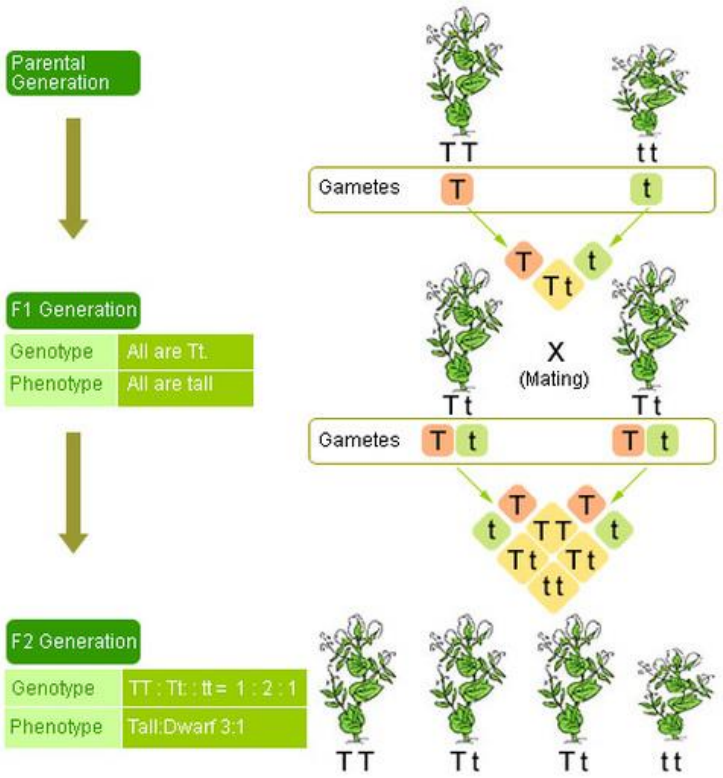


Fig 1. Monohybrid Cross. Source: http://kmbiology.weebly.com/

Mendel proposed that something was being stably passed down, unchanged, from parent to offspring through the gametes, over successive generations. He called these inheritance units as 'factors' (now termed as genes). They contain the information that is required to express a particular trait in an organism.

On the basis of his observations on monohybrid crosses Mendel proposed two general Principles or Laws of Inheritance: Law of Dominance and the Law of Segregation.

I. Law of dominance (Principle of dominance):

i) Genetic characters are controlled by discrete units called factors (genes). There is a pair of factors for each character (alleles).

ii) When two dissimilar pair of factors is present in a single individual (heterozygous), only one gets expressed while as the other remains hidden. The one that gets expressed is called dominant and the hidden one is known as recessive.

II. Law of Segregation (Principle of segregation):

This law states that the pair of factors for a trait separate or segregate from each other during gamete formation and each gamete receives only one factor for each character.

Since each gamete contains only one factor, they are always homozygous or pure. Hence this law is also called **law of purity of gametes.**

Reasons for Mendel's Success: Mendel was successful partly due to his careful choice of experimental plant, *Pisum sativum* (the garden pea). He established various advantages of garden pea over other species.

1. The plant was easily available.
2. Flowers were bisexual and self pollinating
3. Several varieties with quite distinct characteristics (contrasting characters) were available.
4. The plant could be raised, maintained and handled easily and conveniently.
5. Large number of seeds were produced in one generation so conclusions could be drawn more accurately.
6. Life cycle was short and many generations could be studied in a short time period.
7. Pure lines for all the characters were available because of closed flowers and self pollinating nature.
8. Artificial cross-breeding between varieties was possible and the hybrids produced were fertile.

Mendel's work remained hidden: Mendel presented his work before the Natural History Society at Brunn, Austria (Brno Czech Republic) in 1865 and was published in the Annual Proceedings of Natural History Society in 1866, however it remained hidden for about 35 years because of the following reasons:

1. Mendel's ideas were ahead of time.
2. He published his findings in a local journal which had limited circulation.
3. The statistical calculations were complicated.
4. Mendel was a monk and not a member of any University or scientific society. Therefore his reputation as a scientist was not recognised.
5. Mendel's work coincided with Darwin's evolutionary theory in which he proposed the evolution of man from apes. This had created hue and cry and no attention was given to Mendel's work.

Rediscovery of Mendel's work: In the year 1900, three scientists independently rediscovered the research work of Mendel. They were Hugo de Vries of Holland (Netherlands), Carl Correns of Germany and Eric Von Tschermak of Austria. These scientists came across Mendel's publication and realised the importance of his work. They are hence called Rediscoverers of Mendelism.

Incomplete Dominance: In Mendelian inheritance, the F_1 plants resembled only that parent whose factor for that character was dominant. The other remained completely masked. When similar experiments were repeated on other plants, it was found that sometimes the F_1 had a phenotype that did not resemble either of the two parents but was an intermediate phenotype. This phenomenon was termed as Incomplete dominance.

The inheritance of flower colour in the snapdragon (*Antirrhinum majus*) and Four O' Clock plant (*Mirabilis jalapa*) are good examples to understand incomplete dominance. In snapdragon, a cross between true-breeding red-flowered plant (RR) and true breeding white-flowered plants (rr), yielded pink flowered F_1 (Rr).When the F_1 was self-pollinated the F_2 resulted in the following ratio 1 (RR) Red: 2 (Rr) Pink: 1 (rr) White. Here the genotypic ratios were exactly as we would expect in any Mendelian monohybrid cross, but the phenotype ratios had changed from the 3:1 to 1:2:1. Here the factor R was not completely dominant over r and this made it possible to distinguish Rr as pink from RR (red) and rr (white).

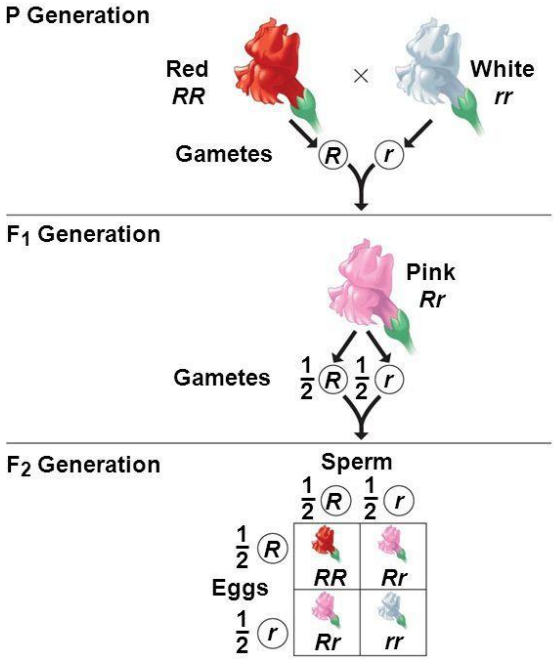


Fig. 2. Incomplete dominance. Source: slideplayer.com

INHERITANCE O F TWO GENES: DIHYBRID CROSS

In another set of experiments, Mendel cross hybridised pea plants that differed from each other by two traits. A cross in which two pairs of contrasting characters are taken into consideration simultaneously is called a Dihybrid cross. In his dihybrid cross experiment, Mendel crossed a pea plant grown from a seed yellow in colour and round shaped with the one from green seed with wrinkled shape. Mendel found that all the seeds resulting from the crossing of the parents were yellow in colour and round in shape. Thus F_1 generation again resembled only one parent. So, yellow colour was dominant over green and round shape

dominant over wrinkled. These results were identical to those that he got when he made separate monohybrid crosses between yellow and green seeded plants and between round and wrinkled seeded plants. When Mendel self hybridised the F_1 plants, he got four different phenotypes in the F_2 generation, two parental types (round yellow and wrinkled green) and two new ones (round green and wrinkled yellow). The ratio in which the four seed phenotypes separated was **9/16** Round Yellow; **3/16** Round green; **3/16** Wrinkled yellow; **1/16** wrinkled green.

Let us use the genotypic symbols **R** for round shaped seeds (dominant) and **r** for wrinkled seed shape (recessive), **Y** for dominant yellow seed colour (dominant) and **y** for recessive green seed colour (recessive). The genotype of the parents can then be written as **RRYY** and **rryy** (factors are paired in parents). The gametes produced by parent plants can be written as **RY** and **ry** because gametes contain only one factor for each character.

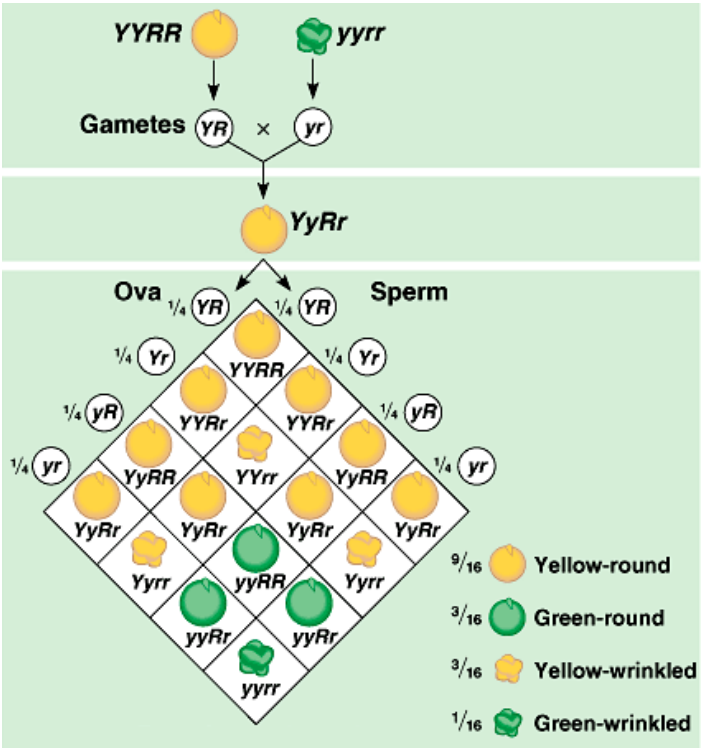


Fig.3. Dihybrid cross. Source: Hammiverse.com

These gametes unite on fertilisation to produce the *F1* hybrid **RrYy** (Round Yellow). When Mendel self hybridised the *F1* plants he got four different phenotypes in *F_2 generation*. These phenotypes *round yellow, wrinkled yellow, round green and wrinkled* green appeared in the ratio of **9:3:3:1**.

The ratio of **9:3:3:1** can be derived as a combination series of 3 yellow: 1 green, with 3 round: 1 wrinkled. This derivation can be written as follows:

(3 Round : 1 Wrinkled) (3 Yellow : 1 Green) = 9 Round, Yellow : 3 Wrinkled, Yellow: 3 Round, Green : 1 Wrinkled, Green

Law of Independent Assortment: It states that '*when two pairs of contrasting characters are combined in a hybrid, segregation of one pair of characters is independent of the other pair of characters*'.

The formation of four different types of phenotypes in F_2 generation was possible only due to the reorganisation of the factors during gamete formation. If the factor for yellow colour always transmitted with the factor for round seed shape or the factor for green colour always got carried with the factor for wrinkled seed shape, such phenotypic variation in F_2 were impossible. Law of independent assortment explains that each factor has an independent identity. Let us consider the gamete formation in the F_1 plant **RrYy**. Fifty per cent of the gametes will have the allele R and the other 50 per cent, r. Now besides each gamete having either R or r, it should also have the allele Y or y. The law of independent assortment says that the segregation of 50 per cent R and 50 per cent r is independent from the segregation of 50 per cent Y and 50 per cent y. Therefore, 50 per cent of the gametes having r allele has Y and the other 50 per cent has y. Similarly, 50 per cent of the gametes bearing R allele has Y and the other 50 per cent has y. So we get four combinations of gametes, RY, Ry, rY and ry resulting in four different phenotypes in F_2. If the assortment of different factors was dependent on each other, the allele R would always transmit with allele Y and allele r with allele y, thereby forming only two types of gametes (RY and ry). This would result in only two phenotypes round yellow 75% and wrinkled green 25%. Thus the dihybrid phenotypic ratio would have been 3:1 but Mendel got 9:3:3:1 which is possible only when the alleles assort independent of each other during gamete formation.

Codominance: When both the alleles of a gene express in a F_1 heterozygote, the phenomenon is called codominance. In contrast to incomplete dominance (where the F_1 individuals are different from both the parents), the F_1 generation in codominance show resemblance with both the parents. These codominant alleles do not show Mendelian dominance-recessive behaviour and are able to express themselves independently when present together in a hybrid. The phenotype of the heterozygous individual is different from both the homozygous parents. Codominance should not be confused with incomplete dominance. In incomplete dominance the effect of one allele on the phenotype is more pronounced compared to other. But codominant alleles are equally potent in expressing themselves in the phenotype. Both the codominant alleles are represented by a single letter in upper case but with different superscripts. e.g., $\mathbf{I^A}$, $\mathbf{I^B}$ for codominant alleles of blood groups in humans. Sometimes two different letters, both in capital are used to symbolise the two

codominant alleles. e.g., R and W for codominant alleles for red and white colour respectively.

A common example of codominant alleles is the alleles for blood groups in human beings. ABO blood groups are controlled by the gene **I**. The plasma membrane of the red blood cells has sugar polymers that protrude from its surface and the kind of sugar is controlled by the gene.

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 the sugar while allele *i* does not produce any sugar. Because humans are diploid organisms, each person possesses any two alleles (I^AI^A, I^Ai, I^AI^B, I^BI^B, I^Bi *or* ***ii***). I^A and I^B are completely dominant over ***i***, in other words when I^A and ***i*** are present only I^A expresses and when I^B and *i* are present I^B expresses. But when I^A and I^B are present together they both express their own types of sugars. Thus I^A and I^B are co-dominant alleles as both express in the phenotype. Hence red blood cells have both A and B types of sugars.

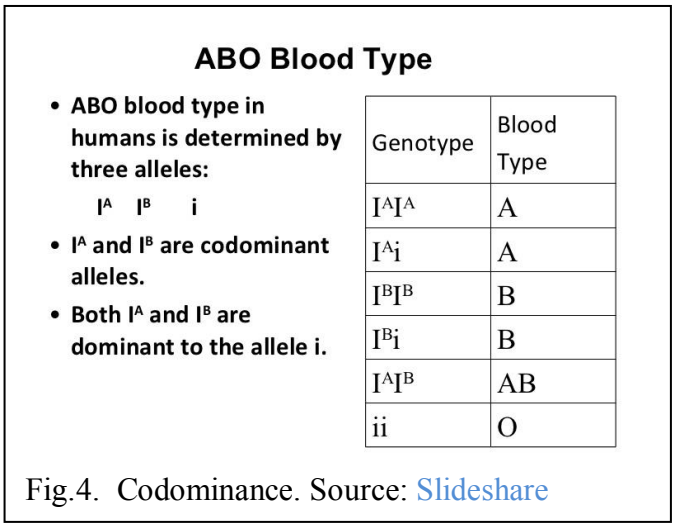


Fig.4. Codominance. Source: Slideshare

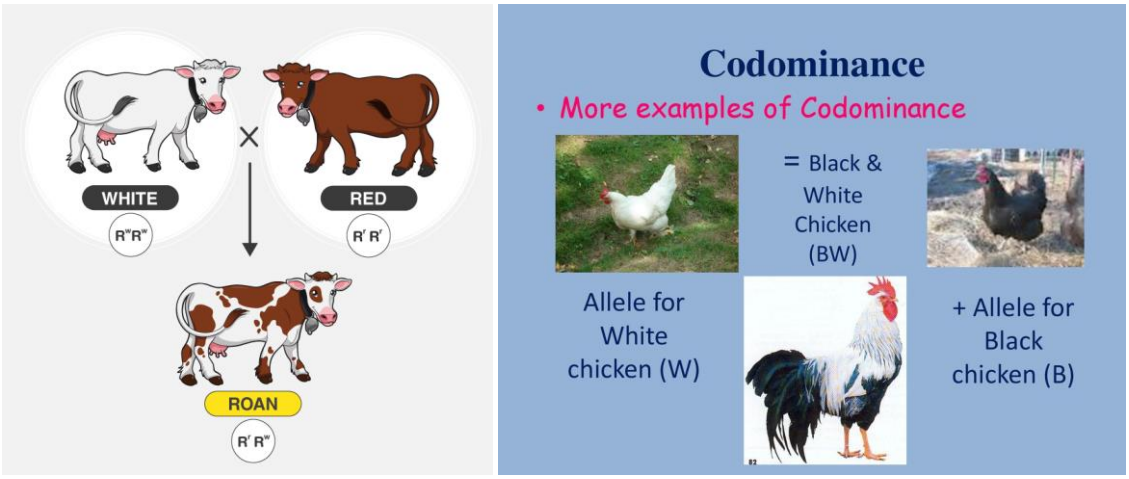


Fig.5. Examples of Codominance. Source: byjus.com; slideplayer.com

MULTIPLE ALLELISM: The presence of more than two alleles of a gene within a population is called multiple allelism and such alleles are called multiple alleles. Despite the presence of many alleles for a gene within a population, a diploid organism can possess only two alleles. Multiple alleles arise as a result of repeated mutation of the same gene in different directions. The main features of multiple allelism are:

i) There are more than two alleles of a gene in a population. *Drosophila* possesses 15 alleles for eye colour.
ii) Multiple alleles occupy the same gene locus of homologous chromosomes.
iii) A diploid individual possesses only two alleles and each gamete carries only one allele.
iv) Multiple alleles express different alternatives of the same character.
v) Different alleles of multiple allele series show codominance, dominance-recessive behaviour or an incomplete dominance amongst themselves.

The most common example of multiple allelism is Human blood groups which are determined by three alleles. Humans have four blood group phenotypes – A, B, AB and O. These blood groups are determined by two antigens (A and B) present on the surfaces of red blood corpuscles. The antigens occur on sugar rich head regions of glycophorin. Persons with blood group A have antigen A, group B have antigen B, AB persons have both A and B antigens while as blood group O people have no antigen on their erythrocytes. These antigens are determined by three immunogen alleles – I^A, I^B and i. Allele I^A forms antigen A, I^B forms antigen B and i (I^0) forms no antigen at all.

S.No.	Genotype	Antigen (sugar)	Blood Group
1.	I^AI^A	A	A
2.	I^Ai	A	A
3.	I^BI^B	B	B
4.	I^Bi	B	B
5.	I^AI^B	A & B	AB
6.	Ii	None	O

TEST CROSS: To determine the genotype of an organism for a trait, a test cross is an important tool. A test cross is a cross in which an organism (test organism) showing dominant phenotype for a trait is crossed with its recessive parent. The progenies of such a cross can easily be analysed to predict the genotype of the test organism.

e.g. A plant with dominant phenotype for height can be having TT genotype or Tt genotype. To determine whether the tall plant is homozygous or heterozygous, let us cross it with recessive parent i.e. dwarf phenotype (tt). If the ratio for this cross is 1:1, the plant is heterozygous tall and if all the plants after crossing resulted in same phenotype, the plant is homozygous for tallness.

TT x tt gives 100% Tall phenotype. But Tt x tt gives 50% tall and 50%dwarf.

TT **Tt**	**Tt** **tt**
(T) **(t)**	**(T), (t)** **(t)**
Tt (tall 100%)	**Tt** (tall 50%) **tt** (dwarf 50%)

Tt and tt 50% tall and 50% dwarf in the ratio 1:1. Monohybrid test cross ratio for a heterozygous trait is always 1:1.

Dihybrid test cross: A cross of an unknown individual having dominant phenotype for two characters is crossed with its recessive parent. The unknown individual with Round Seeds and Yellow colour can have two genotypes, RRYY or RrYy. An individual RRYY when crossed with rryy yields all Round and Yellow seeds. But if the genotype is RrYy, the cross with rryy will yield four different phenotypes in the ratio of 1:1:1:1

	RrYy Round yellow	**rryy** wrinkled green
Gametes	**RY, Ry, rY, ry** (four types of gametes)	**ry** (one type of gamete)

Progeny	**RrYy**	**Rryy**	**rrYy**	**Rryy**
	Round yellow 25%	Round green 25%	Wrinkled yellow 25%	Wrinkled green 25%

Importance of test cross: It is used to determine whether an individual is homozygous or heterozygous for a trait or a pair of traits. Heterozygous monohybrid test cross ratio is 1:1 and dihybrid heterozygous test cross ratio is 1:1:1:1.

Back Cross: A cross in which an individual with unknown genotype is crossed with either of its two parents (dominant or recessive) is called a back cross. Back crossing with its dominant parent is used to make a desired stock homozygous. Test cross is actually a type of back cross. Every test cross is a back cross.

Reciprocal cross: A cross in which the sex of the parent plants is changed. Suppose in one cross pollen is taken from parent with dominant phenotype (male) and deposited on the stigma of plant recessive for that trait (female). Now if we want to conduct a reciprocal cross, we have to take pollen from plant with recessive phenotype and deposit it on stigma of plant with dominant trait.

Reciprocal cross is used to determine the effect of cytoplasmic genes (genes present in chloroplast and mitochondria) on the phenotype of organisms. Since the male gamete is devoid of cytoplasmic organelles in plants only female gamete contributes to cytoplasmic inheritance.

PLEIOTROPY: It is the phenomenon in which a single gene simultaneously controls the phenotypic expression of two or more characters. Such genes having multiple phenotypic effects are called pleiotropic genes. Normally one gene controls the effect of a single character but pleiotropic genes influence many traits at the same time. Pleiotropy is due to the effect of a gene on two or more inter-related metabolic pathways. Usually the effect of pleiotropic genes is more prominent on one trait called Major effect, and less evident on the others called secondary effect.

Examples of pleiotropy:

i) In garden pea, the same gene controls flower colour and also seed coat colour and red spots in the leaf axils.
ii) In drosophila, the same gene which controls white eye colour also leads to depigmentation in other parts of the body.
iii) In Sickle cell anaemia of human beings, the same gene alters the type of haemoglobin as well as the shape of RBCs from normal to sickle like.

iv) The gene that causes Phenylketonuria (an inborn autosomal recessive metabolic disorder) also exhibits pleiotropic effect. In PKU the homozygous recessive individual lacks the enzyme phenylalanine hydroxylase needed to change aminoacid phenylalanine to tyrosine in liver. It results in phenylalaninemia which is characterised by accumulation and excretion of phenylalanine and related compounds. Lack of the enzyme is due to abnormal autosomal recessive gene on chromosome 12. The pleiotropic effects of this gene include hyperalaninemia, impaired brain development, mental retardation, defects in speech and walking, decreased hair pigmentation. Heterozygous individuals are normal but act as carriers. One in about 18000 births among white Europeans has PKU.

Chromosome Theory of Inheritance:

Walter Sutton found a close parallelism between the transmission of genes (Mendelian factors) and the chromosomal behaviour during sexual reproduction. This includes:

i) The chromosomes occur in homologous pairs in diploid cells. Similarly each gene also exists as allele pairs.
ii) Chromosomes segregate at the time of gamete formation such that only one of each pair is transmitted to a gamete. Genes also segregate during gamete formation and only one of each pair is transmitted to the gamete.
iii) During gamete formation, homologous chromosomes of one pair segregate from each other independently to that of other pairs. A pair of genes (alleles) also segregates independently of other allele pairs.
iv) Chromosomes again pair during fertilization and similarly genes again pair at the time of fertilization.

These similarities between the behaviour of chromosomes and genes led to the postulation of chromosome theory of inheritance by Walter Sutton and Theodore Boveri independently in 1902. This was later confirmed by T.H. Morgan. This theory states that:

The Mendelian factors or genes are located on chromosomes and it is the chromosomes that segregate and assort independently during meiosis and recombine at the time of fertilization.

Explanation: The chromosome theory of inheritance can be understood with the help of a dihybrid cross between a pea plant homozygous for Yellow seed colour and round shape and a plant homozygous for green seed colour and wrinkled shape. The figure shows the

segregation of alleles of the gene for seed shape and seed colour during gamete formation for F_1 generation. It shows that there can be two possible ways in which the two replicated homologous chromosomes segregate in the metaphase of Meiosis-I. This leads to the independent assortment of the chromosomes and the genes located in them in four possible ways. This produces four types of gametes each with half the number of chromosomes and genes. Of these four types, two are parental and two are recombinants i.e. with new combinations of chromosomes and genes. All the four types of gametes have equal proportion of chromosomes and genes. They have equal proportion i.e. 25% each. These gametes by random fusion give rise to F_2 generation of four types of pea plants in the ratio 9:3:3:1.

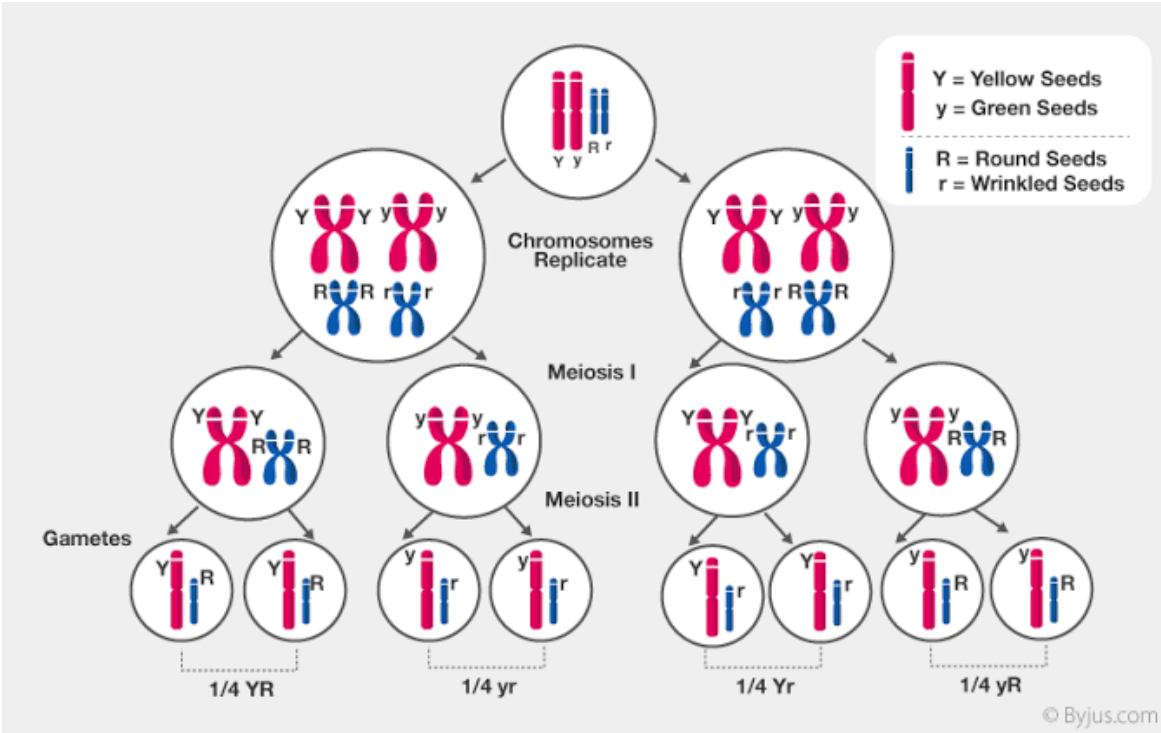


Fig. 6. Chromosome Theory of Inheritance. Source: byjus.com

Polygenic inheritance: When a single phenotypic trait is governed by more than one pair of genes, it is called a polygenic trait and the inheritance pattern of such genes is called polygenic inheritance. Such a group of genes are called **polygenes**, polygenic set or polygenic system. The various genes of a polygenic set may be located on separate chromosomes and at different loci. Each gene of a polygenic system contributes a small degree to the overall phenotype. Since polygenic traits are quantitative, presence of more

dominant genes makes the phenotype more prominent and when the recessive genes are more, the phenotype is less conspicuous. The dominant genes add up their effect

Quantitative inheritance: the inheritance of quantitative traits also called polygenic traits is known as quantitative or multifactor or polygenic inheritance. In this kind of inheritance, F_1 individuals are very similar to one another and are usually intermediate between the two parents. A cross between two F_1 individuals yields a widely variable F_2 generation having a few individuals like one grandparent, a few like the other grandparent, and the rest ranging between the two. A good example of quantitative inheritance is seen in the human skin colour.

Human skin colour is controlled by at least three of genes viz. A, B and C. located on different chromosomes and at different loci. They are inherited independently. The genes for dark colour A, B and C are incompletely dominant and the darkness of skin is proportionate to the sum of the dominant genes present. A pure Negro has six dominant genes (AA BB CC) for skin colour and a pure white person (very light skinned) has six recessive genes (aa bb cc).

A cross between a pure Negro and a completely white produces F_1 hybrid offspring with skin colour intermediate between the two parents. It is called mullato and has the genotype Aa Bb Cc. This F_1 hybrid makes eight kinds of gametes in each sex, thus forming 64 possible combinations in F_2 generation having seven (7) phenotypes and several genotypes. The skin colour of F_2 offspring varies according to the number of dominant genes they inherit for pigmentation. The skin colour ranges from Black in 1/64, very dark brown 6/64, dark brown 15/64, mulatto 20/64, light brown 15/64, very light brown, 6/64 and pure white 1/64.

Qualitative traits vs Quantitative traits: Polygenic traits are usually quantitative unlike Mendelian traits which are qualitative. In quantitative traits, the degree of phenotypic expression depends on the cumulative number of dominant genes (alleles). However in qualitative traits, a single dominant allele is enough to show full phenotype and any additional allele does not increase the expression. There are only two phenotypes in qualitative traits but in quantitative traits numerous phenotypes exist depending on the number of dominant alleles.

Parents **AABB CC** **aabb cc**
Negro **White**

Gametes **ABC** **X** **abc**

F_1 gen. **Aa Bb Cc**

Mulatto

Aa Bb Cc **AaBbCc**
♀ ♂

Gametes ♀ ♂	**ABC** ●●●	**ABc** ●●○	**AbC** ●○●	**Abc** ●○○	**aBC** ○●●	**aBc** ○●○	**abC** ○○●	**abc** ○○○
ABC ●●●	●●●	●●●	●●●	●●●	●●●	●●●	●●●	●●●
	●●●	●●○	●●○	●○○	●●○	●○○	●○○	○○○
ABc ●●○	●●●	●●●	●●●	●●●	●●●	●●●	●●●	●●○
	●●○	●○○	●○○	○○○	●○○	○○○	○○○	○○○
AbC ●○●	●●●	●●●	●●●	●●●	●●●	●●●	●●●	●●○
	●●○	●○○	●○○	○○○	●○○	○○○	○○○	○○○
Abc ●○○	●●●	●●●	●●●	●●○	●●●	●●○	●●○	●○○
	●○○	○○○	○○○	○○○	○○○	○○○	○○○	○○○
aBC ○●●	●●●	●●●	●●●	●●●	●●●	●●●	●●●	●●○
	●●○	●○○	●○○	○○○	●○○	○○○	○○○	○○○
aBc ○●○	●●●	●●●	●●●	●●○	●●●	●●○	●●○	●○○
	●○○	○○○	○○○	○○○	○○○	○○○	○○○	○○○
abC ○○●	●●●	●●●	●●●	●●○	●●●	●●○	●●○	●○○
	●○○	○○○	○○○	○○○	○○○	○○○	○○○	○○○
Abc ○○○	●●●	●●●	●●●	●●○	●●●	●●○	●●○	○○○
	○○○	○○○	○○○	○○○	○○○	○○○	○○○	○○○

● represents a dominant gene and ○ represents recessive gene.

Negro: 01; Very dark brown: 06; Dark brown: 15; Mulatto: 20; Light brown: 15; Very light brown: 06; white: 01

Phenotypic ratio: 1:6:15:20:15:6:1

MOLECULAR BIOLOGY

Chromosomes:

Chromosomes were discovered by Hofmiester as nuclear filaments in *Tradescantia sp.* in 1848. These nuclear filaments were later termed as chromosomes by Waldeyer. Chromosome literally means '*coloured body*' as they readily take biological stains. Chromosomes are the carriers of genetic information among living organisms. Different groups of organisms have different chromosomes:

Prokaryotic chromosomes (*Bacterial chromosome*): Prokaryotes like bacteria have a single circular chromosome. In *Escherchia coli* the chromosome consists of a circular DNA molecule 1100 microns in length. This bacterium has 3000 to 4000 genes on this single chromosome. Prokaryotic chromosomes are simple both genetically and biochemically. They carry only one set of genes (monoploid). The genetic information is stored in a single molecule of nucleic acid (DNA). A prokaryotic chromosome is in direct contact with the cytoplasm as there is no membrane enveloping it and is without any nucleolus. Even the proteins that form essential part of eukaryotic chromosomes, are absent in bacterial chromosomes. They are as such referred sometimes as prochromosomes. A Prokaryotic chromosome is also called a nucleoid.

Replication of DNA in prokaryotes is normally continuous throughout the cell cycle. The replicated chromosomes are distributed between the daughter cells by cell membrane because the mitotic spindle is not formed in prokaryotes. Having a single chromosome, the bacteria genome is limited and the amount of DNA content is very less and therefore codes for fewer proteins compared to eukaryotes.

Plasmids: Many bacteria may also contain extra chromosomal circular DNA molecules capable of autonomous replication. They are called Plasmids or F-factors. The plasmids are not essential for bacterial survival but they carry genes for sexuality and antibiotic resistance.

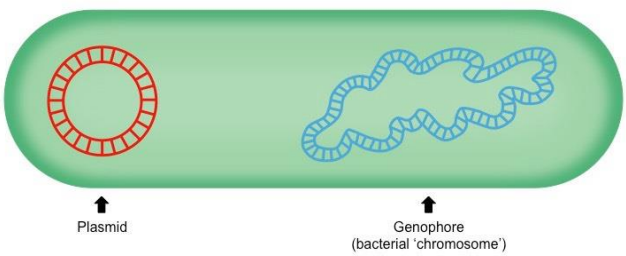


Fig. 7. Diagram of Bacterial cell with chromosome and plasmid. Source: Bioninja.com.au

Viral chromosome: A viral chromosome consists of a single molecule of nucleic acid. The nucleic acid may be DNA or RNA and may be linear of circular. The viruses having DNA or RNA as genetic material are respectively called DNA viruses and RNA viruses. The viruses infecting plant cells usually have their chromosomes formed of linear RNA molecules. However animal and bacterial viruses have RNA or DNA molecules of linear or circular form. In Tobacco Mosaic Virus (TMV), the chromosome consists of a single stranded linear RNA molecule coiled into a regular spiral. Retroviruses have double stranded RNA as their chromosomes. In bacteriophages (a type of viruses that infect bacteria), the chromosome is composed of double stranded DNA molecules. SARS-Cov-2 (novel coronavirus), a member of corona group of viruses is also an RNA virus. These RNA viruses use their RNA to direct the synthesis of DNA by the process of Reverse Transcription inside host cells. This DNA then transcribes RNA for making new virus particles. Such RNA viruses are called Retroviruses. Novel Coronavirus and HIV are examples of retroviruses.

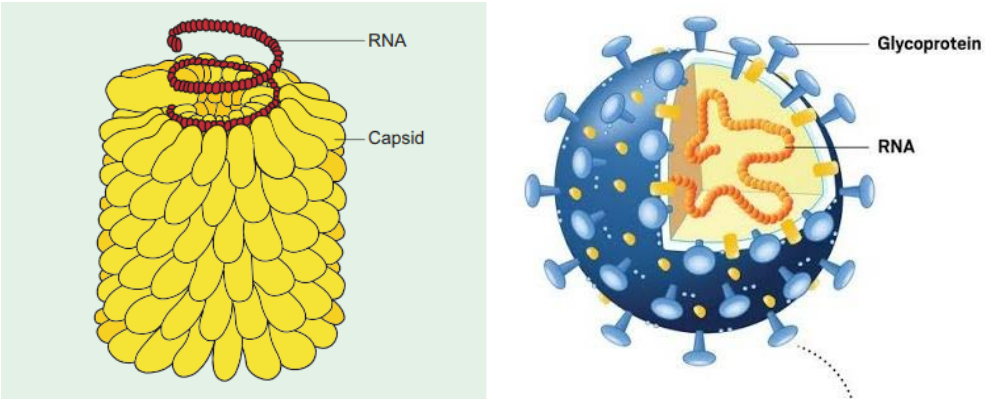


Fig. 8. Tobacco mosaic virus. Source: brainkart.com; Corona virus. Source: cen.acs.org

Eukaryotic Chromosome: The genetic information in the nucleus of eukaryotic cell is found on chromosomes. Each chromosome is a thread like structure containing DNA, basic proteins (histones) and acidic proteins. The composition of chromosome is 40 % DNA, 50% histones, 8.5% acidic proteins and 1.5% RNA.

The eukaryotic chromosomes are usually many and occur in the nucleus of the cell. Some extra-nuclear chromosomes are sometimes present in cell organelles like mitochondria and plastids. However mitochondrial and plastid DNA is usually circular and resembles bacterial chromosomes.

Number: The number of chromosomes varies in different species; however chromosome number is constant for a species. The number varies from 2 to few hundred but is usually

between 8 – 50. In human beings the number of chromosomes is 23 pairs of which 22 are autosomes and the remaining one pair is called sex chromosomes. Adders tongue fern, *Ophioglossum* possesses 1260 chromosomes within a cell.

Shape: The shape of chromosome changes from phase to phase. In the interphase stage the chromosome occurs in the form of thin coiled, elastic thread like structures. In metaphase and anaphase the chromosomes become thick and rod-like and may be straight or curved, J-shaped or V-shaped.

Size: The size of the chromosome varies from species to species. The length of the chromosome ranges from 0.1 to 33 microns. The diameter of eukaryotic chromosomes may vary from 0.2 to 2 microns. Human chromosome is 6 micron in length. The largest chromosomes are polytene chromosomes (2000µ long) and lampbrush chromosome (5900 µ).

Types of chromosomes: Depending upon the position of centromere, chromosomes are of following types:

a) **Telocentric chromosome**: When the centromere of a metaphase chromosome is terminal i.e. on the telomere, the chromosome is called telocentric chromosome.
b) **Acrocentric chromsosome**: When the centromere is located near the telomere, but just below it, the chromosome is acrocentric.
c) **Sub-metacentric chromosome (heterobranchial)**: The chromosome is somewhere between the centre and the tip of the chromosome. Such chromosomes appear L-shaped or J-shaped during anaphase of cell cycle.
d) **Metacentric chromosome (Isobranchial)**: When the centromere is located at the centre of the chromosome, it is called metacentric chromosome. Such chromosomes appear V-shaped during anaphase.
e) **Diacentric**: A chromosome having two centromeres is called a diacentric chromosome.
f) **Acentric chromosome**: When the centromere is absent in the chromosome, it is called an acentric chromosome.

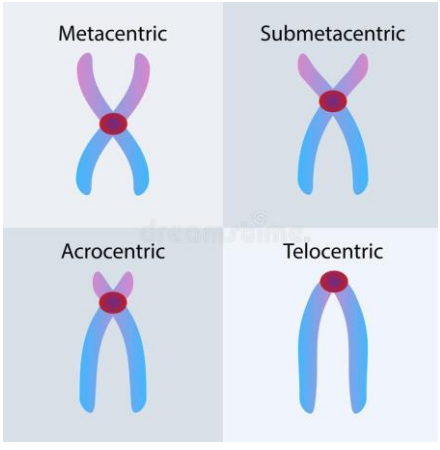


Fig. 9. Types of Chromosomes. Source: dreamstime.com

Structure of a Chromosome:

The structure of chromosomes can be best visualised during metaphase stage of mitosis. At metaphase stage, the chromosomes show the following structures:

a) **Chromatids**: Each metaphase chromosome appears longitudinally split into two halves. These identical split linear structures that are joined at the centromere are called chromatids.

b) **Chromosomal arms**: The centromere may also divide the chromosome transversally into two equal or unequal parts. These are called arms.

c) **Chromonema** (chromonemata): In less compact chromosomes under optical microscope, a coiled filament is seen in the chromosome called chromonema. The chromonema may contain 2, 4 or many fibres according to species type. The chromonema may contain thick and thin regions.

d) **Centromere and Primary constriction**: It is the constricted non-stained portion of the chromosome. Centromere is the specific part of chromosome where spindle fibres get attached during metaphase. Position of centromere is constant for a particular chromosome.

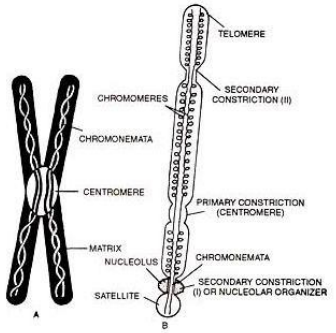


Fig. 10. Structure of Chromosome.
Source: biologydiscussions.com

e) **Secondary constriction**: Additional constriction present outside the region of centromere is called secondary constriction. It is located at the point where the nucleolus is attached with the chromosome and is therefore also called Nucleolar organiser region.

f) **Satelite**: Sometimes extra chromosomal parts are present at the terminal portions of chromosomes beyond the secondary constriction region. These are called satellites. Satellites are elongated, round shaped or variable in size. Those chromosomes which possess satellite DNA are called SAT-chromosomes.

g) **Telomere**: The terminal ends or tips of the chromosome are called telomeres. They differ in structure and composition from the rest of the chromosome. It is unique in that it prevents the ends of chromosomes from sticking to each other.

Genetic Material: The substance which stores inheritable biological information in coded form, transfers it to the next generation and causes its expression in the offspring is called genetic material. The main ***properties of genetic material*** are:

i). Genetic material should be present in every cell in the same amount.

ii). It should be structurally and chemically stable.

iii) Genetic material should be able to replicate (duplicate) and be inherited by the progeny faithfully.

iv). It should be susceptible to an occasional change by way of mutations and such changes should be inherited.

v). It should be able to carry all the coded information for controlling biological functions of the cells.

Search for Genetic Material: In 1902, **Sutton** and **Boveri** proposed the chromosomal theory in which they asserted that genetic information is passed between the generations by chromosomes. But it took many years to clarify which component of the chromosome acts as the genetic material. Both protein and nucleic acids have diversity huge enough to act as the genetic material.

Griffith's Transformation Experiment: Frederick Griffith, a British bacteriolgist in 1928 conducted some experiments on *Diplococcus pneumoniae* (Pneumococcus), a bacterium that causes pneumonia in mammals. There are two strains of this bacterium. One is a smooth, virulent strain (S-type) which is protected by a polysaccharide capsule and causes pneumonia. The other is rough, non-virulent strain (R-type) without a capsule and is harmless.

In one of his experiments, Griffith injected live S-type bacteria in mice. The mice suffered from pneumonia and died. In another, he used R-type bacteria, the mice didn't suffer from pneumonia therefore survived. In the next step, he killed Smooth (S-type) bacteria by heating them at high temperature and injected these dead S-type bacteria in mice. The animals survived. In his final experiment, Griffith injected a mixture of heat killed S-type and live R-type bacteria and the mice died. Although neither

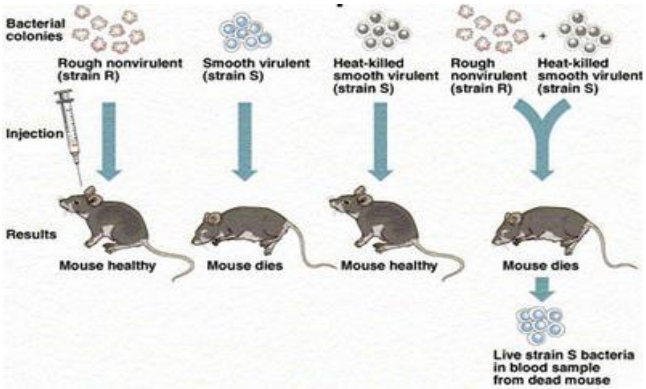


Fig. 11. Transformation experiment. Source: digitalcommons.rockefeller.edu

dead S-type nor live R-type was harmful to mice individually, but the mixture caused pneumonia in mice that lead to their death. When autopsies were conducted, live S-type bacteria were found inside the dead mice. Griffith concluded that some genetic material from dead S-type bacteria entered the living R-type bacteria and transformed it into smooth bacteria.

Griffith concluded that the R strain bacteria had somehow been transformed by the heat-killed S strain bacteria. Some 'transforming principle', transferred from the heat-killed S strain, had enabled the R strain to synthesise a smooth polysaccharide coat and become virulent. This must be due to the transfer of the genetic material. However, the biochemical nature of genetic material was not defined from his experiments.

Oswald Avery, Colin MacLeod and Maclyn McCarty (1944) purified biochemicals (proteins, DNA, RNA, etc.) from the heat-killed S cells to see which ones could transform live R cells into S cells. They discovered that DNA alone from S-type bacteria caused R-type bacteria to get transformed into S-type. Protein-digesting enzymes (proteases) and RNA-digesting enzymes (RNases) did not affect transformation, so the transforming substance must not be a protein or RNA. When DNA of S-type was digested with DNAase enzyme, transformation ceased, suggesting that the DNA caused the transformation. Thus they concluded that DNA is the hereditary material.

Bacteriophage experiment: Alfred **Hershey** and Martha **Chase** (1952) conducted experiments with bacteriophages to confirm whether protein or DNA is the genetic material. Bacteriophages are viruses that infect and parasitize bacteria. They are made of only two components, DNA and protein. The bacteriophage attaches to the bacteria and sends its genetic material inside the bacterial cell. The bacterial cell treats the viral genetic material as if it was its own and subsequently facilitates the production of more virus particles inside it. Hershey and Chase used T_2 phages which use *Escherichia coli* as their host. They received **Nobel Prize in Physiology and Medicine** in 1969.

In their experiment, Hershey and Chase grew some phages on a medium containing radioactive phosphorus (^{32}P) and some others on medium that contained radioactive sulphur (^{35}S) for many generations. Viruses grown in the presence of ^{32}P contained radioactive DNA but not radioactive protein because only DNA contains phosphorus and proteins don't. Similarly, viruses grown on ^{35}S contained radioactive protein but not radioactive DNA because Sulphur is a component of proteins and not DNA.

Radioactive phages were allowed to attach to *E. coli* bacteria. Then, as the infection proceeded, the bacteria along with phage particles were agitated in a blender. The virus particles were separated from the bacteria by spinning them in a centrifuge. Bacteria which were infected with viruses that had radioactive DNA were radioactive, indicating that DNA was the material that passed from the virus to the bacteria. Bacteria that were infected with viruses that had radioactive proteins were not radioactive. This indicates that proteins did not enter the bacteria from the viruses. Only the DNA entered inside *E. coli* and DNA is therefore the genetic material that is passed from virus to bacteria.

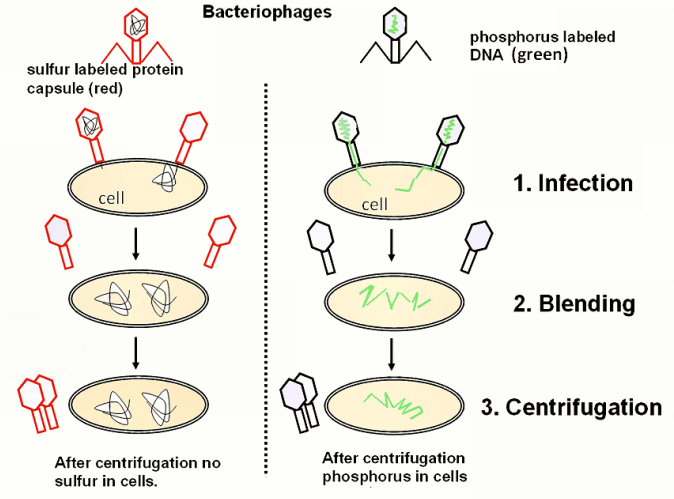


Fig. 12. Bacteriophage experiment. Source: Wikipedia.com

STRUCTURE OF NUCLEIC ACIDS:

Nucleic acids are chains of nucleotides that store genetic information in biological systems. They are located inside the nucleus and have acidic nature because of the presence of phosphate group (Phosphoric acid). The two major nucleic acids are Deoxyribonucleic acid and Ribonucleic acid.

Deoxyribonucleic acid: DNA was discovered by Friedrich Meischer in 1860 and he named it as '*Nuclein'* because of its presence inside nucleus. Richard Altmann (1889) used the term Nucleic acids.

DNA is a long polymer of deoxyribonucleotides. Nucleotides are the basic units of nucleic acids and each nucleotide is made up of a pentose sugar, a nitrogen base and a phosphate group. The length of DNA is usually defined as number of nucleotides (or a pair of nucleotide referred to as base pairs) present in it. Nucleotide count is also the characteristic of an organism. In the virus $\phi\times174$ there are 5386 nucleotides, Bacteriophage lambda has 48502 base pairs (bp), *Escherichia coli* has 4.6×10^6 bp, and haploid content of human DNA is 3.3×10^9 bp.

A Deoxyribonucleotide has three components viz a deoxyribose sugar, a nitrogenous base, and a phosphate group. Though the sugar and phosphate groups have no variants, the

nitrogenous bases are of four types – Adenine, Guanine, Cytosine and Thymine. Adenine and Guanine are called purines while cytosine and thymine are known as pyrimidines. A nitrogenous base is linked to the pentose sugar through an **N-glycosidic linkage** to form a nucleoside (DNA nucleosides include deoxyadenosine, deoxyguanosine, deoxycytidine and deoxythymidine). The phosphate group is linked to 5' -OH of a nucleoside through a linkage called **phosphoester bond**. Two nucleotides are linked through **3'-5' phosphodiester** bond to form a dinucleotide. More nucleotides can be joined in such a manner to form a polynucleotide chain. A polymer thus formed has at one end a free phosphate moiety at 5' -end of pentose sugar, which is referred to as 5′-end (5 prime end) of polynucleotide chain. Similarly, at the other end of the polymer the sugar has a free 3′ -OH group which is referred to as 3'-end of the polynucleotide chain. The backbone in a polynucleotide chain is formed by sugars and phosphates. The nitrogenous bases linked to sugar moiety project from the backbone.

How RNA is different from DNA: Apart from being single stranded, the RNA differs from DNA in the type of pentose sugar and a nitrogen base. In RNA, every pentose sugar has an additional –OH group present at 2' -position. Thus RNA has ribose sugar instead of deoxyribose sugar and also uracil as nitrogenous base in place of thymine. Thus the nucleosides of RNA include adenosine, guanosine, cytidine and uridine. However the phosphate group is same in both DNA and RNA.

DNA DOUBLE HELIX:

DNA was discovered in 1860 but it took around a century to understand its architecture. In 1953 James Watson and Francis Crick, based on the X-ray diffraction data produced by Maurice Wilkins and Rosalind Franklin, proposed a very famous Double Helix model for the structure of DNA. Base pairing concept is considered the hallmark of double helix model of DNA. The idea was base pairing was taken from the observations of Erwin Chargaff that for a double stranded DNA, the Adenine – thymine ratio and guanine – cytosine ratio are constant. The base pairing confers a very unique property to the polynucleotide chains. They are complementary to each other, and therefore if the sequence of bases in one strand is known then the sequence in other strand can be predicted. Also, if each strand from a parent

DNA acts as a template for synthesis of a new strand, the two daughter double stranded DNA molecules thus, produced would be identical to the parental DNA molecule.

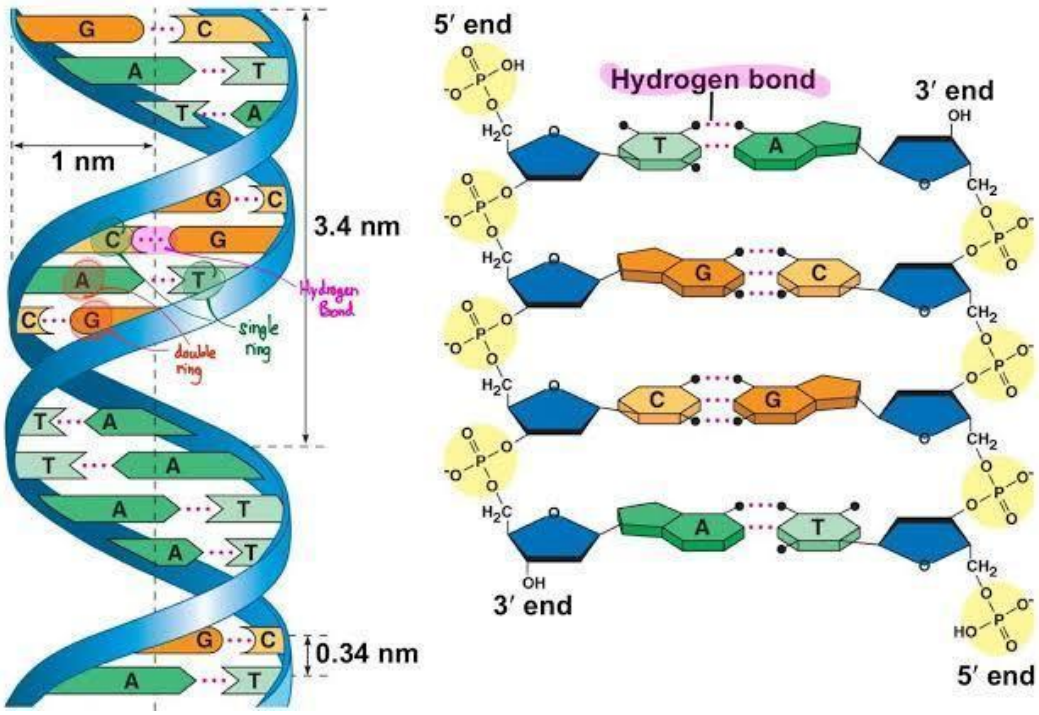


Fig. 13.Structure of DNA double helix. Source: Pinterest

The Original two page research publication by Watson and Crick in which they proposed Double Helix Model for DNA structure.

No. 4356 April 25, 1953 NATURE 737

equipment, and to Dr. G. E. R. Deacon and the captain and officers of R.R.S. *Discovery II* for their part in making the observations.

[1] Young, F. B., Gerrard, H., and Jevons, W., *Phil. Mag.*, **40**, 149 (1920).
[2] Longuet-Higgins, M. S., *Mon. Not. Roy. Astro. Soc., Geophys. Supp.*, **5**, 285 (1949).
[3] Von Arx, W. S., Woods Hole Papers in Phys. Oceanog. Meteor., **11** (3) (1950).
[4] Ekman, V. W., *Arkiv. Mat. Astron. Fysik. (Stockholm)*, **2** (11) (1905).

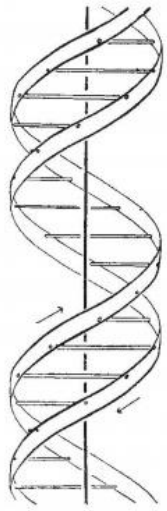
MOLECULAR STRUCTURE OF NUCLEIC ACIDS

A Structure for Deoxyribose Nucleic Acid

WE wish to suggest a structure for the salt of deoxyribose nucleic acid (D.N.A.). This structure has novel features which are of considerable biological interest.

A structure for nucleic acid has already been proposed by Pauling and Corey[1]. They kindly made their manuscript available to us in advance of publication. Their model consists of three inter-twined chains, with the phosphates near the fibre axis, and the bases on the outside. In our opinion, this structure is unsatisfactory for two reasons: (1) We believe that the material which gives the X-ray diagrams is the salt, not the free acid. Without the acidic hydrogen atoms it is not clear what forces would hold the structure together, especially as the negatively charged phosphates near the axis will repel each other. (2) Some of the van der Waals distances appear to be too small.

Another three-chain structure has also been suggested by Fraser (in the press). In his model the phosphates are on the outside and the bases on the inside, linked together by hydrogen bonds. This structure as described is rather ill-defined, and for this reason we shall not comment on it.



This figure is purely diagrammatic. The two ribbons symbolize the two phosphate—sugar chains, and the horizontal rods the pairs of bases holding the chains together. The vertical line marks the fibre axis

We wish to put forward a radically different structure for the salt of deoxyribose nucleic acid. This structure has two helical chains each coiled round the same axis (see diagram). We have made the usual chemical assumptions, namely, that each chain consists of phosphate di-ester groups joining β-D-deoxy-ribofuranose residues with 3′,5′ linkages. The two chains (but not their bases) are related by a dyad perpendicular to the fibre axis. Both chains follow right-handed helices, but owing to the dyad the sequences of the atoms in the two chains run in opposite directions. Each chain loosely resembles Furberg's[2] model No. 1; that is, the bases are on the inside of the helix and the phosphates on the outside. The configuration of the sugar and the atoms near it is close to Furberg's 'standard configuration', the sugar being roughly perpendicular to the attached base. There is a residue on each chain every 3·4 A. in the *z*-direction. We have assumed an angle of 36° between adjacent residues in the same chain, so that the structure repeats after 10 residues on each chain, that is, after 34 A. The distance of a phosphorus atom from the fibre axis is 10 A. As the phosphates are on the outside, cations have easy access to them.

The structure is an open one, and its water content is rather high. At lower water contents we would expect the bases to tilt so that the structure could become more compact.

The novel feature of the structure is the manner in which the two chains are held together by the purine and pyrimidine bases. The planes of the bases are perpendicular to the fibre axis. They are joined together in pairs, a single base from one chain being hydrogen-bonded to a single base from the other chain, so that the two lie side by side with identical *z*-co-ordinates. One of the pair must be a purine and the other a pyrimidine for bonding to occur. The hydrogen bonds are made as follows: purine position 1 to pyrimidine position 1; purine position 6 to pyrimidine position 6.

If it is assumed that the bases only occur in the structure in the most plausible tautomeric forms (that is, with the keto rather than the enol configurations) it is found that only specific pairs of bases can bond together. These pairs are: adenine (purine) with thymine (pyrimidine), and guanine (purine) with cytosine (pyrimidine).

In other words, if an adenine forms one member of a pair, on either chain, then on these assumptions the other member must be thymine; similarly for guanine and cytosine. The sequence of bases on a single chain does not appear to be restricted in any way. However, if only specific pairs of bases can be formed, it follows that if the sequence of bases on one chain is given, then the sequence on the other chain is automatically determined.

It has been found experimentally[3,4] that the ratio of the amounts of adenine to thymine, and the ratio of guanine to cytosine, are always very close to unity for deoxyribose nucleic acid.

It is probably impossible to build this structure with a ribose sugar in place of the deoxyribose, as the extra oxygen atom would make too close a van der Waals contact.

The previously published X-ray data[5,6] on deoxy-ribose nucleic acid are insufficient for a rigorous test of our structure. So far as we can tell, it is roughly compatible with the experimental data, but it must be regarded as unproved until it has been checked against more exact results. Some of these are given in the following communications. We were not aware of the details of the results presented there when we devised our structure, which rests mainly though not entirely on published experimental data and stereo-chemical arguments.

It has not escaped our notice that the specific pairing we have postulated immediately suggests a possible copying mechanism for the genetic material.

Full details of the structure, including the conditions assumed in building it, together with a set of co-ordinates for the atoms, will be published elsewhere.

We are much indebted to Dr. Jerry Donohue for constant advice and criticism, especially on inter-atomic distances. We have also been stimulated by a knowledge of the general nature of the unpublished experimental results and ideas of Dr. M. H. F. Wilkins, Dr. R. E. Franklin and their co-workers at

King's College, London. One of us (J. D. W.) has been aided by a fellowship from the National Foundation for Infantile Paralysis.

J. D. Watson
F. H. C. Crick

Medical Research Council Unit for the Study of the Molecular Structure of Biological Systems,
Cavendish Laboratory, Cambridge.
April 2.

[1] Pauling, L., and Corey, R. B., *Nature*, **171**, 346 (1953); *Proc. U.S. Nat. Acad. Sci.*, **39**, 84 (1953).
[2] Furberg, S., *Acta Chem. Scand.*, **6**, 634 (1952).
[3] Chargaff, E., for references see Zamenhof, S., Brawerman, G., and Chargaff, E., *Biochim. et Biophys. Acta*, **9**, 402 (1952).
[4] Wyatt, G. R., *J. Gen. Physiol.*, **36**, 201 (1952).
[5] Astbury, W. T., Symp. Soc. Exp. Biol. 1, Nucleic Acid, 66 (Camb. Univ. Press, 1947).
[6] Wilkins, M. H. F., and Randall, J. T., *Biochim. et Biophys. Acta*, **10**, 192 (1953).

Molecular Structure of Deoxypentose Nucleic Acids

While the biological properties of deoxypentose nucleic acid suggest a molecular structure containing great complexity, X-ray diffraction studies described here (cf. Astbury[1]) show the basic molecular configuration has great simplicity. The purpose of this communication is to describe, in a preliminary way, some of the experimental evidence for the polynucleotide chain configuration being helical, and existing in this form when in the natural state. A fuller account of the work will be published shortly.

The structure of deoxypentose nucleic acid is the same in all species (although the nitrogen base ratios alter considerably) in nucleoprotein, extracted or in cells, and in purified nucleate. The same linear group of polynucleotide chains may pack together parallel in different ways to give crystalline[1-3], semi-crystalline or paracrystalline material. In all cases the X-ray diffraction photograph consists of two regions, one determined largely by the regular spacing of nucleotides along the chain, and the other by the longer spacings of the chain configuration. The sequence of different nitrogen bases along the chain is not made visible.

Oriented paracrystalline deoxypentose nucleic acid ('structure *B*' in the following communication by Franklin and Gosling) gives a fibre diagram as shown in Fig. 1 (cf. ref. 4). Astbury suggested that the strong 3·4-A. reflexion corresponded to the inter-nucleotide repeat along the fibre axis. The ~ 34 A. layer lines, however, are not due to a repeat of a polynucleotide composition, but to the chain configuration repeat, which causes strong diffraction as the nucleotide chains have higher density than the interstitial water. The absence of reflexions on or near the meridian immediately suggests a helical structure with axis parallel to fibre length.

Diffraction by Helices

It may be shown[5] (also Stokes, unpublished) that the intensity distribution in the diffraction pattern of a series of points equally spaced along a helix is given by the squares of Bessel functions. A uniform continuous helix gives a series of layer lines of spacing corresponding to the helix pitch, the intensity distribution along the nth layer line being proportional to the square of J_n, the nth order Bessel function. A straight line may be drawn approximately through

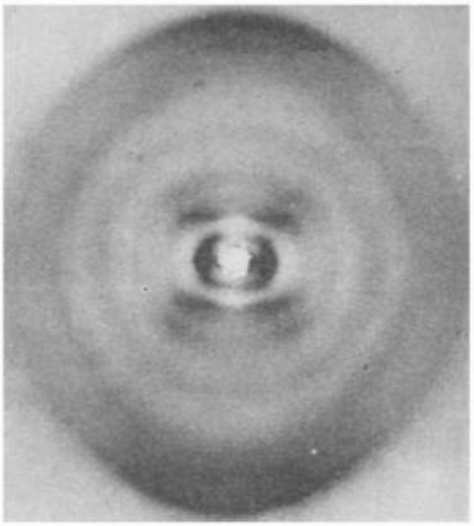


Fig. 1. Fibre diagram of deoxypentose nucleic acid from *B. coli*. Fibre axis vertical

the innermost maxima of each Bessel function and the origin. The angle this line makes with the equator is roughly equal to the angle between an element of the helix and the helix axis. If a unit repeats n times along the helix there will be a meridional reflexion (J_0^2) on the nth layer line. The helical configuration produces side-bands on this fundamental frequency, the effect[5] being to reproduce the intensity distribution about the origin around the new origin, on the nth layer line, corresponding to *C* in Fig. 2.

We will now briefly analyse in physical terms some of the effects of the shape and size of the repeat unit or nucleotide on the diffraction pattern. First, if the nucleotide consists of a unit having circular symmetry about an axis parallel to the helix axis, the whole diffraction pattern is modified by the form factor of the nucleotide. Second, if the nucleotide consists of a series of points on a radius at right-angles to the helix axis, the phases of radiation scattered by the helices of different diameter passing through each point are the same. Summation of the corresponding Bessel functions gives reinforcement for the inner-

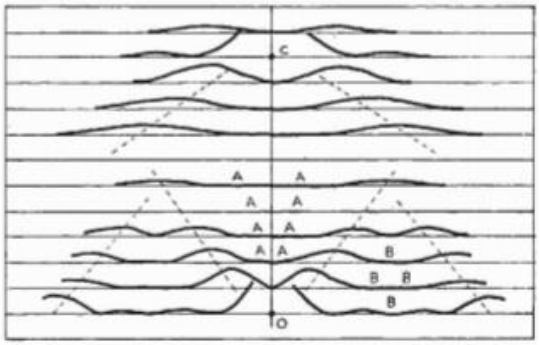


Fig. 2. Diffraction pattern of system of helices corresponding to structure of deoxypentose nucleic acid. The squares of Bessel functions are plotted about 0 on the equator and on the first, second, third and fifth layer lines for half of the nucleotide mass at 20 A. diameter and remainder distributed along a radius, the mass at a given radius being proportional to the radius. About *C* on the tenth layer line similar functions are plotted for an outer diameter of 12 A.

The main features of the DNA Double-helix include:

i) DNA is made of two polynucleotide chains, where the backbone is constituted by sugar-phosphate, and the bases project inside. Sugar and phosphate are joined together by **Phosphodiester bond** while as sugars and nitrogenous bases are held together by **glycosidic bonds**.

ii) The two chains have **anti-parallel** polarity. It means, if one chain has the polarity **5'→3'**, the other has **3'→5'**.

iii) The nitrogenous bases in two strands are paired through **hydrogen bond**s (H-bonds) forming base pairs (bp). Adenine forms two hydrogen bonds with Thymine from opposite strand and vice-versa. Similarly, Guanine is bonded with Cytosine with three H-bonds. As a result, always a purine comes opposite to a pyrimidine. This generates approximately uniform distance between the two strands of the helix.

iv) The two chains are coiled in a right-handed fashion. The pitch of the helix is 3.4 nm ($34A^0$) and there are roughly 10 bp in each turn. Consequently, each nucleotide pair (base pair) is approximately equal to 0.34 nm ($3.4A^0$). The diameter of the helix is $20A^0$

v) The plane of one base pair stacks over the other in double helix. This, in addition to H-bonds, confers stability of the helical structure.

DNA Packaging: The DNA is not present inside the cell nuclei as extended fibres but as highly condensed form. If the distance between two consecutive base pairs is 0.34nm (i.e. 0.34×10^{-9}m), the length of DNA double helix in a typical mammalian cell can be calculated simply by multiplying the total number of base pairs with distance between two consecutive base pairs. In a typical mammalian cell, there are $6.6x10^9$ base pairs. Therefore $(6.6 \times10^9) \times (0.34\times10^{-9})$ comes out to be approximately 2.2 metres. This length is far greater than the dimension of a typical cell nucleus (approx. 0.000001m or 10^{-6} m). The paradox is how these 2.2 meters of DNA can be accommodated within a small space, 1 micron (1µ) in diameter. This has been solved by the proposal that DNA remains highly condensed and supercoiled inside nucleus as chromosomes and this condensation is made possible by various supporting proteins.

Nucleosome Model of DNA packaging: This model was proposed by Roger Korenberg in 1974 after Don and Ada Olins observed beaded structures in DNA under electron

microscope. In the eukaryotic nucleus, there is a set of positively charged, basic proteins called histones. Histones are rich in the basic amino acid residues lysines and arginines both of which carry positive charges in their side chains. These histones are organised to form a unit of eight molecules called as histone octamer. A histone octamer consists of two proteins each of H_2A, H_2B, H_3 and H_4. Since the DNA is negatively charged, it is wrapped around the positively charged histone octamer to form a structure called nucleosome. A typical nucleosome contains 200 bp of DNA helix wound around the histone octomer. Another histone H_1 is present on the outer side of the nucleosome. Nucleosomes constitute the repeating units of chromatin – thread-like stained (coloured) bodies seen in nucleus. Two repeating nucleosomes are separated by a DNA stretch called linker DNA that is about 80bp long. These nucleosomes in chromatin are seen as 'beads-on-string' structure when viewed under electron microscope (EM). The beads-on-string structure in chromatin is packaged to form chromatin fibers that are further coiled and condensed to form chromosomes. The supercoiled packaging of chromatin at higher level is done with the help of additional set of proteins collectively referred to as Non-histone Chromosomal (NHC) proteins.

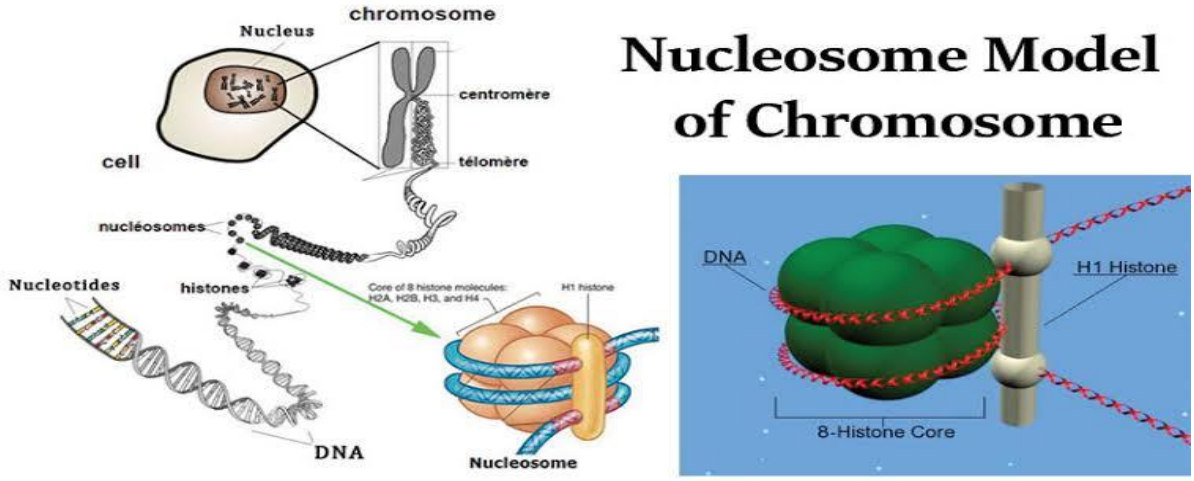


Fig. 14. Structure of nucleosome. Source: microbenotes.com

Central Dogma of Molecular Biology:

Generally, dogma refers to a set of principles laid down by an authority as intercontrovertibly true and the core principle in that set of rules is called central dogma. Soon after the successful elucidation of structure of DNA in the form of Double Helix

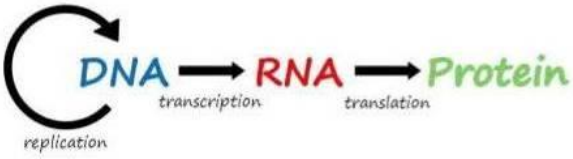


Fig. 15. Crick's Central Dogma of Molecular Biology. Source: slideshare.com

model, Francis Crick proposed the central dogma in molecular biology. It states that the flow of genetic information takes place from DNA to RNA and then RNA to Proteins. The coded genetic information flows from DNA located inside the nucleus to RNA present in the cytoplasm by the process of transcription. The coded information on RNA is later decoded in the form of proteins by the process of translation. Crick proposed that the flow of genetic information is unidirectional from DNA to RNA and to Proteins.

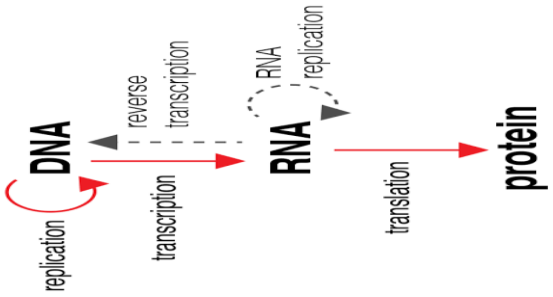


Fig. 16. Modified Central dogma. Source: Quora.com

This central dogma was however modified later with the discovery of retroviruses. Howard Temin and David Baltimore (1970) isolated an enzyme from some RNA viruses that help them to generate DNA from their RNA (genetic material). They called the phenomenon of turning RNA into DNA as **Reverse Transcription** and named this enzyme **Reverse Transcriptase**. Now as per the modified central dogma, the flow of information is not universally unidirectional but bidirectional for transcription in some cases.

DNA REPLICATION:

The process by which DNA forms its own carbon copies is called replication. DNA replication takes place during the **S-phase** of cell cycle. The process of replication requires parent DNA as template and a set of biocatalysts (enzymes). The main enzyme is referred to as DNA-dependent **DNA polymerase (DNA pol III)**, since it uses a DNA template to catalyse the polymerisation of deoxynucleotides. These enzymes are highly efficient catalysts and they catalyse polymerisation of a large number of nucleotides in a very short time. *E. coli* with 4.6 $\times 10^6$ bp completes the process of replication within 18 minutes; that means the average rate of polymerisation has to be approximately 2000 bp per second.

The mechanism of DNA replication was put forth by **Watson** and **Crick** immediately after their proposition of Double Helix model. They suggested that the two strands of DNA would separate and act as a template for the synthesis of new complementary strands. After the completion of replication, each DNA molecule would have one parental and one newly synthesised strand. Watson and Crick used the term semi-conservative DNA replication for this method of DNA copying.

Experimental Evidence for Semi-conservative replication of DNA: Watson and Crick proposed that replication of DNA is semiconservative but had no proof. However it was Matthew Messelson and Franklin Stahl who in 1958 proved that DNA replication is semi-conservative and not conservative or disruptive.

Messelson and Stahl grew *Escherichia coli* bacteria in a medium containing heavy isotope of nitrogen ($^{15}NH_4Cl$) as the only nitrogen source for many generations. The result was that heavy nitrogen ^{15}N was incorporated into newly synthesised DNA. This heavy DNA molecule could be distinguished from the normal DNA by centrifugation in a cesium chloride (CsCl) density gradient. Since ^{15}N is heavier than ^{14}N, it can be separated from ^{14}N based on the difference in their densities. These bacteria (with all heavy nitrogen in the DNA) were transferred into a medium with normal nitrogen ($^{14}NH_4Cl$) and took samples at definite time intervals as the cells multiplied, and extracted the DNA that remained as double-stranded helices. The various samples were separated independently on CsCl density gradients to measure the densities of DNA. When the DNA was extracted from the culture one generation (20 minutes, 1st generation) after the transfer from ^{15}N to ^{14}N medium, it had density intermediate between ^{15}N DNA and ^{14}N DNA. DNA extracted from the culture after another generation (40 minutes, 2nd generation) was composed of equal amounts of this hybrid DNA (^{15}N-^{14}N) and of 'light' DNA (^{14}N-^{14}N).

If the replication was conservative, after one generation two bands would have formed during centrifugation one heavy (^{15}N-^{15}N) and one light (^{14}N-^{14}N) instead of intermediate band hybrid ^{15}N-^{14}N. Similarly presence of two bands of Hybrid (^{15}N-^{14}N) and two bands of light (^{14}N-^{14}N) after second generation proves that the replication is smi-conservative.

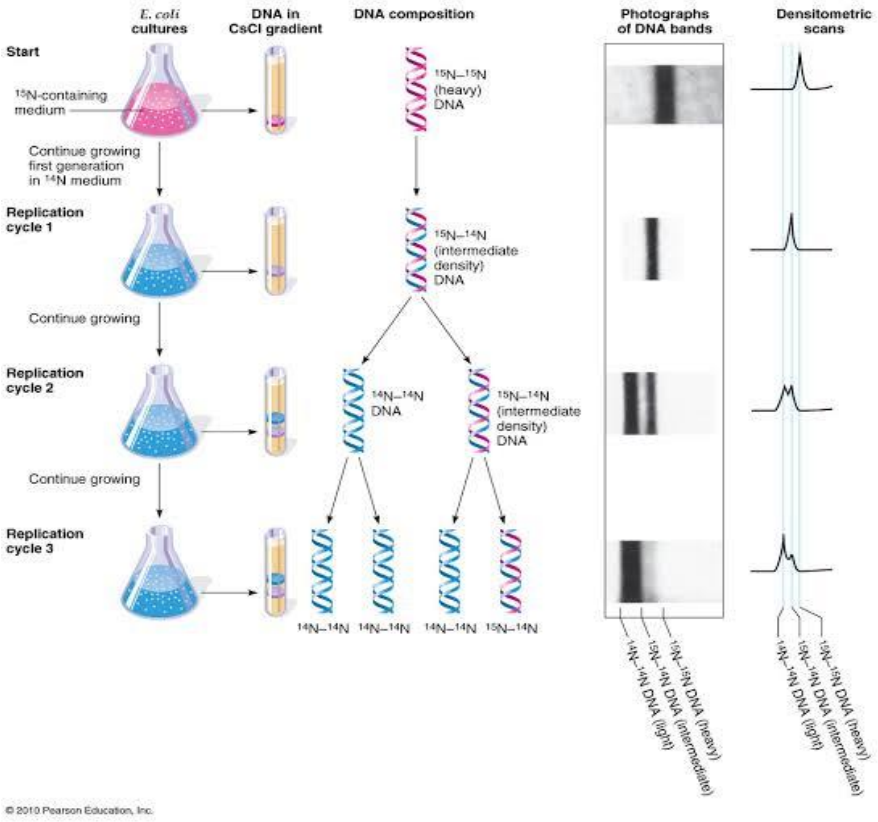


Fig. 17. Meselson and Stahl experiment. source: www.mun.ca

Mechanism of DNA replication: The semiconservative mode of DNA replication is a complex, multistep process that requires the involvement of many enzymes, factors and metal ions. The various steps involved in the process of replication are:

1. **Activation of deoxyribonucleotides**: The main raw materials for replication process are the inactive nucleotides or more specifically deoxyribonucleoside monophosphates that are floating free in the nucleoplasm. These include – deoxyadenosine monophosphate (deAMP), deoxyguanosine monophosphate (deGMP), deoxycytidine monophophate (deCMP) and deoxythymidine monophosphate (deTMP). These nucleoside monophosphates or nucleotides are activated by reacting with ATPs (phosphorylation). This phosphorylation reaction is catalysed by the enzyme **phosphorylase**. The result is the production of deoxyribonucleoside triphosphates viz. deoxyadenosine triphosphate (deATP), deoxyguansine triphosphate (deGTP), deoxycytidine triphosphate (deCTP) and deoxythymidine triphosphate (deTTP).
2. **Unwinding of DNA double helix and exposure of strands**: The coiled DNA double helix uncoils and the two DNA strands get separated from each other at some

specified location. The unwinding and strand separation is done by an enzyme ***Helicase*** (unwindase) with the help of energy released by breakdown of ATP. The two separated DNA strands are prevented from rejoining and recoiling by a set of proteins called SSBs or single stranded DNA binding proteins (Helix stabilising proteins). The whole DNA of a chromosome does not split simultaneously but at a specific point called origin of replication or ***Ori*** site. There is only one Ori – site in prokaryotes, but in eukaryotes there can be multiple Ori-sites in a single DNA molecule.

Unzipping of the double stranded DNA forms a Y-shaped region called a Replication fork. The replication fork is a short DNA segment beginning from Ori-site towards the overall direction of replication.

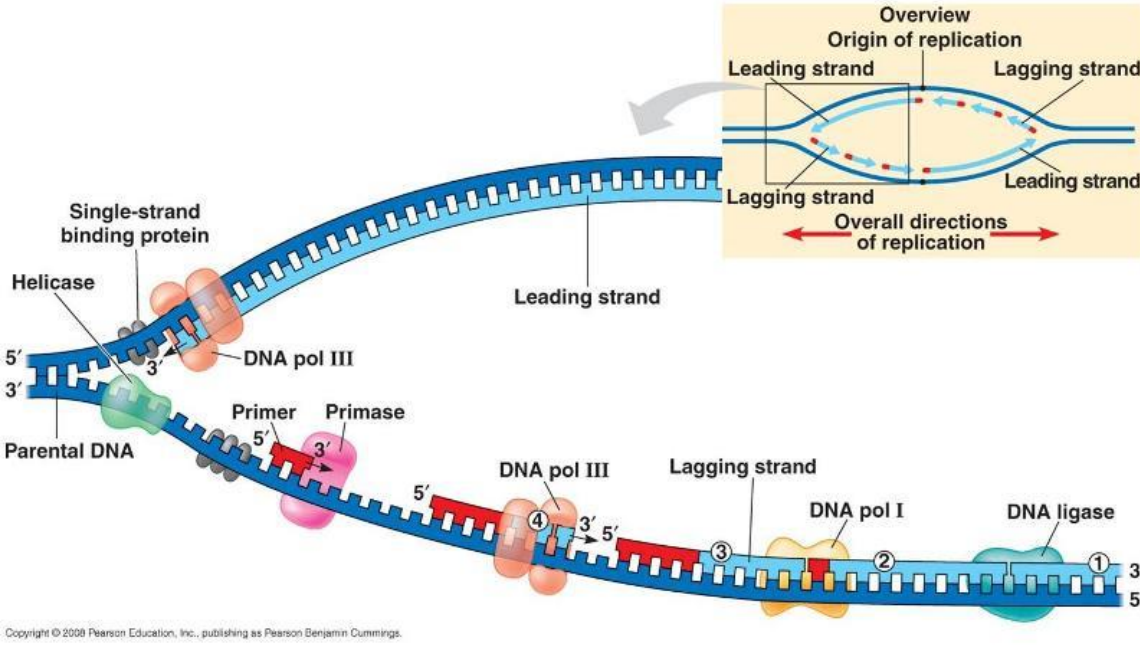


Fig. 18. Mechanism of DNA replication. Source: mun-ib/biology

3. **Formation of RNA primer**: Primer is a short stretch of RNA formed on the DNA template at the 5′ end. An RNA polymerase called **Primase** catalyses the polymerisation of RNA nucleotides as a primer on single stranded DNA. RNA primer is necessary because DNA polymerase (DNA pol III) can catalyse the growth of DNA chain only in the presence of already existing nucleotide stretch. So RNA polymerase (primase) prepares that RNA stretch to which DNA polymerase binds and repeatedly adds DNA nucleotides. Since the primers are formed in 5′→3′ direction and the two DNA strands are antiparallel, one RNA primer is formed at the wider end of replication fork and the other at its base.

4. **Growth of new DNA strands**: The activated nucleoside triphosphates join with their complimentary nitrogen bases (A-T; C-G) by forming hydrogen bonds. The high energy bonds of deoxyribonucleoside triphosphates break to form nucleoside monophosphate, and the energy released in the process is used to form phosphodiester bonds with the primer and subsequent DNA nucleotides. This polymerisation of deoxyribonucleotides is catalysed by DNA polymerase.
5. **Leading and lagging strands**: At the time of replication, the two DNA strands do not separate in the entire length but opens up as short stretches. The DNA polymerase can add deoxyribonucleotides unidirectionally in 5′→3′ only. Because the two parental strands are antiparallel, the new strands must be formed on the old strands in opposite directions. One new strand is formed in a continuous stretch in the 5′→3′ direction (because it proceeds towards the overall direction of replication or towards direction of unzipping of DNA) and this strand is called ***Leading*** strand. The other new strand (whose direction is opposite to the overall direction of replication) is formed on second parent strand as short segments and this strand is therefore called ***Lagging*** strand. The leading strand requires only one RNA primer but the lagging strand has to form a new primer every time the fork unzips further. DNA replication on the leading strand is continuous but on the lagging strand it is discontinuous and new DNA strand is formed as fragments called Okazaki fragments. The Okazaki fragments are later linked together with the help of DNA ligase which replaces RNA nucleotides of primer with DNA nucleotides. The process of replication continues till the whole DNA molecule gets replicated.

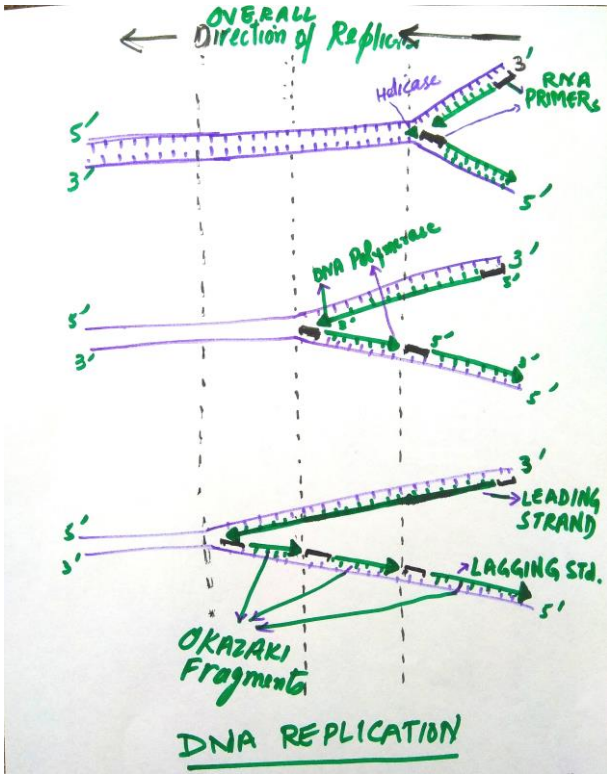


Fig. 19. Process of replication.

6. **Proofreading:** The specificity of base pairing ensures accurate replication. However occasionally wrong bases get incorporated which are later removed by another type of

DNA polymerase called DNA pol I. This process of correction is called DNA proofreading. When the whole process of replication is complete, two DNA molecules each with one parental strand and one new strand is formed. They again coil to form typical double helices.

Ribonucleic Acid (RNA): Although RNA acts as a genetic material in many viruses, in cellular organisms, RNA is produced from DNA. RNA is a single stranded polynucleotide that functions as carrier of coded genetic information from DNA inside nucleus to ribosomes in the cytoplasm. RNA is made of ribonucleotides and each nucleotide consists of a ribose sugar, a phosphate group and one of the four nitrogen bases (adenine, guanine, cytosine and uracil). There are three categories of RNA viz messenger RNA (mRNA), ribosomal RNA (rRNA) and transfer RNA (tRNA). All these three RNAs are required to synthesize proteins.

Messenger RNA: mRNA accounts for about 5% of the total RNA in the cell. It is the most heterogeneous among the three types of RNA both in terms of base sequence as well as size. It carries the coded genetic information copied from the DNA during transcription. As part of post-transcriptional processing in eukaryotes, the 5' end of mRNA is capped with a guanosine triphosphate nucleotide, which helps in mRNA recognition during translation or protein synthesis. Similarly, the 3′ end of an mRNA has multiple adenine nucleotide residues added to it (called a poly A' tail), which prevent enzymatic degradation of mRNA. The main function of mRNA is that it carries the coded genetic information from nucleus to the ribosomes in cytoplasm.

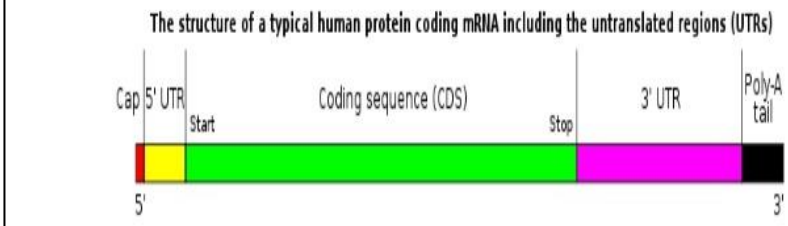


Fig.20. Structure of mRNA. Source: slideshare.net

Ribosomal rRNA (rRNA): rRNA is present in the ribosomes and account for almost 80% of the total RNA present in the cell. Depending upon the sedimentation coefficient, rRNA of eukaryotes are of four types – 28s, 18s, 5.8s and 5s. However in prokaryotes there are three types, 23s, 16s and 5s. 18s of eukaryotes and 16s of prokaryotes are located in the small subunits while the rest are found in the larger subunits. The main function of rRNA is that it directs the translation of mRNA into proteins inside ribosomes.

Transfer RNA (tRNA): Transfer RNA is the smallest of the 3 type of RNA having about 75-95 nucleotides. It forms about 15% of total cellular RNA. tRNAs have been named so because they are involved in transfer of aminoacids from cytoplasm to ribosomes during protein synthesis. Each of the 20 amino acids has a specific tRNA that binds with it and transfers it to the growing polypeptide chain. The tRNA molecules act as adaptor molecules between mRNA and amino acids.

***Structure of tRNA*:** Although RNAs are single stranded, a tRNA molecule has a clover leaf structure which is stabilized by strong hydrogen bonds between the nucleotides. Three structural loops are formed via hydrogen bonding. tRNA has the following regions:

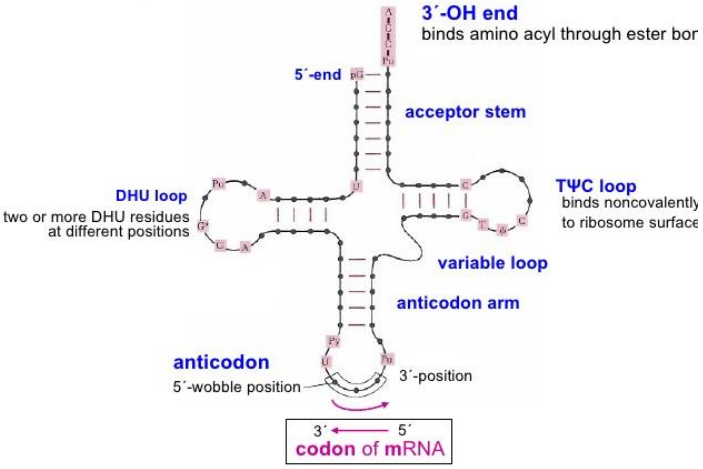


Fig. 21. Structure of tRNA. Source: eduladder.com

a) *Anticodon loop*: It is made up of seven nucleotides of which three form the anticodon for recognising and attaching to the codon of mRNA.
b) *Amino acid Binding Site*: This site lies at the 3' end and possesses a CCA – OH group. Aminoacids that are carried by tRNA get attached to this site.
c) *TΨC loop*: It is made of seven bases and two of them pseudouridine and ribothymidine are unusual. It helps in binding with ribosome.
d) *DHU loop* (Aminoacyl synthetase enzyme site): This loop contains 8-12 bases and forms the binding site for enzyme aminoacyl synthetase enzyme.

The transfer RNA transfers amino acids to the ribosomes that correspond to each three-nucleotide codon of mRNA. The amino acids then can be joined together and processed to make polypeptides and proteins.

TRANSCRIPTION:

The process by which genetic information present on DNA is used to form RNA is called transcription. Transcription can also be defined as the formation of RNA from a portion of one strand of DNA. As the DNA is located inside nucleus and the ribosomes are present in

the cytoplasm, there must be some agency to carry the coded information from DNA to ribosomes, where protein synthesis takes place. This agency is the RNA that is formed via Transcription. Various types of RNAs – mRNA, t-RNA and r-RNA are formed from DNA template using different enzymes.

Transcription also works on the principle of complementarity except than nitrogen base adenine pairs with uracil instead of thymine. In transcription only one strand acts as a template for RNA synthesis. This by convention is called non-coding or anti-sense strand because its bases are complementary to the RNA formed from it. The other strand which does not participate in RNA formation is called coding strand or sense strand as its sequence resembles the RNA formed from template strand. In transcription only a segment of DNA gets transcribed unlike replication where in the whole DNA gets copied.

Transcription Unit: A transcription unit consists of three regions in the DNA. They are promoter, the structural gene and a terminator. The promoter and terminator sequences flank the functional structural gene. The promoter is located towards 5′ -end (of coding strand/ non-template strand) of the structural gene. It is actually a DNA sequence that provides a binding site for RNA polymerase. The direction of transcription is again 5′→3′ direction. The structural gene is a long DNA sequence that contains the real information required for transcription. The terminator is located towards the 3′ -end of the coding strand and signals the end of transcription. Some additional DNA sequences located randomly and called regulators may also be connected with the process.

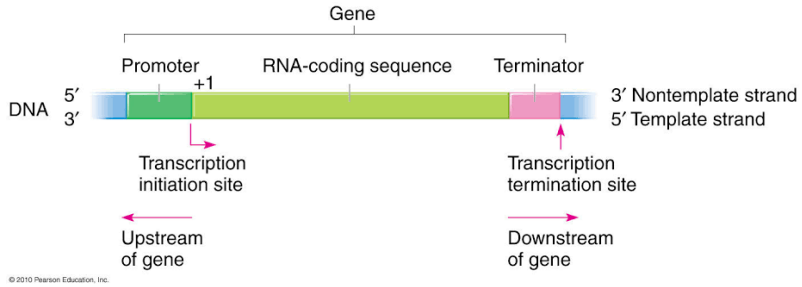


Fig. 22. Transcriptional unit. Source: mun.ca

Process of Transcription:

Initiation of transcription: The DNA dependent RNA polymerase binds with the double stranded DNA at a specific site called promoter sequence. Basically, the job of promoter is to tell the polymerase where to "sit down" on the DNA and begin transcribing. Promoter is

located towards the 5′ end of the sense strand (non-template strand). The promoter also determines which among the two strands will act as template for transcription. Once the RNA Polymerase gets bound with the promoter, it is ready to unzip the DNA duplex and add RNA nucleotides to it. This part of transcription is called Initiation. For binding and unzipping, the RNA polymerase associates itself temporarily with *initiation factor* **σ**.

***Elongation of RNA Chain*:** Elongation is the stage when the RNA strand gets longer due to the addition of new ribonucleotides. During elongation, RNA polymerase unzips the DNA duplex and "walks" along one strand of DNA, known as the template strand, in the 3' to 5' direction. Only one strand acts as template and this strand is called anti-sense or non-coding strand. The other non-template strand is called sense strand or coding strand.

Fig

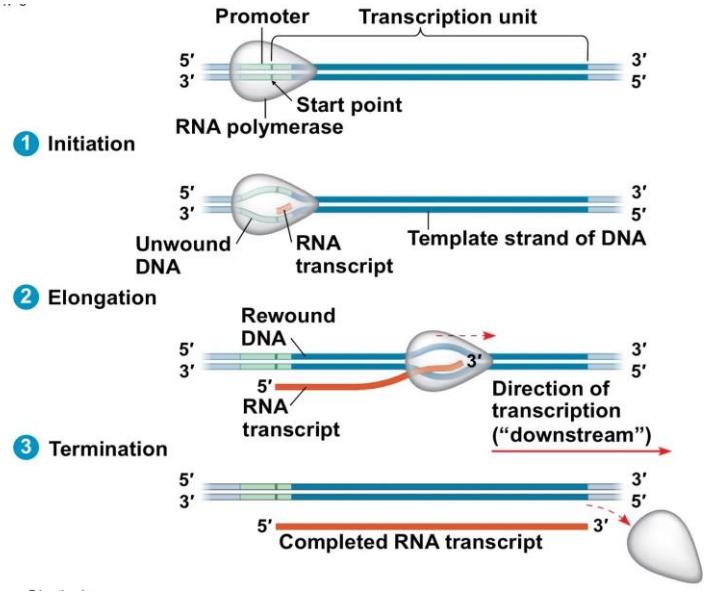


Fig. 23. Steps of Transcription. Source: learnexpert

For each nucleotide in the template, RNA polymerase adds a complementary RNA nucleotide (ribonucleotide) to the 3' end of the RNA strand (i.e. 5′→3′ direction). Ribonucleoside triphosphates that are present in the nucleus in the form of Adenosine triphosphate (ATP), guanosine triphosphate (GTP), cytidine triphosphate (CTP) and uridine triphosphate (UTP) serve as the raw material for transcription. These nucleoside triphosphates on linking to the DNA template break off into ribonucleoside monophosphates (ribonucleotides) and pyrophosphates releasing ample energy. This energy is utilized in adding the ribonucleotides to the developing RNA chain.

Termination of transcription: RNA polymerase will keep on polymerizing RNA nucleotides until it gets signals to stop. The process of ending transcription is called **termination**, and it happens once the polymerase transcribes a sequence of DNA known as a **terminator**. For this culmination of transcription, RNA polymerase binds with another factor called ***termination factor ρ***. At this point the RNA molecule as well as the RNA polymerase enzyme is released from the DNA template.

Processing of RNA: The RNA transcribed from the DNA template is termed as Primary transcript. It undergoes several post transcriptional modifications before becoming functional. The main modification involves the removal of those unnecessary portions that are transcribed from intron regions of DNA (which are useless to carry forward). Other modifications depends upon the type of RNA it has to form. There are different modifications for mRNA, rRNA and tRNA already described in their structures.

GENETIC CODE:

The genetic code is the set of rules by which information encoded in genetic material (DNA or RNA sequences) is translated into proteins (amino acid sequences) by living cells.

Translation requires transfer of genetic information from nucleotides to amino acids but no complementarity can be seen between the two. So a genetic code was proposed that could direct the sequence of amino acids during synthesis of proteins. Deciphering of genetic code was one of the most challenging jobs in molecular biology. It required involvement of scientists from varied disciplines – physics, organic chemistry, biochemistry and genetics. It was **George Gamow**, a physicist, who argued that genetic code should be in triplet form (three nucleotides forming one codon) as there are only four types of nucleotides and twenty types of aminoacids. **Har Gobind Khorana** developed the chemical method for synthesizing RNA molecules with defined combinations of bases (homopolymers and copolymers). Marshall **Nirenberg's** cell-free system for protein synthesis finally helped the code to be deciphered. **Severo Ochoa**'s discovery of enzyme polynucleotide phosphorylase was also helpful in polymerising RNA with defined sequences in a template independent manner (enzymatic synthesis of RNA).

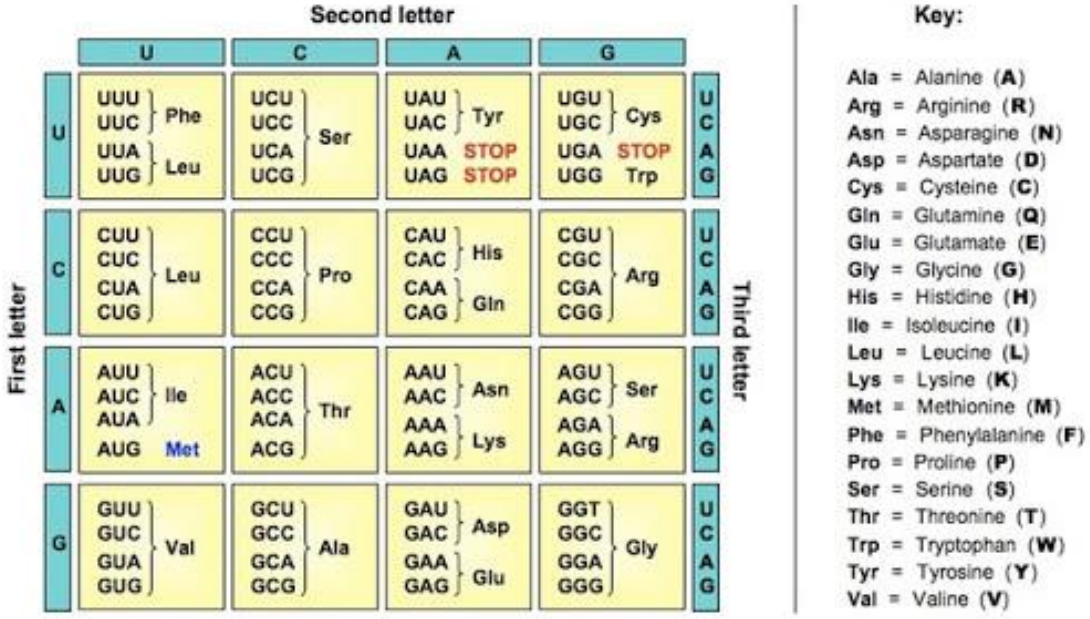


Fig. 24. Genetic code chart. Source: bioninja.com

Properties of Genetic code:-

1. **Genetic code is a triplet**: As pointed out earlier, the coding units or codons for amino acids comprise three letter words (triplets). Each codon is actually a three nucleotide sequence of mRNA.
2. **The Code is Degenerate:** The occurrence of more than one codon for a single amino acid is referred to as degeneracy. Out of 61 functional codons, AUG and UGG code to one amino acid each. But remaining 18 amino acids are coded by 59 codons.
3. **The Code is Non-overlapping**: In a non-overlapping code, the same letter (i.e., base) is not used in the formation of more than one codon.
4. **The Code is Comma Less**: A comma less code means that no nucleotide or comma (or punctuation) is present in between two codons. Therefore, code is continuous and comma less and no letter is wasted between two words or codons.
5. **The Code is Unambiguous**: There is no ambiguity in the genetic code. A given codon always codes for a particular amino acid, wherever it is present.
6. **The Code is Universal**: The genetic code has been found to be universal in all kinds of living organisms — prokaryotes and eukaryotes.
7. **Start and Stop codons:** There is a specific codon AUG that initiates the process of protein synthesis (initiation codon) and there are three codons that signal the termination of polypeptide synthesis (Termination codons; non-sense codons).

8. **Colinearity**: DNA is a linear polynucleotide chain and a protein is a linear polypeptide chain. The sequence of amino acids in a polypeptide chain corresponds to the sequence of nucleotide bases in the gene (DNA) that code for it. Change in a specific codon in DNA produces a change of amino acid in the corresponding position in the polypeptide. The gene and the polypeptide it codes for are said to be co-linear.
9. **Gene-polypeptide Parity**: A specific gene transcribes a specific mRNA that produces a specific polypeptide. On this basis, a cell can have only as many types of polypeptides as it has types of genes. However, this does not apply to certain viruses which have overlapping genes.

TRANSLATION (Protein Synthesis):

The ultimate goal of the DNA is realised when its information gets expressed in the form of proteins. Since the DNA is located inside nucleus and ribosomes (protein factories) are present in the cytoplasm, the coded information on DNA is first converted into RNA via transcription. The RNA then leaves nucleus and carries the information to ribosomes where it is decoded into proteins. During translation, the genetic code in mRNA is used to arrange amino acids in a particular sequence thereby making a protein.

In translation, the four letter language of DNA (deoxyribonucleotides) that was converted into four letter language of RNA (ribonucleotides) during transcription is translated into 20 letter language of proteins (amino acids).

Process of Translation: Protein synthesis within a cell takes place in a step wise manner. The main steps for translation include:

i) Activation of Amino Acids: Amino acids present in the cellular pool react with ATP molecules in presence of enzyme aminoacyl-tRNA synthetase to form Amino acid – AMP- Enzyme complexes releasing pyrophosphates. There is a separate aminoacyl tRNA synthetase enzyme for each kind of amino acid. This **amino acid – AMP – Enzyme complex** is called activated amino acid.

ii) **Charging of tRNA**: The Amino acid – AMP –Enzyme complex joins with the amino acid binding site of its specific tRNA and forms **tRNA-Amino acid complex**. The enzyme aminoacyl-tRNA synthetase that also catalyses this step is finally released along with AMP.

iii) **Activation of Ribosomes**: Ribosomes, the factories for protein synthesis must be activated to begin translation. For activation, the smaller and large subunit are joined together with the help of mRNA. mRNA first joins with the smaller subunit by base pairing with appropriate sequences on rRNA present inside small subunit. The large subunit later joins the small subunit to form active ribosome.

iv) **Formation of polypeptide**: This process takes place in three steps viz. initiation, elongation and termination.

a) **Initiation of polypeptide**: For initiation, there is a start codon at the 5′ end of the mRNA chain. This initiation codon **AUG** lies at the **P-site** (peptidyl site) of the ribosome and codes for methionine (formyl methionine in prokaryotes). Activated

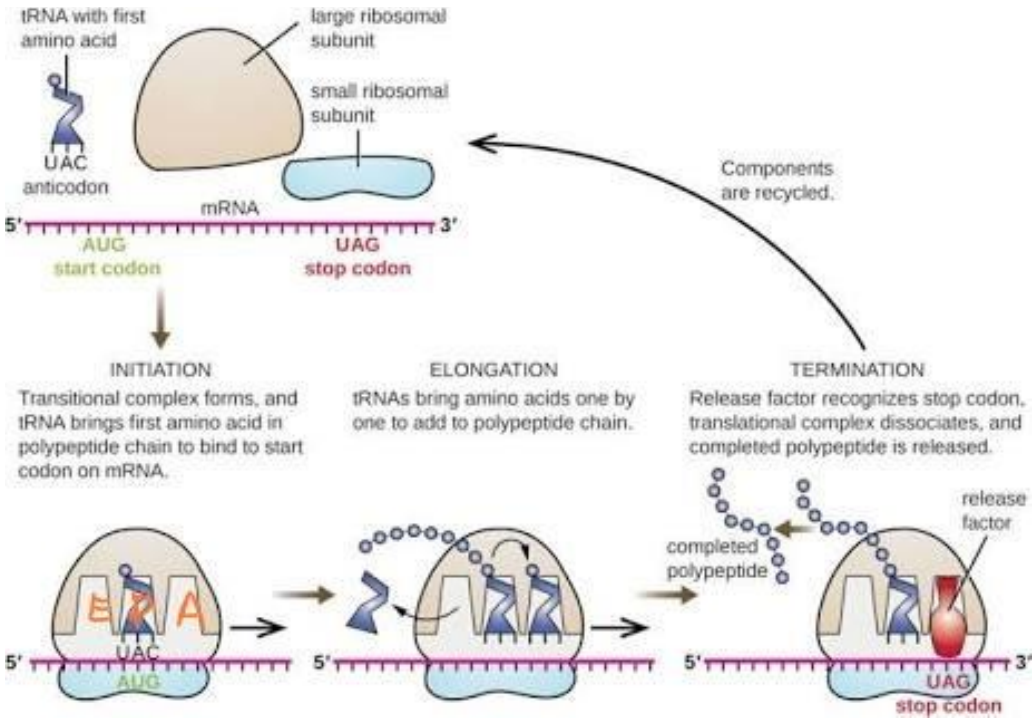


Fig. 25. Process of Translation. Source: lumenlearning.com

Methionine is carried from amino acid pool by tRNA having the anticodon UAC that bonds with **AUG** codon by hydrogen bonds. This step is promoted by three initiation factors namely IF_1, IF_2 and IF_3 and a GTP. The other site called A-site (aminoacyl site) remains vacant to accommodate the next charged tRNA. It is at this point the larger ribosomal subunit actually joins the small subunit to complete the ribosome.

b) **Elongation of polypeptide chain:** A charged tRNA molecule carrying an aminoacid enters the ribosome at the vacant A-site. After finding its complementary codon, its anticodon makes hydrogen bonds with the codon. Now the aminoacid Methionine is

transferred to newly arrived charged tRNA forming peptide bond with the free NH_2 group of its amino acid. This process is catalysed by **peptidyl transferase**. So the second tRNA now carries a dipeptide. The ribosome moves 3-nucleotides (one codon) along the mRNA towards 3′direction. The first tRNA after transferring its amino acid dissociates from the mRNA through E-site (exit site) and returns back to amino acid pool. With this the tRNA carrying dipeptide at the A-site is pulled to the P-site. This is called Translocation and it is carried by Translocase enzyme with GTP as energy source. As the A-site becomes vacant, a third specific charged tRNA fills the gap. Again peptidyl transferase shifts the dipeptide from P-site tRNA to third tRNA (tRNA at A-site). Uncharged tRNA leaves the mRNA via E-site of ribosome, to carry another amino acid from the pool. Translocation of ribosome towards next codon leaves the A-site empty for next tRNA. This process of arrival of charged tRNAs onto mRNA, peptide bond formation and translocation of ribosomes to the next codon is repeated several times. After every translocation, the previously formed amino acid chain is shifted to the newly arrived tRNA. Thus the amino acids are linked up into a polypeptide in a sequence already communicated by DNA via mRNA. Elongation of polypeptide is assisted by three elongation factors namely **EF-Tu**, **EF-Ts** and **EF-G**.

c) **Termination and Release of Polypeptide chain:** The process of chain elongation continues till a stop signal is arrived. Three codons UAA, UAG and UGA respectively named as **Ochre**, **Amber** and **Opal** act as stop signals or termination codons because they do not code for any amino acid. At this point the linkage between the last tRNA and the polypeptide chain is broken with the help of three release factors, $\mathbf{RF_1}$, $\mathbf{RF_2}$ and $\mathbf{RF_3}$ and GTP. This reaction is also catalysed by peptidyl transferase enzyme. At this point the ribosome also separates from mRNA and dissociates into its two subunits.

The released polypeptide has a primary structure. It gets modified by folding and coiling into various types of secondary and tertiary proteins.

REGULATION OF GENE EXPRESSION:

Genes are not transcribed or translated continuously within a cell. A continuous expression of different genes would be a waste of energy. Therefore expression of genes takes place as and when the need arises. If we consider that the gene expression results in the formation of a polypeptide, it can be regulated at several levels. In eukaryotes particularly, the regulation could be applied at (i) transcriptional level (formation of primary transcript), (ii) processing level (regulation of splicing), (iii) transport of mRNA from nucleus to the cytoplasm, (iv) translational level (polypeptide synthesis level).

In prokaryotes, gene expression is regulated predominantly at the transcriptional level. Within a transcriptional unit, the activity of RNA polymerase at a promoter site is regulated by interactions with accessory proteins. These proteins affect the ability of RNA polymerase to recognise initiation sites and bind with them. These regulatory proteins act either positively (inducers or activators) or negatively (repressors). The accessibility of promoter regions of prokaryotic DNA is usually regulated by the interaction of proteins with sequences termed operators that lie adjacent to promoter sites.

Regulation of gene expression in prokaryotes is exerted by a set of genes together called an operon. An Operon can be defined as a cluster of functionally related genes that are controlled by an operator. It can be defined as a segment of DNA containing adjacent genes that include structural genes, an operator gene and a regulatory gene. The operator region is adjacent to the promoter elements in most operons. Each operon has its specific operator and specific repressor.

The *Lac* Operon: The pioneering work in gene regulation was done by two French scientists Francios **Jacob** (Geneticist) and Jaque **Monod** (Biochemist). They put forth the *Lac* Operon model of gene regulation in 1961 for which they received **Nobel Prize** for Physiology and Medicine in 1965. In *Lac* Operon (here lac refers to lactose), a polycistronic structural gene is regulated by a common promoter and regulatory genes.

The *Lac* Operon consists of one regulatory gene (***i*** *gene - inhibitor*) and three structural genes (cistrons) namely Lac **Z**, Lac **Y**, and Lac **A**). The ***i*** gene codes for the repressor of the Lac Operon. The Z gene codes for an enzyme beta-galactosidase (also called Lactase), which hydrolyses disaccharide lactose into monosaccharides galactose and glucose. The Y gene codes for β-galactoside permease, which increases permeability of the cell to β-galactosides (lactose). The A gene encodes another enzyme, transacetylase. Hence, all the three gene products in Lac operon are enzymes required for metabolism of Lactose.

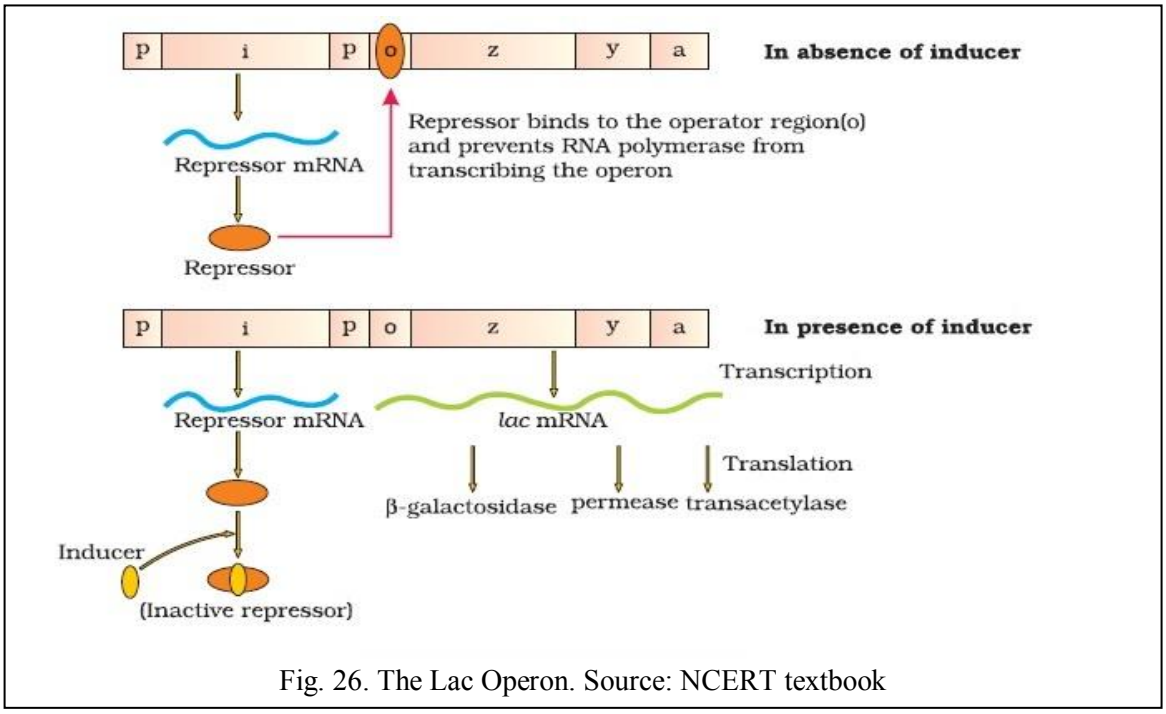


Fig. 26. The Lac Operon. Source: NCERT textbook

Lac Operon is an Inducible Operon: Inducible operon is one that is switched on by the presence of substrate (inducer). In Lac Operon, Lactose is the substrate for the enzyme beta-galactosidase and it regulates switching on and off of the operon. Hence lactose is termed here as inducer.

In this Operon, The regulator gene '***i***' is a constitutive gene (house keeping gene) and forms its product **repressor** all the time. In the absence of Lactose (inducer), the *Repressor* protein remains attached to the Operator region of the Operon and thereby prevents RNA polymerase from binding to the Promoter thus ceasing any transcription of structural genes Z, Y and A.

If lactose (inducer) is provided in the growth medium (when it's preferred source i.e. glucose is absent), the repressor gets inactivated because it binds with the inducer

(lactose) owing to its affinity towards it. The repressor-inducer complex is unable to bind with operator gene. This allows RNA polymerase access to the promoter (Operon is Switched on) leading to transcription of Lac Z, Lac Y and Lac A. Transcription is followed by translation of the three structural genes which are the three enzymes (β-galactosidase, β-galactoside permease and transacetylase). β -galactosidase breaks down Lactose into its monosaccharide units-glucose and galactose and permease transports more lactose into the cell to get metabolised by β-galactosidase. Process of transcription and translation of structural genes continues till most of the lactose is metabolised. Now there are no more inducers to bind with continuously forming repressors and therefore repressor again binds with the operator. The RNA polymerase cannot bind with promoter now and transcription of structural stops again, thus the Operon is switched off.

Evaluation

Q1. Mendel was lucky in selecting Pea plants for his hybridisation experiments. What were the advantages of selecting pea plant over other plants?

Q2. Differentiate between the following:

(a) Dominance and Recessive

(b) Homozygous and Heterozygous

(c) Monohybrid and Dihybrid.

Q3. Explain the Law of Dominance using a monohybrid cross.

Q4. What is a test-cross?

Q5. When a cross in made between tall plant with yellow seeds (TtYy) and tall plant with green seed (Ttyy), what proportions of phenotype in the offspring could be expected to be

(a) tall and green.

(b) dwarf and green.

Q6. Two heterozygous parents are crossed. If the two loci are linked what would be the distribution of phenotypic features in F1 generation for a dibybrid cross?

Q7. A child has blood group O. If the father has blood group A and mother blood group B, work out the genotypes of the parents and the possible genotypes of the other offsprings.

Q8. Explain the following terms with example

(a) Co-dominance (b) Incomplete dominance

Q9. Who had proposed the chromosomal theory of the inheritance?

Q10. What is the difference between a test cross and a back cross?

Q11. If a double stranded DNA has 20 per cent of cytosine, calculate the per cent of adenine in the DNA.

Q12. Group the following as nitrogenous bases and nucleosides: Adenine, Cytidine, Thymine, Guanosine, Uracil and Cytosine.

Q13. If the sequence of one strand of DNA is written as follows:

5'-CATGGCATGCATGCATGCATGCATGCATGCA-3'.

Write down the sequence of complementary strand in 5'→3' direction.

14. If the sequence of the coding strand in a transcription unit is written as follows:

5' -GAATGCTATGCATGCATGCATGCATGCATGCT-3'.

Write down the sequence of mRNA.

15. Which property of DNA double helix led Watson and Crick to hypothesise semi-conservative mode of DNA replication? Explain.

16. How did Hershey and Chase differentiate between DNA and protein in their experiment while proving that DNA is the genetic material?

17. Differentiate between the followings:

(a) mRNA and tRNA

(b) Template strand and Coding strand

18. List two essential roles of ribosome during translation.

19. What do you understand by:

(a) Transcription (b) Translation

20. Briefly explain the function of the followings:

(a) Promoter (b) tRNA

UNIT – III **BIOLOGY IN HUMAN WELFARE**

Plant breeding: Introduction, steps in plant breeding and application of plant breeding, and Single cell protein, Biofortification.

Food Production

The pre-historic man was a hunter of animals and would also gather plants from his surroundings. He lived in small groups and nomadic life. The history of life and food habits of pre- historic man can be obtained from the plant and animal fossils, archaeological sites, radioactive carbon dating technique etc.

The agriculture originated some 7000 – 13000 years ago somewhere in the well watered high lands of **Euphrates** (Iraq), **Neil** (Egypt) and **Indus River Valley** civilization etc. The fossil records of some seeds and food grains from **Chinese River Valley** civilization, **Tarucan Valley** in Mexico have been obtained to give an idea about their pre-historic agricultural activity.

The first relationship between man and plants was established when he started gathering fruits, seeds and roots of wild plants for food purposes. The earliest human civilizations consumed cereals, wheat, barley, rice, rhizomes, wild plants, seeds and weeds.

The archeological evidences for the origin of cereals points to mountainous areas of old world (Asia, Europe and Africa) and new world (North and South America). The Chinese and Indians have been reported to be the older rice eaters (7000 years). The earlier cultivated plants include Hemp (Canabis sativa), potato, rhizomes etc.inAfrica and Mulbery, loquat and litchi in China. The coconut, wheat and some rhizomes in Mexico and tobacco and potato in South America etc. Due to human migrations the cultivation techniques was carried from across the continents with the development of human civilizations.

The oldest stone age (Paleolithic) shows absence of agriculture, the Mesolithic shows scanty agriculture and new stone age (Neolithic) shows fully developed agriculture (3000-4000 BC) with polished stone tools.

Nowadays with the help of metals (Iron age) and scientific techniques, agriculture has developed enormously. The multiple cropping, use of fertilizers,

hybrids and pest resistant varieties all over the world has helped in raising quality and quantity of agricultural practices (products).

Plant Breeding

Crop improvement means to develop crops with desired characters such as higher yields, better qualities, resistance to different stresses (disease, pests, drought, frost, salinity etc.) and shorter duration of maturity. It is done by improving the genetic potentialities in crop plants. The scientists who are concerned with improvement of plants are *Plant breeders* and the science of improvement of crop varieties is called *Plant breeding*. Plant breeders select different plants with desired characters and cross them to obtain the desired characters in the offsprings. The offsprings (hybrids) with desired characters are then selected, tested, multiplied and supplied to farmers, grower or planters.

Improved Variety: An improved variety is superior to the other existing varieties of the same crop in respect of one or more characters like high yield, superior quality, early maturity, resistance to important diseases and insect pests.

Crop improvement can be achieved on the following grounds:

- Selection
- Introduction
- Acclimatization
- Hybridization
- Polyploidy
- Breeding
- Induced mutations
- Tissue culture
- Genetic engineering

  1. **Selection**: The process by which suitable and desired varieties of plants are chosen for breeding or improvement is called *Selection*. The selection in a population can be achieved by preserving desirable traits like early ripening, disease resistance, better quality, high yield and drought and water resistance etc. The selection can be of two types;

  a). Mass selection

  b). Pure line selection

a). **Mass selection:** Mass selection means when desirable individual plants are selected solely on the basis of their phenotypes, grown in mass.

b). **Pure-line selection**: Pure-line selection is the collection of true breeding plants resulting from self pollination of a single homozygous individual.

Some varieties have got selected naturally during the course of evolution, while as by human efforts various varieties have been selected artificially.

2. **Introduction**: The process of introducing various plants from their growing localities to new places is called introduction. The varieties of potatoes,tobacco,maize were introduced in Asia from America, The dwarf rice variety (Taichung) was introduced from Taiwan to India. The international rice variety -8 (IR-8) has been introduced from Phillipines to India. The simple gene dwarf variety of wheat (Sonora,Lerma,Roja) is introduced from Mexico to India. It has proved extensively useful. Similarly litchi fruits and loquats have been introduced from China. Through the sameway , various useful animal varieties have been introduced from Europe to India.

3). **Acclimatization**: The process of adjustment and adaptation of different plants under changed climatic conditions is called acclimatization. When these varieties and their cross breeds get hybridized then their seeds are given to farmers and growers for further cultivation and propagation.

4).**Hybridisation**: Hybridisation is the process of making a cross between the two genetically different parents to obtain a progeny with the desired traits. OR

Hybridisation is the method of producing new crop varieties in which two or more plants of unlike genetical constitution are crossed together to produce new improved and genetically desired variety.

Hybridisation may involve a ***single cross*** (two plants) or ***multiple cross*** (more than two plants).

Hybridisation may be;

i. Intravarietal: Cross between two plants belonging to same variety. E.g;
ii. Intervarietal (Intraspecific): Cross between two varieties of the same species.e.g; Different varieties of wheat.

iii. Interspecific: Cross between the different species of the same genus. E.g; Rice variety ADT-37 from *Oryza japonica* and *Oryza indices* and all sugarcane varieties

iv. Intergeneric: Cross between two different genera.e.g;

Hybridisation is the most common method of creating genetic material. It brings about useful genetic or heritable variations of two or more lines together.

Process of Hybridisation

The process of hybridisation involves the following steps:

- Selection of parents with desired characters
- Selfing
- Emasculation
- Bagging
- Tagging
- Crossing (Artificial pollination)

Line – a group of individuals related to descent and have similar genotype.

Lines used in hybridisation are called **parents**.

Emasculation prevents self pollination in flowers.

Bagging prevents the contact between unwanted pollens and emasculated flowers.

a). Selection of parents with desired characters: Firstly all the required desired characters (traits) in a new crop variety are selected.

b). Selfing: The selected parents are allowed to undergo self breeding to bring about homozygosity of the desired traits.

c). Emasculation: Emasculation is the removal of anthers (male parts) from a bisexual flower before maturity.

d). Bagging: The emasculated flowers are immediately covered by paper, plastic or polythene bags, it is called bagging.

e).Tagging: The emasculated and bagged flowers must be tagged by writing every step with date and time.

f). Crossing (Artificial pollination): Controlled pollination by bringing selected pollens in contact with stigma through human efforts is called artificial pollination. When the stigma of the emasculated flower of female parent matures, the covering bag is removed for a short while. The stigma is dusted with already collected pollens by means of a clean brush.

After pollination the emasculated flower is covered again till the stigma remains receptive. When the begin to develop, the bags are removed. The seeds formed by such flowers are termed as *hybrid* or F_1 *seeds* which later on give rise to F_2 *seeds* and so on.

The process of hybridisation has played a vital role in better crop yield, better quality and quantity, early ripening and disease resistance, thus enhancing *green revolution*.

Heterosis – the superiority of the hybrid over either parent in one or more traits is called heterosis or *hybrid vigour*. Heterosis is commercially exploited in maize,sugarcane,rice,wheat,barley,petunia cabbage,tomato and cucumber etc.

5). **Polyploid Breeding**: When an organism has more than two sets of chromosomes or genomes in their cells, its termed as *polyploid* and the condition is reffered as *Polyploidy*.Any organism which has two sets of chromosomes is called as *diploid*, when three sets *triploid*, with four sets or two diploid sets is *tetraploid*. When such variations arise within a species, it is called as *auto-polyploidy*.

Polyploidy may be;

a): **Autopolyploidy**: Polyploidy in which there is a numerical increase of the same genome,e.g; AAA(autotriploid), AAAA(autotetraploid).

e.gs; maize, rice, gram.

b).**Allopolyploidy**: It is the doubling of chromosomes by a cross between two species.e.g; AABB(allotetraploid).Allopolyploids function as new species,e.g; Wheat, American cotton, *Nicotiana tabtcum*

Autoallopolyploid is a type of allopolyploids where one genome

Autopolyploidy induce *gigas effect*.

Gigas effect- Polyploidy is the increase of chromosome set than the diploid plants which results in increase in cell size due to which stem becomes thicker,plant size increases, flower,seed and leaves becomes larger than the corresponding diploid plant, is called gigas effect.

Triticals – a hybrid of wheat (*Triticum turgidum*) and rye (*Secale cereale*) is the first man made crop derived by crossing wheat and rye.

is in more than diploid state. These are commonly hexaploids (AAAABB), e.g; *Helianthus tuberosus*

Colchicine drug is used for inducing polyploidy artificially. Fusion of multiple sets of chromosomes (separate gametes) and failure of meiosis results in polyploid species, which when found agriculturely better, are preserved and cu Triticum turgidum ltivated.

Karpechenko-1927- crossed *Raphanus sativus* (2N=18) with *Brassica oleraceae* (2N=18), the intermediate hybrid was called *Raphano brassica*, had all characters between radish and cabbage. The hybrid was sterile and meiosis did not take place because chromosomes were different. Similarly hexaploid wheat and seedless triploid watermelons have been produced.

Thus, triploids, tetraploids, pentaploids, hexaploids, heptaploids and octaploids produced have different chromosome numbers and give more crop and field yield. This process plays an important role in the formation of new species and natural selection besides crop improvement.

6). **Mutational Breeding**: A sudden change or abrupt rearrangement of genes or in number and size of chromosomes causes *mutation*, resulting in enormous variations in plant growth and behavior.

Mutation can be due to a change in;

a). Base sequence of the concerned gene

b). Chromosome structure and number

Mutation can be induced by short-wave electromagnetic radiations,U.V rays, X-rays, α,β,γ-rays etc.

Mutations may be:

A). Spontaneous Mutation

B). Induced Mutations

A). Spontaneous Mutation: The naturally occurring mutations is called as *spontaneous mutation*.

Spontaneous Mutation are both germinal as well as somatic. Somatic mutations can be incorporated in crop improvement only in vegetatively propagated plants e.g;seedless grape, naval orange,Bhaskara banana.

Germline variations got through sexual reproduction can be maintained by vegetative means, e.g; apple, mango, potato, sugarcane.

Nowadays, spontaneous mutations are the source of all the genetic variations occurring in living things.

B). Induced Mutations: Mutations produced in response to mutagens are *induced mutations*.

Induced mutations were first produced by Muller(1927) by X-rays on Drosophila and by Stadler in maize.

Lerma Rozo-64 $\xrightarrow{\gamma\text{-rays}}$ Pusa Lerma

Sonora - 64 $\xrightarrow{\gamma\text{-rays}}$ Sharbati
Sonora

Mutagens are used to

increase the rate of mutations.

Mutagens may be chemical or physical.

i) *Chemical mutagens*: Chemicals used as mutagens, e.g; Ethylmethane sulphonate (EMS),Sodium azide, Nitrous acid,Mustard gas, Caffein.

ii) *Physical mutagens*: Here different kinds of radiations are used as mutagens, e.g; X-rays,alpha,beta, gamma rays, UV rays, Cosmic rays. These mutagens induce changes in DNA and chromosomes.
Mutation breeding : It is the use of induced mutations in plant breeding to develop improved varieties.

Somaclonal Variation

Somaclonal variation is the genetic variation among plant cells during tissue culture. It is used to develop several useful varieties.e.gs; resistance to diseases and pests, stress tolerance, male sterility, early maturation, better yield, better quality etc.Somaclonal variations also proved useful against several diseases such as;

- Wheat tolerance to rust and high temperature
- Rice to leaf ripper and Tungro virus
- Potato to *Phytophthora infestans* (late blight of potato)
  It also leads to high protein content of potato, short duration sugarcane and increase shelf life of tomato.

Many barley varieties were subjected to γ-radiations, which in future generations produced mildew resistance and high yield. A mutant gene that induces semi-dwarfism in rice by X-rays have proved useful. This way, some 45-mutant varieties with high yielding capacities have been produced by *gamma ray treatment*. The crop plants of sugarcane,potato,cotton and fruit crops have increased their breeding quality and disease resistance by *induced radiation* treatment. Sometimes a mutation can be lethal so a plant breeder has to be cautious, if he gets a new beneficial character in a million.

-In microorganisms- various mutant strains have been raised with better fermentation capacity and better yield of commercial *antibiotics.*

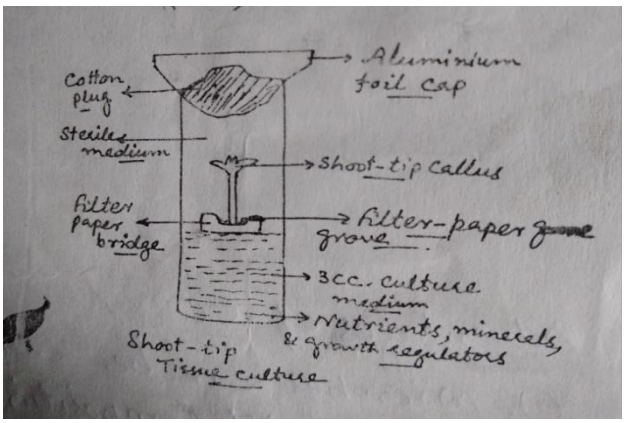
-*Neurospora* mutants have proved helpful in molecular biology of gene action and enzyme function.

-**Indian Agricultural Research Institute** {IARI} New Delhi has a large field shielded by a high wall and having a source of gamma rays using Cobalt-60, referred to *Gamma garden*. Here the mutants are raised by exposing the desired crop to the scattered dosage of gamma rays for a specific period of time.

7). **Tissue Culture**: The technique of tissue culture involves in the isolation of cells, tissues or organs from plant body and growing them especially in the suitable containers on an artificial nutrient medium under controlled environmental conditions.

Methodology/ **Procedure**:The technique of tissue culture was first used by a German botanist **Gottlieb-1902**, then **Gautheret** and **White-1939** and **Steward-1964**, laid its importance in crop improvement.

A culture vessel in complete aseptic condition is filled with nutrients,sugars,vitamins,minerals,growth hormones,IAA,IBA,Gibbrellins,coconut milk factors etc. The complete sterilization of medium is achieved by autoclaving at $120C^0$ for 15 minutes. The ultra- violet radiation are used for killing microbes in this chamber.The culture vessel, test tube is filled with liquid culture medium having a filter paper bridge in the form of M.A terminal shoot apex 0.5mm tissue is placed carefully in filter paper depression. The test tube is plugged with non-absorbant cotton and an aluminum foil is placed over.The entire test tube is placed in a plant growth chamber at $27C^o$ at an intensity of 1000 lux and a photoperiod of 16 hours. A small plantlet is formed from these **totipotent** cells which is transferred to a green house and then in open fields.



Schematic Representation of Tissue Culture (shoot tip)

When one cell or many totipotent cells are cultured,it soon gets organized in to a mass sprouting roots, shoots and embryo-like structures hence called *embryoids*.

Accorrding to **Steward**, from one carrot embryo nearly 100000 embryoids can be obtained by tissue culture method and can grow in to numerous flowering plants,producing viable and desired seeds.

In somatic hybridisation, the leaf cells are taken, their cellulose of cell wall is removed by cellulases and protoplast of two different cells is brought in contact in cultured medium (polythene glycol) then polyploid somatic hybrids are produced.

Tissue culture phenomenon can be utilized for crop improvements by the following methods;

a).**Micropropagation**: It is the vegetative multiplication of the valuable plant material for agriculture, horticulture and forestry.

-Totipotency is the ability of a single cell to divide and produce all of the differentiated cells in an organism e.g; spores and zygotes or Totipotency is the genetic potential of a plant cell to produce the entire plant.

-Test tube forests can be raised through tissue culture technique.

- Maheshwari and Guhu-1966, produced a single celled cultured embryoid from one pollen grain(1N)plant.

b).**Multiple shootlet production**: Technique used to multiply a rare hybrid or a pathogen free rare plant or a sterile plant with valuable features or for getting plants with only one sex.

c). **Somatic embryogenesis**: Differentiation of somatic tissues to produce artificial embryos is called *embryoid differentiation* and embryos as *embryoids*.Embryoids can be produced from the callus, raised from the somatic cells which finally develop in to a complete plant. Production of somatic hybrids help in making new useful polyploid varieties with better yield and bettely adapted.

d).**Production of disease-free plants**: The healthy shoot tips of infected plants, if grown in culture media, can produce healthy and disease free plants.

e). **Androgenic haploids**: Androgenic haploids, as successful hybrids have been produced by tissue culture process which has proved an important variety. Haploid plants are always pure because they possess only one set of

chromosomes. Moreover, the mutations are expressed very easily in haploid plants as compared to diploid plants.

f).**Embryo Rescue for successful Hybridisation**: It deals with culturing of young embryo, fertilized ovule or pollinated ovary to protect the development of embryo. It is used in case of Interspecific or intergenic crossed where embryos abort at an early stage of development and mature seeds are not formed. In such cases, the immature hybrid embryos are excised and cultured on a nutrient medium.

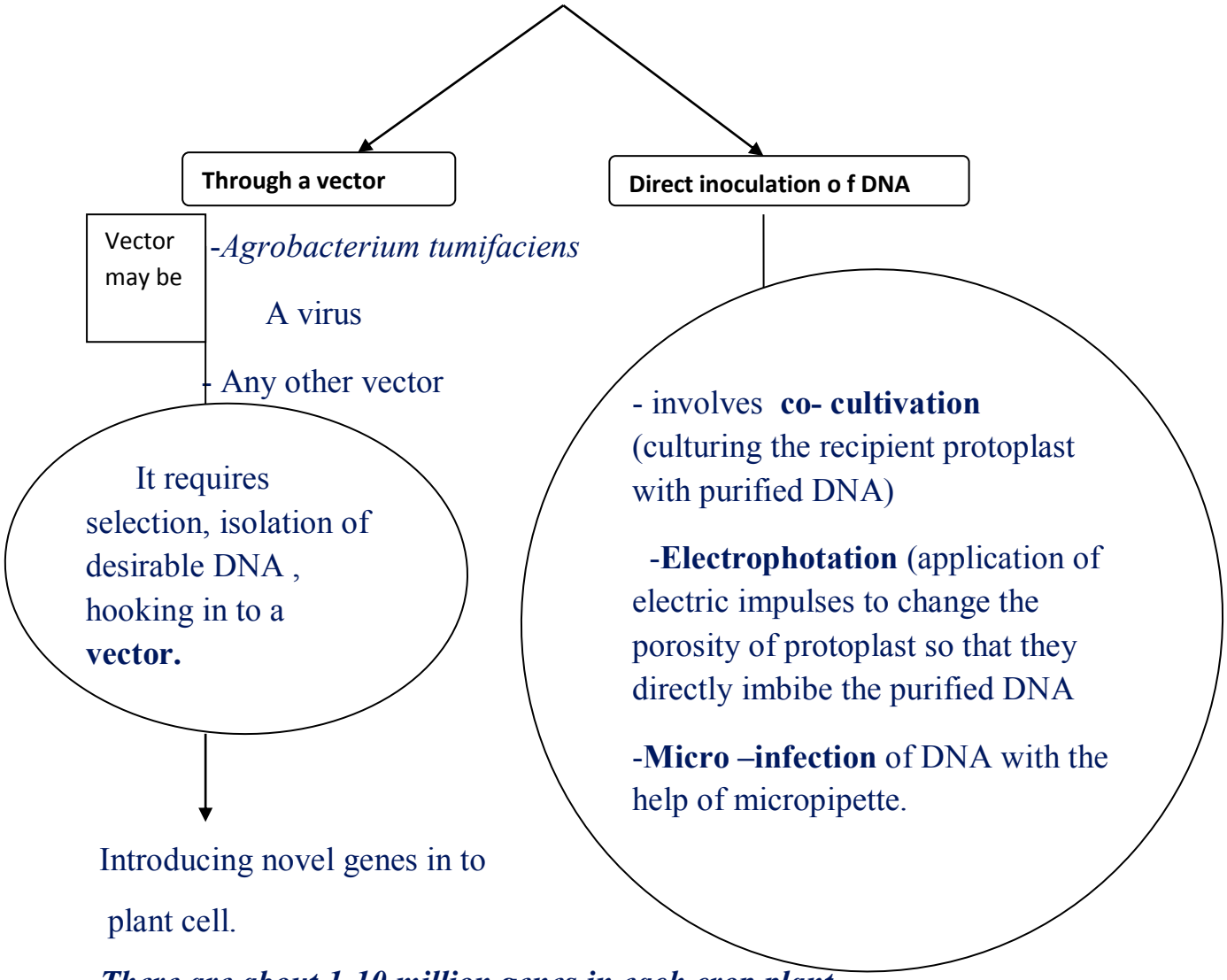
vii). **Protoplast Technology**: The isolated protoplasts are purified and then tested for their viability. The viable protoplasts are cultured in liquid medium or on a solid medium. Within 2-4days, cell wall develops and it divides to form colonies. Each colony of cells may develop in to callus or somatic embryo. The naked protoplasts isolated from different plant cells have been used by plant breeders for various genetic manipulations.

8). **Genetic Engineering**: Genetic engineering is the method of changing the phenotype according to the will by the removal, repair or addition of the genetic material with regard to the requirement of that organism. OR

The *recombinant DNA technology* or *genetic engineering* involves transfer of one or more genes (i.e; re DNA fragment) from one plant to another. The plant in which a foreign gene has been introduced is called *transgenic plant*.

-Some methods of gene transfer are *Liposome mediated gene transfer*, *Ca3(PO4) precipitation method*, *Trnsformation by ultrasonication* and *transformation* using pollen/pollen tube.

Foreign gene (DNA) introducing technique



There are about 1-10 million genes in each crop plant.

Some examples of genetic engineering / recombinant DNA technology are as;

In plasmid engineering, the insulin gene of rats is isolated and then fixed in the plasmid ring of *E.coli*, so that bacterial progeny induce numerous insulin genes. These insulin producing genes can be supply for Diabetic patients, if found viable. Nowadays, human insulin is cloned in *E.coli* which is highly useful and efficient in action.

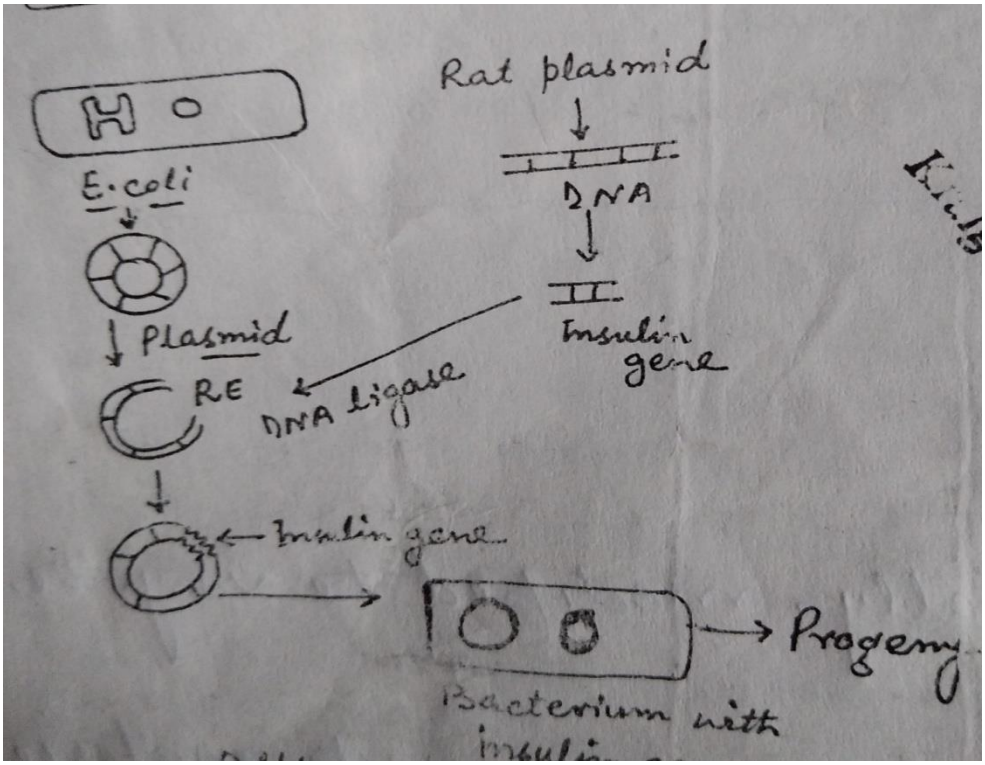
Similarly, the lac genes of *E.coli* (genes concerned with localization of lactose) have been successfully transferred to the haploid cells of Arabidopsis and tomato using lambda (ʎ) bacteriophages by the process of transduction.

Various variations and antibiotics synthesized by bacteria may be transduced to higher plants through genetic engineering devices. The nitrogen fixing genes of *Rhizobium leguminosarum* have been found possible to transfer into non-nitrogen

fixing bacteria, their aim is to transfer these genes to crop plants like rice, maize, wheat etc; so that crop yield and quality is improved.

An intresting success in the field of genetic engineering has been made by **Dr A.M.Chakarvarthy** from Calcutta (working in USA) developing a new type of bacterium *Pseudomonas*, which can feed on hydrocarbons, so can be used for cleaning accidental oil leakages on ocean surfaces.

Human growth hormone (**HGH**) could be synthesized and cloned in *E.coli*, where it is expressed under the control of a lac-promoter. During viral infection, human immune cells (leucocytes) produce a substance called **interferone**,that gives immunity or resistance against that particular virus. Bacteria have now been used to produce interferons with its specific coded DNA by genetic engineering devices. Curing of certain diseases by genetic engineering is called **Medical engineering** or **Euphenices**.



Different events of plasmid engineering

Role of Genetic Engineering in the crop improvement:

i). **Herbicide tolerance**: Through genetic engineering many herbicide tolerant plants have been produced,e.g; tomato, tobaco,potato,soyabean etc.

ii). **Insect / pest resistance**: Through transgenic technique many insect and pest resistant plants have been developed, e.g; tomato, cotton, tobaco etc.

iii). **Resistance against stress**: Many stress (heat, cold, salt, heavy metals etc.) resistant genes are manipulated to produce stress resistant crop plants.

iv). **Plants suitable for food processing**: Genetic engineering is helpful in producing tomatoes with delayed ripening, mangoes with less ethylene production and potatoes with 20-40% more starch content.

v). **Male sterility**: Transgenic plants with male sterility are helpful in production of hybrid seeds without manual emasculation and controlled pollination, e.g; *Brassica napus*.

Benefits of crop improvement

(As amelioration to human problems)

Modern man has brought more and more land under cultivation. The irrigation of this land has been enhanced by making canals, dams and streams. The development and extension of high yielding varieties in farms has proved extensively useful. The production of hybrids, polyploids, disease-resistant and pest-resistant varieties of cereals, pulses, millets, sugar and cotton etc. have proved useful in raising quality and quantity of these crops. For instance, cereal crops like rice in 1960 had 700kg per hector yield which has now become more than 2400 kg per hecter.Similarly wheat production has increased four times more than what was reported in 1960 in India. The same is case with sugarcane, oil seeds, millets, pulses etc. The mechanized agriculture with the use of tractors, helicopters, spring machines etc; has provided base for increased crop yield to an extent so that India has now become self sufficient in its food grain consumption and vegetable growth. This process can rightly be called as "green revolution". The use of fertilizers has been exploited by man in a scientific manner, which has added to soil fertility and increased crop yield.

Some chemical fertilizers added to the soil in the form of Potassium chloride, Pot. Sulphate, Ammonium sulphate, Calcium nitrate, Sodium nitrate, Pot. Nitrate, Urea, Diammonium phosphate, Calcium phosphate etc. to increase crop yield and soil fertility.

Green manures, vegetable garbages, farmyard manure, decomposed animal dung, decomposed plant remaining, humus, human faeces, nitrogen fixing algae and bacteria can increase nitrogen fixation and crop yield.

Role/ Advantages/Achievements of Plant Breeding

Some important adventages of plant breeding are as;

a). Development of *Triticale* from *Triticum turgidum* (wheat) and *Secale cereal* (rye).

b). Development of Shakti, Rattan, Protina - Lysine- rich maize varieties.

c). Development and introduction of disease resistant varieties.

d). Development of Interspecific hybrid varieties of sugarcane.

e). Several other varieties of crops like Sonara-64 (wheat), Taichung (rice) etc. are also developed by hybridisation.

Single Cell Protein (SCP)

The cells from microorganisms such as bacteria,yeasts,filamentous algae, treated in various ways and used as food, are called as single cell protein.

Thus, single cell protein is produced by using bacteria, algae and fungi. For the production of SCP the substrates used range from CO_2 through industry effluents like whey (water of curd) to low cast organic materials like saw dust and paddy straw.

Commercial production of SCP is mostly based on yeasts and some other fungi like Fusarium gramiearum.

It has been estimated that a 250kg cow produces 200g of protein per day where as 250g of Methylophilus methylotrophus(microorganism) produces 25 tonnes of protein per day.

Some common microbes used as SCP producers are as ;

i).Cyanobacteria- *Spirulina*

ii).Bacteria- *Methylophilus methylotrophus*

iii).Yeasts- *Candida utilis*

iv).Filaments fungi- *Fusarium gramiearum*

Adventages of SCP:

a).Microbes can be easily genetically modified to vary the aminoacid composition.

b).Large variety of raw materials, including waste materials can be used as a substrate, thus, minimize the number of pollution.

c).Its production is independent of climatic conditions.

d).Some microorganisms are used as human foods SCP technique such as blue-green algae-*Spirulina* and mushrooms-fungi.

e).Fermented foods- cheese, butter, idlis etc; are also produced.

f). It is rich in high quality protein and poor in fat.

g). Cost of production is very less as compared to the amount of protein produced.

Biofortification

It is the breeding of crops to increase their nutritional value. Or

It is a process to increase the bioavailability and the concentration of nutrients in crops. Or

The process of breeding staple crops to have higher levels of essential nutrients.e.gs; Biofortification of wheat with zinc, golden rice.

Biofortification can be done through conventional breeding and recombinant DNA technology (genetic engineering).

Biofortification differs from fortification because it focuses on making plant foods more nutritious as the plants are growing, rather than having nutrients added to the foods when they are being processed.

Objectives of Biofortification

The main objective of biofortification is to develop micronutrients dense staple crops to achieve provitamins A, iron and zinc concentrations that can have a measurable impact on nutritional status.

Advantages of Biofortification

a). It adds the nutrients to foods particularly staple foods so increase intakes among most of the population.

b). It helps to reduce nutrient deficiency diseases. e.g; the addition of iodine to salt to decrease iodine deficiency disorders.

Test your Understanding

What is plant breeding? Mention the different steps of plant breeding.

1. What is heterosis? What role has it played in crop productivity?
2. Define hybridisation? What are intravarietal, Intervarietal, Interspecific and Intergeneric hybridisations?
3. Differentiate: (a). Spontaneous and Induced mutations
   (b). Chemical and physical mutagens
4. Define recombinant DNA technology. Write down its role in the plant breeding?
5. What is a plant breeder, hybrid, gamma garden, totiopotency, SCP and biofortification.
6. Write a note on plasmid engineering and emasculation.

MCQs

i). Nowadays, the quicker method of plant breeding is:

(a). Introduction (b). Selection (c). Hybridisation (d). Mutation breeding

ii). Homozygosity of characters can be achieved by:

a) Mass selection (b). Hybridisation (c). Inbreeding (d). Pure line selection

iii).The removal of anthers from an intersexual flower is commonly known as :

(a). Cytosterility (b). out breeding (c). Emasculation (d). Induced mutation

iv). Male sterility has been largely studied in:

(a). wheat and maize (b). maize and sunflower (c). tomatoes and potatoes (d).sugarcane

v). Sonora and Lerma Rojo are variations of:

(a). Barley(b). Sugarcane (c). Rice (d). Wheat

vi). Sonora-64 is an example of:

a). Polyploidy b). Mutation c). Hybridisation d). Quarantine

vii). Which part of the plant should be used to produce disease free potato:

(a). Seeds (b). Tubers (c). Shoot apices (d). Phloem

viii). Haploids are of great importance in crop improvement programmes because they:

(a). Are useful in meiotic studies (b). Grow better under stress conditions

(c). Give homozygous lines after diploidisation (d). Do not express mutations induced in them

ix). Somaclonal variations are those which appear during:

(a). Hybridisatio b). Induction of polyploidy (c). Tissue culture

(a). Emasculation (c). Parasexual hybridisation

(d). Inbreeding

xi). The first man made crop triticale has evolved by Intergeneric crossing between:

(a). wheat and mustard (b). wheat and rye (c). maize and rice 9d). wheat and rice

xii). Hybrid vigour is due to:

(a). chiasma (b). linkage (c). crossing over (d).heterozygosity

xiii). Genetic variability can be induced by:

(a). Mass selection (b). Clonal selection (c). Back crossing (d). Mutation

xiv). Father of green revolution is:

(a). A. Howard (b). N.J.Borlaug (c). M.S. Swaminathan (d). R. Rhoades

x). Saffron is obtained from:

(a). Seeds (b). Style and stigma (c). Petals (d).Roos

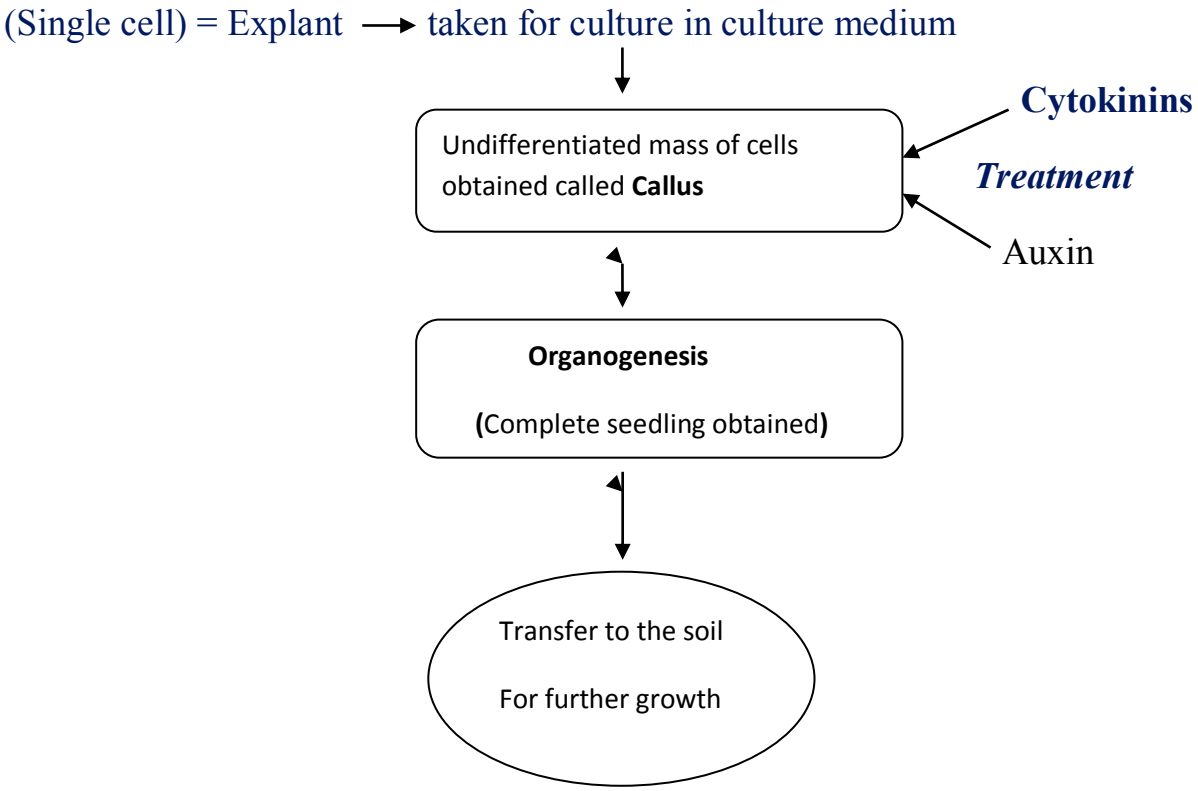
Tissue Culture

Tissue culture: Cellular totipotency, technique and application of tissue culture

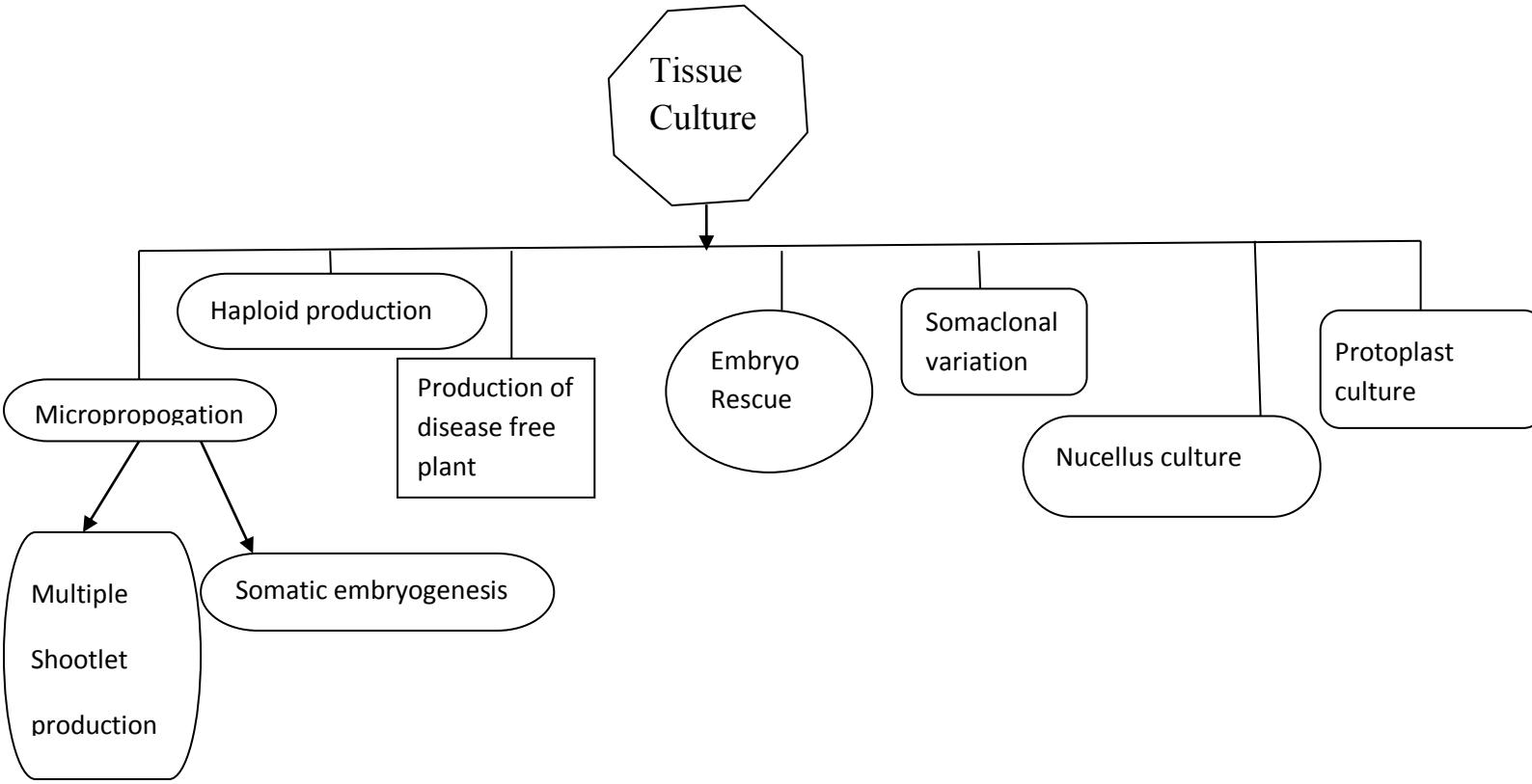
Tissue culture is the latest method of crop improvement, based on totipotent nature of plant cell (Totipotency), i.e; each cell has capacity to develop in to complete plant.

The concept of Totipotency was given by Haberlandt-1902 but the practical application was shown by Steward in 1932.

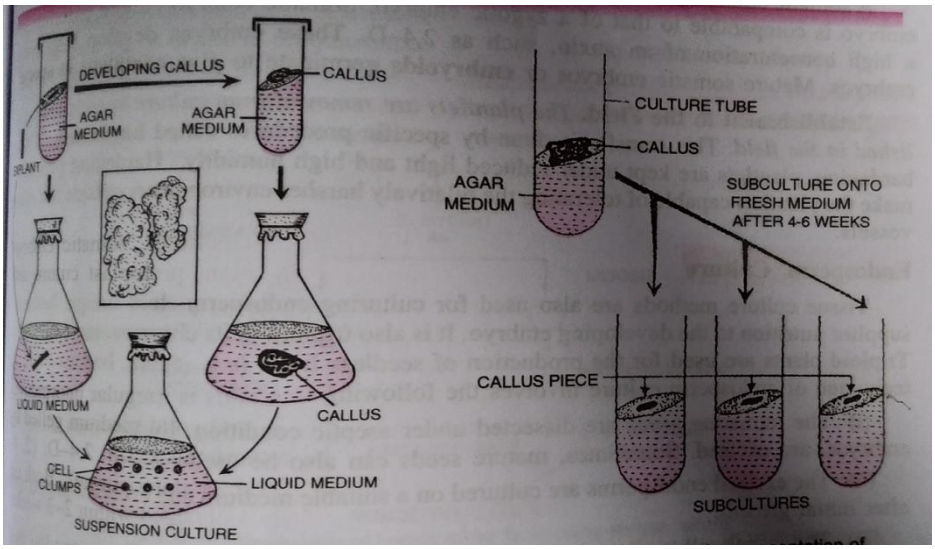
In tissue culture technique a single cell or group of cells taken for culture is called ***explants.***



Adventages of Tissue Culture: Go through the given chart



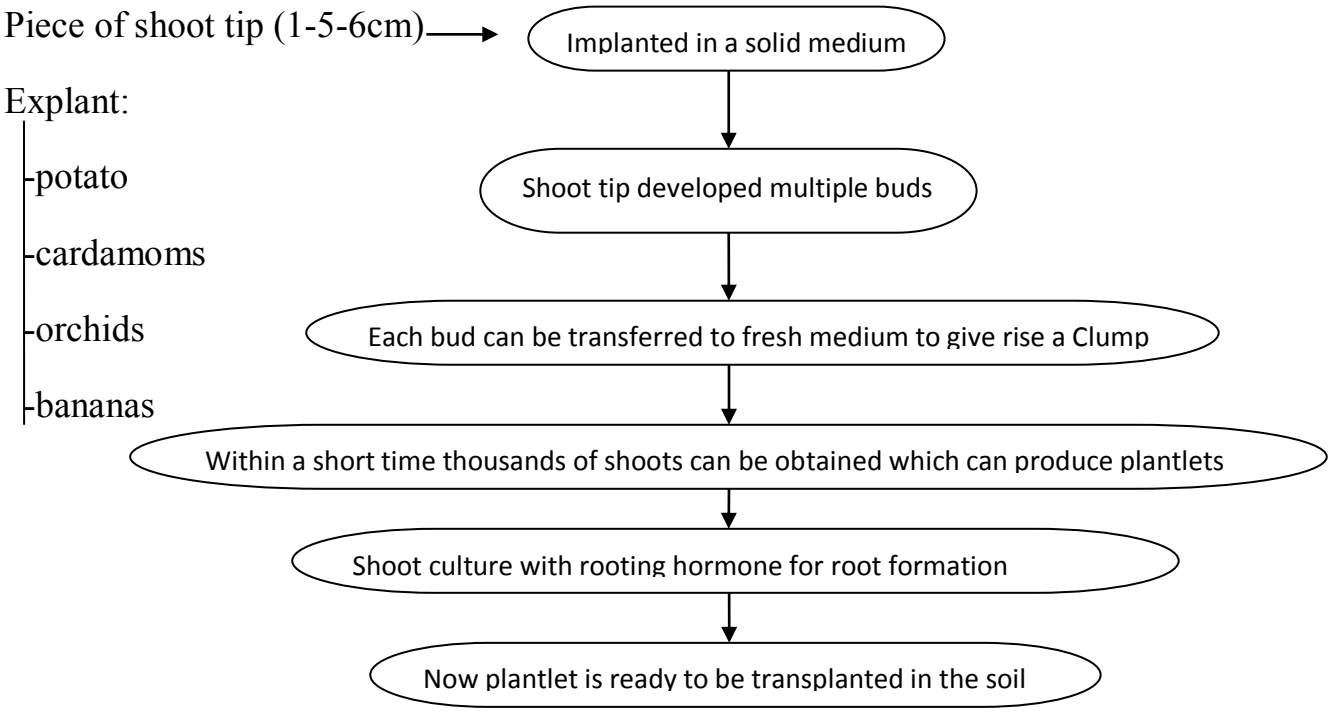
(Callus and suspension cultures) (Subculturing)



A). **Micropropagation** (*Cloning*): It is a technique to produce strong and healthy plantlets by rapid vegetative multiplication under controlled condition. It may be;

(i). **Multiple shoot production** – It is used to multiply a rare hybrid or pathogen free plants or a sterile plant with valuable feature or for getting plants with only one sex.

Multiple shoot production can be done by the following methodology;



Use of Meristem Culture:

1. For rapid clonal multiplication
2. For production of virus free plants
3. For germplasm conservation
4. For production of transgenic plants

(ii). **Somatic Embryogenesis**: Here embryoids are produced by the following method;

Somatic tissues $\xrightarrow{\text{Differentiation}}$ Artificial embryos or embryoids

Tissue culture ⟶ Somatic cells ⟶ Callus ⟶ Embryoid formation

(Globular ⟶ Heart shaped ⟶ Torpedo shaped stages)

Embryoids remain attached with callus but separate on shaking in liquid medium. Now floating embryoids separate and grown separately with a tap root in culture tubes. e.g; Somatic embryos in carrot, celery and alfa-alfa.

B). **Haploid production**: In this tissue culture technique the haploids are produced from immature pollens or microspores (male gametophytic cells) . These pollens are cultured on a solid or liquid medium, then the callus / embryo formed is transferred to a suitable medium to finally produce a haploid plant and then a diploid plant (on colchicine treatment).

Pollen culture is first carried out by **Guha** and **Maheshwari** on *Datura innoxia*/ **Jimson weed**.

Sometimes diploid plants are formed among haploid plants and source of these diploid plants is **anther wall** (2n).

Androgenic haploid and their use in breeding:

Self pollination plant ⟶ Production of homozygous (pure line).

Haploid plants are always pure (having one set of chromosome). They are always required for cross breeding and improvement of crop. Mutations can be detected in haploids as mutations are expressed easily in haploids as compared to diploid plants.

Anther Culture is used for the production of haploids in more than 250 plant species.e.g; wheat, rice, tomato, asparagus etc. The methodology of anther culture is as follows;

Advantages of Haploids in Crop improvement:

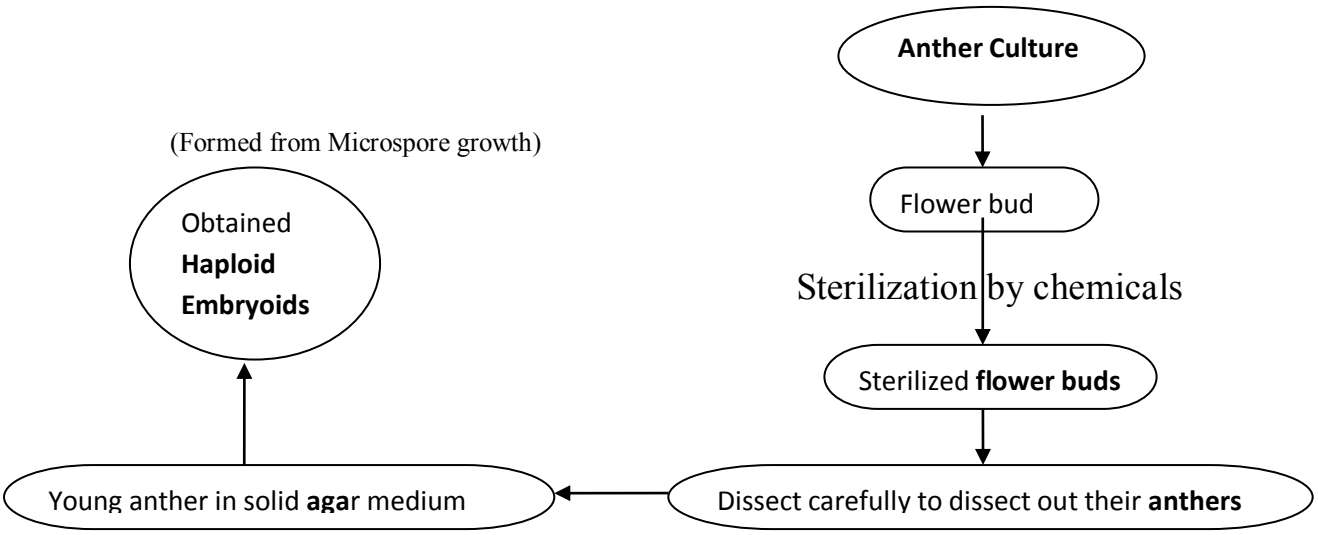
Various advantages of haploids in crop improvement are as;

i). Induction of mutations

ii). Leads to hybridisation

iii).Production of homozygous pure line by doubling of chromosome number.

iv). Production of diploids by Colchicine treatment. e.g; In China varieties of rice, wheat, maize, rubber are developed from haploids.



Flow chart of Anther culture

C). **Production of disease free plants**: By shoot meristem culture as shoot apical meristem is free from pathogen, specially viruses, e.g; potato, sugarcane, sweet potato, dahlia.

D). **Embryo rescue for successful hybridisation**: This technique is used to assist in the development of plant embryos that might not survive to become viable plants. This technique nurtures the immature or weak embryo, thus allowing it the chance to survive.

Here embryo in a seed abort at an early stage thus seed does not reach maturity. Such an embryo can be rescued or saved if taken out from the seed and then cultured.

Significance: (a). It is used in Interspecific hybridisation.

(b). Embryo rescue had been of great value in growing embryo from hybrids. E.g; cross between common bean (*Phaseolus vulgaris*) and its wild species (*Phaseolus angustissinus*).

(c). Hybrids developed by this technique was disease tolerable and had other desirable characters.

E). **Somaclonal variation**: Somaclonal variation is the genetic variation among plant cells during tissue culture. It is used to develop several useful varieties.e.gs; resistance to diseases and pests, stress tolerance, male sterility, early maturation, better yield, better quality etc.

Advantages: (a). Such variations are stable and have agronomic character (pest and disease resistance).

(b). Such variations causes rust resistance and high temperature tolerance (wheat) and increase shelf life (tomato).

F). **Nucellus Culture**: In nucellus culture embryos are derived from the Nucellus or integument. Such cultures are taken from pollinated flowers and widely reported in the genus of citrus.

Importance: (a). By this culture we can obtain plants of parental type.

(b). Such plants are free from microorganisms – bacteria, virus.

(c). These plants can maintain their characters for a long period.

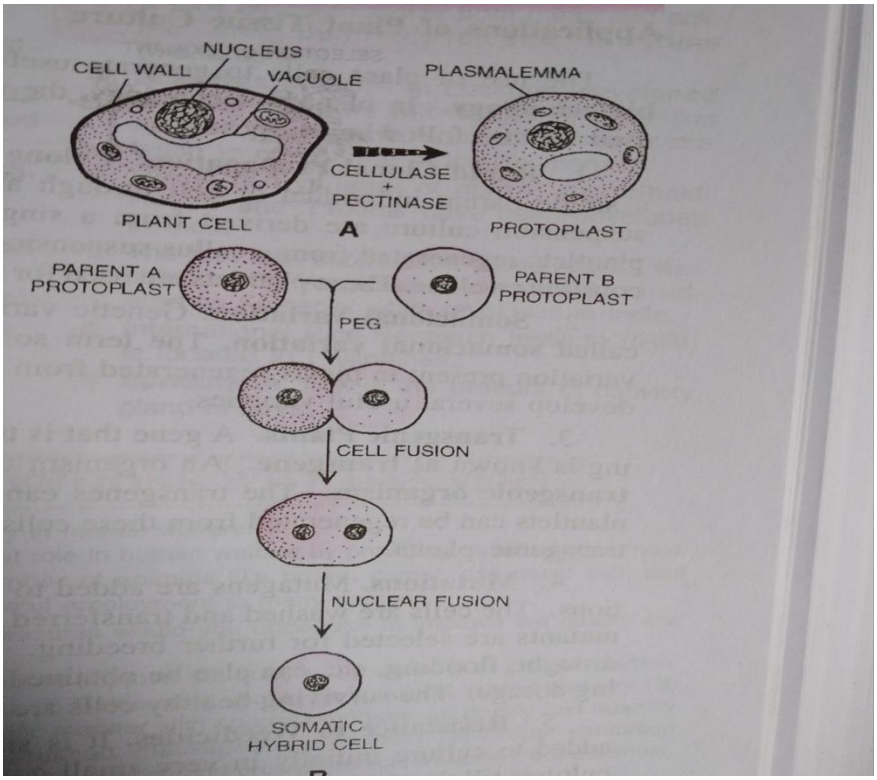
G). **Protoplast Culture**: In this culture, the aseptic isolation of large number of intact living protoplasts removing their cell wall and cultures them on a suitable nutrient medium for their requisite growth and development. Protoplast can be isolated from varieties of plant tissues.

Uses: several uses of protoplast culture are as;

Protoplast Culture

Somatic Hybridisation ↙ ↘ Genetic Engineering

- By PEG (PolyEthylene Glycol) - Due to fragile nature of protoplast.
- Separation of protoplast by treating cells with Pectinase and Cellulase enzymes - E.g; POMATO obtained by somatic hybridisation between tomato and potato (intergenic hybrid). - Certain bacteria, portion of DNA and Nitrogen fixation gene (NIF genes) can be easily incorporated.
- Bomato = hybrid between Brinjal and Tomato



SOMATIC HYBRIDISATION

A. PRODUCTION OF PROTOPLASTS USING

Parasexual Hybridisation: It is a non-sexual mechanism of transferring genetic material by fusion of cells of different characteristics. Protoplast fusion technique is used for hybridisation.

- Protoplast fusion or somatic hybridisation become in use when sexual reproduction is absent.

- Cell suspension preparation preparation with the help of Pectinase enzymes.
- Protoplast can be induced to fuse by exposure to PEG or by Electrofusion carried out in a fusion cell (at low voltage between two electrodes causes protoplast to clump followed by coalescence of cytoplasm of two protoplast).
- Fusion of protoplast of same genotype - fused cell called **Homokaryocytes.**
- Cells contain non- identical nuclei called **Heterokaryocytes.**
- If 2 nuclei of heterokaryocyte fuse, a true hybrid protoplast Synkaryocyte is formed.*POMATO (Hybrid of potato and tomato).
- First somatic hybrid were obtained by somatic hybridization between *Nicotiana glauca* and Nicotiana *longsdorfit* by **CARLSON** (1972) et al.
- Somatic hybridisation is used for producing hybrids of those species which cannot be sexually hybridized.
- Somatic hybrids are used for;
- Gene transfer
- Cytoplasm transfer
- Production of useful Allopolyploids.
- **Subculturing**: If tissue cultures are kept in the same culture vessel, they die in due course of time. Therefore, cells/ tissues are regularly transferred in to new culture vessels containing fresh media. This process is called **subculturing**. It is important to note that during subculture, only a part of the culture from a vessel is transferred in to the new culture vessel.

Single Cell Culture [Cell Cloning]

Single cells can be isolated either from suspension culture or directly from explants by mechanical or enzymatic methods.

(i). **Mechanical method**: In this method the leaf pieces (other plant parts) are grind in a pestle and morter so making a uniform homogenous culture medium. It is then filtered through muslin cloth.The filtrate is centrifuged to obtain free cells in the pellet.

(ii). **Enzymatic method**: It involves the treatment of macrozyme (0.5%) or Pectinase with the plant tissues. In this method, the epidermal peels are removed from the leaf surfaces. The pieces of leaves (mesophyll) are incubated in a

mixture of enzyme solution, culture solution and 0.3 M mannitol. The enzyme breaks down middle lamella and causes maceration, in this way the cells are separated.

Single cells obtained through these methods can be cultured in liquid or semi-liquid medium.

The two popular techniques for single cell culture are as;

A). Filter paper raft – nurse tissue technique

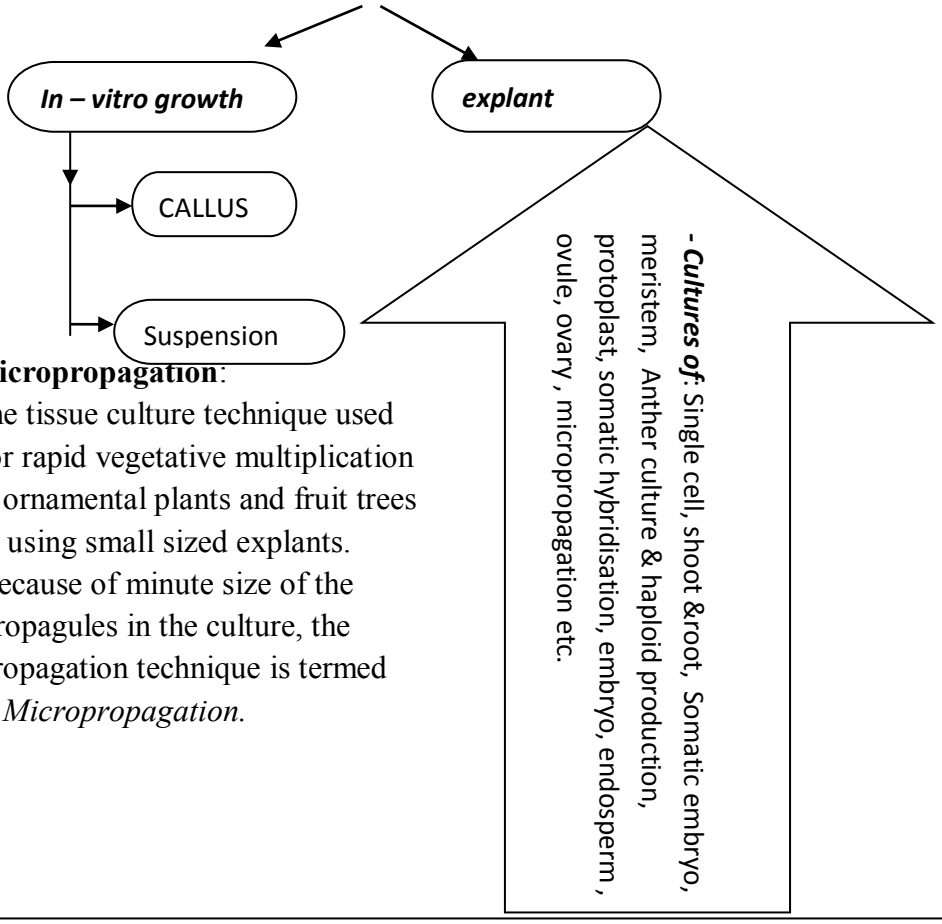
B). Bergman's plating technique

A). **Filter paper raft – nurse tissue technique**: Here callus is allowed to grow on a semi- sold medium in a petri dish. After having moderate growth, a circular filter paper is placed over the callus culture. The filter paper becomes wet by the liquid exudate of callus tissue . A liquid suspension containing single cells is poured over filter paper, close the disc and allow the culture to grow inside a culture chamber. The cells divide and develop into macroscopic colonies on the filter paper which are later on sub- cultured on a fresh medium.

B). **Bergman's plating technique**: It is commonly used technique. Here cells are suspended in a liquid medium at a cell density that is twice the desired density in the plate. Sterilized agar (Ca 1%) medium is kept malted in a water bath at 35^{o}C. Equal volumes of the liquid and agar media are mixed and spread in Ca 1% mm thick layer in a petridish.The cells remain embedded in the soft agar medium which are observable under a microscope. The large colonies developed are isolated are cultured separately.

Significance: Single cell cultures can be used for induction of somatic embryos, regeneration of roots, shoots and flower buds, in – vitro mutagenesis and mutant selection , genetic transformation and production of secondary metabolities.

Methods of Plant Tissue Culture



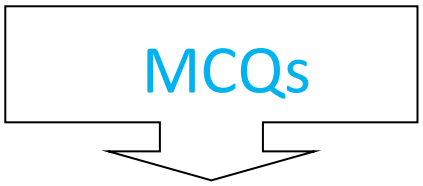
Micropropagation:
The tissue culture technique used for rapid vegetative multiplication of ornamental plants and fruit trees by using small sized explants. Because of minute size of the Propagules in the culture, the propagation technique is termed as *Micropropagation.*

Gene bank: The institution where seeds and vegetative materials of those plants which are likely to become irretrievably lost in the wild or in cultivation are conveniently stored and maintained.

Artificial Seeds: Plants which either do not bear seeds or produce small quantity of seeds propagate generally by vegetative means. Now biotechnologists provided a technique through which artificial seeds of such plants may be produced.The process involves artificial somatic embryos which are encapsulated in gels like Na-alginate, Ca- alginate, polyacralamide gel etc.which prevent them from desiccation. These are water soluble gels and have been found suitable for making artificial seeds.

Test Your Understanding

1. Define Tissue culture. Write down its various advetages.
2. What is Micropropagation? Write down its methodology.
3. Mention different types of Micropropagation.
4. Writedown the applications of haploids in crop improvement.
5. What is an explant?
6. What is Parasexual hybridisation?
7. What is POMATO?



1) Colchicine is employed to diploid or haploid cells, as it:
(a). Inhibits formation of centromere (b). Inhibits formation of spindle formation
(c). Inhibits mitosis or meiosis (d). Allows DNA replication twice in one cell cycle

2). In order to obtain virus-free plants through tissue culture the best method is:

(a). Embryo rescue (b). Anther culture (c). Meristem culture (d). Protoplast culture

3). Two microbes found to be very useful in genetic engineering are:

(a). *Vibrio cholera* and tailed bacteriophages (b). *Diplococcus* sp. And *Pseudomonas* sp.

(c). Crown gall bacterium and *Caenorhabditis elegans* (d). *Escherichia coli* and *Agrobacterium tumefaciens.*

4). Somaclonal variation is seen in :

(a). Tissue culture grown plants (b). Apomicts (c). Polyploids (d). Vegetatively propagated plants

5). The technique of obtaining large number of plantlets by tissue culture method is called:

(a). Plantlet culture (b). Organ culture (c). Micropropagation (d). All of the above.

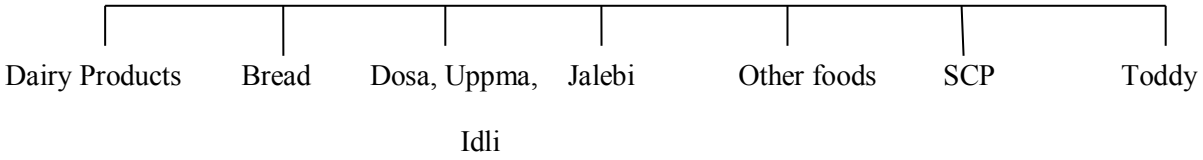
Microbes in Human Welfare

In household food processing, industrial production, sewage treatment, production of energy (Biogas), biocontrol agent (Biopesticides) and Biofertilizers.

In household food processing

Various food products prepared at homes by the help of microbes are as;

Microbes in Household Products



1) **Dairy Products**: Several dairy products (curd, yoghurt, cheese) are prepared at home from milk using Lactic acid bacteria (LAB). Lactobacillus bacteria are added to milk which converts lactose sugar of milk into lactic acid.Lactic acid causes coagulation and partial digestion of milk protein casein. Milk is changed in to curd, yoghurt and cheese. In this methodology, the inoculums used in preparation of milk products actually contain millions of Lactic acid bacteria.

a). **Curd:** Curd is prepared by inoculating skimmed and cream milk with *Lactobacillus acidophilus* at a temperature of ± 40°C . It is more nutritious than milk as it contains a number of organic acids and vitamins (B_{12}). Curd is eaten as such, salted or sweetened or churned to prepare lassi.It is also used to obtain butter and butter milk.

b). **Yoghurt**: Yoghurt is produced by curdling milk with the help of *Streptococcus thermophilus* and *Lactobacillus bulgaricus*, temp; is

maintained about (40 – 46°C) for four hours. It is often sweetened and mixed with fruit.

c). **Butter Milk**: It is formed by inoculating skimmed milk with culture of *Streptococcus cremoris*. *S. lactis*, *Lactobacillus acidophils*, *Leuconostoc* species at 22°C for 18 hours. Acidulated liquid left after churning of butter from curd is also called butter milk.

d). **Sour Cream**: Here the cream obtained by churning of milk is inoculated with *Streptococcus lactis* for producimg lactic acid and *Leuconostoc cremoris* for imparting the characteristic flavour.

e). **Cheese**: Cheese is one of the oldest milk products prepared with the help of microbes. Here curd is separated from liquid part or whey to form cheese.

Steps of Cheese manufacturing:

i) To milk, *Streptococcus lactis* or *Streptococcus cremoris* bacteria are added.

ii) Milk becomes acidic by bacteria.

iii) Rennin enzyme is added for curdling of milk.

iv) Now Whey liquid (93% water, 5% Lactose) is separated by removing curd.

v) Lactose of Whey is used for making Lactic acid (Ist fermented acid).

vi) Cheese at this stage called as Cottage Cheese (Unripe stage).

vii) Cottage cheese + salt put into frames and pressed so as to allow removal of Whey.

viii) The frames are removed as soon as the cheese maintains its shape.

ix) The ripening period is 1 – 16 months.

x) This ripened cheese (hard) contains about 20-30% fat, 20-35% protein and small amount of minerals and vitamins.

xi) Cheese prepared at home with the help of lemon juice is called **Raw Cheese.**

Depending upon water content, cheese is of three types such as;

(a). **Soft cheese** – 50-80% water

(b). **Semihard cheese** – 45% water

(c). **Hard cheese** – less than 40% water

Cheese	Microorganisms used	Reaction
1). **Soft**		
A. Camembert	*Pencillium camemberti*	Ripened by action of
	Brevibacterium	microorganisms on the
B). Limburger	*Streptococcus liquifaciens*	surface of curd.
	Brevibacterium	
2). **Semi-hard**		
A. Roquefort	*Pencillium roqueforti*	Combination of surface
B). Blue		and interior growths.
3). **Hard**		
A). Swiss	*Propionibacterium* sp.	Inoculating the organisms
B). Cheddar	*Geotrichum*	throughout the curd.

2). **Bread**: Here the powder or cakes of selected strains of Baker's Yeast, *Saccharomyces cerevisiae* is added to wheat flour. The kneaded flour is kept at a warm temperature for a few hours. It swells up, (**Leavening**). Leavening is caused by secretion of enzymes by yeast such as amylase, maltase and zymase. **Amylases** break down small quantity of starch in to maltose sugar. **Maltase** converts maltose in to glucose. Glucose is acted upon by **zymase**, which is a complex of several enzymes of respiration so causes fermentation. As a result of fermentation, glucose forms ethylalcohol and carbondioxide which cause swelling or leavening of the dough. The dough is backed whereas ethylalcohol and carbondioxide evaporates making the bread porous and soft.

3). **Dosa, Uppma** and **Idli**: These are fermentation preparation of rice and black gram (Urad). The two are allowed to ferment for 3-12 hours with air borne *Leuconostoc* and Streptococcus species of bacteria.Here the puffing up of the dough is caused due to Co_2.

4). **Jalebi**: Here the semi-liquid dough of fine flour of wheat is fermented and then fried in the form of coils and dipped in sugar syrup to get **Jalebi**. Same preparation is of **Imriti** from black gram flour.

5). **Other foods**: Several other foods prepared as a result of fermentation are as;

- Temph (Indonesia), Tofu (Japan), Sufu (China) ⟶ Soyabean
- Bamboo Vegetable & other products ⟶ Bamboo shoots
- Sausages ⟶ Fish and Meat
- Sauerkraut ⟶ Cabbage

6). **SCP** (single cell protein): SCP is the production of microbial biomass as supplementary food for humans and animals. *Spirulina*, Yeast and *Fusarium graminearum* are some common SCP. It is rich in high quality protein vitamins and minerals but poor in fat. As oftenly grown on organic wastes, SCP also reduces the environmental pollution.

7). **Toddy**: Toddy is a traditional drink of some parts of South India made by fermentation of sap of palms. Its common source is tapping of unopened spadices of coconut. If left for a few hours, it under goes fermentation with the help of naturally occurring yeast, it form beverage containing about 6% alcohol. After 24 hours toddy becomes unpalatable and can be used for producing vinegar.

Probiotic Bacteria – *Consumption of fermented milk products introduce special kind of microbes in our digestive system,such live microorganisms are called* ***Probiotics. e.g;Lactic acid bacteria(LAB),Bifidobacteria, Saccharomyces*** *etc;*

Probiotics influence the intestinal microflora and bring about desirable alternations in food quality.

Lactobacillus*- immune enhancement, prevents intestinal disorders and reduces viral diarrhea.*

Bifidobacteria*-(Produced from the fermentation of carbohydrates, acetic acid and lactic acid)= increase acidity, reduce colonies of disease causing bacteria by producing toxic metabolites.*

Saccharomyces boulardii *prevents antibiotic-induced diarrhea and other digestive*

Dairy Products ⟶ curd, cheese, lassi, buttermilk, shrikhand etc;

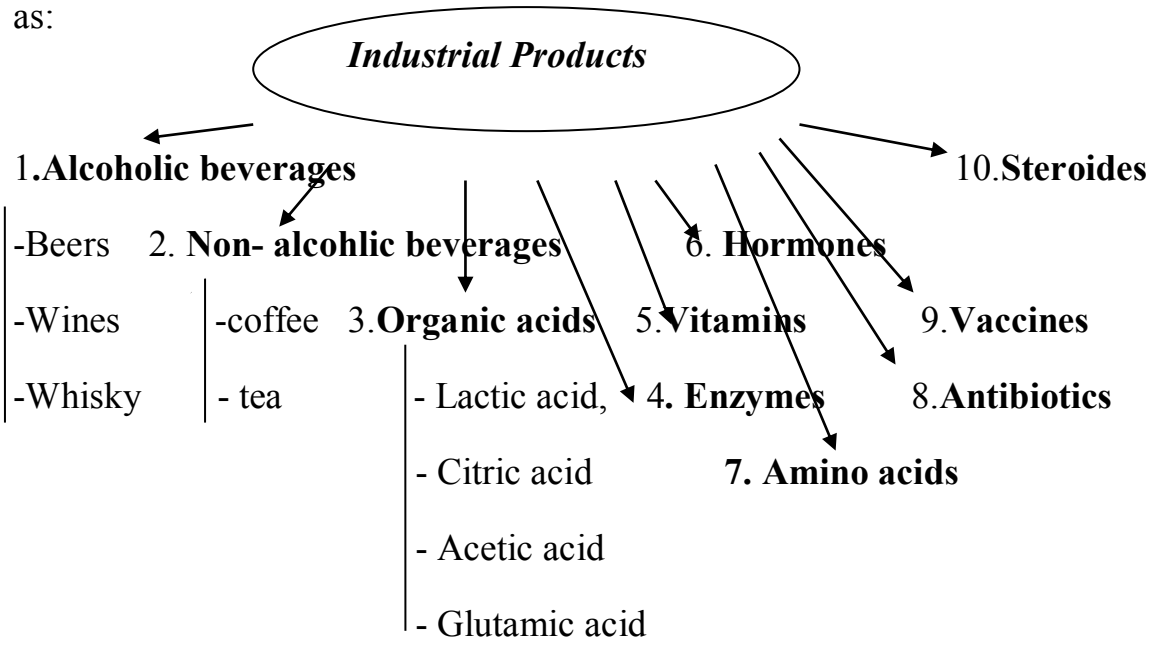
Fermented foods ⟶ idli, dosa, dhokla, khaman, bhatura, kulcha etc;

- reduction of blood cholesterol, BP, prevention of alcohol – induced liver damage, promotion of the health of the gastrointestinal tract, prevention of constipation, promotion of bone density by absorption of calcium and magnesium from the intestine etc;

m

Industrial Production

A number of powerful industries based on beneficial activities of microbes have been set up in recent years to produce a variety of products. Some of these are as:



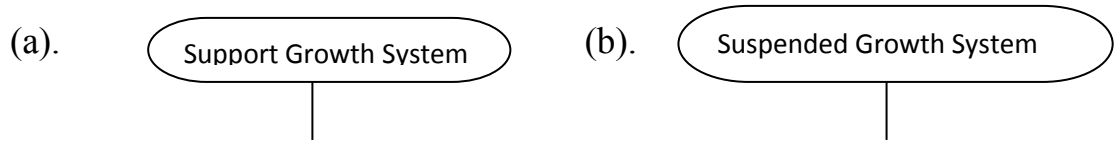
Methodology: Industrial utilization of microbial activity (Fermentation) involves three processes such as laboratory scale processes, pilot plant scale and manufacturing unit. The development of such processes is called **Scaling Up**.

(i). **Laboratory Scale**:

- Maximum number of strains (microorganisms) are searched and the most suitable strain is selected and multiplied.
- A laboratory scale apparatus/plant is manufactured. It has a glass fermenter. All the parameters of the process are worked out like nutrient for the microbe,pH, aeration, disposal of CO_2 if evolved, optimum temp;, by products,product- inhibitionor stimulation, time of optimum production, separation of product and its purification; ultimately the laboratory scale process is finalized.

(ii). **Pilot Plant Scale**:

- Working in laboratory scale is tested in this intermediate stage.In this stage cost and quality of product is thoroughly checked. Glass apparatus are replaced by steel equipment or container called as **bioreactor**.
- Microorganisms can be grown in two systems.



In this method microorganisms grow as a thin layer or film in the solid medium

By suspending cells in the liquid medium

(iii). **Manufacturing Unit**:

- During the designing of bioreactor for the process often very large size of reactor is created to accommodate huge amount of medium.

1). **Alcoholic Beverages** (Yeast Fermentation):

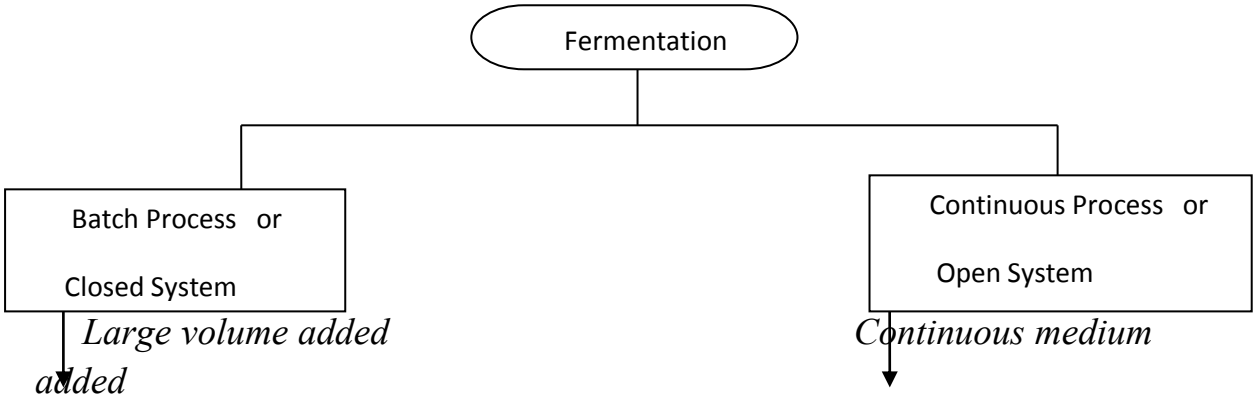
Yeast can be grown on a no. of substrates such as starch, sugar, glucose, sucrose and lactose, also on straight chain hydrocarbons (obtained from petroleum)or on ethanol and methanol.

The yeasts used in the brewing industry for the preparation of alcoholic drinks are called **Brewer's yeast**. Different kinds of alcoholic beverages are produced by using different brewer's yeast and the fermenting medium. Some known alcoholic beverages are as;

a). **Beer**: An undistilled product of grain-mesh fermentation brought about by *Saccharomyces cerevisiae* and *S. carisbergensis*

b). **Wine**: An undistilled product of fruit-juice fermentation brought about by *Saccharomyces ellipsoidens*.

Whisky, Rum, Gin, Brandy, Vodka and Fenny are distilled beverages. Distilled beverages are called "**hard liquors**" as they contain higher percentage of alcohol.



-Medium with organism removed after with

Each charge. The bioreactor (fermentation

tank) is cleaned & fresh batch started.

- A portion of the medium

fermented product is drawn off

Regular intervals & fresh medium

is introduced in to Reactor.

Fermentation is carried out in the following steps:

I. Sterilization of fermentor and medium in steam under pressure.
II. Inoculation of the selected strain of yeast.
III. Product recovery.

Growth Condition:

- Sterilized nutrient medium, pH, O_2,CO_2 conc. is easily provided on a laboratory scale so that micro organism can be cultured in small glass fermenters.
- Now from this stage the process is raised to the Pilot Plant Stage.
- Now glass vessel is replaced by steel vessel.
- Beer, buttermilk are product of fermentation brought about by yeast (*Saccharomyces cerevisiae*).
- Alcohol was the Ist product of ancient biotechnology.
- Yeast are basically of two types:
  (i). Baker's yeast; (ii). Brewer's yeast(Alcohol Yeast)
  - Baker's Yeast: Used in making food material to increase the taste and flavour of food.
  - It is also utilized as leavening agent.

- Incomplete degradation of sucrose:

$$C_{12}H_{22}O_{11} + H_2O \xrightarrow[\text{Invertase}]{\text{Yeast}} \underset{\text{Glucose}}{C_6H_{12}O_6} + \underset{\text{Fructose}}{C_6H_{12}O_6}$$

$$C_6H_{12}O_6 \xrightarrow[\text{Zymase}]{\text{Yeast}} \underset{\text{Ethylalcohol}}{2C_2H_5OH} + 2CO_2$$

Products of Brewer's yeast (Alcohol Yeast):

I. **Beer**: Product from *Hordeum vulgare* (Barley Malt.) Alcohol content is 3-8%.

II.**Wine**: Produced from grapes. Alcohol content is 10-20%.

III.**Brandy**: Produced by distillation of wine and alcohol content is 43-70%.

IV.**Gin**: Alcohol content 40%, produced from European Rye-Secale cereals.

V.**Rum**: Produced from molasses of sugarcane and alcohol content is 40%.

The bye – products of alcoholic fermentation are CO_2 and Yeast. A number of other chemicals can be formed with the change of nutrient medium, pH and aeration. Various products of yeast fermentation are as;

i). Ethanol ii). n- propanol iii). Butanol iv). Amyl alcohol v). Phenyl ethanol

vi). Glycerol vii). Acetic acid viii). Lactic acid ix). Pyruvic acid x). Succinic acid xi). Caproic acid xii). Caprolic acid xiii). Ethyl acetate xiv). CO_2 xv). H_2S

xv). Acetaldehyde xvi). Diacetyl

- A large quantity of yeast is utilized for the production of yeast extract, animal feed, food supplements and vitamins.

2). **Non - alcoholic beverages**: A non – alcoholic beverage is an alcohol – free or a non – alcoholic drink, also called as **temperance drink** is a version of an alcoholic drink made without alcohol, or with the alcohol removed or reduced to almost zero.

Non – alcoholic beverages can be broadly classified in to three types as:

I. Stimulating beverages – Tea, Coffee.
II. Refreshing beverages – Mineral water, Syrup.
III. Nourishing beverages – Milk, Malt based drinks.

3). **Organic Acids**: It is known that certain microorganisms have ability to convert carbohydrates in to organic acid. E.g;

- *Clostridium* ⟶ Acetic acid, Butyric acid
- *Lactobacillus*, *Streptococcus* sps. ⟶ Lactic acid
- *Acetobacter* sp. ⟶ Acetic acid

Some commercially important organic acids are as:

Acetic acid, Citric acid, Lactic acid, Fumaric acid, Gluconic acid, Itaconic acid, Kojic acid, Butyric acid, ,Gibberellic acid etc.

I). **Acetic Acid**: Commercial acetic acid is produced from fermented alcohol with the help of acetic acid bacteria, Acetobacter aceti. Alcoholic fermentation is an aerobic process whereas the conversion of alcohol to acetic acid is aerobic process.

$$C_6H_{12}O_6 \xrightarrow[\text{complex}]{\text{Zymase}} 2C_2H_5OH + 2CO_2$$

$$C_2H_5OH + O_2 \xrightarrow{\text{Acetobacter}} CH_3COOH + H_2O$$

It is used in the preparation of "Vinegar" (French, vin = wine + aigr=sour, sour/ wine), pharmaceuticals, colouring agents, insecticides, plastics etc.

II). **Citric Acid**: It is obtained by *Aspergillus niger*, yeast (*Candida lipolytica*) and Carbohydrates like cane and beet molasses, commercial glucose starch etc. It is used in medicines, dyeing, mirror silvering, inks, flavouring and preservation of food and candies.

III). **Lactic Acid**: It is the first acid to be produced microbially by *Lactobacillus* sps. (*L.delbrueckii*, *L. bulgaricus*) produced by fermentation by *Streptococcus lactis* and fungi *Rhizopus*.

$$2\,C_6H_{12}O \longrightarrow \underset{\text{Lactic acid}}{4C_3H_4O_3} + 4H_2 \longrightarrow 4CH_3.\,CHOH.\,COOH$$

It is used in confectionary, fruit juices, essences, pickles, curing of meat, lemonades, canned vegetables, fish products, tanning, printing of wool, pharmaceuticals, plastics etc.

IV). **Gluconic Acid**: It is manufactured through the agency of *Penicillium purpurogenum* and *Aspergillus niger* for use in pharmaceuticals and as calcium gluconate for infants, cows, and lacting mothers.

V). **Butyric Acid**: It is obtained by bacterium *Clostridium acetobutylicum*.It causes the rancidity of butter.

VI). **Cyclosporin- A**: It is produced by the fungus *Trichoderma polysporum*.It is used as an immune suppressive agent in organ transplantation as inhibits activation of T-cells. It has also antifungal, anti-inflammatory and immunosuppressive properties.

VII). **Alcohols**: Various alcohols such as ethanol, methanol, propanol and Butanol are produced commercially by some fungi (Yeast, *Mucor*, *Rhizopus*) and bacteria (*Clostridium acetobutylicum, C. saccharotobutylicum*). Alcohols are important industrial solvents.

VIII). **Statins**: They are produced by yeast *Monascus purpureus* and are used as blood cholesterol lowering agent (lovastatin, pravastatin, simvastatin)

IX). **Gallic Acid**: It is obtained with the help of Aspergillus *niger* used in ink industry.

4). **Enzymes**: (Gk, en = in, zyme = yeast)

- Payen and Persoz – 1833, identified and name enzyme Diastase.

- the term *Enzyme* was coined by Willium Kuhne – 1867.

- Buchner – 1901, found yeast extract to have enzymatic action.

Enzymes are proteinaceous substances of biological origin , capable of catalyzing bio - chemical reactions without themselves under going any change.

- All enzymes are macromolecules with three-dimensional shape, substrate specific, carryout a specific catalytic action.
- Out of about 2200 enzymes, only near about 300 are being used in industry.
- Enzymes may be constitutive and adaptive;
  Constitutive – produced in amounts usable by the cell
  Adaptive - produced by microbial cells in response to the presence of particular enzyme substrate.

Uses/ Adventages: Some important uses of enzymes in the industries are as;

i). Proteases (proteolytic enzymes): They are obtained from *Mortierella renispora*, *Aspergillus* and *Bacillus* sps. Several uses are as-

- Cleaning beer and whisky(chill proofing).

- Cleaning of hides.

- Softening of bread and meat.

- Degumming of silk.

- Manufacture of liquid glue.

- Manufacture of detergents.

ii). **Amylases**: Amylases are obtained from *Aspergillus,Rhizopus* and *Bacillus* sps. Several uses are as;

- Softening and sweetening of bread.
- Production of alcoholic beverages.
- Cleaning of turgidity in juices, corn, chocolate syrups.
- Separation and desizing of textile fibres.

iii). Amylase, glucoamylase and glucoiso-merase: Used as;

- Converts corn starch into fructose rich corn syrup.
- Sweetening and flavouring of soft drinks.
- Sweetening and flavouring baked products.

iv). **Rennet**: Extract from the stomach of calf that contains enzyme rennin/ chymosin. Now obtained from *Mucor* and *Endothio* sps. It is (Ficin) also extracted from several fruits like Withania, fig etc.Several uses are as;

- Cheese making
- Digester

v). **Lactases**: It is obtained from *Saccharomyces fragilis* and *Torula cremoris*. Some uses are;

- Coagulation of milk protein (Casein).

- Prevents crystal formation (sandiness) in dairy preparations.

vi). **Streptokinase** (*TPA*- **Tissue Plasminogen Activator**) : It is obtained from some haemolytic bacterium *Streptococcus*. It functions as **clot buster** as helps in clearing blood clots inside the blood vessels.

vii). **Pectinases**: Obtained from Byssochlamys fulvo. Uses as;

- Clearing of fruit juices.
- Retting of fibres.
- Preparation of green coffee.

viii). **Lipases**: Obtained from Candida lypolytica and Geotrichum candidum. Used as;

- In detergents for removing oily stains.
- Flavouring cheese.

Major Disadvantages of Enzymes:

I. They are more fragile than inorganic catalysts.
II. More expensive.
III. Rapidly deactivated at high temperature by exposure to air. Organic solvent, by acidic or by basic conditions.

5). **Vitamins**: vitamins are essential dietary factors required in small amounts. They influence on growth and metabolism and their deficiency may cause scurvy, beriberi, rickets, pellagra, night blindness etc. Most of the vitamins needed by the body of animal and humans are to be found in body tissues although the main supplements come through food.

- Discovery of vitamin by Kasimir Funk 1911, he succeeded in isolating Vit.B_1.
- Mac Collum isolated Vit.A
- Mellanby isolated Vit.D
- Albert Van SzentGyorgy isolated Vit.C
- Vit.C (first vitamin) produced during a fermentation process using a wild bacterium – *Acetobacter.*

Out of all vitamins produced during the normal metabolism by

microorganisms, only Vitamin B_{12} and Vitamin B_2 are manufactured biotechnologically. Whereas Vit.A as beta-carotene can be produced by various organisms.

i). **Vitamin B_{12}** (*Cobalamine*): It is isolated by **Rickes** in 1948 from liver extract and as a by-product during production of antibiotics by fermentation. The chief source of Vit. B_{12} is liver whereas milk, meat, eggs and fish also produce small amount of it. Within the body, it is produced by the microflora of the digestive system obtained mainly from meat, fish, brewer's yeast.

Commercially, Vit.B_{12} is produced by Propionibacteria (*Propionibacterium freudenreichii* and *P.shermanii*) and *Pseudomonas denitrificans*.

ii). **Vitamin B_2** (Riboflavin): It is produced for the Ist time in 1038 using a microbe. Its natural sources are cereals, green vegetables, brewer's yeast, egg white, milk and liver.

Commercially, Vit.B_2 is produced by *Ashbya gossypii* and *Ermothecium ashbyii*.

6). **Hormones**: Hormones are chemical substances that act like messanger molecules in the body. After being made in one part of the body, they travel to other parts of the body where they help to control how cells and organs do their work. E.g;

Insulin made by beta-cells in the pancreas.

Hormones may be;

i). Protein hormone (Polypeptide): Hormones made of chains of aminoacids.e.g; ADH(Antidiuratic hormone) which decreases blood pressure.

ii). Steroid hormone: Hormones derived from lipids.e.g; Glycocorticoids,sex hormones.

iii). Amine hormones: Hormones derived from amino acids.e.g; Melatonin (regulate circadian rhythm).

On the basis of secretion, hormones may be;

Exogenic: Ducted glands secrete their products through well defined ducts like liver, salivary glands, sweat glands,pancreas etc. e.g; FSH,LH.

Endogenic: Ductless glands secrete their products directly in to the blood stream. E.g; pineal gland,pituitary gland, pancreas, ovaries,testes, thyroid. Adrenal etc.

-Pancreas is both exogenic as well as endogenic in nature.

-Exogenous phytohormone supplementation has been adopted to improve growth and metabolism under stress conditions. Recent investigations have shown that phytohormones produced by root-associated microbes may prove to be important metabolic engineering targets for inducing host tolerance to abiotic stress especially in crop plants.

-Hormone imbalances: It occur when there is too much or too littleof a hormone in the bloodstream. Beccause of their essential role in the body, even small hormonal imbalances can cause side effects throughout the body.

Insulin: Insulin is an important hormone secreted by pancreas.

- Sir Edward Sharpy-Shafer (1921), thought that origin of certain forms of diabetes lay in the failure of pancreas to secrete a substance called Insulin (Lt. insula = island).

- Banting and Best isolated insulin from a dog's pancreas and demonstrated its effectiveness against diabetes.

- It is a polypeptide consisting of 51 amino acids, arranged in two chains – 21AA and 30AA. The chains are interconnected by S-S-disulphide bridges.

- The insulin used for treatment of diabetes are extracted from the pancreas of slaughter cattle and pigs.

- This extracted insulin differs slightly from human insulinin their aminoacid sequence.

- Animal insulin was useful in controlling diabetes but they produce afew undesirable side effects.

- On 5th july 1983, the American firm Eli Lilly launched HUMULIN (Ist genetically engineered human insulin).

  - Such human protein that are manufactured by the application of appropriate recombinant E.coli clones have been approved for clinical use.

- A new insulin product is Humalog – analog of human insulin.

- Plant growth regulators that aid in crop yield and management fall into several categories.
  - First, plant growth regulators are used to increase crops yield including horticultural crops.
  - Second, plant growth regulators can be used as direct replacement for hand labor in horticultural crop production other than harvesting.
  - Third, plant growth regulators can be used for manipulation of the harvest date in a wide range of crops.
  - Fourth, reducing the fruit removal force by the application of growth regulator allowed the development and use of mechanical harvesting equipment.
- Plant growth regulators are useful because they can in some way modify plant development. This may occur by interfering with the biosynthesis, metabolism, or translocation of plant hormone, or applied plant growth regulator may increase or decrease the endogenous levels of plant hormone when their endogenous levels are below or exceed that needed to change the course of plant development.

Some examples ofPractical application of phytohormonal compounds in Horticulture Crops:

Maleic hydrazide

Maleic hydrazide has been used since the 1950s for tobacco sucker growth control, the prevention of bud sprouting in onions and potato, and for the control of turf grass growth. At one time, maleic hydrazide accounted for almost 90% of the sales of plant growth regulators. In tobacco production, the terminal

bud is removed from the plant after a selected number of leaves has been produced. This practice. called topping, increases the size, weight, and quality of the cured leaf. Axillary buds, which develop as a result of topping, will reduce the effect of terminal bud removal on leaf yield and quality. Maleic hydrazide will provide excellent control of axillary bud growth when applied as a foliar spray to the upper two-thirds of the plant, after terminal bud removal. Maleic hydrazide is also used to control storage sprouting of onions and potatoes. The compound is applied as a pre-harvest foliar spray, since it is rapidly translocated to storage organs. Maleic hydrazide inhibits cell division in a wide range of plants, and the ability of the compound to be translocated to meristematic tissue probably accounts for the effect of the compound on axillary bud growth in tobacco and the sprouting of tubers and bulbs.
Maleic hydrazide is an analog of uracil and may inhibit cell division by reducing nucleic acid biosynthesis in shoot and root meristems.

Citrus Abscission Agents*:*

Several abscission agents are being developed for the mechanical harvesting of oranges intended for processing use rather than fresh market. The products, Release (5-chloro-3-methyl-4-nitro-1-pyrazole) and Pik-Off (ethandiol dioxime), induce abscission by causing superficial injury to the rind of the fruit. Wound ethylene is synthesized and presumably is the cause of the reduction in fruit removal force. Application of ethephon to trees with mature fruit will also induce abscission, however, significant defoliation often will occur with this chemical

Sugarcane Ripeners:

One of the more useful compounds for increasing yield in sugarcane is glyphosine, which in Hawaii, has increased sucrose yield by 10-15%. The herbicide Glyphosate, an analog of glyphosine, is also effective, and lower rates of application can be used. Glyphosine will decrease terminal growth of the cane, and other workers lave shown that removing the upper leaves of the stalk increases sucrose translocation from lower leaves into the stem and ripening joints. In addition, these compounds apparently alter the partitioning of carbohydrate
in the sugarcane internode. More carbohydrate goes into sucrose storage at the expense of fiber production.

Cotton defoliats*:*

The organophosphate DEF and Folex are used as leaf abscission agents before mechanical harvesting of cotton. Two new compounds are being evaluated for this purpose. Dimethipin (2,3-dihydro-5,6-dimethyl-1,4-dithiin-1,1,4,4-tetraoxide) and thidiazuron (1-phenyl-3(1,2,3-thiadiazol -55-yl)urea) induce defoliation and provide control of regrowth vegetation other leaf abcissions.

7). **Amino acids:** Amino acids are the basic building blocks of proteins and fulfill multiple functions
in the plant—structural, metabolic and transport**:**

- Amino acids for the production of biostimulants are obtained by chemical synthesis, from plant

proteins (e.g., algae, corn, and soybean), as well as from animal proteins by chemical or enzymatic
hydrolysis

- Plants can produce amino acids, but this synthesis is highly energy consuming. Therefore,

the application of ready for uptake amino acids allows plants to save energy and increase the pace of
their development or reconstruction, especially in critical times of plant development.

- Amino acids are also known in the industry of products for agriculture as chelates of metal

ions.

- Microelements chelated with amino acids form very small, electrically neutral molecules,

which accelerate their absorption and transport within the plant.

- These types of products are beneficial for plants that require supplementation with microelements. For example,
- the cultivation of high quality winter wheat varieties needs an adequate supply of micronutrients, which act as stimulants for macronutrients, in particular nitrogen.
- The sensitivity of winter wheat to the micronutrient deficiencies in soil, mainly copper and manganese, is higher than in other crops.
- The availability of micronutrients is limited by the characteristics of the soil (pH and sorption capacity), as well as the agricultural practices (fertilization system without plowing, and intensification of crop production and consequently uptake of nutrients).
- Therefore, foliar application of micronutrients seems to be more effective than soil and it is becoming more popular Fundamental functions of the amino acids in plants.

Function	**Amino Acid**
Anti-stress agent	Hyp, Pro
Chelating agent	Cys, Glu, Gly, His, Lys
Cold weather resistance	Ala,
Generative development of plants and improvement of plant pollen fertility	Hyp, Pro

Growth stimulator	Glu
Precursor of auxin	Ser, Trp, Val
Precursor of chlorophyll	Gly
Precursor of polyamines: necessary to start the cell division	Arg
Precursor to the formation of lignin and woody tissues	Phe
Regulation of the water balance	Hyp, Pro, Ser
Reserve of organic nitrogen necessary for the synthesis of other amino acids and proteins	Glu
Stimulation of the chlorophyll synthesis	Ala, Lys. Ser
Stimulation of the ethylene synthesis	Met
Stimulation of the germination	Asp, Glu, Lys, Met, phe, Thr
Stimulation of the hormone metabolism	Ala
Stimulation of the resistance mechanism to viruses	Ala

- The application of amino acids helps to reduce the amount of chemicals used in agriculture and plant protection.
- One type of biostimulants are preparations based on amino acids.
- Products on the market containing amino acids include:
  i. Metalosate Calcium and Metalosate Fe (Albion Minerals, Layton, UT, USA);
  ii. Agrocean B (Agrimer, Plouguerneau, France);
  iii. Tecamin Brix, Tecamin Max, Tecnokel Amino Mix, and Terra Sorb Foliar (Agritecno Fertilizantes, Valencia, Spain);
  iv. Amino Quelant Ca (Bioibérica, Barcelona, Spain);
  v. Bosfoliar Activ (COMPO EXPERT, Münster, Germany);
  vi. NaturalCrop SL (NaturalCrop Poland Sp. z o.o., Warszaw, Poland);
  vii. Delfan Plus (Tradecorp, Madrid, Spain).

8). **Antibiotics:** (Gr. *anti*- against, *bios*- life)

An antibiotic is an organic compound /substance produced by a microorganism that inhibits the growth of or kills another microorganism.

- **Penicillin** was the first antibiotic to be discovered by **Alexander Fleming**-1928,when he found that a fungus *Penicillium notatum* or its extract could inhibit the growth of bacterium *Staphylococcus aureus*.
- **The term antibiotics was coined by Waksman – 1942.**
- **Fleming, Chain** and **Florey** were awarded **Nobel Prize** in **1945.**
- **Waksman** and **Woodruff-** isolated Actinomycin in1941 and Streptomycin in 1942.
- **Waksman** and **Albert-** 1943 and **Waksman-** 1944 discovered Streptomycin.
- **Burkholder** – 1947, isolated Chloromycin.
- Every year some 300 new antibiotics are discovered, so for over 7000 antibiotics are known.
- *Streptomyces griseus* produces more than 41 antibiotics whereas *Bacillus subtilis* forms about 60 antibiotics.
- **Broad Spectrum Antibiotics** - ability to act on several pathogenic species differing from each other.
- **Limited Spectrum Antibiotics** – act on similar species.

Types of Antibiotics

There are five basic types of antibiotics such as;

Betalactans, Amino-glycosides, Quinolenes, Sulphonamides and Glycopeptides.

Mode of action of antibiotics:

a). Bactericides – by killing bacteria.

b). **Bacteriostatic** – by inhibiting growth of bacteria.

The action is done by the following ways;

1). Disruption of wall synthesis – penicillin, cephalosporins, bacitracin.

2). Disruption of plasma lemma repair and synthesis – polymixin, nystatin, amphotericin,

3). Inhibition of 50S ribosome function – erythromycin.

4). Inhibition 30S ribosome function – streptomycin, neomycin.

5). Inhibition of aa-tRNA binding to ribosome – tetracycline.

6). Inhibition of translation – chloramphenicol.

Resistance to Antibiotics:

a). reduction or elimination of antibiotic ability to penetrate bacteria through modification of the cell wall membrane.

b). spontaneous mutation, altering the antibiotics target so that it no longer recognizes its point of impact.
c). modification of the antibiotics by the bacterial enzyme.

Properties of a good Antibiotic:

i). harmless to host with no side effect.
ii). harmless to normal microflora of alimentary canal.
iii). Ability to destroy pathogen as well as show broad spectrum.
iv). effective against all pathogenic strains.
v). have quick action.

Monoclonal Antibodies:

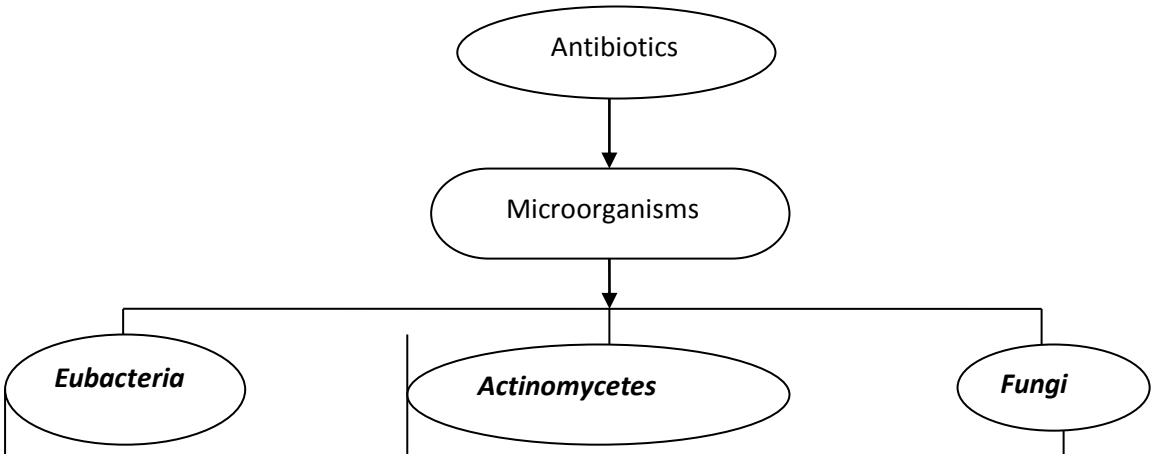
- In 1974, **George Kholer** and **Cesar Milstein** hit upon the idea of fusing normal antibody- producing cells with cells from the cancerous tumours Known as **Myeloma Hybridoma Technique**.
- Monoclonal antibodies are called as Magic Bullets and reproducible than the antibiotics produced by conventional technique.

  Clinical use of Monoclonal Antibodies:
- Pregnancy test, diagnosis and treatment of disease.
- It is feasible in immune suppression for **Kidney Transplantation**. This antibody is directed against the immune cells that mediate graft rejection and is normally administrated to kidney transplant patient experiencing a rejection episode.

  Sources of Antibiotics:

  All the antibiotics are obtained from the microorganisms (bacteria, fungi).However, the bulk of antibiotics are produced mainly from three groups of microorganisms.

  a). **Eubacteriales** – True/ Simple bacteria
  b). **Actinomycetales** – Ramified bacteria
  c). **Fungi**



-Bacillus Penicillin,

Produce 70% cephalosporin

Antibiotics used

-Pseudomonas Produce 30% Antibiotics

-B.subtilus Produce more than 60 diff. Antibiotics

-Branched-mycelium narrow hyphae, mostly saprophytic but few are parasitic.

Streptomyces sp. Yields anti-biotics as; Chloramphenicol, erythromycin, neomycin, novobiocin, oxy- tetracycline, spira Beside *Streptomyces*, micro-monospora, nocardio, streptoverticillium, actinoplanes, and streptosprangium are chief source of antibiotics.

-

widely in **Chemo-therapy**.

9). **Vaccines: Vaccine** is a liquid containing dead, attenuated form of antigen or pathogen which can be injected or taken orally to provide immunity (acquired active, long or short duration) towards the pathogen.

- Jenner's work on small pox leads to the concept of vaccination.

- Technique of attenuating or weakening the pathogen was discovered by L.Pasteur-1879 against cholera.

- Pasteur established the scientific basis of vaccination.

- Ist generation vaccine: These are obtained by conventional technique by killing or weakening the pathogen. These may have side effects.

- Recently new vaccine known as **IInd generation vaccine** was produced by genetic engineering for **Hepatitis B** and **Herpes Virus.**

- IInd generation vaccine: These are made up of pure surface antigen of the pathogen only which are multiplied through genetic- engineering or DNA - recombination technique. e.g; Vaccine for Hepatitis B.

- Even **IIIrd generation vaccines (**synthesized vaccine**)** have been produced**.**

- IIIrd generation vaccine: These are purest and highly potent vaccine. These are synthetic in nature.

10). Steroids: Steroids are chemicals, often hormones made by body naturally.Steroids include drugs used to relieve swelling and inflammation such as prednisone, cortisone, Vit.D and some sex hormones.

- All steroids have the same core structure that consists of one- 5carbon and three - 6 carbon rings.

- Cholesterol is an essential component of animal cell membrane and is the starting point for the synthesis of the body's steroid hormones.

- These are constituents of cell membrane, hormones, cholesterol, progesterone, estrogen, testosterone, antifertility drugs.

- Testosterone – male hormone (steroid)

- Esterogen – female hormone (steroid)

- Progestrone – female hormone (steroid)

- Patient with auto immune disease or who have organ transplant are treated with steroids to suppress immune response.

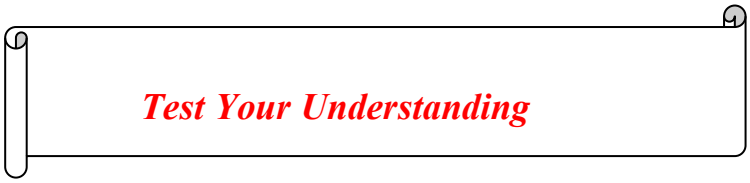
- Most widely used steroids are **Esterogen** and **Progesterone** that go in to the composition of **birth control pills**.

- **Murray** and **Peterson** – 1950 extracted steroid from ***Rhizopus stolonifer***;

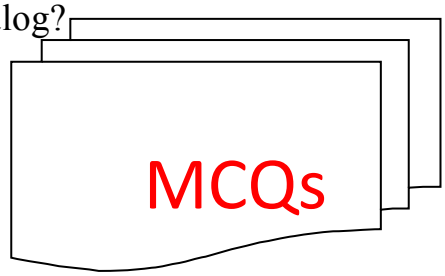
Rhizopus stolonifer ——***Hydroxylation***→ ***Steroid synthesis***

- Microbiological hydroxylation of steroid has been one of the best example of the superiority of biological catalysis over chemical catalysis.

Cortexolone (steroid) —Microbes→ Prednisolone (used as anti-inflamatory drug involving first a hydroxylation than a dehydrogenation)



1. How cheese is prepared? What are its different types?
2. Mention various methods of fermentation.
3. Name didderent organic acids with their sources.
4. What do you meam by LAB and Probiotics?
5. Define Leavening and Scalling Up.
6. What do you mean by Brewer's yeast?
7. Give uses of some important enzymes.
8. Write a not on Vitamin B_{12}.
9. Define Monoclonal antibiotics with example.
10. What is Humalog?



i). 'Hard liquors' are :

a. Undistilled beverages b. Distilled beverages c). Non-alcoholic beverages

d). All the above

ii). Cobalamine is:

a). Enzyme b). Hormone c). Bacteria d). Vitamin

iii). 'HUMULIN' is:

a). Steroid b). Insulin c). Both d). None

iv). Bacillus are main source of:

a). Enzymes b). Vitamins c). Antibiotics d). Hormones

v). Most widely used steroids are:

a). Testosterone b). Estrogen c). Progestrone d). a &b

Sewage treatment, production of energy (Biogas)

Sewage is the municipal waste water (both liquid and solid wastes) generated in cities and towns which is carried off in sewers.

-Sewage contains huge amount of domestic water borne wastes including human and animal excreta, washing waters, microbes and everything that enters in to the sewerage system.

- Sewage should not be directly passed into the waterbodies because it not onlt contains human excreta and other organic wastes but a large number of pathogenic microbes.

- In India about 800 million gallons of sewage is produced per day of which 20% is disposed properly whereas rest remains untreated.

- Chemically, sewage consists 90% water and 10% solid waste (inorganic and organic matter).

- Microorganisms present in sewage are mainly – ***coli***- forms, ***streptococci***, ***clostridia***, ***lactobacill***i etc.

- So in order to make sewage water less harmful, it is passed through **Sewage Treatment Plants** (***STPs***).

Sewage Treatment Plant

In a Sewage Treatment Plant heterotrophic microbes naturally present in sewage carryout the process of decomposition.

-There are three stages of a Sewage Treatment Plant, such as;

i). **Primary Treatment** – *Physical*

ii). **Secondary Treatment** – *Biological*

iii). **Tertiary Treatment** - *Chemical*

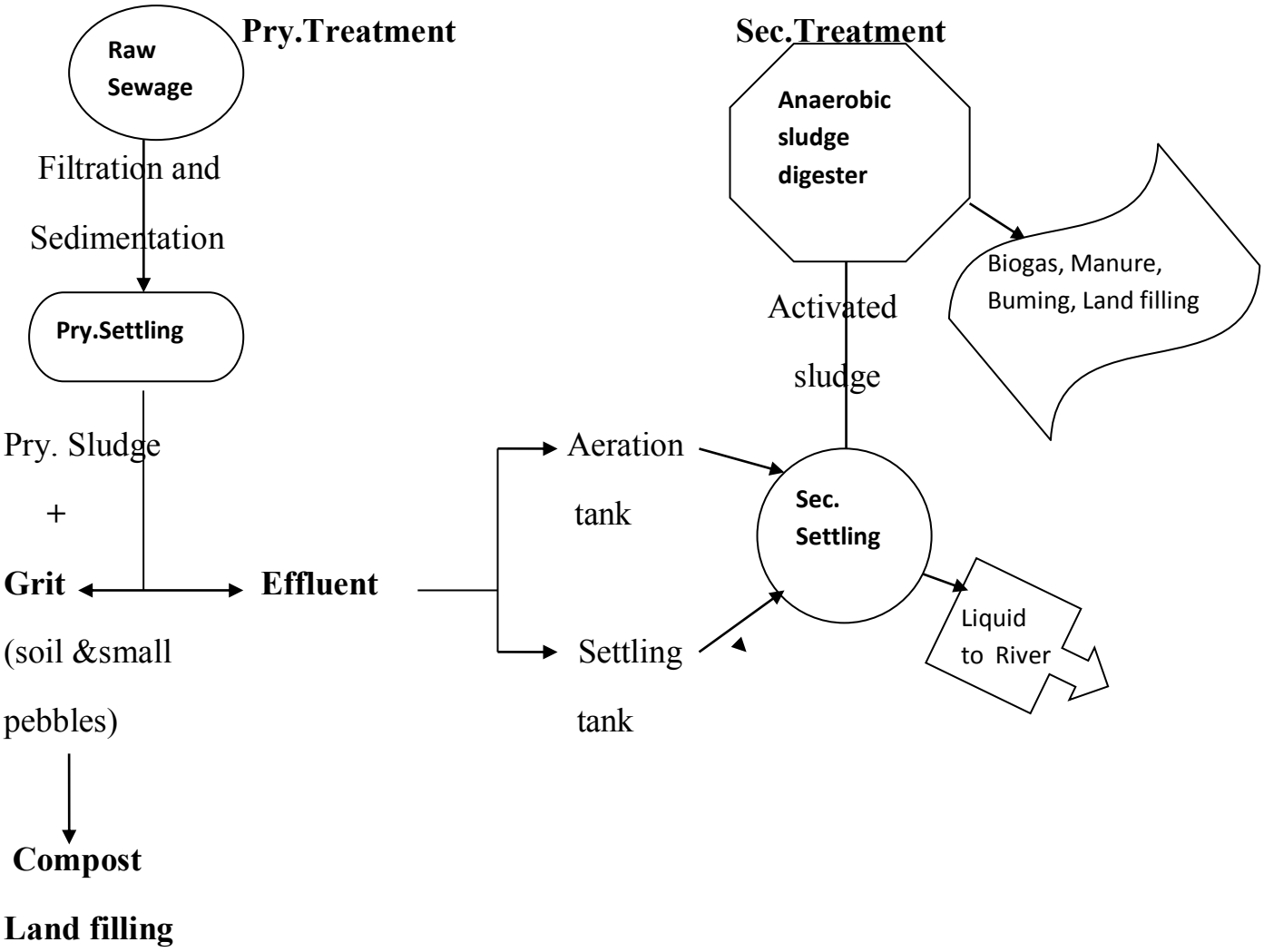
i). **Primary Treatment:** Primary Treatment is the process of removal of small and large, floating and suspended solids from sewage through filtration and sedimentation. The **filtration** is done through progressively smaller pore filters. For sedimentation, the filtrate is kept in settling tanks where grit (sand, silt, small pebbles) settles down with the addition of aluminium or iron sulphate. This sediment is called **primary sludge** and supernatant is **effluent**. The primary sludge traps a lot of microbes and debris.

- Primary sludge is used to composting, land filling or anaerobic digestion to produce biogas and manure.

ii). **Secondary Treatment:** Secondary Treatment can be done by various methods like oxidation tanks, trickling filter system and activated sludge system. In activated sludge system, the primary effluent is taken to aeration tanks where liquid is constantly agitated with air. In order to fasten the process of decomposition some primary sludge is also added. A large number of aerobic heterotrophic microbes growing the aeration tank resulting in the formation of **flocs.** These microbes digest a lot of organic matter thus converting it into microbial biomass and releasing a lot of minerals so reducing BOD (Biochemical Oxygen Demand). In settling tank. The flocs are allowed to settle down (sedimentation) whereas effluent or supernatant is generally passed in to natural waters like rivers and streams after chemically treated for disinfection.

- Flocs are the masses of bacteria held together by slime and fungal filaments to form mesh - like structures.
- The sediment of settling tank is called activated sludge.
- The activated sludge is used as inoculums in aeration tanks and as anaerobic sludge digesters in large tanks.
- Anaerobic microbes are methanogenic as well as non-methanogenic.
- Methanogenic microbes produce **marsh gas –** mixture of **methane, H_2S and CO_2.**The mixture is also called **Biogas**.

Flow chart of Sewage treatment

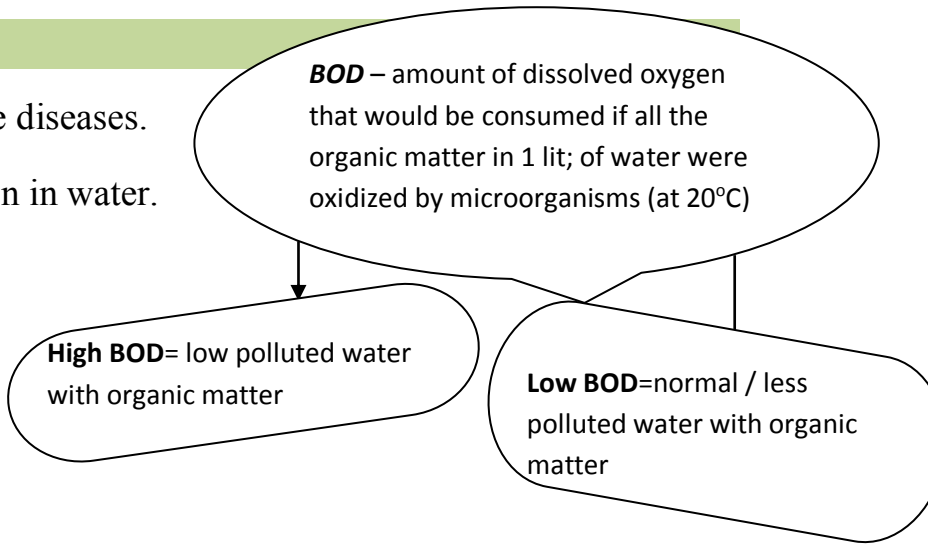


Harmful Effects of Sewage:

i.Dissemination of water- borne diseases.

ii.Depletion of dissolved oxygen in water.

iii.Produce offensive odour.



- ***River Action Plans***:

Due to the huge production of sewage and less number of sewage treatment plants, the untreated sewage is being discharged into the water bodies thus leading to the water pollution and increase in the

incidence of water – borne diseases. Realizing the importance of microbes in pollution control and to protect the major rivers of India from sewage pollution , the Ministry of Environment and Forests , has initiated the development of sewage treatment plants under the National River Conservation Authority e.g; **Ganga Action Plan (GAP), Yamuna Action Plan, Sutlej Action plan, Gomati Action Plan.**

BIOGAS (Gobar Gas)

***Mixture of gases produced from degradable organic matter by the activity of various anaerobic bacteria offering a low cost alternative for energy requirements*. OR**

A methane rich fuel gas produced by anaerobic breakdown or digestion of biomass with the help of methanogenic bacteria.

- The microorganisms mainly invove in biogas production are facultative as well as strict anaerobes such as *Methanobacterium,Bacillus, Cellulomonas, Clostridium, Eubacterium and Rumenococcus.*
- Biogas mainly contains;
- Methane = 50 – 70%
- Carbondioxide = 30 – 40%
- Hydrogen = 1 – 5%
- Nitrogen = 0 – 0.1%
- Hydrogen sulphide = traces
- 50% of the combustible energy present in the organic waste can be changed in to the methane gas.
- The calorific value of biogas is 23 – 28 MJ/m3.
- The residue left after the fermentative generation of biogas is rich in minerals, lignin, and cellulose.
- The biogas technology was developed by the collaboration of Khadi and Village Industries Commission (**KVIC**) and Indian Agricultural Research Institute (**IARI**).

Biogas Plant

Biogas Plant mainly build in the rural areas are used to produce **Biogas.** A biogas plant consists of a concrete tank (10-15 feet deep) covered by a floating lid. The plant is fed with a mixture of dung and water (1:1 ratio). Apart from dung, many other raw materials can be used for biogas production such as waste materials available from kitchen and **night soil**. After filling the slurry of the dung , the biogas tank is covered by a floating lid which keeps on rising as the gas is produced from the slurry. The tank has two outlets, one is used to supply **biogas** to nearby houses through a pipe and another to remove leftover slurry used as **fertilizer**.

Production of Biogas

Biogas generation is a three- stage anaerobic digestion of animal and organic wastes like lignin, cellulose, hemicelluloses, lipids and proteins.

These three-stages are:

i). Solubilisation

ii). Acidogenesis

iii). Methanogenesis

Lignin-cannot be broken-down under anaerobic conditions.

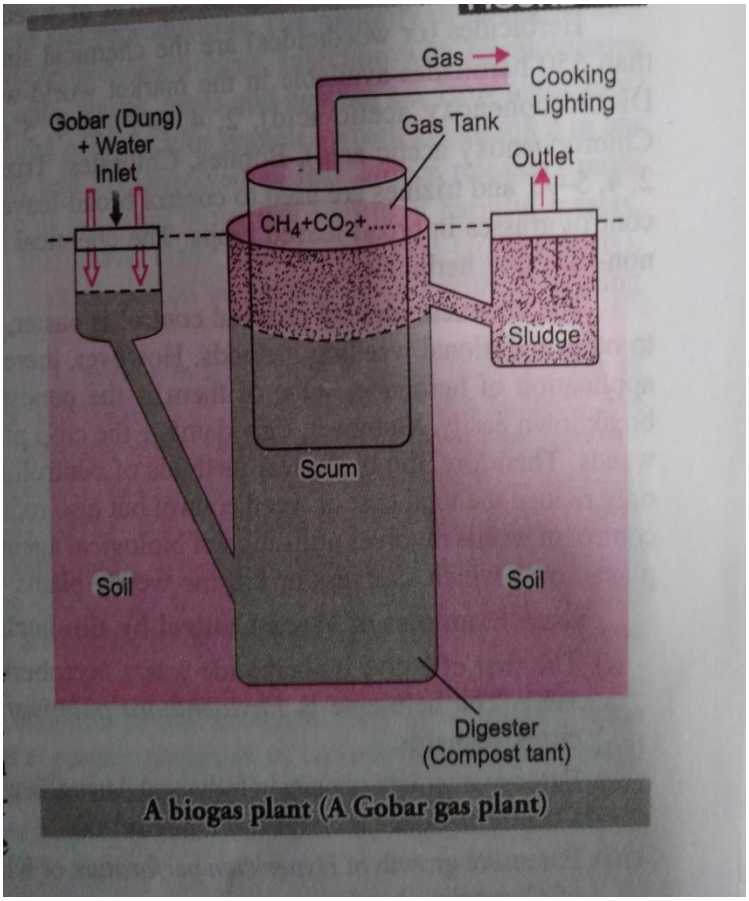
Cellulose- digest in slower than that of other substances.

i). **Solubilisation**: In this stage, facultative anaerobic decomposer microbes (*Clostridium*, *Pseudomonas*) bring about enzymatic breakdown of complex organic compounds in to simpler and soluble compounds – ***monomers.*** The decomposer microbes secrete cellulases, proteases and lipases (cellulolytic, proteolytic and lipolytic enzymes).

ii). **Acidogenesis**: In this stage, the monomers are acted upon by fermentation causing microbes (*Acetovibrio*, *Propionibacterium*) so change monomers in to organic acids.

iii). **Methanogenesis**: Now the organic acids (as well as carbondioxide) are acted upon by methanogenic bacteria and converted into **Methane.**

Thus, the biogas formed is stored in to tanks for supply.



A biogas plant (A Gobar gas plant)

Complex Organic Complex —*Solubilisation*→ **Monomers**

Clostridium, Pseudomonas

Organic Acids + ←*Acidogenesis*—

CO_2

Acetovibrio, Propionibacterium

Methane ←*Methanogenesis*—

Methanogenic bacteria

Flow Chart of Biogas Production

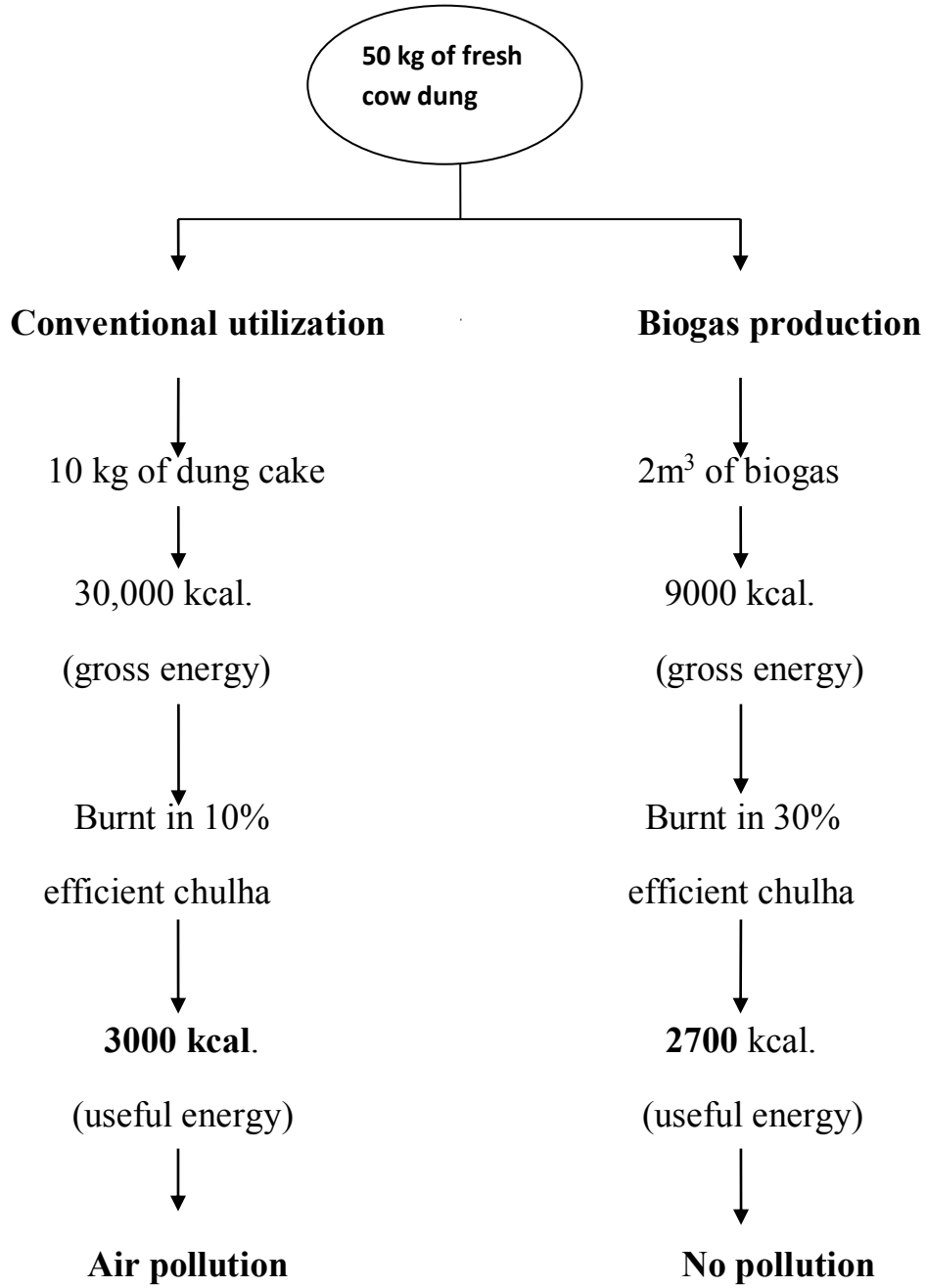
Gobar Gas Plant – as the cattle/cow dung is commonly known as **"Gobar"** so the plant is called Gobar gas plant.

-It is the rich source of cellulosic material from plants.

Advantages of Biogas: Various advantages of **Biogas/ gobar gas** are as**:**

I. Provides both fuel and manure.
II. Storable form of energy can be used more efficiently and economically.

III. Minimizing the spread of infectious diseases.
IV. Minimizes the pollution as different types of organic wastes are used as raw material.
V. Cheap and easy availability especially in rural areas.
- 50 kg of fresh cowdung can produce manure for 2 acre.
- 10 kg of dried dung cakes have 30,000 kcal.



Alternate ways of Using Cow dung

Have A Look

- Energy Croping/ fuel alcohol – Growing crops for production of alcohol and other energy fuel.
- Energy Fuel - Sugarcane,Sugarbeet,potato,Tapioca & Maize used to produce alcohol and other fuel.
- Energy Plantation – Growing of more trees for fuel wood and biofuels.
- 90% of the cars in Brazil are running on mixture of Petrol and Alcohol.
- Gasohal programme of USA uses mixture of alcohol (10-15%) & petrol (85-90%).
- Petroleum plants/ Petrocrops – plants which can yield large amount of latex having long chained liquid hydrocarbons [(*Jatropa*, *Euphobia lathyris* (Euphorbiaceae), *Brickellia* (Compositae)]. Some other Petroleum plants are- *Euphorbia antisyphilitica*, *Calotropis procera*, *Copaifera langsdorfi*, *Cryptostegia grandiflora*, *Pittosporum resiniferum*.

Biocontrol Agent (Biopesticides) and Biofertilizers

PESTICIDES

PEST: A pest is defined as an organism that can damage human cultivated crops between sowing, growing, harvesting, storage and whose population often increases beyond certain level so that its existence endangerous human welfare, profits and convenience. OR

PESTS are the organisms which may damage the economic and physical well-being of humans.

A variety of unwanted and even harmful insects, weeds, microorganisms (fungi, bacteria) rodents, nematodes and other organisms come under the category of pests.

PESTICIDES: Pesticides are those substances which are used to kill, control or repel pests.

Depending upon the type of target organisms, the pesticides may be;

i) **Fungicides:** used against fungal diseases(fungi).
ii) **Herbicides:** used against weeds(unwanted plants).
iii) **Insecticides:** used against insectal diseases (insects).
iv) **Nematicides:** used against nematode diseases (Nematodes).
v) **Bactericides:** used against bacterial diseases (bacteria).
vi) **Rodenticides:** used against rodents (rodents).

An ideal pesticide possesses the following characteristics:

a). easily available in the market and cheap.

b). control only the specific target organisms.

c). non- persistent.

d). non- toxic to other living organisms.

e). biodegradable.

BIOPESTICIDES

Biopesticides are living organisms or their products used for killing pests or interfering with their biological process. OR

Biopesticides involve use of one kind of animals for destruction of others hence pest population can be checked.

Biopesticides have not hazardous effects on environment.

Biopesticides are of two-types;

I. Bioherbicides
II. Bioinsecticides

I). **Bioherbicides**: The biological methods of controlling weeds are more effective and economic which not only reduce the total cost of weed control but also reduce the residue and pollution problems.

Bioherbicides involves utilization of biological agents such as insects, fungi, bacteria, nematodes, parasitic agents etc. which suppress or kill the weedy plants without causing significant injuries to other plants.

Some examples of bioherbicides are as;

i). The Cochineal insect (*Cactoblastis cactorum*) is used to reduce growth of cacti and the fungus (*Phytophthora pulmivora)* is used to control the growth of milk weed vines in citrus orchids.

ii). The commercial products like Devine and Callego are developed from fungal spores and suitable for weed control.

iii). *Cercospora rodmanii* & *Alternaria eichhomiae* are used to control growth of water hyacinth (*Eichhomia crassipes*).

iv). Use of smoother crops (Alfalfa, Soyabean, Sunflower, Sweet clover etc.) which do not allow the weeds to grow nearby.

v). Introduction of specific insects which feed on the weeds.

vi). Various insects destroy a noxious weed Lanthana.

vii). Genetic engineering – some identical genes known for herbicidal resistance are incorporated in to the desirable crop plants which are named as ***Transgenic plants*** and are resistant to weeds e.g; tomato, tobacco. The herbicide kill the weeds selectively and the transgenic crop plants remains healthy.

II). **Bioinsecticides**: Various methods of biological control are being used in the pest management to keep the environment pollution-free and yield clean, non-toxic and good quality products for human consumption.

Some examples of bioherbicides are as;

i). **Pathogens, Parasites and Predators**: Harmful insects are controlled by the application of their natural parasites and predators.

a**. Baculo viruses**: (a group of rod shaped viruses) These viruses infect the larval stages of many harmful insects like ants, wasps, gnats and beetles.

b. Use of different strains of bacteria ***Bacillus thuringiensis*** (**crystal protein -thurioside**) to kill insects like moths, flies, beetles, mosquitoes, ants, aphids, termites, butterflies etc., animal and plant nematodes, protozoa, snails and cockroaches.

Baculo viruses-may be *Nucleopolyhedroviruses* (NPV) and *Granulovir-uses* (GV), generally ***Nucleopolyhedra virus*** are used as bio- control agents.

c. Introduction of predator, lady bird beetle has proved successful in feeding on scale insect pest.

d. Fungus (*Beauveria barsiana*) is used to control potato beetle and codling moth.

e. Ampleomyces parasitise the conidia of some powdery mildews.

f. Uredospores of some rusts (*Melampsora* spp.) are attacked by *Cladosporium* spp.

g. Crown gall disease of apple (*Agrobacterium tumefaciens*) is controlled by treating the seeds, seedlings and cuttings with the non-pathogenic strain of *Agrobacterium radiobacter* (K84).

h.*Trichoderma* (produces trichodermin,gliotoxin, viridian & gliovirin) inhibits growth of several soil borne diseases like root rot, foot rot, rhizome rot, collar rot, stem rot, dumping off wilts etc;

ii). **Sterilization Strategy**: Release of sterile individuals so that insect mating, breeding and propagation is biologically controlled.

iii). **Insect Hormones**: Use of insect hormones for controlling the insect pests. Two types of insect hormones are employed for this purpose. These are:

a). ***Pheromones***: These are highly volatile chemicals produced by animals which are odourless so undetectable but influence animals. These are used by insects for communication. These may be;

i. **Signaling pheromones**: Substances which alter the behaviour of animal's social or sexual partner are primer pheromones.

- Sex pheromones are released by female to signal male that where she is and is ready to be mated. The male moths fly from kilometers around to visit female.

Now these chemicals are artificially synthesized in order to trap insects in the hallow cylinders coated with sticky substances and synthetic sex pheromones.

ii. **Confusion technique**: A large number of hydrophobic paper pieces containing synthetic sex pheromones are dropped around the crop fields as a result the smell of sex pheromones spread all over that causes the confusion among male insects so they fail to locate their partner.

iii. **Juvenile hormone and Molting hormone**: (*Ecdysone*): These hormones play their role in the metamorphosis of the insect. Juvenile induce growth and prevents early maturation where as Ecdysone causes molting (periodical

shedding of cuticle in the process of growth). If these hormones are given at late stage, the larvae become giant and die.

b). ***Natural Insecticides***: These are obtained from some plants and microbes having low toxicity to mammals.

Some important bioinsecticides are;

- **Rotenones** – obtained from roots of *Derris elliptica* & *Lonchocarpus nicou*.
- **Pyrethroids and Cinerin** – obtained from dry inflorescence of *Chrysanthemum cinerarifolium*.

c). **Nicotine**: obtained from leaves of *Nicotiana tabacum*.

d). **Azadirachtin**: obtained from different parts of Neem (*Azadirachta indica*).

e). The extracts of various parts of ginger are also used as bioinsecticides.

f). **Squill**: red variety of Sea Onion (Red Squill, *Ureginea maritime*) produces a radicide which does not have any harmful effe other anim

IMP – Integrated Pest Management

A "perfect target" crop may be grown to attract the insects away from the economic crop sometimes as economic crop is grown mixed with a target weed which is preferred by the insect-pest over the desired crop (Starvation method).

IPM involves use of different pest control methods which are ecologically good e.g; bio-control methods, crop rotation, use of very low and most effective concentration of chemicals which are pollution free.

BIOFERTILIZERS

Biofertilizers are microorganisms which bring about nutrient enrichment of soil by enhancing the availability of nutrients to crops. OR

Biofertilizers are microorganisms which bring about soil enrichment, maximize the ecological benefits and minimize the environmental hazards.

Various microorganisms which act as biofertilizers are bacteria, Cyanobacteria (blue green algae) and mycorrhizal fungi. Bacteria and Cyanobacteria have the property of nitrogen fixation whereas mycorrhizal fungi provides minerals from organic matter to their associates.

Preparation:

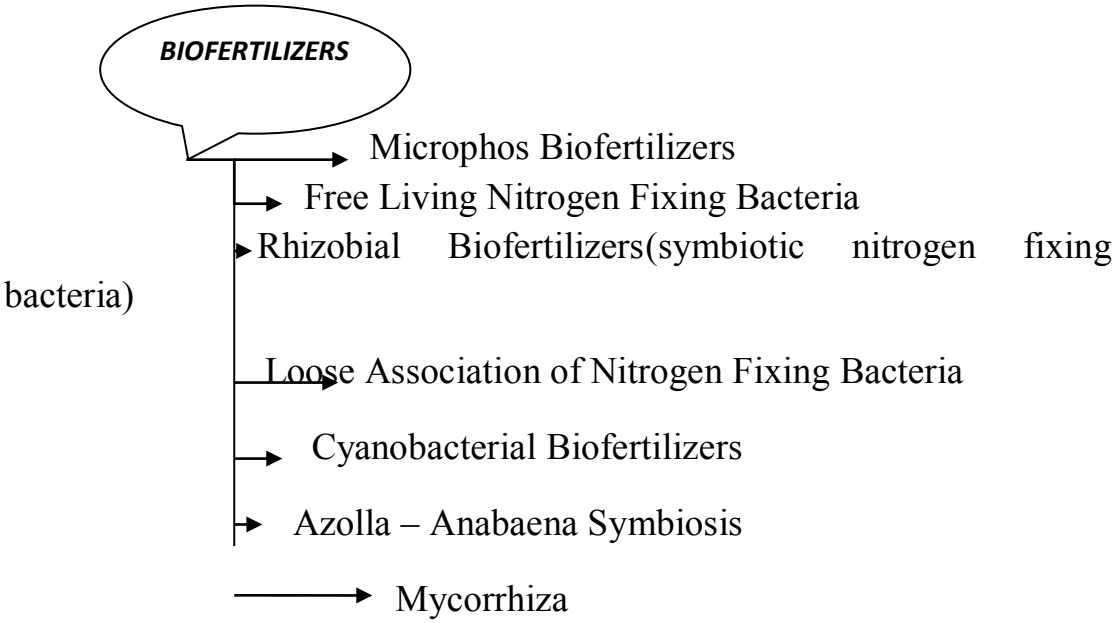
- A suitable medium is prepared by mixing powdered coal, lignite soil and essential nutrient elements in 10% aqueous mixture.

- Mixture is autoclaved to destroy any contamination.
- Sterilized medium is cooled for about 48 hours.
- Specific microorganisms are mixed in the medium and filled in pockets.
- Pockets are kept under controlled temp; conditions for a week.
- Microorganisms multiply and increase in population
- Biofertilizer pockets are supplied to farmers.

Types of Biofertilizers

Some of the important biofertilizers recommended for the use in agriculture are as follows:

1. Microphos Biofertilizers
2. Free Living Nitrogen Fixing Bacteria
3. Rhizobial Biofertilizers (Symbiotic Nitrogen Fixing Bacteria)
4. Loose Association of Nitrogen Fixing Bacteria
5. Cyanobacterial Biofertilizers
6. Azolla – Anabaena Symbiosis
7. Mycorrhiza



1. **Microphos Biofertilizers**: These biofertilizers are used when plants show deficiency of phosphorus. In the soil when the phosphates are present in the form of immobile or bound forms, these micro-organisms make it available to the plants in the soluble form. They reduce their pH by secretion of a number of organic acids such as formic acid, acetic acid, propionic acid, lactic acid, glycolic acid, Fumaric acid and Succinic acid. These cause release of soluble inorganic phosphate (H2PO4) into soil through decomposition of phosphate-rich compounds. Main Microphos biofertilizers are *Pseudomonas striata* and *Bacillus polymyxa* and fungi *Aspergillus*.
2. **Free Living Nitrogen Fixing Bacteria:** These **bacteria** live freely in soil, may be saprotrophic (living on organic remains) such as *Azobacter*, *Bacillus polymyxa*, *Clostridium*, *Beijerinckia* and photoautotrophic like *Rhodopseudomonas*, *Rhodospirillum*, *Chromatium* besides some other aerobic and anaerobic forms. Inoculation of these bacteria helps in increasing yield and saving of nitrogen fertilizers. E.g; application of **Azotobactrin** (trade name of *Azobacter* biofertilizer) saves 10-25 kg/ha tunes of nitrogen fertilizer in the fields of cotton, maize, jowar and rice.
3. **Rhizobial Biofertilizers** (**Symbiotic Nitrogen Fixing Bacteria**): These micro-organisms show mutual beneficial association with the plants. The bacteria get food and shelter from plants whereas the plants in return get nitrogen/nitrogenous compounds from these bacteria. The most important

of the symbiotic nitrogen fixing bacteria is Rhizobium which forms nodules on the roots of legume plants. There are about a dozen species of ***Rhizobium*** which form association with different legume roots like ***R.leguminosarum***, *R. lupine*, *R.trifolii*, *R.meliloti*, *R. phaseoli* etc. In the nodule cells, bacteria (bacteroids) lie in groups surrounded by membrane of the host which is lined by a pink-red pigment called ***leghaemoglobin***. Rhizobium can fix up to 100-500 kg nitrogen per hectare o land. The fixed nitrogen is used up by the leguminous plants. However, a sizeable amount of fixed nitrogen is left behind in the soil in the form of residue which can be utilized by the succeeding crops.

- *Frankia* a nitrogen fixing mycelia bacterium (actinomycete) is associated symbiotically with the root nodules of several nonlegume plants like *Casuarina*, *Alnus* (Alder), *Myrica, Rubus* etc.
- Leaves of a few plants like *Ardisia* develop special internal cavities for providing space to symbiotic nitrogen fixing bacteria- *Xanthomonas*, *Mycobacterium*.
- Application of Rhizobium inoculums needs proper storage and selection of strains based on soil characteristics, the pH of the soil, moisture content and the temperature of the area.

4. **Loose Association of Nitrogen Fixing Bacteria**: It was observed that in some nitrogen fixing bacteria (*Azospirillum*) live around the roots of higher plants without developing any intimate relationship (**Rhizosphere Association**). Both are benefited, the phenomenon is termed as **Associative mutualism** (**associative symbiosis**).
5. **Cyanobacterial Biofertilizers**: It is the most suitable source of biofertilizer achieved by the use of Cyanobacteria (Blue-green algae), particularly in rice fields. These bacteria form symbiotic association with several plants such as Cycad roots, lichens, liverworts, Azolla.

   More than one hundred species and strains of nitrogen fixing Cyanobacteria are known. Some of them are as *Anabaena, Nostoc, Anabaenopsis, Aulosira, Chlorogloea, Cylindrospermum, Calothrix, Scytonema, Tolypothrix, Fischerella, Haplosiphon, Mastigocladus, Westiellopsis, Wollea, and Gloeotrichia.etc*.

   - **Azolla-Anabaena** association is of great importance in agriculture.
   - ***Azolla pinnata*** a small free floating fresh water fern coexisting with rice plants, multiplies rapidly, doubling every 5-7 days.

- In **China**, the rice fields are regularly provided with Azolla.
- The techniques of inoculating the rice fields with Cyanobacteria is being extensively practiced in Tamil nadu

Anabaena azollae* resides in the *leaf cavities of the fern and fixes nitrogen. A part of the fixed nitrogenis excreted in the cavities and becomes available to the fern. The decaying fern plants releases the same for utilization of the rice plants.When field is dried at the time of harvesting, the fern functions as the green manure, decomposing and enriching the field for the next crop.

6. **Azolla – Anabaena Symbiosis**: An attractive Cyanobacterial biofertilizer is Azolla-Anabaena symbiosis. It is very efficient biological nitrogen fixer. Azolla a small water fern consisting of branched floating stem with deeply bilobed leaves and true roots. Anabaena azollae grows as symbiont within the cavities formed in the upper lobes of the aerial chlorophyllous leaves of Azolla.

   It is usually inoculated at the rate of 300-500kg fresh weight per hectare of land about a month before rice plantation. Its inoculation improves the height of rice plants, number of tillers, grains and straw yield causing 50% higher in yield.

***Azolla pinnata*—grows abundantly in ponds, ditches and stagnant water bodies in India.**

7.Mycorrhiza (Mycorrhizae): The term was coined by Frank-1885. Simply means an association of fungi with the roots of higher plants. This association is true symbiosis (mutualism) . Here mycorrhizal roots lack root cap and root hairs. They change their shapes and may become irregular, tuberous, nodulated or coralloid. Depending upon the residence of the fungus, mycorrhizae is of two types:

1. **Ectomycorrhizae**
2. **Endomycorrhizae**

1. **Ectomycorrhizae:** (Ectotrophic mycorrhiza) Here the fungus forms a mantel on the surface of the root. The hyphae penetrate the

intercellular spaces of the root cortex to form a network called the "***Hartig net***". The root cells secrete sugars and other food ingredients in to the interstellar spaces for feeding the fungal hyphae. The exposed fungal hyphae increases the surface of the root to several times and performs the several functions such as;

I. Absorption of water

II. Solubilisation of organic matter of the soil humas, release of inorganic nutrients, absorption and their transfer to roots.

III. Direct absorption of minerals from the soil over a large area to provide it to root.

IV. It protects plants from disease – inciting pathogens by secreting antimicrobial substances –

Ectomycorrhizae occur in several plant genera like *Pinus, Quercus* (Oak), *Betula, Alnus, Salix, Populus, Eucalyptus, Peach* etc.

The fungal partner is generally specific – **Basidiomycetes.**

Complex organic food ⟶ Simple organic food⟶ Absorb by root hairs

2. **Endomycorrhizae**: (Endotrophic mycorrhiza) Here thee fungus does not form an external mantel but lives in the intercellular spaces as well as intracellularly in the cortical cells of roots. Only a small portion of fungus lives outside the root. This type of association generally occurs in roots of grasses, cereals, legumes, rubber, tea, tobacco, soybean, citrus and in several orchids like *Neottia, Vanilla, Galeola, Epipogon* etc.

- In seedling stage of orchids, the fungal hyphae also provide nourishment by forming nutrient rich cells called ***pelotons.***
- Pelotons degenerate and supply the orchid with carbohydratesand other nutrients obtained from the outside.

One of the important type of Endomycorrhizae is **Vesicular – arbuscular mycorrhizae** (***VAM***). In this association, the fungal mycelium forms some special kind of organs called vesicles and arbuscules, either within the intercellular spaces or cortical cells of the root. They serve as the food storage organs of the fungus.

- **Vesicles** are thick-walled, swollen structures

- **Arbuscules** are dichotomously branched haustorial branches of mycelium

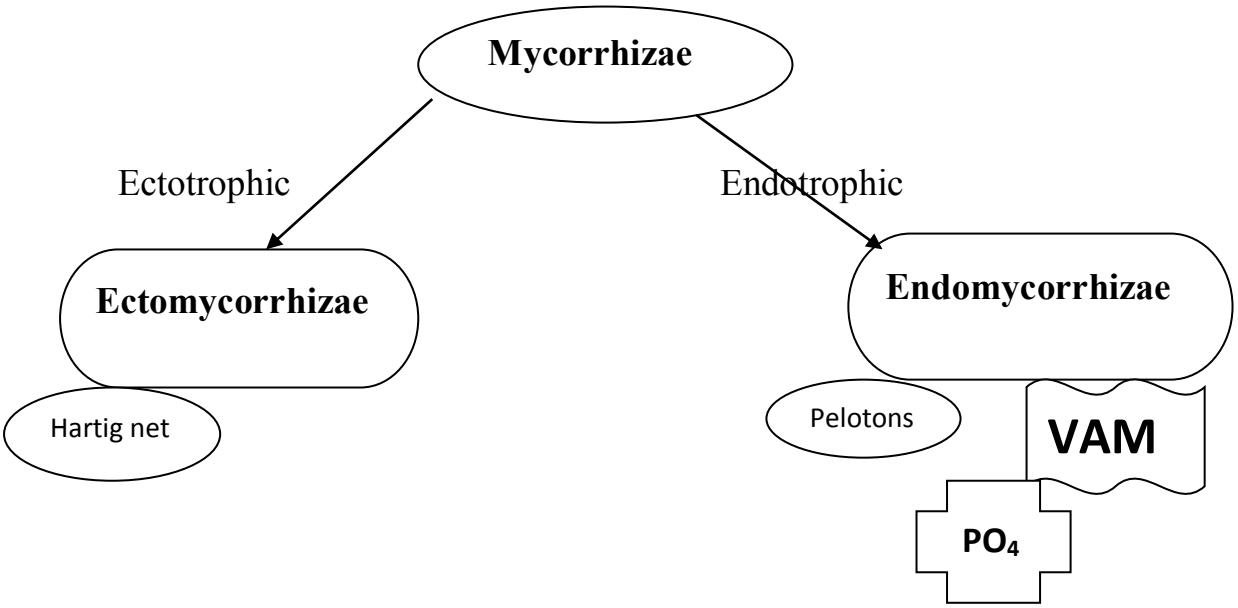
Endomycorrhizae :

a). stimulate the absorption of phosphorus, zinc, copper, sulphur, potassium and various other elements by roots.

b). enhance nodulation in legumes.

c). secretion of antimicrobial substances.

d). VAM is very important in PO_4 nutrition of plants.



- The most common fungal partners of mycorrhiza are *Glomus* sps.
- Plants with ectomycorrhiza are known to absorb 2 – 3 times more of nitrogen, phosphorus, potassium and calcium.

Advantages of biofertilizers:

- Increase the yield of plants by 15 – 35 %.
- Effective even under semi - arid conditions.
- Preparation of inoculam is easy.
- Improve soil texture (water holding and buffer capacity).
- Release antimicrobial secretions so harm pathogens.

- Produce vitamins (Vit.B12) and growth promoting biochemicals (Auxins, Ascorbic acid).
- Cheap and economical.
- Excrete antibiotics so act as biopesticides.
- Pollution free, causing no atmospheric pollution.

- **Genetically Modified Organism – Bt crops**
- **Biopiracy and patents**

Biotechnology and Agriculture

Biotechnology is the technological utilization of biological process including genetic engineering for obtaining useful materials for agriculture, industry, domestic, medical and other uses.

- The first use of biotechnology must have been in pre-historic time when humans discovered fermentation for alcoholic beverages and dairy products.

Biotechnological applications were applied in agriculture with the option to increase food production. There are three options to increase the food production as:

1. Agrochemical based Agriculture
2. Organic Agriculture or organic Farming
3. Genetically Engineered Crop based Agriculture

1. **Agrochemical based Agriculture**: The green revolution succeeded in increasing the yield of crops mainly due to;

- Use of improved varieties of crops.
- Use of agrochemicals i.e; fertilizers and pesticides.

- With the passage of time it was found that it is not sufficient to feed the fast growing human population.

3. **Organic Agriculture or organic Farming**: In this type of farming, farmers use manures, biofertilizers, biopesticides and biocontrol measures in order to increase the crop production.

- Later on it was also noticed that organic farming cannot increase the yield of crop up to mark.

3.**Genetically Engineered Crop based Agriculture**: In order to increase the plant yield/ food production, the genetically modified crops were introduced by incorporating the desired genes from the genetically modified organisms.

Genetically Modified Organisms (*GMOs*)

Plants, bacteria, fungi and animals whose genes have been changed by manipulation are called Genetically Modified Organisms (GMOs)

A **genetically modified organism** (**GMO**) is any organism whose genetic material has been altered using genetic engineering techniques.

A wide variety of organisms have been genetically modified (GM), from animals to plants and microorganisms. Genes have been transferred within the same species, across species (creating transgenic organisms) and even across kingdoms. New genes can be introduced, or endogenous genes can be enhanced, altered or knocked out.

- Herbert Boyer and Stanley Cohen made the first genetically modified organism in 1973, a bacteria resistant to the antibiotic kanamycin.
- The first genetically modified animal, a mouse, was created in 1974 by Rudolf Jaenisch, and the first plant was produced in 1983.
- In 1994 the Flavr Savr tomato was released, the first commercialized genetically modified food.
- The first genetically modified animal to be commercialized was the Goldfish (2003) and the first genetically modified animal to be approved for food use was the Aqua vantage salmon in 2015.

Creating a genetically modified organism (GMO) is a multi-step process. Genetic engineers must isolate the gene they wish to insert into the host organism. This gene can be taken from a cell or artificially synthesized. If the chosen gene or the donor organism's genome has been well studied it may already be accessible from a genetic library. The gene is then combined with other genetic elements, including a promoter and terminator region and a selectable marker.

- In plants the DNA is often inserted using *Agrobacterium*-mediated recombination, biolistics or electroporation.

- As only a single cell is transformed with genetic material, the organism must be regenerated from that single cell. In plants this is accomplished through tissue culture

- The first medicinal use of GM bacteria was to produce the protein insulin to treat diabetes. Other medicines produced include clotting factors to treat haemophilia, human growth hormone to treat various forms of dwarfism, interferon to treat some cancers, erythropoietin for anemic patients, and tissue plasminogen activator which dissolves blood clots.
- Outside of medicine they have been used to produce biofuels

Genetically Modified Crops – *GM Crops*

Crops in which foreign genes have been introduced through genetic engineering are called Genetically Modified Crops (GM Crops). OR Genetically modified crops are transgenic crops that contains and expresses a transgene.

The adventages of transgenic crop techniques are:

i). any gene of an organism or synthetic gene can be used for transfer.

ii). the addition of transgene in to the crop genome can control the change in the genotype.

Adventages of the transgene are:

a). It can produces a protein that is the product in which we are interested.

b). It can produce a protein that can produce desired phenotype.

c). It can modify an existing biosynthetic pathway as a result new end-products are produced.

d). It can prevents the expression of an existing native gene.

Transgenic Plants

Plants in which foreign genes have been introduced through genetic engineering.

There are two techniques for introducing foreign genes (Transgenes) in to the plant cell genome:

1. Through Vector
2. Direct introduction of DNA

Production of transgenic plants

Interspsecific gene transfer is now carried out through genetic engineering. It can be done by:

I. Ti plasmid (Tumour inducing).

II. Ri plasmid (Hairy Root inducing/ Crown gall tumour inducing).

I.Ti plasmid: Ti plasmid from the soil bacterium Agrobacterium tumefaciens is used as vector for gene transfer to plant cells (tomato, tobacco and soybean) because having ability to transfer DNA in to plant cells.This bacterium is also called **natural genetic engineer** because the genes carried by its plasmid produce effect in several parts of the plant.

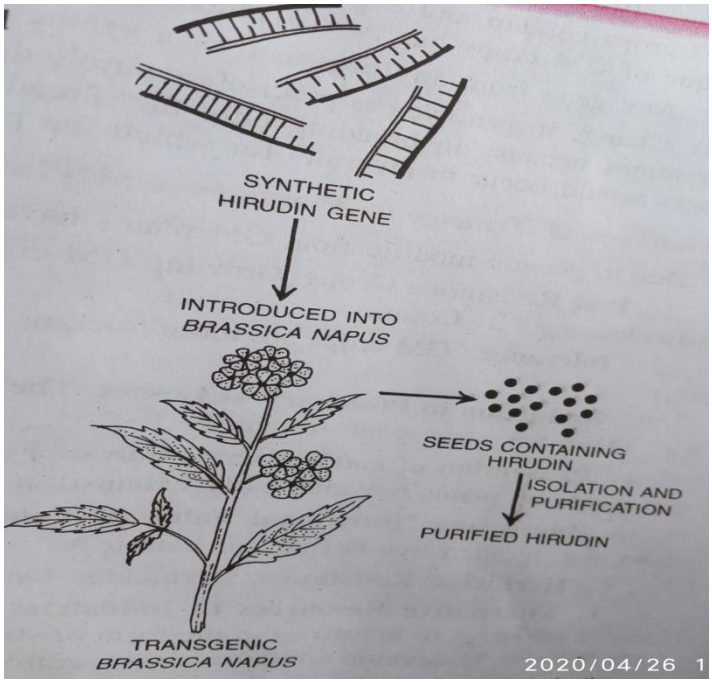
II. Ri plasmid: Ri plasmid of *Agrobacterium* ***rhizogenes*** is also being used as vector for gene transfer in plants including tomato, soybean, sunflower and cotton.

- **For the genetic engineering purposes, Agrobacterium strains are developed in which tumour forming genes are deleted**.

The part of Ti plasmid transferred in to plant cell DNA is called T-DNA. This T-DNA with desired DNA spliced in to it, is then inserted in to the chromosomes of the host plant where it produces copies of itself by migrating from one chromosomal position to another without inducing tumors. Such plant cells are then cultured induced to multiply and differentiate to form plantlets. Then transferred in to soil, as to grow as plants with desired foreign gene (character).

Some achievements of transgenic techniques are:

A. **Hirudin**: Hirudin is a protein that prevents blood clotting. The gene encoding hirudin was synthesized chemically and then incorporated in to the ***Brassica napus***, where it accumulates in seeds. It is used as medicine after purification.

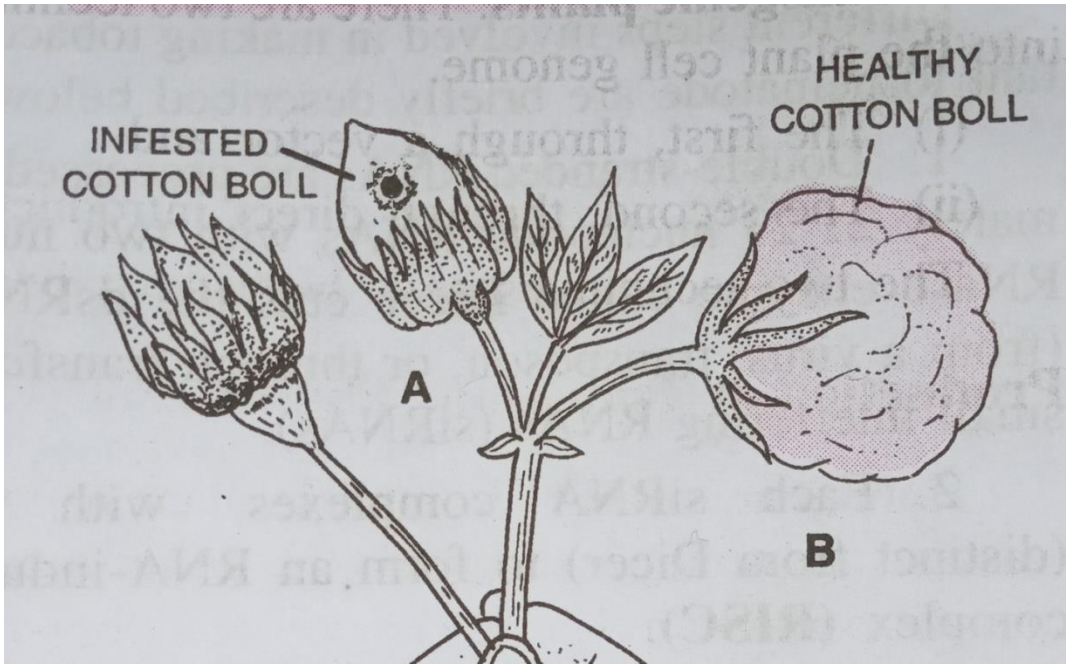


Production of Hirudin from transgenic ***Brassica napus*** seeds

B. **Cry Protein**: Cry or Crystal protein is produced by soil bacterium ***Bacillus*** *thuringiensis*. It is toxic to larvae of certain insects. The gene encoding cry protein (Cry gene) has been isolated and incorporated in to the several crops,such as **Cotton** resulting in the resistance to the group of insects for which cry protein is toxic.

Two cry genes, *cry* IAc and *cry* IIAb have been incorporated in cotton. The genetically modified crop is called **Bt cotton** as it contains Bt toxin genes.

- The genes *cry* IAc and *cry* IIAb control cotton **bollworms.**
- Cry IAb has been introduced to control cotton from **corn borer.**
- *cry* proteins kill various insects like Lepidopterans (order of class insect—tobacco budworm, army worm), Coleopterans (beetles) and Dipterans (flies, mosquitoes).



Cotton boll – A. Destroyed by boll worms **(B).** A fully mature cotton boll-uninfected

Why does Bt-toxin not kill the *Bacillus* bacterium-Bt.

The Bt toxin proteins exist as inactive protoxins but once an insect ingests the inactive toxin it is converted in to an active formof toxin due to the alkaline pH of the alimentary canal that solublizes the crystals. The activated toxin binds to the surface of midgut epithelial cells and create pores which cause cell swelling and lysis and finally death of the insect.

Bt cotton farming has shown good results in Malwa region of Punjab. It will save water starved Malwa region from turning into desert as cotton which needs much less water, will replace paddy.

C. **Flavr Savr**: Flavr savr is a transgenic variety of **tomato** where expression of a native tomato gene has been blocked. Fruit softening is promoted by the enzyme ***polygalacturonase,*** which degrades pectin. As in this transgenic tomato variety, production of ***polygalacturonase*** was blocked, therefore, its fruits remain fresh and retain their flavour much longer than other normal tomato varieties. The fruits have also superior taste and increased total soluble solids.

D. **Tobacco**: *Meloidogyne incognitia* (Root-knot nematode) infect the roots of tobacco plants so causes great yield reduction. Through RNA interference technique a transgenic variety of tobacco plant was developed which was highly resistant to the nematode infection.

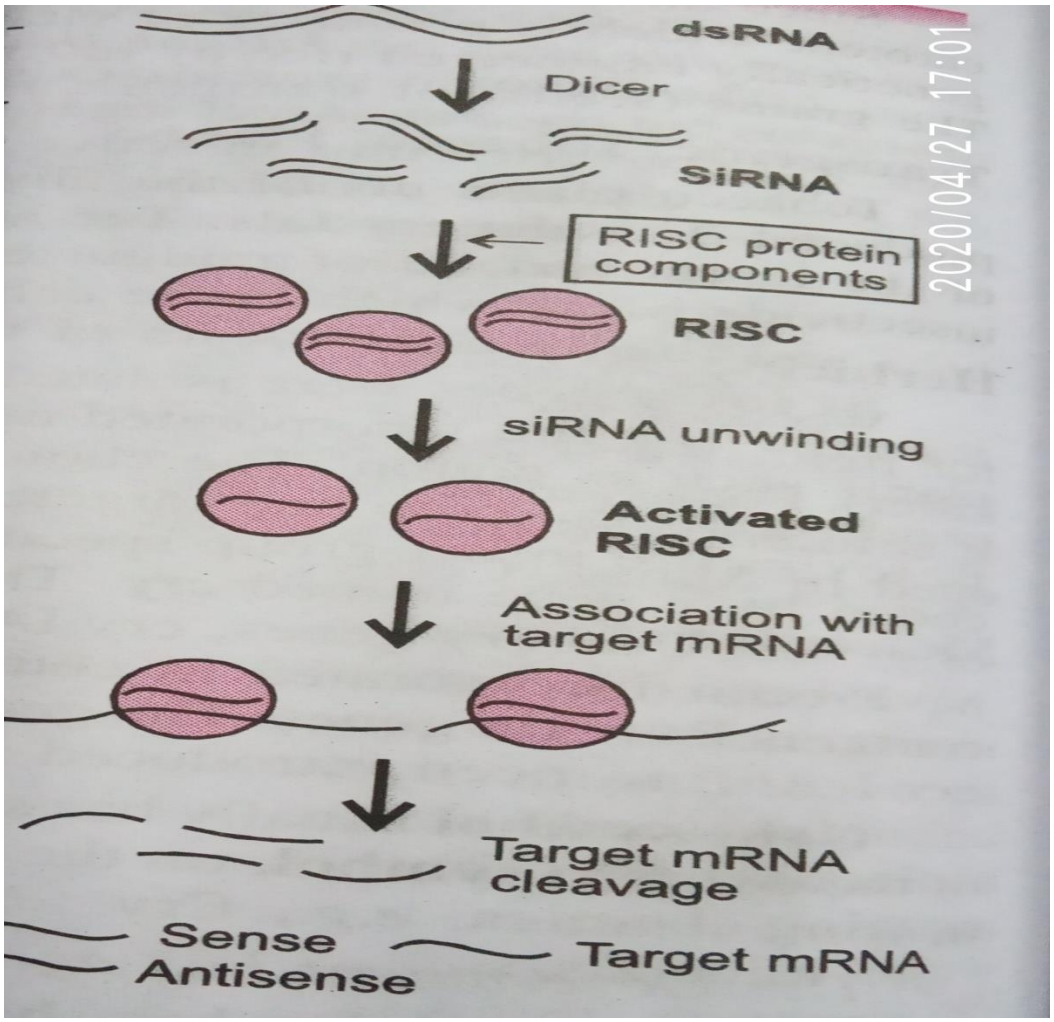
- **Fire and Mello- 1998,** developed a strategy to prevent this infestation that was based on the process of RNA interference (RNAi).RNAi takes place in all eukaryotic organisms as a method of cellular defense. This method involves silencing of a specific mRNA.
  - Using Agrobacterium vectors, nematode specific genes are introduced in to the host plant (tobacco plant). This produces both sense and anti-sense RNA in the host cells. These two RNAs being complementary to each other formed a dsRNA (double stranded RNA) that initiated RNAi**.**
  - **[Fire and Mello-** were awarded **Nobel Prize in 2006** for their discovery of ***RNA interference gene silencing*** by double stranded RNA (dsRNA)].

RNA interference in Tobacco plant

It can be done as following:

1. First double stranded RNAs are processed in to approximately 21-23 nucleotide RNAs with two nucleotides. An RNase enzyme called Dicer cuts the dsRNA molecules in to small interfering RNAs (siRNAs)
   - Dicer cuts the dsRNA molecule through a virus, transposon or transformation.
   - Transposon - A DNA sequence that can change its position within a genome, sometimes creating or reversing mutations and altering the cell's genetic identity and genome size.
2. Each siRNA complexes with ribonucleases to form RNA- induced silencing complex (**RISC**).
3. The siRNA unwinds and RISC is activated.
4. The activated RISC targets complementary mRNA molecules. The siRNA strands act as guides where the RISCs cut the transcripts in an area where the siRNA binds to the mRNA. This destroys the mRNA.
5. When mRNA of the parasite is destroyed no protein was synthesized. It results the death of the parasite/ nematode in the

transgenic host. Thus, in this way the transgenic plant got itself protected from the parasite.



Steps in RNA interference (RNA)---[Tobacco plant]

6. Golden Rice: Golden rice is a transgenic variety of rice (Oryza sativa) which contains good quantity of **β- carotene** (provitamin A – inactive state of Vit.A) . **β- carotene** is a principal source of Vit.A. Since the grains (seeds) of the rice are yellow in colour due to β- carotene , the rice is commonly called ***Golden rice***. The genetically engineered rice was produced by introducing three genes associated with synthesis of carotene. The seeds / grains of transgenic rice are rich in provitamins.

- As β- carotene is converted in to vitamin A so golden rice is rich in Vit.A.
- The transgenic golden rice was produced by **Prof. Ingo Potrykus** and **Peter Beyer**.

- Deficiency of Vit.A causes night blindness and skin disorder.

Seeds of Golden Rice

. Crops have been inoculated with Rhizobia (and more recently *Azospirillum*) to increase their production or to allow them to be grown outside their original habitat. Application of *Bacillus thuringiensis* (Bt) and other bacteria can help protect crops from insect infestation and plant diseases. With advances in genetic engineering, these bacteria have been manipulated for increased efficiency and expanded host range. Markers have also been added to aid in tracing the spread of the bacteria. The bacteria that naturally colonize certain crops have also been modified, in some cases to express the Bt genes responsible for pest resistance. *Pseudomonas* strains of bacteria cause frost damage by nucleating water into ice crystals around themselves. This led to the development of ice-minus bacteria, that have the ice-forming genes removed. When applied to crops they can compete with the non-modified bacteria and confer some frost resistance

[143]

Come to Know!!!

- Tobacco was the first plant to be altered using genetic engineering and is considered a model organism for not only genetic engineering, but a range of other fields.[140] As such the transgenic tools and procedures are well established making tobacco one of the easiest plants to transform.
- Another major model organism relevant to genetic engineering is *Arabidopsis thaliana*. Its small genome and short life cycle makes it easy to manipulate and it contains many homologues to important crop species.[142] It was the first plant sequenced, has a host of online resources

Some genetically modified plants are purely ornamental. They are modified for flower color, fragrance, flower shape and plant architecture.The first genetically modified ornamentals commercialized altered color. Carnations were released in 1997, with the most popular genetically modified organism, a blue rose (actually lavender or mauve) created in 2004. The roses are sold in Japan, the United States, and Canada.[148][149] Other genetically modified ornamentals include *Chrysanthemum* and *Petunia*.As well as increasing aesthetic value there are plans to develop ornamentals that use less water or are resistant to the cold, which would allow them to be grown outside their natural environments

Blue Rose

Drawbacks of Transgenic Plants

The drawbacks may be Environmental as well as Human health risks.

A. Environmental hazards:

1. The transgene may be transferred through pollen from these crops to their wild relatives, thus making the weeds more persistent and damaging.
2. The transgenic crops may themselves become persistent weeds – Super weeds i.e; become herbicide tolerant.
3. The transgenic plants may damage the environment in some yet known manner. It is feared that these transgenic plants may lead to the huge loss to the various important species of insects.

B. Human health risks:

1. The transgenic foods may cause toxicity and allergies.
2. Transgenic foods may cause incorporation of antibiotic resistance gene in to the bacteria present in the human alimentary canal.
3. Transgenic foods have economic concerns due to their high cost (lengthy and costly process).

YES

- First transgenic plant was a tobacco plant which was created in 1983; it was resistant to an antibiotic.
- First transgenic cereal plants were maize and wheat, produced in 1990 – 92.
- Flavr Savr tomato was the first transgenic variety to reach the market.

BIO – PIRACY

(Stealing of Natural Patrimony)

***Bio-piracy* is the use of biological and genetic resources indigenous to a country by another (pirate) country. OR**

Many organizations and multinational companies exploit and or patent biological resources of other nations without proper authorization from the countries concerned, this is known as ***Bio-piracy*.**

TO Steal the bio-resources of a country is unethical

The process of biopiracy involves collection of samples of bio-sources, which can be done unnoticed. The biological material is the subjected to product development for use on a commercial scale. A wide range of bio resources are facing biopiracy. These include;

- **Soil microorganisms**
- **Plants**
- **Animals**
- **Genetic material**

  These resources are being used in research work besides in making new products. It is a concrete fact that;

The industrial nations are rich in technology and financial resources but poor in biodiversity and tradtional knowledge related to the utilization of the bio-resources, in contrast , The developing countries are poor in technology and financial resources but are rich in biodiversity and traditional knowledge related to bio-resources.

The worldwide opposition to bio-piracy is rapidly building up as the people become aware that the western countries are getting great benefits from using

the knowledge and experiences of the 3rd. world communities.e.g; Asia, Africa and Latin America have the highest biological resources while the companies make huge revenues from the process and the local communities have to pay high prices in order to get products of these companies.

Exploitation of Bioresources

Institutions and companies of industrialized nations are collecting and exploiting the bio resources, as follows:

- **Collection and patenting the genetic material by themselves**. E.g; A patent granted in U.S.A. covers the entire ***Basmati*** rice germplasm indigenous to our country.
- **Patenting the bio- molecules and using them for commercial activities.**
- **Patenting the useful genes, isolated from the Bioresources which are used to generate commercial products.**
- **Traditional knowledge related to Bioresources is utilized to achieve the above mentioned objectives.**

  So in some cases the traditional knowledge itself may be the subject of a patent**.**

  - A west African plant, ***Pentadiplandra brazzeana,*** produces a protein called ***brazzein,*** which is approximately 2000 times as sweeter as sugar and is a low calorie sweetener. Local people have known and using the super berries of this plant from centuries.
  - The protein brazzein was patented in U.S.A. Subsequently, the gene encoding brazzein was also isolated, sequenced and patented in USA.
  - It is proposed to transfer the brazzein gene in to maize and express it in maize kernels. These kernels will then be used for the extraction of brazzein.
  - This development could have serious implications for countries exporting large quantities of sugar.

  Bio-resources of the developing world have always been commercially exploited by the industrialized nations without an adequate compensation to the developing world. This exploitation has dramatically increased in pace.with the development of powerful analytical tools and techniques. There has been a growing realization of this injustice and demands are

being made for adequate compensation and benefit sharing. Some nations are developing compensive laws to prevent unauthorized exploitation of their Bioresources and traditional knowledge.

- India "***Sparrow of Spices***" being rich in tradition, communal knowledge and expertise in natural medicines, spices, food preparations, biopesticides and diverse agriculture is now under siege from biopirates. Through patenting without consent, foreign companies have collared at least 22 plants for their beneficial derivatives. Patents have been taken out on plants such as **black pepper** (*Piper nigrum*), **basmati rice** (*Oryza sativa*)**, Indian mustard** (*Brassica campestris*), **pomegranate** (*Punica granataum*), **turmeric** (*Curcuma longa*) and **neem** (*Azadirachta indica*).
- US, Japan and German companies are the principal patenting pirates.

Control Measures

Although our government has passed a law to regulate accesses to our biological and genetic resources but it lacks steps to implicate and prosecute the biopirates.

The non-seriousness of our governments about checking bio-piracy can be guessed when they signed the **General Agreement on Tariffs and Trade** (GATE) , which open country's natural resources for foreign exploitation.

PATENT

Patent is the right granted by to an inventor to prevent others from commercial use of his invention.

Bio-patent is a government protection to an inventor of a biological material, securing to him for a specific time the exclusive right of manufacturing, exploiting, using and selling an invention.

Primarily, industrialized countries such as USA , Japan and members of European Union are awarding bio-patents.

- The effect of biopiracy will be maximum on india as it is the worlds richest country in biodiversity (81000 sps. Of fauna, 47000 sps. of flora).

Biopatents are awarded for the following:

i. Strains of microorganisms.
ii. Cell lines.
iii. Genetically modified strains of plants and animals.
iv. DNA sequences.
v. Proteins encoded by DNA sequences.
vi. Biotechnological procedures.
vii. Production processes.
viii. Products and product applications.

Bio-patent Abuse

The companies are being granted patents for products and technologies that make use of biological resources such as genetic materials, plants and animals, which have long being identified, developed and used by farmers and indigenous peoplepeople.T TheThe

people**.** The transnational companies are rushing to get patents of new products derived from bio-resources as to prevent competitors from using them so they reap larger profits by hiking of prices of the products or by charging royalities to other firms according to their wishes.

Opposition to Bio-patent

There are strong protests from farmers and indigenous people against the grant of patent rights to companies. As it is of course these communities which originally identified and evolved use of plants for food, medicines and other purposes. So they are the right owners. The companies are now claiming exclusive right to produce and sell these materials.

U.S Patent Law

It does not recognize technologies and methods in use in other countries as "prior art" if knowledge is new for U.S, it is novel, even if it is a part of an old practice in other countries.

Success in compaign against Bio-piracy -- *Control measures*

i). With the efforts of "Vandana Shiva" the leaders of "Neem Compaign". The European patents to transnational corporation for pesticides were cancelled.

ii). Success of Indian govt. in getting cancelled the patent granted by the U.S patent office on the healing properties of turmeric.

1. Define Sewage. Write down its harmful effects.
2. What is bio-gas. Write down its composition.
3. Write a note on bioinsecticides.
4. Define biofertilizer. Briefly mentions various types of biofertilizers.
5. What is Azolla-Anabaena symbiosis?
6. Bio-patents are opposed. Why?
7. Write a note on River Action Plan.
8. What is Acidogenesis?
9. Define sterilization strategy.
10. Write a note on Mycorrhiza.
11. Write a note on Bt-cotton.
12. Name the institutions who developed biogas technology in India.
13. Name some petroleum plants.
14. List various types of pesticides.
15. What are transgenic plants?

1. In India, per day production of sewage is:

(a). 1000 million gallons (b). 8oo million gallons (c). 500 million gallons

(d). None of above

2. The calorific value of biogas is:

(a). 20 -25 Mj/m3 (b). 15 -20 Mj/m3 (c). 23 – 28 Mj/m3 (d). None

3. Conversion of organic acids into Methane:

(a). Solubilisation (b). Acidogenesis (c). Methanogenesis

(d). All above

4. Baculo viruses:

(a). bio-herbicides (b). bio-insecticides (c). both a and b (d). only b

5. Ecdysone:

(a). molting hormone (b). juvenile hormone (c). growth hormone

(d). None

6. VAM:

(a). Ectomycorrhizae (b). Endomycorrhizae (c). Vesicular- Arbuscular Mycorrhiza (d). Both a and b

7. "Hartig net":

(a). Ectomycorrhiza (b). Endomycorrhiza (c). Both a and b (d). None

8. Hirudin was incorporated in:

(a). Cotton (b) . *Brassica napus* (c). golden rice (d). Tomato

9. Polygalacturonase degrades:

(a). Cellulose (b). Pectin (c). Lignin (d). All above

10. *brazzein* is:

(a). Enzyme (b). Protein (c). Sugar variety (d). Vitamin

UNIT - IV

ECOLOGY AND ENVIRONMENT

Ecology:- No organisms lives by itself alone. It most lives in the same environment with organisms of its kind and with those of other species. It may eat other organisms and be eaten by other organisms and compete with other for the necessities of life such as space, food etc. it most also continuously make adjustments' to its physical environment. Every organism is thus influenced by the life and physical environment around it. Eernest Hackel a germen naturalist defines ecology as total relationships of organisms with the living and non living environment is called ecology. Today ecology is defined as the total relationships of organisms to each other, to environment end to each. The entire energy within a given ecosystem.

Ecology is derived from a Greek word '*okios'* meaning house or place to live and '*logos'* to study. It is also called bionomics or bionomies (French). So it is the study of organisms at home.

Scope:- Ecology is the science that needs minimum time and labor for introduction

Environment:-the term Environment etymologically means surroundings .thus environment is a complex of so many things (light, soil, light plants and Animals temperature water etc) which surrounds organisms.

So environment can be defined as the aggregate of all the external conditions and influences(both biotic and abiotic) affecting the life and develop. of an organism in its natural habitat, and in absence of organisms it is meaningless to speak of environment and vice versa.

Habitat:- the term habitat indicates a specific place where a species or population normally lives in nature. It is physical area, some particular part of earth's surface soil water air. Habitat includes both living and non living objects e.g Rabbits habit is grass land and open wood land. Animals often live in specific habitat having a particular environment conditions e.g Amphibians occur in fresh water only.

Ecological Niche:- The word niche literally means a specific place. However ecologists use it in place of habitat along with role of species (e.g What is eats, its range of movement etc.)

Gauses principle:- There is usually a single species in an ecological niche of any given habitat. If two species are sharing the same ecological niche. The direct competition between two will eliminate one of them which is called competitive exclusion.

Habitat	Ecological niche
1. It refers to specific physical area Where a species lives.	1. It refers to the role a species plays in its habitat.
2. A habitat has number of niches.	2. A niche does have components
3. Habitat of species usually does not change.	3. . A species may change its niche with due course of time.

Organism and Environment:- the place of living of organism is called habitat. Every habitat has its own peculiar type of environment. The individual conditions such as light, air water, minerals, pre, and predator around an organism are termed as factors of the environment. The environmental factors can be classified into two types abiotic (physical) and biotic factors. The biotic factors are living creators present in the organism's environment and are referred as biological factors. The biotic factors influence both the physical environment as well as the other organisms living in it.

Environment means surrounding of an organism. Also our earth have three components namely atmosphere, hydrosphere and lithosphere where living and non-living organisms interact with each other. In words the different components of the environment are interlinked and interdependent.

Ecology is basically concerned with four level of biological organization - organism, population, communities and ecosystem and biomes.

Hirechery in nature:- the word "Hierarchy" means any gradual organization of persons, principles or things ranked one above the above in ascending order type.

1. **Physical Hierarchy**:- Every organisms consists of particles that can be arranging in progressing increasing level of organization complexity. Each higher level is more complex than the one below it. The smallest unit of matter both living and non living are protons neutrons and electrons → Atoms etc.

2. **Biological Hierarchy**:-Biological Hierarchy begins with cell. A cell forms tissues → organs → I Organ → system → organism. At each level there is exchange of material between the unit and its environment.

3. **Ecological Hierarchy**:- Ecology level begins at the level of organism and proceeds to the level of grater complexity namely population, biotic community and ecosystem and biomes.

1. **Organism**:- an organism is a living unit in nature. It has the following characteristics. (A)One organism is as growing, self regulating, self repairing capable of autonomous movement. (B) Organism are fully adopted to their environment and acts as the smallest unit of ecological hierarchy

2. **Population:-** a group of individual of the same species living together in a common area of a particular time forms population fo that area. Members of the local population are adapted to their specific environmental conditions such a population is called ecotype.

3. **Biotic community**:-Groups of organisms belonging to several different specie that live together in the same area or habitat interact through tropic spatial relationships to form a self sustained unit is called biotic community. A biotic community has three sub units called an animal community, plant community and microbial community. A forest is the example of biotic community.

4. **Ecosystem and biome:-** living organisms cannot be isolated from their non living environment because two interacts to produce a stable unit. A natural self sufficient unit comprising of biotic community and its abiotic environment is called ecosystem. And the geographical area producing uniform conditions for life is called biotype ie. An ecosystem has two parts viz physical part or biotype and living part or biotic community.

5. Biome: - very large ecosystems are termed as biomes. Infact variation in intensity and duration of temperature and annual precipitation accounts for the formation of biomes e.g desert, rain forests etc.

6. Biosphere (Ecosphere). All the ecosystems of the world together constitute biosphere or ecosphere.

Non living:

Protons → neutrons → electrons

↓

Atoms → Compounds

↓

Complex of compounds

↓

Cells

↓

Living:-

Tissues → organs

↓

Organism ← Organ system

↓

Ecological Hierarchy:-

Population

↓

Biotic Community + Abiotic factor

↓

Ecosystem → Biosphere

Environmental Factors:- some ecologists classifying various environmental factors into two major groups.

1. **Direct factors:-** As the name indicated these factors have direct influence on the organism eg. Temperature, light, Humidity, Soil nutrients, soil moisture, air etc.
2. **Indirect factors**:-these factors have indirect influence on organisms. Thus modify other factors eg. Soil texture, precipitation. Soil organisms etc.

On the basis of their nature, ecologists classify various environmental factors into two major categories.

1. **Abiotic factors**: Non living factors which includes temperature, light, atmosphere. Air, humidity, rain water, soil texture, substratum, topography etc.
2. **Biotic factors**:- These include all the living organisms.

1. **Atmosphere (Air)**:- atmosphere is thick gaseous envelop called air, which surrounds the hydrosphere and lithosphere. Composition of air remains quit uniform up to the height of 89 km. higher levels has lighter gases. The atmosphere has a layer of ozone between altitudes of 16-50km. Air contains different gases in different amounts

Nitrogen = 78%
oxygen = 20.95%
CO2 = 0.03%
Tracer gases = 0.04% ,
It is invisible, order less, tasteless mixture of gases.

Direct and indirect effects of air on organisms

I. **Plants:-** 1. Pollination: - air helps wind pollinated flowers in pollination 2. Dispersal of seeds 3. Transpiration:- wind increases transpiration . hot dry wind kills young shoots and damages the buds and fruits. Persistent strong wind reduces the height of plants.

II. **Animals**:- The wind effects the distribution of small animals.

Indirect effects:- indirect effects of wind is in the form of desiccation of plants and dwarfism.

2. **Water**:- water is a retable source. Snow rain, sleet are the natural sources of water.

   Importance:- water is considered as matrix for life. All the metabolic process of plants and animals carry water as a constituent.
3. **Humidity**:-Humidity refers to the moistures content of air. It determines the formation of clouds, dew and fog. It affects the land organism's bearing loss of water as vapours from their bodies through evaporation, transpiration. Humidity effects animal life indirectly also by influencing plant life. Higher humidity provides more normal life.

4. **Precipitation**:- precipitation means fall of rain, snow, sleet, hail or dew. Total annual rainfall, seasonal distribution of rain, humidity of air, amount of retained in the soil is the main aquatic criteria that limit the distribution of plant and animals on land.

5. **Light:-** we all know that sun is ultimate source of energy to all living organisms. Light in the visible range of electromagnetic spectrum (390- 680) and light of (400 – 700) is effective in photosynthesis and is called photo synthetically active radiation (PAR) . Impact of intensity. , duration and quality of light controls number of process organism's in many respects.

1. **Sources of energy**:- Light is the source of all energy synthesized for photosynthesis in green plant.

2. **Photoperiodsm**:- a response of plants to day light is controlled by light.

3. **Pigmentation:-** intensity of light changes the skin color of various organisms. Bright results in light color and dim light in darker colors in frog and lizard. Light is also responsible for the synthesis of pigments.

4. **Phototropic movements**:- Motile algae shifts towards surface water in dim light and to deep water in strong light.

5. **Temperature**:- Degree of hotness, or coldness of a body or subastance is called temperature. Animals are adopted in various ways to temperature.

    1. **Homeotherms**:- Animals with constant body temperature eg. Whales and features in birds

    2. **Poikilotherms:-** organism which maintain their body temperature with the environment temperature.

Adaptation for temperature in plants

1. Plants have certain adaptation for protection from temperature in the form of thick corky bark, thick leaves, high cuticle. Rate of transpiration increase in temperature.

2. **Soil:** the fertile surface layer of earth capable of supporting plant growth is called soil. It is dynamic layer in which many chemical, physical and biological activities are going on constantly. Study of soil is called pedology.

3. **Weather:-** The short term properties of atmosphere such as temperature , rain fall, pressure, sunshine , cloud cover, wind at a given place and time is called weather. Weather reflects the hourly, deeply or weakly changes in these factors.

4. **Climate:-** it refers to average weather conditions of a particular area. Temperature and rain fall are the most important factors which determine the diameter of an area.

Population Characteristics

Population characteristics:- the population is a group of organisms of the some species occupying a particular has the following characters.

1. **Size and density:-** Total size is generally expressed as the number of population in a population. More information are estimated of density which is defined as the number of individuals per unit area(volume) of the environment eg.

    Large organisms as trees may be expressed as 500 trees per hectare whereas as the smaller ones like phytoplankton as 2 million algae per cubic meter of water.

TYPES:

i. **Crude density**:- it is the density (number or biomass) per unit space.

ii. **Specific density or ecological density**:-it is the density per unit of habitat space i.e available area or volume that can actually colonized by the population.

2. **Growth rate**:-since population Is changing entity. As varying from place to place population density may remain constant, fluctuate or may steadily increase or decrease. Natility and immigration lead to an increase in density, while motility and emigration lead to decrease in population density. This growth rate is defined as the average rate of change in the number of organisms per unit of time and given by

2: Age structure or distribution:- in most of cases individuals are of different ages. The proportion of individuals in each group is called age distribution or structure of the population. e g. A palm tree population in an evergreen forest of Mexico had 50% of individuals as seedlings (less then 2yrs old) 19% as saplings (8 year old) 5% as 30 years old adults and so 9% up to 70 years old .

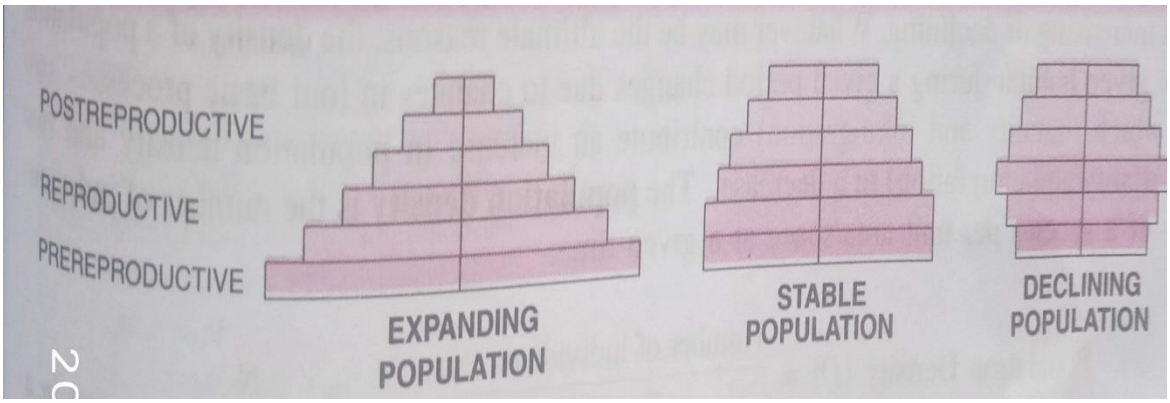
Age distribution is important as it influences both natility and motility of the population. The ratio of the various age groups in a population determines the current reproductive status of the population. Thus anticipating its future. From an ecological point of view, there are three major ecological age group in any population i.e; pre reproductive, reproductive and post reproductive. The relative duration of these age groups in proportion to life span varies greatly with different organisms. In man three ages are relatively equal in length. Some insects have very long pre- reproductive periods, very short reproductive periods and no post reproductive periods.

3: Age pyramids:- The modal representing geometrically the proportions of different age groups in the population of any organism is called age pyramids. 3 hypothetical types are used to indicate the proportions of individuals of various age groups.

1. **A pyramid with board base**: - it indicates a high percentage of young individuals. In rapidly growing young populations birth rate is high and population may be exponential as in yeast, housefly paramecium etc.

2. **A bell shaped polygon**: - it indicates a moderate proportion of young to old. As the rate of growth become slow and stable i.e; Pre reproductive and reproductive age group becomes more or less stable and equal in size, post reproductive remaining as the smallest. There results a ball shaped pyramid.

3. **An urn- shaped figure**: - it represents the low percentage of young ones. If the birth rate is drastically reduced the pre-reproductive phase becomes short in proportion to other two which indicates that population is dying off.



Age Pyramids for human population

3: Natality: it is simply a broader term covering the production of new individuals of any organism. The new individuals are born , hatched, germinated , arise by division etc. in human population, however, the Natality rate is equivalent to the birth rate. Natality rate is the number of offspring produced per female per unit time. Natality is of two types.

(i) **Maximum Natality (absolute or potential Natality):** it is the theoretical maximum production of new individuals under ideal conditions I,e no ecological limiting factors, reproduction being limited by physiological factor. it is constant for a given population. It is also called fecundity rate.

(ii) **Ecological or realized Natality**: it is also known as simply Natality, which refers to the population increase under an actual existing environmental Conditions. Thus it takes into account all possible existing conditions. It is also called as fertility rate. Natality is expressed as

Three population characteristics determine rate at which females produce offspring.
(a). Clutch size which refers to the number of young ones produced at each event.
(b). The time between one reproductive event and the next.
(c). The age of first reproduction.
Natility usually increases during the period of maturity and then falls again as the organisms gets older.

4: **Mortality:** it refers to the death of individuals in the population. Like Natality mortality is of two types.

(i) **Minimum mortality**: it is also called specific or potential mortality. It represents the theoretical minimum loss under ideal env. Conditions, and individuals would die off only because of their old age.

(ii) **Ecological or realized mortality:** it is the actual loss of individuals under given existing env. Conditions. It is like natility not constant and varies with populations and env. Conditiond

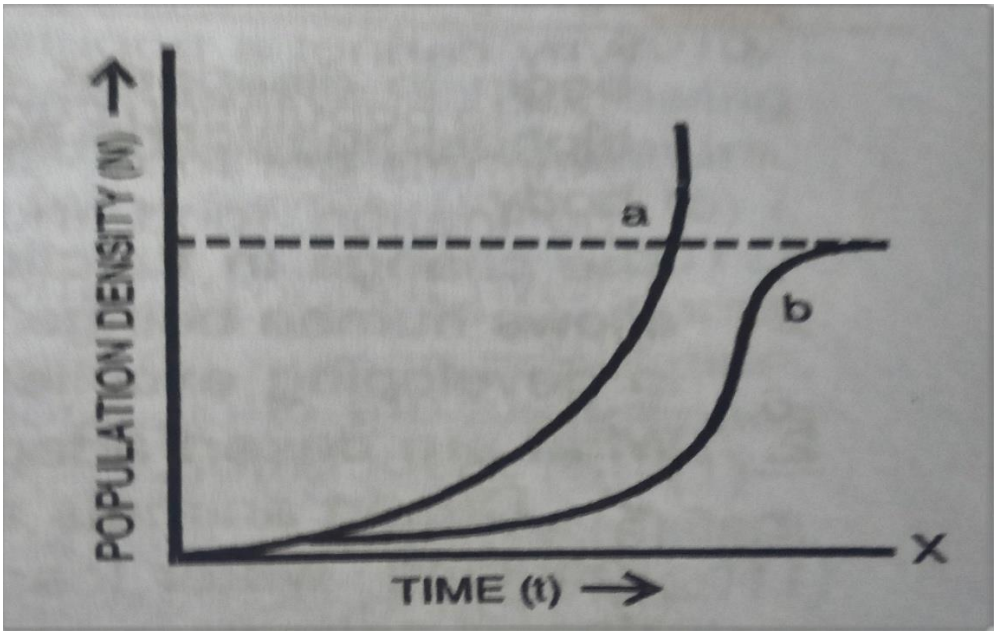
Like natility mortality may be expressed as no. of deaths in a given period of time. A birth- death ratio(100 births/ deaths) is called vital index. For a given population the imp. Thing is not which members are dying off but which nmembers survive. Thus survivou rates ore of much interst than the death rates. Survivor rates are generally expressed by survivorship curves.

Survivorship curves: the pattern of mortality with age is best illustrated with survivorship curves which plots the no. of surviving to a particular age. There are three general types of survivorship curves.

(i) **highly convex curve**: curve A in the figure is the characteristic of the populations in which the mortality rate is low until near the end of lifespan. Here individuals die only of old age e,g Man, deer etc.

(ii) **highly concave curve**: curve C is the characteristic of such species where mortality rate is high during the young stage. E,g oysters, shell fish, oak trees etc.

(iii) **diagonal curves**: if age specific survival is more nearly constant , the curve approaches a diagonal straight line (B2) . it thus shows a constant proportion of organisms dying per unit time. Probably no population in the real world has a constant age specific survival hence the curve becomes somewhat sigmoid (B3) characteristic of many birds, mice and rabbits.



POPULATION INTERACTIONS:

In an ecosystem populations of various species interact with each other in order to form a stable ecosystem Odum 1971 introduced the term symbiosis for all those interactions between the organisms and divided all the relationships into two groups: (i) Positive (ii) Negative Interactions

1. **Positive interactions**:- where populations help one another , interaction being either one way or the reciprocal these include

   (i) **Mutualism** :- Here both the species derive benefit . In such assocociations there occurs a close and often permannant and obligatory contact more or less essential for the survival of each. The two populations enter in somne sort of physiological exchange eg. Pollination by animals as they deriove food i.e nectar and in return bring about pollination. (ii) Dispersal of fruits and seeds by animals while getting food (iii) Lichens (iv) Symbiotic nitrogen fixation and Micorrhizae.

   (ii) **Commensalism:-** It is an association of different species where only one is benefitted and none is harmed eg. Lianas (vascular plants rooted in ground and maintains erectness of their stem by making use other objects for support), Epiphytes etc

   (iii) **Protocooperation**:- It is an association where both the populations are benefitted but the relationship is not obligatory eg. Sea Anemone attached to the shells of the hermit crab.

2. **Negative Interactions** :- where member of one population may eat the members of other population , compete for food , excrete harmful wastes or otherwise interfere with the other populations. These include

(i) **Exploitation:-** He one species hams the other by making its direct or indirect use for support, shelter or food . thus exploitat5ion may be in respect of shelter or food

   1. **Shelter**: - Among animals there are cases where one species of ants use another species for slave labour eg. Polygus workerts raid nests of Fumica and the later feeds and builds nests for the former. Similarily parasitic birds as Cuckoo and cow bird never build their nests and lay eggs in the nests of other birds .

   2. **Parasitism**:- A parasite is the organism living in or on the body of another organism and deriving its food more or less permanently from the host . The relationship can result in killing or without killing the host eg. Some parasitic plants as cuscuta grow on other plants on which they depend for nourishment, lice and bugs on animals , various bacteria and viruses parasitize plants and animals .

3. **Predation**:- It is an association between the two organisms where one organism called predator catches and kills another species for food called prey. Most of the predatory organisms are animals eg. Lion eating deer while as some plants also catch and kill insects and other worm like animals eg. Number of fungi as zoophagous , Dactylela catches and kills insects other example include Nepenthes , Utricularia. Other example includes browsing and grazing by herbivores that kill plants , herbs and shrubs and trees

Animals only the adults are predatory the young are parasites. Predators feed upon the adults, or larvae or even eggs of their prey . A number of fungi such as species of Dactyllaia, zoophages capture insects, nematodes and other worm like animals, such as fungi are specialized structures, the traps formed on their mycelia to capture the nematodes.

Examples

1. **Browsing and grazing:-**Herbivores kill the plants and use in harvested herbs, shrubs or even trees for their food. Different plants receive varying degrees of harmness as a result of browsing and grazing. Generally animals suffer more than plants.
2. **Seeds and seedling destruction**:- Animals such as insects, squirrels, ,mice, rodents etc consume much quality of seeds as found. More ever browse seedling of shrubs and trees and damage most of them by trapping.
3. **Plants as food:-**Aquatic animals use plants in water as food.
4. **Carnivorous plants**:_ A number of *Nepenthes*, *Darlingtonia* consume insects and small animals for their food. They are adapted to remarkable ways to attract, catch and digest their victims.

Flow chart {Interaction between different species in biotic community}

Berk Holder – 1952, has used three basic symbiosis:

I. **[0]** = No significant interaction or impact.

II. **[-]** = growth, survival, population aspects are inhibited**.**

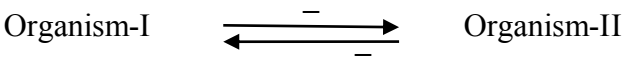
III. **[+]** = growth, survival, population aspects are benefited.

Using these three symbols for two interacting organisms, he classified nine types of interactions;

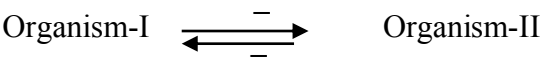
1. **[0,0]** = **Neutralism** = both interacting organisms are neither benefited nor harmed.

Organism-I **0** ⟷ **0** Organism-II

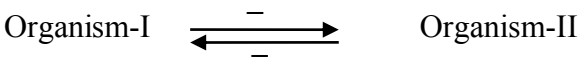
2. **[-,-]** = **Competition** = (Direct competition) = in which whole population directly inhibit each other.



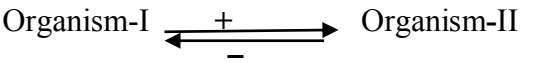
3. **[-.-]** = **Competition** = (Indirect competition) = in which whole population is affected due to resource limitation.



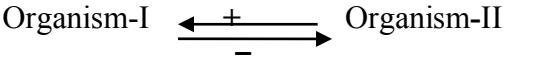
4. **[-,0]** = **Amensalism** = Organism first is inhibited, organism second is unharmed.



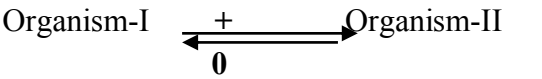
5. **[+,-]** = **Parasitism** = Organism first is benefited, organism second is inhibited.



6. **[+,-]** = **Predation** = Organism first is benefited, organism second is inhibited.



7. **[+,0]** = **Commensalism** = Organism first is benefited, organism second is harmless.



8. **[+,+]** = **Protoco-operation** = Interaction is not obligatory, e.g; Sea anemone and Hermit crab live separately but when come in contact crab takes anemone to new site.

9. **[+,+]** = **Mutualism** = Interaction is obligatory, e.g; Mycorrhizal association.

Antibiosis:- The term Antibiosis generally refers to the complete or partial inhibition or death of one organism by another to the production of some substances or environmental conditions . eg. Production of chemicals called antibiotics by bacteria, actinomycetes and fungi that kill the other organisms causing diseases .

Competition:-Competition occurs when individuals attempt to obtain a resource that is inadequate to support all the individuals seeking it or even if the resource is adequate, individuals harm one another in trying to obtain it, the resource competed for can be divided into two types.

1. Raw materials such as light, organic and inorganic nutrients, water in heterotrophs

2. Space to grow, nest, hide from predators etc. In higher plants it is in the form of specific patterns and in animals too or by movements. It is a important area of population ecology, in both situation intra and interspecific competition.

A: Intra-specific competition:- this is a often known as scramble competition and is important density dependent factor regulating populations. The wild beast population is thought to be regulated by interspecfic competition for as limit supply of grasses of adequate quality seasons. Overcrowding reduces female fecundity which leads to fever successful mating due to mechanical interference between individuals. Subsidiary effects are egg mortality; increased larval competition for food hence reduces population density.

B: Inter-specific competition:- This is also called inter specific competition).Here

1. Only one species survives, it being the one with great negative effect on its competitor. Growth of surviving population to carrying capacity is slower than if the second population had been absent.

2. Both the species co exists independently. This occurs when interspecific competition is less then intraspecific competition in both the species neither of the species can reach the carrying capacity.

3. The species beginning at higher density persists and other eliminated. This is the special case when population has equal negative effects on the growth of each other, but intespecific competition is stronger then intraspecific eg. In labarority experiment mode on two species of paramecium (p. caudatum and p. Aurelia), one species survives and other is eliminated.

Ecosystem

The term ecosystem was proposed by A.G Tansley in 1935 who defined it as the system resulting from the integration of living organisms and non living factor of the environment. Thus he regarded ecosystem as including not only organisms- complex, but also the whole complex of physical factors forming the environment. Ecosystem is derive from “eco” Implies the environment and system implies an interacting, interdependent complex.

Thus any unit that includes all the organisms I,e the communities in a given area , interact with the physical environment so that a flow of energy leads clearly defined as tropic structure, biotic diversity and material cycle (i.e exchange of materials between living and non- living components) within in the ecosystem as ecosystem or ecological system. Keeping this view point in mind, we may think of the earth, we live upon as a giant ecosystem where

abiotic and biotic components are constantly acting and reacting upon each other bringing forth structural and functional changes in it. We generally study nature by subdivisions into unit of smaller ecosystem (terrestrial, forest, desert, grass land) and man engineered as a crop land. Aquatic fresh water etc.)of different sizes. An ecosystem may thus be a small as pond, as crop land or as large as an ocean, desert or forest. It must be it remembered, however that the unit ecosystem are simply separated from each other with time and space only, practically there exists no boundary between them.

So an ecosystem is overall integration of whole mosaic of integrating organisms and their environment . An ecosystem represents the highest level of ecological integration which is energy based and this functional unit is capable of energy transformation, accumulation and circulation.

Kinds of ecosystem:-

Different types of ecosystem in nature constituting the giant ecosystem the biosphere .

1. **Natural ecosystem**:- these operate without any major interferences of man . (a) Terrestrial or forest grassland, desert etc. Aquatic (water –open) which is further of fresh water which lotic (running) lentic stagnant, Marine such as deep bodies as ocean or shallow ones or sea or estuary etc.

2. **Artificial ecosystem**:- these are maintained artificially by man whereby addition of energy and planned manipulation, natural balance is distributed regularly. E.g crop lands , the maize, rice fields etc. where man tries to control the biotic community as well as the Physio-chemical environment are artificial ecosystem

Structure of the ecosystem:- the two major aspects' of an ecosystem are , the structure and the function. By **structure** we mean.

i. the composition of biological community including species, numbers, biomes, life, history and distribution in space etc.
ii. the quantity and distribution of nonliving materials such as nutrients , water etc.
iii. The range of gradient of conditions of existence, such as temperature, light etc.

By **function** we mean

i. The rate of biological energy flow i.e the production and respiration rates of the community.

ii. Rate of materials, nutrients cycles.

iii. Biological or ecological regulation including both regulation of organisms by environment (photoperiodism) and regulation of the environment by the organisms (nitrogen fixing organisms) . Thus in any ecosystem structure andfuction are studies together.

Structure:- An ecosystem has two major components.

1. **Abiotic component**:- it includes

   i. The amount of inorganic substances as P, S, C, N, H etc involved in material cycles . The amount of these inorganic substances present at any given time in ecosystem is designated as the standing state or standing quality .

   ii. Amount and distribution of organic chemicals such as chlorophyll, organic material , proteins, carbohydrates, lipids etc present either in biomes or in environment I,e the biochemical structure that link the biotic and abiotic components of the ecosystem .

   iii. The climate of a given region.

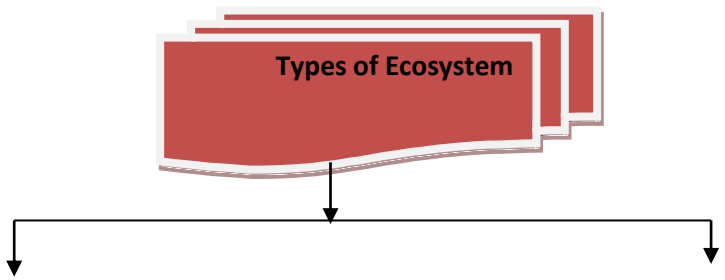
2. **Biotic components:-** this is a indeed the tropic structure of an ecosystem , where living organisms are distinguished on the basis of their nutritional relationships. From the tropic structure and ecosystem has two components.

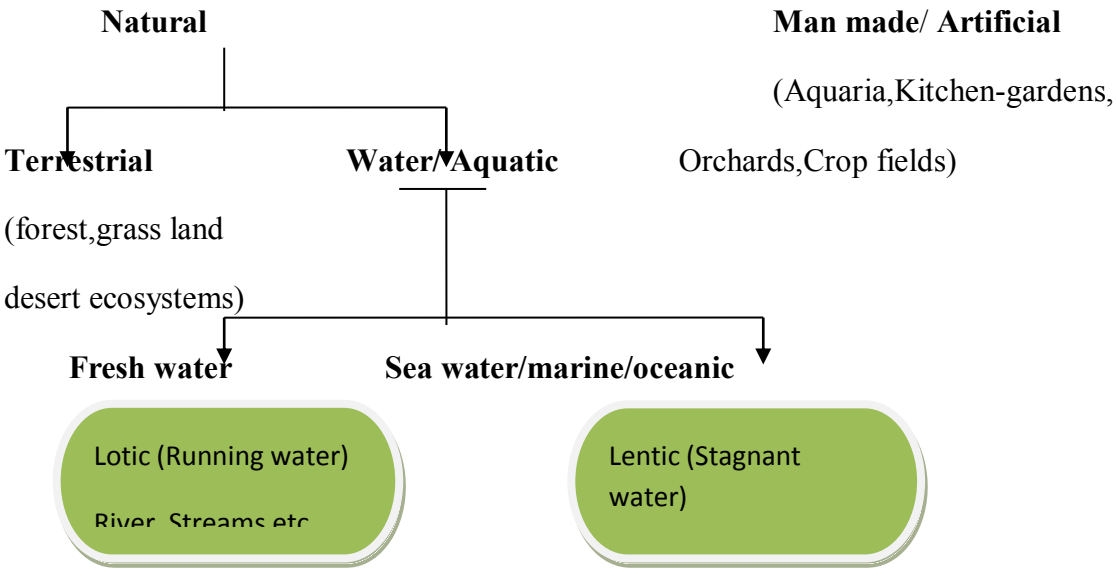
   (a) **Autotrophic component**:- the component in which fixation of light energy, use of simple inorganic substances' and build up complex substances predominate . The component is continued, mainly by green plants. These are also known as producers.

   (b) **Heterotrophic component:-** that component in which utilization, rearrangement and decomposition of complex materials predominate. The organisms involved known as consumers.

      i. Macro consumers:- consumers which in order they occur in a food chain are Herbivorous, Carnivores, omnivores.

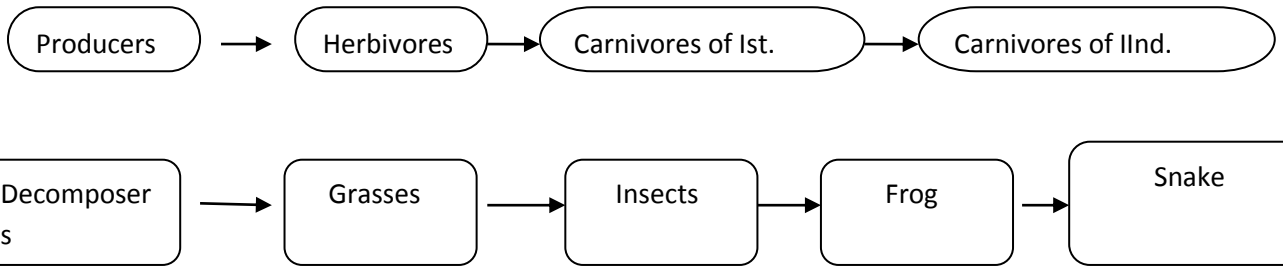
      ii. Micro consumers:- these are popularly known as decomposers.





Food Chain : A sequence of population or organisms of an ecosystem through which the food and its contained energy passes with each member becoming the food of a later member of the sequence. OR

- Food chain is the sequential inter-linking of organisms involving the transfer of food energy from the producers, through a series of organisms with repeated eating and being eaten**.**
- In nature or ecosystem green plants alone are capable of synthesizing their own food by trapping solar energy and converting it into chemical energy in the form of various organic compounds which are eaten up by herbivores. Herbivores are eaten up by carnivores of first order and carnivores of Ist. Order by carnivores of second order and so on.
- In this way food is transferred from one trophic level (feeding level) to other trophic level in the form of a chain called **food chain**.

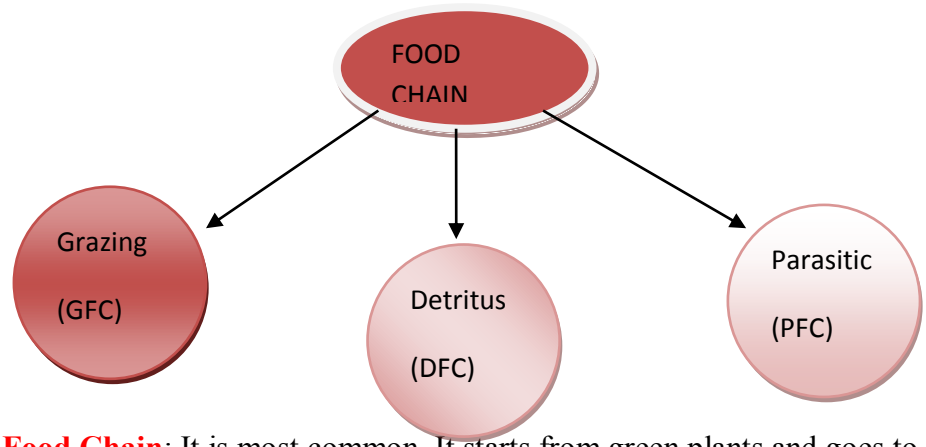


- Green plants are always the first link of food chain because the alone are capable of synthesizing organic food by using light energy.

Characteristics of a food chain:

- Consists a series of populations which are related by eating and be eaten.
- Generally straight.
- No. of trophic level 3 – 6.
- Progressive reduction in available biomass, energy and no. of individuals with the rise in trophic level.
- At each trophic level generally 80 – 90 % of energy is lostas heat.
- Some organisms like human occupy different trophic positions in different food chains.
- Food chains are sustained by producers and decomposers.

TYPES



1.Grazing Food Chain: It is most common. It starts from green plants and goes to grazing herbivores and then to carnivores. Herbivores are also called ***key industry animals [Elton-1927]*** *(because they convert plant matter in to animal matter).*

2. Detritus Food Chain: This type of food chain goes from dead organic matter to detritivores and then to organisms feeding on detritivores. This food chain is also called as **Saprophytic food chain**.

- ❖ **Detritivores** – organisms obtain food from detritus(dead and decayed organic matter).

3.Parasitic Food Chain: This chain goes from large to small parasitic organisms**.**

Trophic Levels

The distinct sequential steps in the straight food chains are referred to as different **trophic (nutritional) levels.** E.g;

I. Green plants (producers) stand for the first trophic level- **Producer level.**

II. The plant eaters (herbivores) also called primary consumers belong to second trophic level**.- Primary consumer level.**

III. The flesh eaters (carnivores) also called secondary consumers represent the third trophic level**- Secondary consumer level.**

- The food chain consists of producers, consumers and decomposers. As the decomposers are operating at all trophic levels in all food chains so are often not considered.

❖ Consumers are often of 3 – 5 types:-

i). First order- Primary
ii). Second order – Secondary
iii). Third order – Tertiary
iv). Fourth order – Quaternary

1.**Producers**: These are green plants who prepared the food by the process of photosynthesis. They are able to convert solar energy in to the chemical energy. Organic food synthesized by producers consists of all essential ingredients like carbohydrates, proteins, fats, vitamins etc.

- Producers constitute the first trophic level or base of a food chain.
- Producers are also known as ***Transducers*** as they are able to change radiant or light energy in to chemical form.

2.**Primary Consumers or Herbivores**:They are animals which feed on plants or plant products.eg; grasshoppers, several insects, rabbit, hare, field mouse, deer, antelope, elephant, zooplankton (*Paramecium*, *Daphnia*,some larvae), some fishes etc.

3.**Secondary Consumers or Pry; carnivores**: They are animals which feed upon herbivores. E.g; frog, predator insects, several birds, fishes, wild cat, fox etc.

4.**Tertiary Consumers or Sec; carnivores**: They are larger carnivores which feed on primary carnivores. e.g; Wolf (feed on fox), snake (prey up on frog).

5.**Top Carnivores**:They are the last order consumers or carnivores which are not killed by/ preyed upon by other animals because of their size, agility and ferociousness. e.g; shark, crocodile, eagle, peacock, tiger, lion.

Termination of a food chain may occur at;

- **Herbivores level**: Vegetation ⟶ Elephant.
- **Pry. Carnivores**: Vegetation ⟶ Squirrel ⟶ Bear.
- **Sec; Carnivore**: Vegetation ⟶ Squirrel ⟶ Wild cat ⟶ Tiger
- **Quaternary consumer/ Tertiary Carnivore**:
- Vegetation ⟶ Rabbit ⟶ Cat ⟶ Wolf ⟶ Tiger

- **Terrestrial food chains**:

i). Grass ⟶ Rabbit ⟶ Cat ⟶ Wolf ⟶ Tiger

ii). Grass ⟶ Grasshopper ⟶ Frog ⟶ Snake ⟶ Peacock ⟶ Falcon

iii). Vegetation ⟶ Insect ⟶ Predator bird ⟶ Hawk

- **Aquatic food chains**:

i). Phytoplanktons ⟶ Zooplanktons ⟶ Crustaceous ⟶ Predator insects ⟶ Small fish ⟶ Large fish

ii). Phytoplanktons ⟶ Zooplanktons ⟶ Crustaceous ⟶ Predator insects ⟶ King fisher ⟶ Stork

FOOD WEB

Food web is a network of food chains which become inter-connected at various trophic levels so as to form a number of feeding connections amongst different organisms of a biotic community. OR
When food chains are inter-connected with each other forming a net work of food transfer or energy flow, it is called as ***Food Web.***

- Food web is meant for increasing the stability of an ecosystem by providing alternate source of food and allowing the endangered population to grow in size.
- Food webs operate because of taste preference for a particular food and unavailability of food.
- One animal may feed upon organism of even different trophic level like—snakes may feed upon mice (herbivore) and frogs (carnivore).
- ***Jackals are both Carnivores and Scavengers.***
- Food web is not straight.
- It provides a number of alternate foods to consumers which, therefore, become ***Polyphagous.***
- ***Function:*** Food webs distinguish levels of producers and consumers by identifying and defining the importance of animal relationships and food sources, beginning with primary producers such as plants, insects and herbivores.

- ***Significance:*** Food webs are important tools in understanding that plants are the foundation of all ecosystems and food chains, sustaining life by providing nourishment and oxygen needed for survival and reproduction.

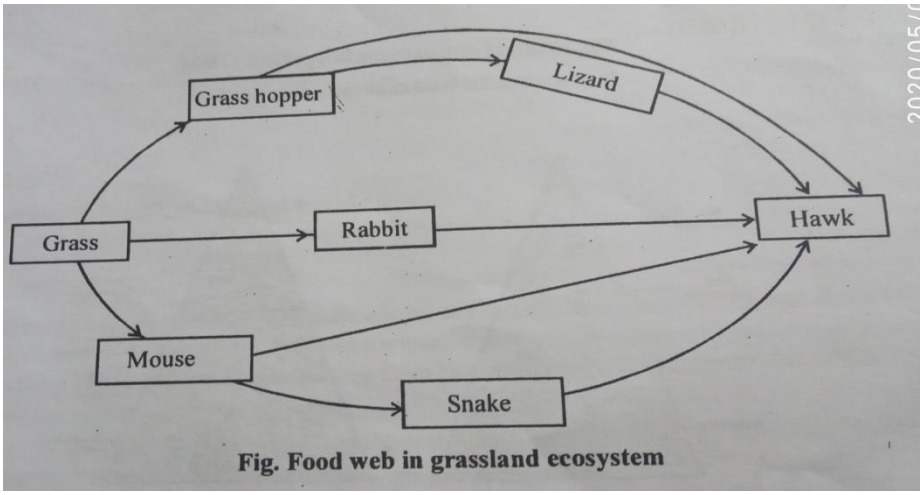


Fig. Food web in grassland ecosystem

Ecological Pyramids

Ecological pyramids:- Tropic structure i.e interaction of food chain and seize relationship between various biotic components on an ecosystem. The tropic level i.e producers herbivores carnivores may be shown graphically by means of ecological pyramids. Where the first are producer; level constitutes the basis of the pyramid and successive level, the tires making the apex. Ecological pyramids' are of three general types.

i.Pyramid of numbers

ii. Pyramid of biomass.

Iii Pyramid of energy.

1. Pyramid of numbers: - showing the number of individuals /organisms at each level. It shows the relationship between producer's,herbivores and carnivores at successive tropic levels in terms of their numbers. The pyramid of number of three different types of ecosystem are shown. In grassland, the producers which are mainly grass are always maximum in numbers. This number then shows decrease towards apex, as the primary consumers (Herbivorous) like rabbits, mice etc. are lesser in numbers then the grasses the secondary consumers snakes and lizards are lesser in number then the rabbits and mice. Finally the top (tertiary) consumer's hawks or other birds are least in number. Thus the pyramid becomes upright. Similarly in a pond ecosystem, the pyramid is upright. In a forest ecosystem however of numbers is somewhat different in shape .the producers , which are

mainly large sized trees are lesser in number and form the base of pyramid. The herbivores which are fruit eating birds, elephants, deer's etc are more in number then the producers. Then there is a gradual decrease in the number of successive carnivores thus making the pyramid again upright. However in a parasite food chain the pyramids are always inverted. Pyramids of number showing the number of individuals/ organisms at each level.

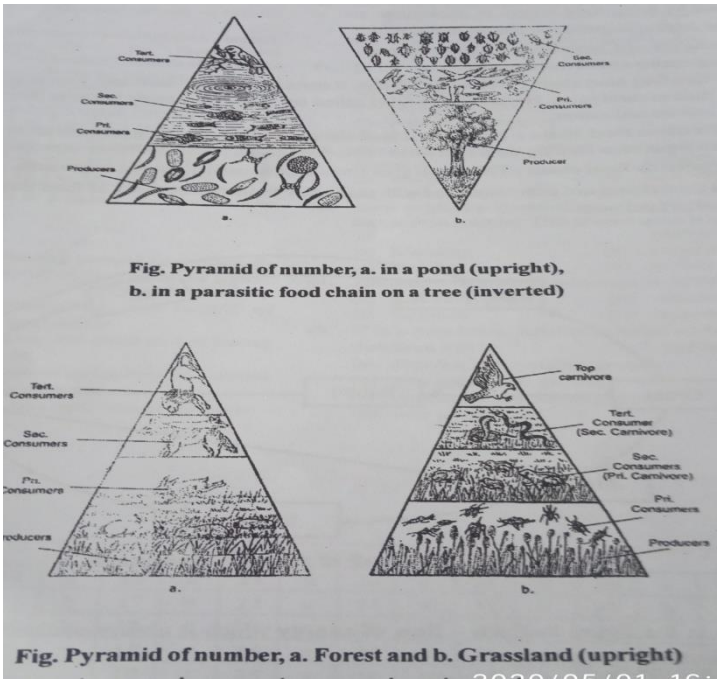


Fig. Pyramid of number, a. in a pond (upright), b. in a parasitic food chain on a tree (inverted)

Fig. Pyramid of number, a. Forest and b. Grassland (upright)

2.Pyramid of biomass :- they are competitively more fundamental, because the qualitative relationship of biomass in grass land and forest ecosystem shows a gradual decrease in biomes of organisms at successive levels from the producers to the top carnivores/. Thus pyramid is upright however in a pond ecosystem the producers are small organisms, their biomass in least and the value gradually shows in increase towards the apex of the pyramid. Thus making the pyramid inverted in shape.

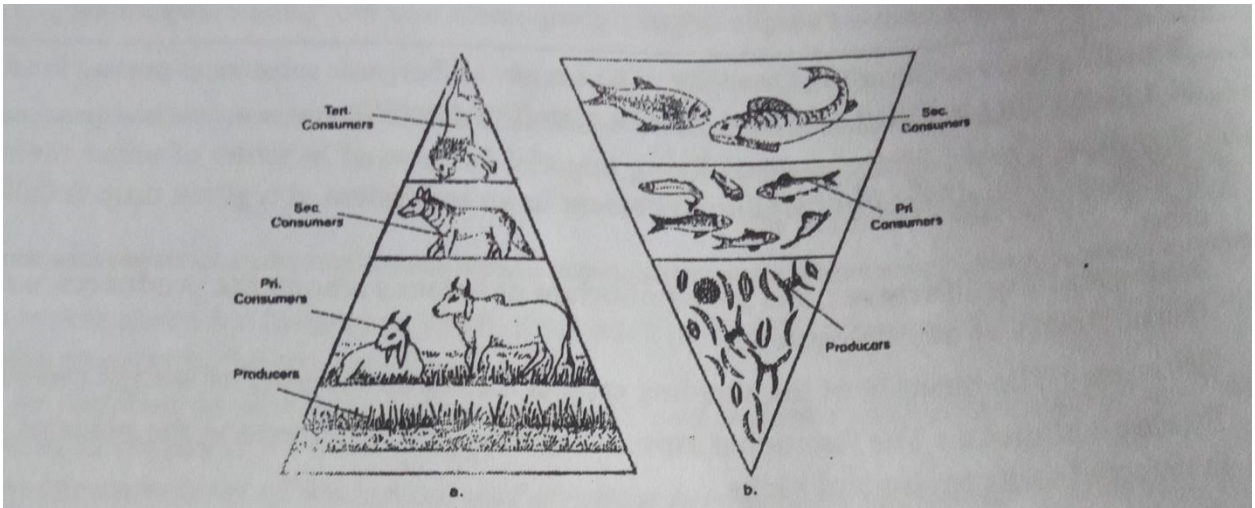


Fig. Pyramids of biomass, a. in a forest (upright) and b. in a pond (inverted)

Pyramid of energy:- of the three types of ecological pyramids, the energy [pyramid gives the best picture of overall nature of the ecosystem. In contrast to the pyramid of numbers and biomass the pyramid of energy is the picture of the rates of passage of food mass through the food chain; in shape it is always upright. because the e is a gradual decrease in the energy content at successive tropic levels from producers to various consumers .

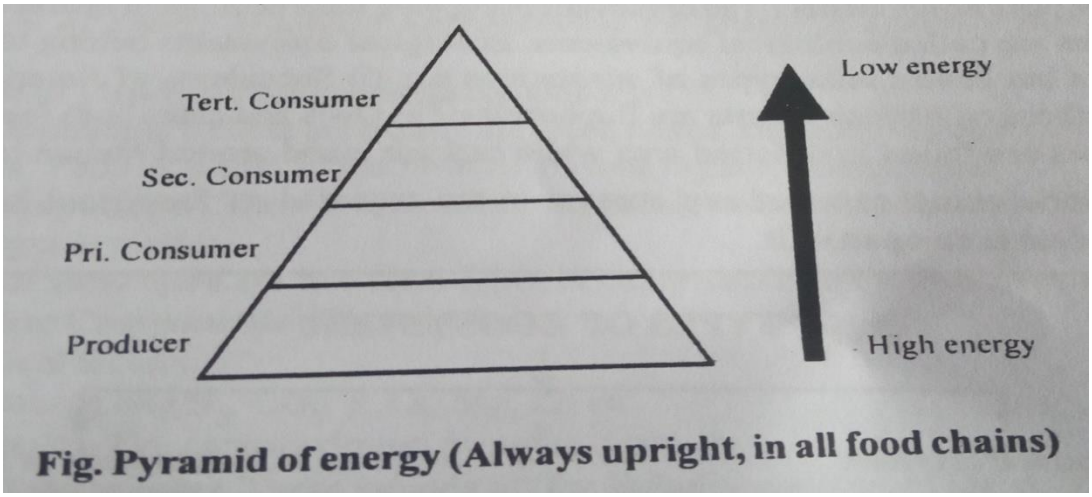


Fig. Pyramid of energy (Always upright, in all food chains)

Functional aspect of an ecosystem:- the living and non living component of an ecosystem, are so interwoven that their separation from each other becomes very much difficult. The mode of movement of materials and energy in an ecosystem is shown in simple modal. The producers green plants fix radiant energy and with the help of minerals (i,e, H, o, n, p , ca, mg, Zn, Fe etc) taken from their soil and aerial environment. Organic matter (carbohydrates, proteins , fats, nucleic acids etc) some ecologists prefer to call green plants as converters or transducers. Two ecological processes of energy and minerals cycles involve interaction between the physic-chemical environment and the biotic communities may be thought of the heart of the ecosystem dynamics. According to the modal shown energy flows in non cyclic

manner (uni-directional) from sun to the decomposers via producers and macro consumers, where as the minerals keep on moving in a cyclic manner.

Productivity of An Ecosystem

Productivity of an ecosystem:- the productivity of an ecosystem refers to the rate of production i.e the amount of organic matter accumulated in any unit time. Productivity is of the following types:-

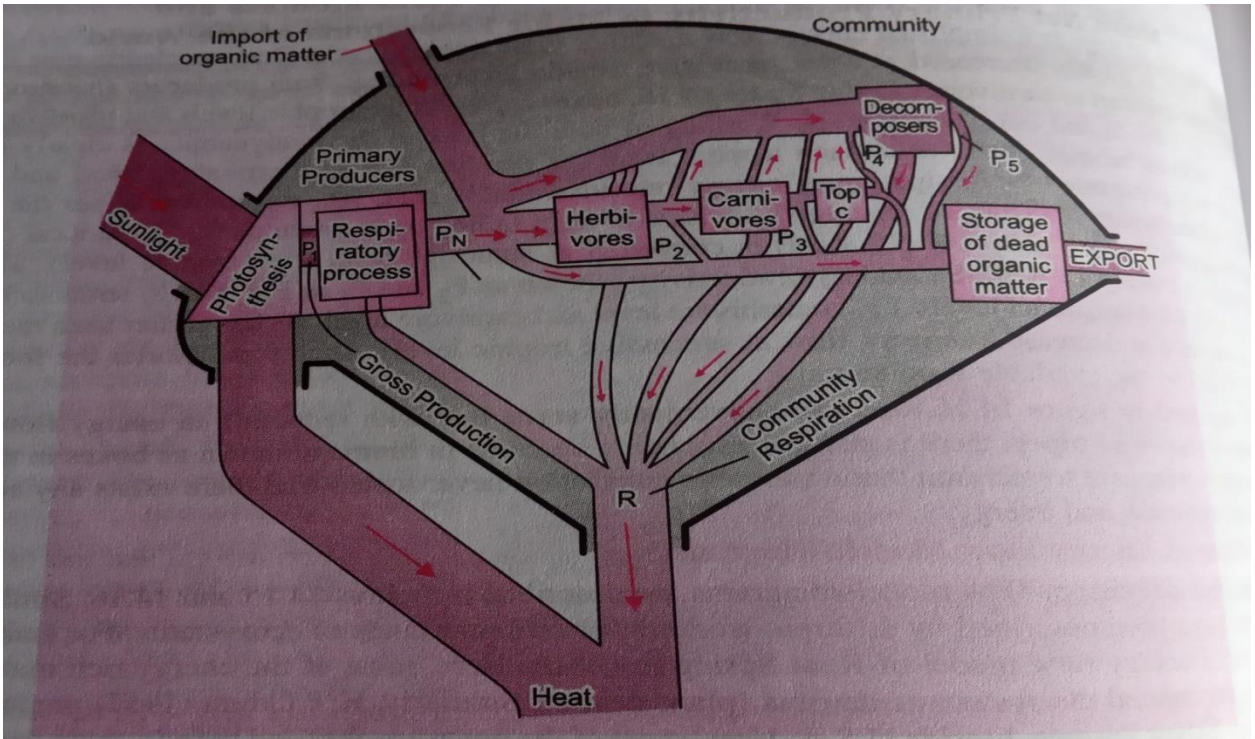
1. **Primary productivity**:- it is associated with he producers which are autotrophic. Most of which are photosynthetic and to a much lesser extent the chemo synthetic organisms. These are green plants, higher macrophytes as well as the lower forms. The phyto-planktons and some photosynthetic bacteria. Primary productivity is defined as the rate at which radiant energy is stored by photo synthetic activity of producers. It may be:

   (a) **Gross primary productivity**:- it is the total rate of photosynthesis including the organic matter used up in respiration during the measurement period . This is also called as total grass photosynthesis or total assimilation and deepens upon chlorophyll content .Te rate of primary productivity is estimated either by chlorophyll content or amount of CO_2 fixed / g.chl/ hour.

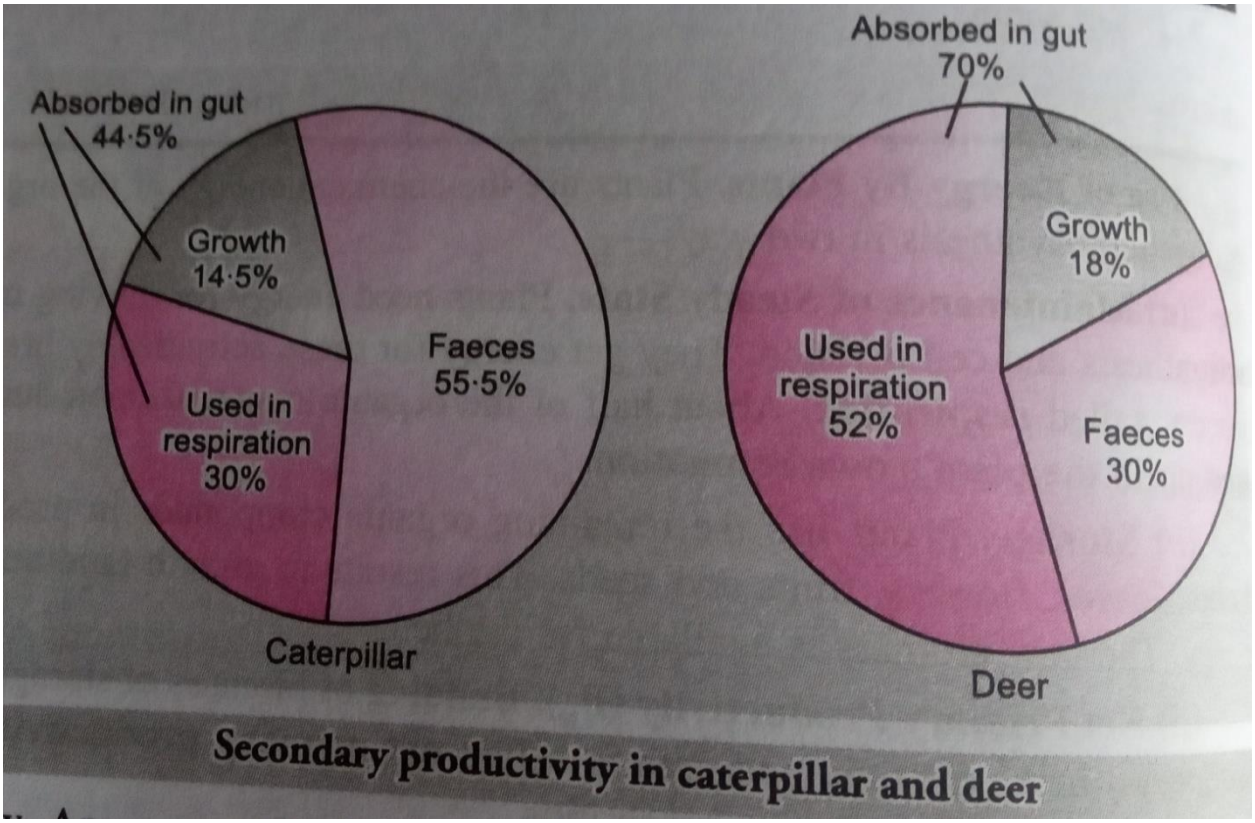
   (b) **Net primary productivity: -** it is the rate of storage of organic matter ion plant tissues in excess of the respiratory utilization by plants during measurement period. This is thus the rate of increase of biomass and is also known as apparent photosynthesis or net assimilation. it refers to balance between gross photosynthesis and respiration and other plant losses .

2. **Secondly productivity**:- it refers to consumers or heterotrophs. These are the rates of energy storage of energy level. Since consumers only utilize food materials in their respiration .ecologists as Odom refers it as assimilation. Rather than production. Secondary productivity remains mobile.

3. **Net productivity:-** it refers to the ate of storage of organic matter not utilized by heterotrophs equivalent to net primary production minus. Consumption by the heterotophs during unit period as season or year etc. it is thus the rate of increase of biomass of primary producers which has been left over by the consumers.



The relationship between flow of energy through the grazing food chain and detritus pathway (After H.T. Odum, 1956)



Secondary productivity in caterpillar and deer

Ecological Energetics

Ecological Energetics:- the energy used for all plants life process is derived from solar radiations. In ecological energetics we study.

i. quantity of solar energy reaching an ecosystem.

ii. Quantity of energy used by the green plants for photosynthesis and

iii the quantity and path of energy flow from producers to consumers.

About 34% of the sunlight reaching the earth's atmosphere is reflected back into the atmosphere. 10% is held by the ozone layer, water vapor and other gases. The rest 56% reaches the earth's surface. Only a fraction of this energy reaching the earth's atmosphere (1-5%) is used by green plants for photosynthesis and rest is absorbed as heat by green vegetation or water.

Energy flow in ecosystem:- the behavior of ecosystem can be termed as energy flow due to un directional flow of energy. From energetics point of view it is essential to understand for an ecosystem.

i. The efficiency of producers in absorption and conversion of solar energy.

ii. The use of this converted chemical energy by the consumers

iii. The total input of energy in the form of food and its efficiency of assimilation

iv. The loss through respiration, heat, excretion etc.

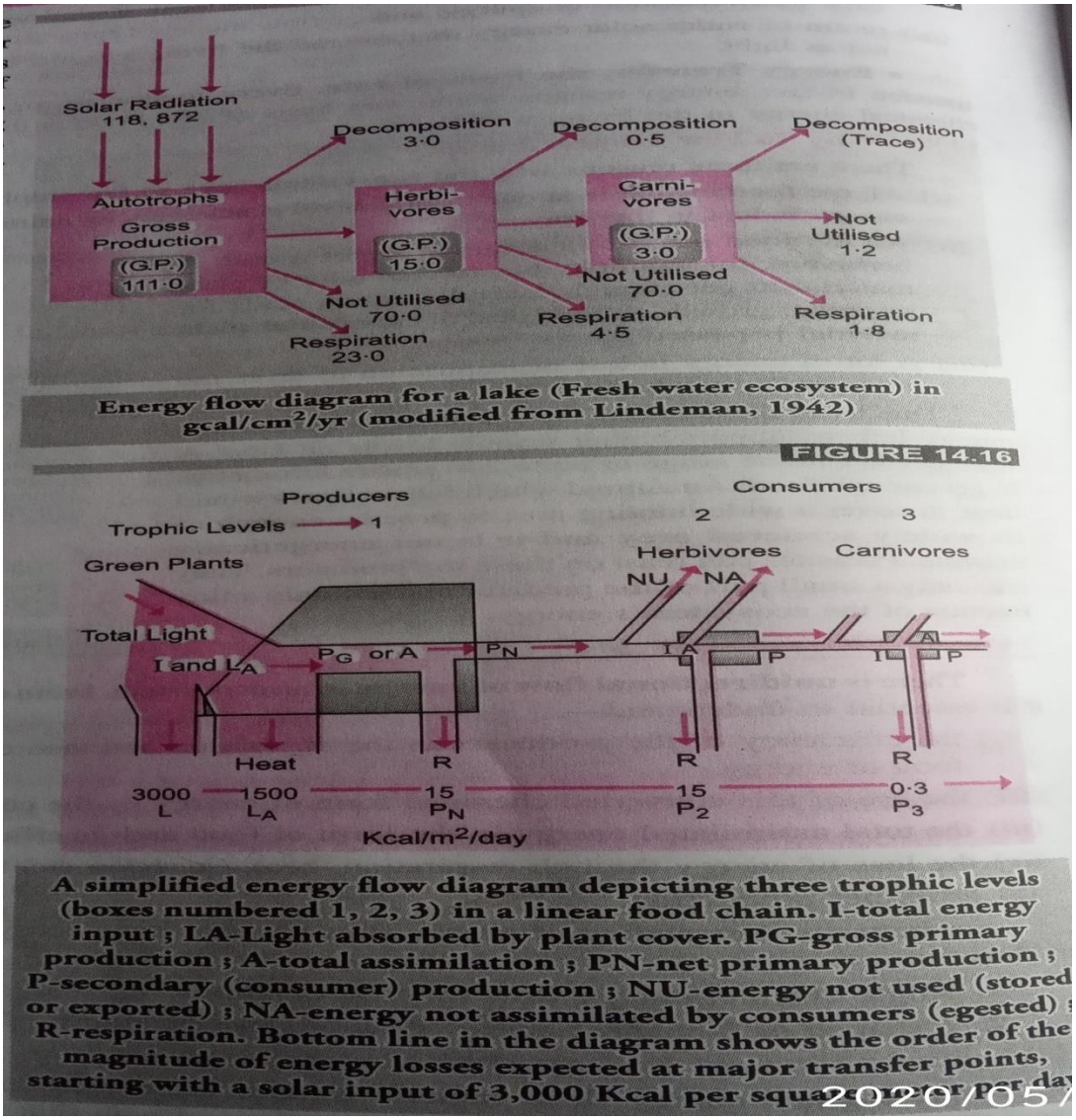
v. The gross net production.

Single channel energy model:- the principles of food chain and the working of two laws of the thermodynamics can be better made clear by means of energy Flow diagram shown in Fig. As shown in figure out of the total incoming solar radiations (118872gcal/ cm^2gm) 118761 gcal/ cm^2gm/yr remain unutilized and the grass production (net productivity plus respiration) in autotrophs is 111 gcal / cm^2/yr with an efficiency of energy capture of 0.10%. It may be further noted that 21% of this energy or 23 g.cal is consumed by metabolic reactions of autotrophs for their growth, development, Maintenance and reproduction. It may be seen further that 15 g.cal/ cm^2 / yr are consumed by herbivores that graze or feed on autotrophs production. The reminder of plant material 70 g.cal/ cm^2yr or 79.5 % of net production is not utilized at all but becomes the part of accumulating sediments. It is oblivious then that much or more energy is available for herbivory than is consumed. It may be also notes that various pathways of loss are equivalent to and account for total energy capture of autotrophs i.e. grass production. Also the tree upper upper fate (Decomposition, herbivory and not utilized) is equivalent to net production. Out of the total energy incorporated at the herbivore level i.e. 15 g.cal/ cam / yr 30% or 45% g.cal is used in metabolic functions . Thus there is considerably more energy loss via respiration by herbivores (30%) then by autotrophs (21%). Again there is considerable energy

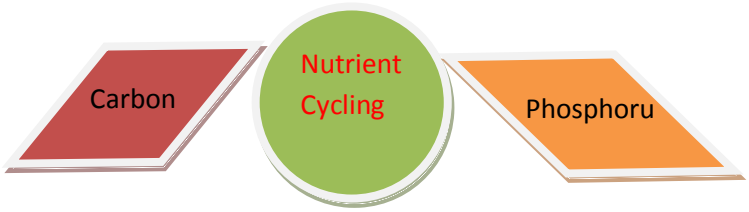
available for the carnivores, namely 10.5 g cal or 70 % which is not entirely utilized. Only 28.6 % of net [production passes to the carnivores.

At the carnivore level about 60% of the carnivores energy intake is consumed in metabolic activity and the reminder becomes part of the not utilized sediments. Only a small amount is subjected to decomposition yearly, thus high respiratory loss compares with 30% by herbivores and 21 % by autotrophs in this ecosystem.

From the energy flow diagram two things become clear, firstly there Is one way street along which energy moves. By the energy that is captured by the autotrophs does not revert back to solar input. That which passes to the herbivores does not back pass back to the autrotops as it move s through various tropic levels, it is no longer available the previous level. This is due to one way flow of energy. Secondly there occurs a progressive decrease in energy level at each tropic level.



Figs. Showing Energy Flow in an Ecosystem



BIO-GEO CHEMICAL CYCLING

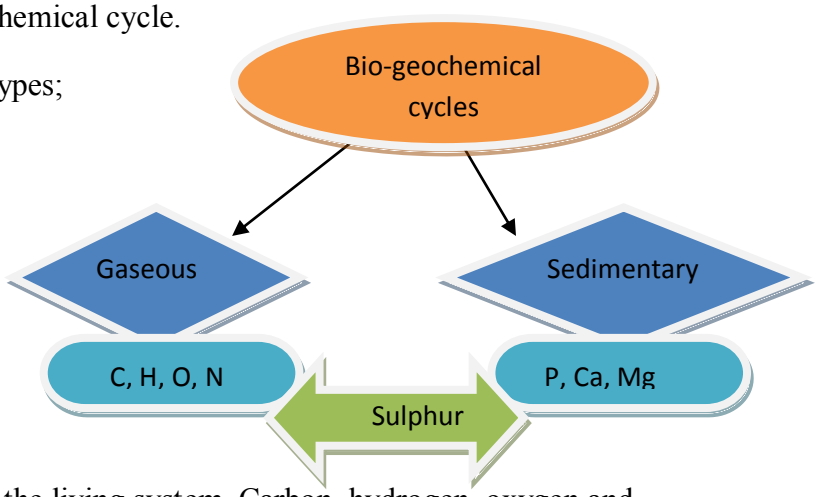
(Recycling of materials)

The movement of materials through the living and the non-living components of biosphere or in any ecosystem is called bio-geo chemical cycle.

Bio-geochemical cycles are of two types;

A. Gaseous cycles

B. Sedimentary cycles



A. Gaseous cycles:

The reservoir for the elements is atmosphere or hydrosphere.

The four most abundant elements in the living system. Carbon, hydrogen, oxygen and nitrogen have predominantly gaseous cycle.

B. **Sedimentary cycles:** The reservoir for the elements is in the sediments of earth (lithosphere) e.g; Phosphorus, calcium, magnesium.

- sulphur has both sedimentary and gaseous phase.

CARBON CYCLE

Carbon is considered the basis of life. Carbon atoms form the back bones for the complex organic molecules like carbohydrates, lipids, proteins and nucleic acids of the cell.

- Carbon is present in the abiotic environment in four forms like CO_2(air), dissolved CO_2(hydrosphere), fossil fuels e.g; coal, petroleum and natural gas are carbonates, graphite in rocks.
- Carbon content of atmosphere is about $6x10^{14}$kg, while that of the hydrosphere is 1.3 – 5 $x10^{15}$kg. Carbon present in lithosphere is 2.8 x 10^{21}kg.About 7 x 10^{13}kg of carbon is fixed by photosynthesis in a year by the producers and this release about 9 $x10^{13}$ kg of Oxygen. One hectare of productive forest produces 10,000 kg (10tonnes) of oxygen and absorbs 30,000kg (30 tonnes) of CO_2 (2000 kg of carbon).
- Carbon fixed by producers enters the food chain and is passed to herbivores, carnivores and decomposers. Carbon dioxide is being added to atmosphere by respiration of organisms , decomposition of dead bodies of organisms, decay of animal excreta by anaerobic organisms, burning,

weathering of carbonate containing rocks, volcanic eruptions and hot springs.

- Combustion of fossil fuels is adding more than 6 x10^{12}kg of carbon annually in to the atmosphere which results in increase in concentration of CO_2 causing rise in atmospheric temperature. **Volcanic eruptions; Acid rain**

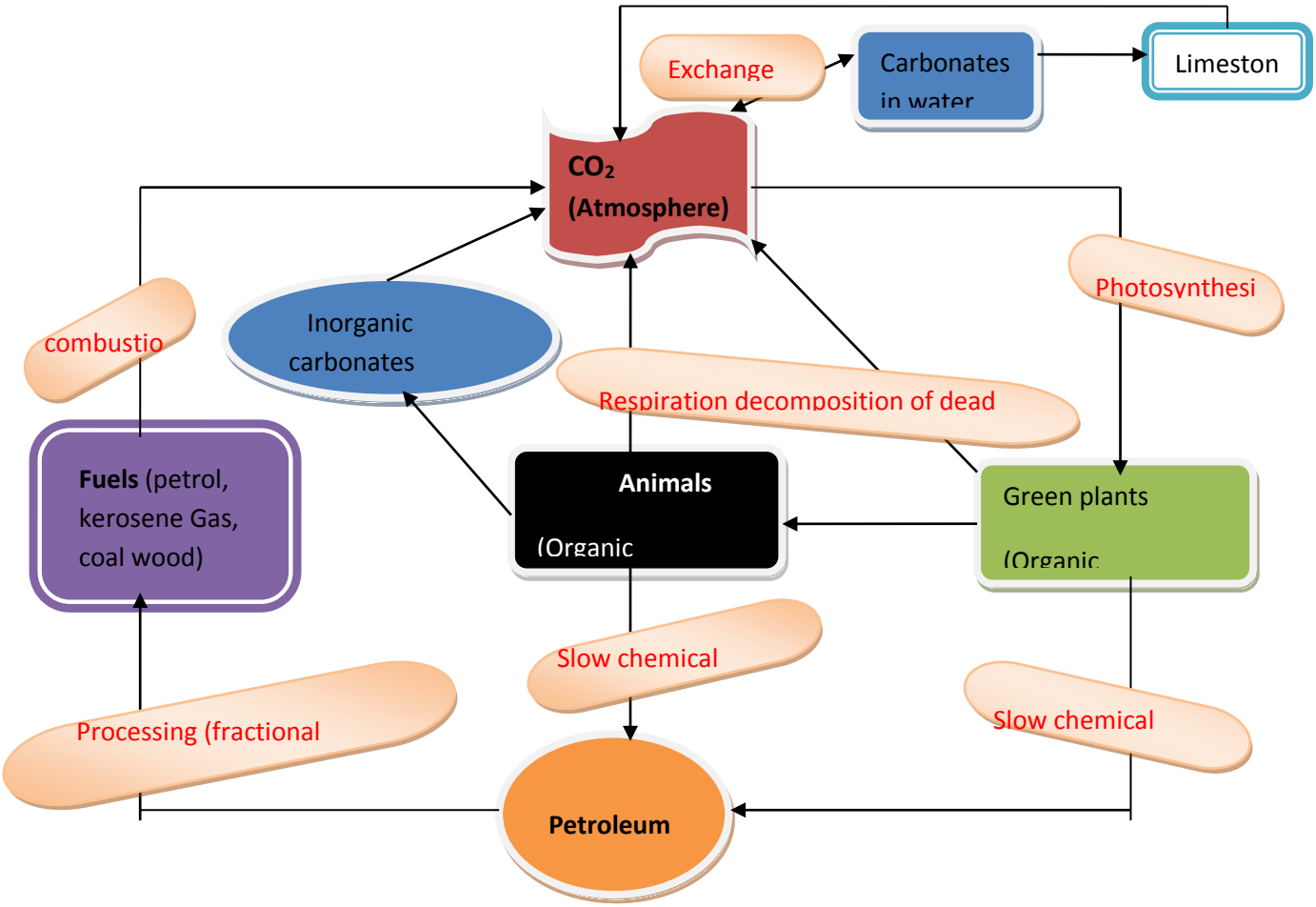


Fig. CARBON CYCLE

PHOSPHORUS CYCLE

Phosphorus is a major constituent of biological membranes, nucleic acids and cellular energy transfer system. Many animals also need large quantities of this element to make shells, bones and teeth, the natural reservoir of phosphorus is rock, which contains phosphorus in the form of phosphates.

- When rocks are weathered, minute amounts of these phosphates dissolve in soil solution and are absorbed by the roots of the plants.

- Waste products and the dead organisms are decomposed by phosphate-solubilising bacteria releasing phosphorus.
- Unlike Carbon cycle, there is no respiratory release of phosphorus in to atmosphere.

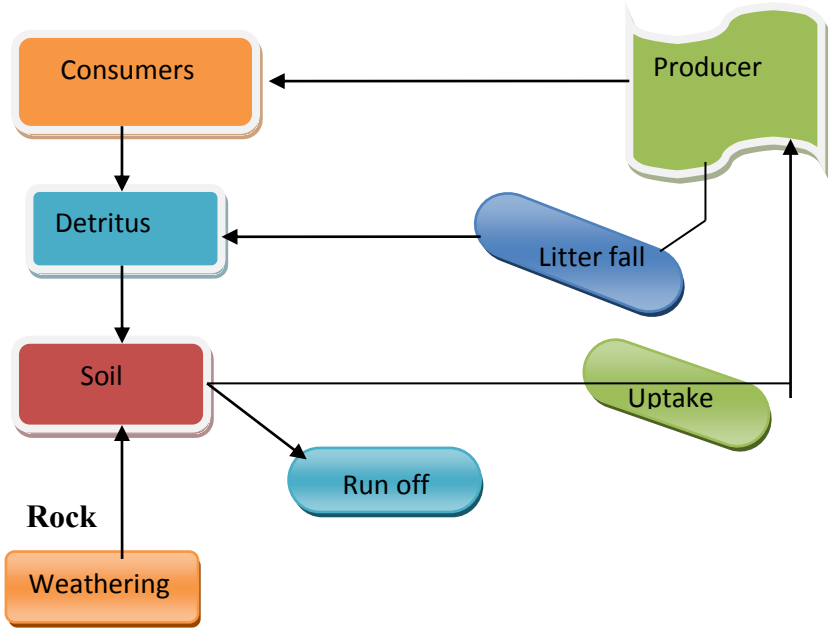


Fig. Phosphorus Cycle

❖ **Difference between carbon and phosphorus cycle is as;**

- Firstly, atmospheric inputs of phosphorus through rainfall are much smaller than carbon inputs.
- Secondly, gaseous exchanges of phosphorus between organism and environment are negligible.

Ecological Succession

Ecological succession:- although a typical community maintains itself more or less in equilibrium with the prevailing conditions of the environment , in nature it is hardly true, communities are never stable, but dynamic, changing more or less regularly over time and space. They are never found in permanently complete balance with their component species or with physical envirionment.

Environment always kept on changing over period of time due to

1. Variations in climate and physic graphic factors
2. The activities of species or the communities themselves. There influences bring about marked changes in the domains of the existing community which is thus sooner or

after replaced by another community at the same place. This process continuous and successive communities develop one after another over the small area , until the terminal final community becomes again more or less stable for period od time. This occurrence of relatively definite sequence of communities over a period of time in the same area is known as ecological succession. Odom prefers to call this orderly process an ecosystem Development rather than ecological succession . Odom defined the ecological succession in three parameters .

1. It is an orderly process of community development, That involves changes in species structure and community process with time.
2. It results from the modification of physical environment by the communities i.e. succession is community controlled.
3. It culminates in a stabilized ecosystem in which maximum biomass and symbiotic factions between organisms are maintained per unit of energy flow.

Causes of succession:-

1. **Initial causes**: - these are climatic as well as biotic, former includes factors such as erosion and deposits , wind, fire etc caused by lightning or volcanic activity and the later includes the activities of organisms. These causes produce the bare areas or destroy the existing population in an area.
2. **Continuing causes:-** these are the process as migration , aggregation, competition, reaction etc.
3. **Stabilizing causes**:- these cause the stabilization of the community. According to the Clements, climate of the areas is the chief cause of stabilization, others are of secondary value.

Types of Ecological succession:-

1. **Primary succession**:- in any of the basic environment (terrestrial , fresh water, marine) one type of succession is primary succession which starts from the primitive conditions of the environment (substratum) where there was no previously any sort of living matter. The first group of organisms establishing there are known as the **pioneers** or primary community.
2. **Secondary succession**:- starts from previously built up substrata with already existing living matter. The action of any external force as a sudden change in climatic factors , biotic intervention, fire etc causes the existing communities to disappear . Thus area becomes devoid of living matter but its substratum instead of primitive is build up. These are more rapid.

3. **Autogenic succession**;- Community itself modifies its own environment as a result of interactions with its environment, this causing its own replacement.

4. **Allogenic Succession**:- Replacement caused by the largely external environment.

General process of succession:- An ecological succession is actually completed through a number of sequential steps.

1. **Nudation**:- The develop. of a bare area without any form of life. The area may develop due to several causes such as land side, erosion, deposition topographic or biological due to which the existing community may disappear.

2. **Invasion:-** This is the successful establishment of a species in a bare area. The species may actually reach the bare area from any other area through migration. After invasion there occurs successful establishment of the species, as a result of adjustment with the prevailing conditions known as **Ecesis (Gk-*oiksis*-**inhibition**)** after ecesis, as a result of reproduction, individuals of species increase in number and they come close to each other. The process is known as **aggregation**.

3. **Competition**:- After aggregation of a large number individuals of the species at the limited place, there develops competition(inter as well as intra specific) mainly for space and nutrition, individuals of a species affects each other life in various ways and this is called **co-action**. The species, if unable to compete with other species if present would be discarded.

4. **Reaction**: - This is the most important stage in succession, the mechanism of the modification of the environment through the influence of the living organisms on it, is known as **reaction.** As a result of reaction, changes take place in soil, water, light, conditions, and temperature etc of the environment. Due to all these the environment is modified becomes unsuitable for the existing community which sooner or later is replaced by another community (seral community) the whole series of communities that replace one another in sequence are referred as **seral communities** or **seral stage**.

5. **Stabilization (Climax):-** Finally There occurs a stage in the process, when the final terminal community becomes more or less stabilized for a longer period of time and it can maintain itself in equilibrium with the climate of the area. The final community is not replaced, and is known as climax community and the stage as **climax stage**.

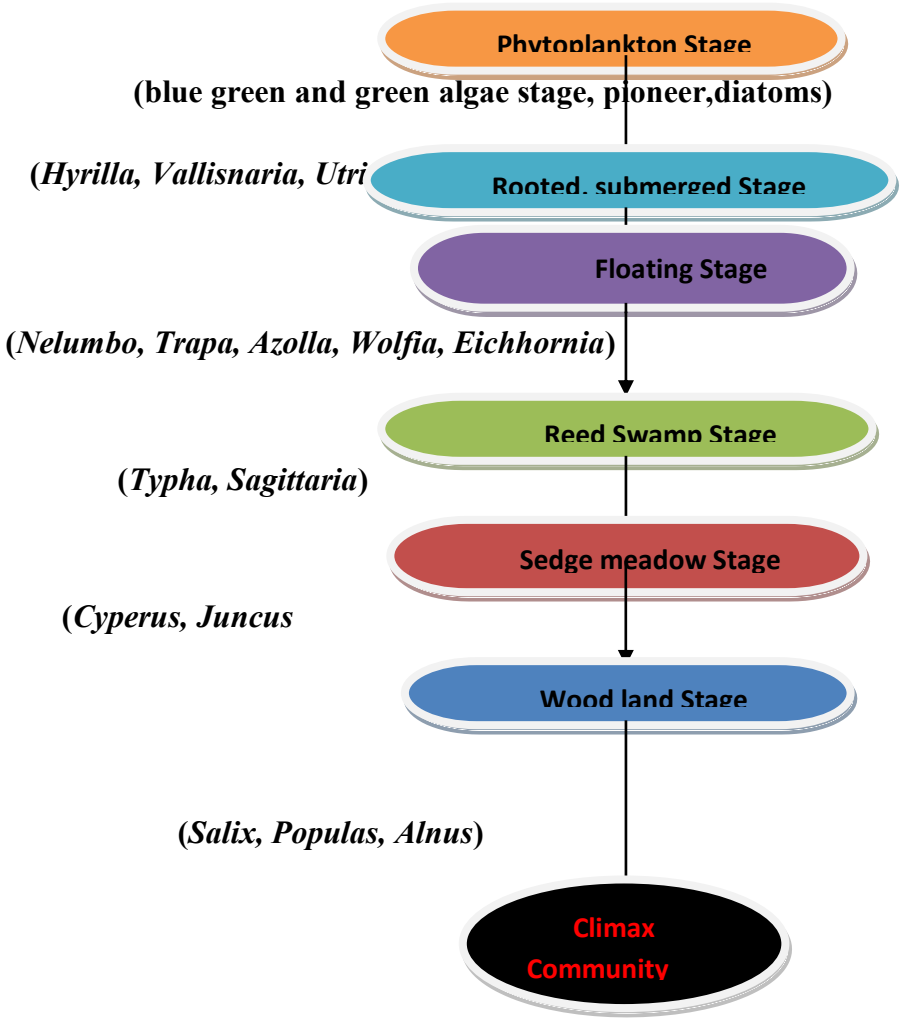
Ecological services:- Human kind benefits from a multiage of resources and process that are supplied by natural ecosystem. Collectively these benefits are known as ecosystem system or ecological services and include the services like drinking water, decomposition of wastes, carbon fixation. Oxygen release, pollination etc.

Oxygen release:- The photosynthesis releases oxygen into the environment. Oxygen is one of the several products of photosynthesis and this is one of reason why plants

and animals are so important in the maintaining the balance of a tropical ecosystem. The water molecules split at the beginning of photosynthesis for the electrons which make their way to electron transport chain and oxygen is released to the atmosphere.

Carbon fixation:- Photosynthesis is the process through which atmospheric carbon is fixed by autotrophs into organic food. Different path ways in photosynthesis have been adopted by various types of plants via C3, C4, and CAM where carbon dioxide is fixed to glucose.

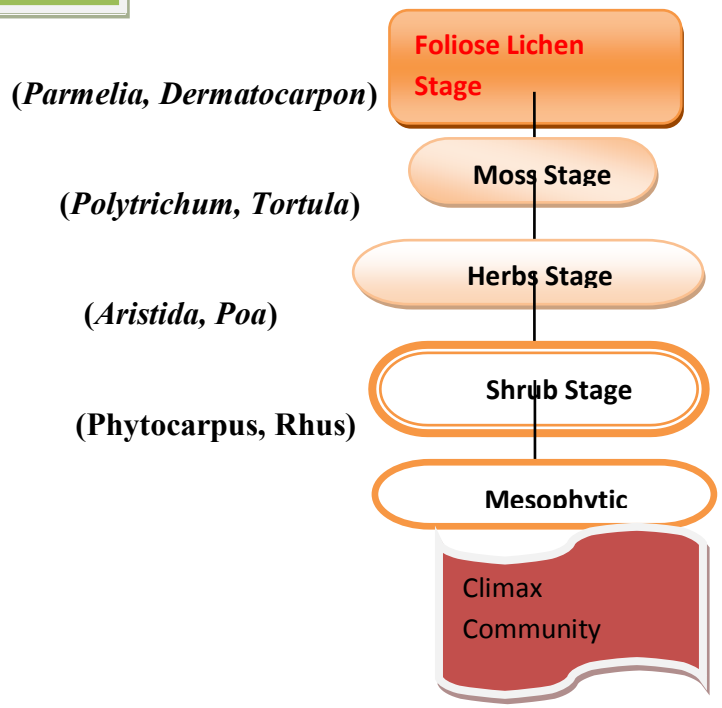
Flow Chart of Hydrosere



Xerosere

Crustose lichen stage(Pioneer),(in Temperate region)

Or blue green algae (in Tropical region)



Homeostasis: An **ecosystem maintains a functional balance or relatively stable state of equilibrium amongst its different components. This phenomenon is called Balance of Nature or Homoeostasis.{Term coined by W.O.Cannon - 1920}.**

Homeostasis is maintained by:

i). Carrying capacity of the environment.

(ii). Recycling of wastes.

(iii). Self regulation (effect of density on reproductive potential. Mortality decreases and natality increases in case of small population while reverse is true in case of large populations).

(iv). Feedback system. It is of two types;

a). Positive feed back: The increased availability of resources increases the no. of utilizers e.g; with increase in population of prey, the predator population will increase.

b). Negative feed back: The increase in no. of members of higher trophic level will decrease the

- Phenology - the science of dealing with the appearance of certain events during life cycle of an organism found in nature.
- Phanerophytes – perennating buds occur at 10 cm or more height above ground level (perennial herbs, shrubs and trees, epiphytes, lianas and succulents).
- Cryptophytes – perennial plants with underground storage parts such as geophytes (root, bulb, stem, tuber, rhizome and corm), helophytes (perennating structures are embedded in mud).
- Therophytes – annual plants which perennate in the form of seeds.
- Euryhaline – organisms which can tolerate wide fluctuation in salt concentration.
- Stenohaline – organisms which cannot tolerate fluctuation in salt concentration and live at nearly constant concentration.

Biodiversity

Biodiversity is the variety of life , there genetic makeup and the natural communities in which they occur. It can be studied on many levels. At the highest level, we can look on different species on the entire earth. On much smaller scale, we can study biodiversity with a pond ecosystem.

According to U.S office Technology Assessment (1987) **biological diversity** is the variety and variability among living organisms and the ecological complexes in which they occur. The term biological diversity refers to the variety of life forms and habitats found in defined area. **UNEP (1992)** defines it as the variety and variability of all animals, plants and micro organisms and the ecological complexes of which they are part. It was coined by W.G Rosen (1985).

Threats to biodiversity:

Extinction is the complete elimination of a wild species , it is natural but slow process but done to unplanned activities of man . The rate of decline of biodiversity has been particularly rapid in last one hundred years. India alone has a total 459 species threatened of which include 86, mammals, 70 birds, 25 reptiles, 3 amphibians, 8 fishes, 23 invertebrates and 244

plants. The biological diversity of **Kohli Hills**, **Lakshadweep Islands** , A.P and Mizoram is under threat because of the conflict between food security of population living there in and biodiversity conservation . There are a number of causes which are known to be threats to the biodiversity.

Habitat Destruction and fragmentation:-

____ it is the most serious threat to wildlife and is due to

(a) **Pollution due to automobiles**.

(b) **Deforestation** which leads to decrease the area of movement so decreasing the reproductive powers. In Tripura forests deforestation id posing a serious threat to shy monkey e. soil erosion.

(c) **Agricultural expansion**

(d) **Increasing urbanization and forest fires.**

Declaration of common Hippopotamuses the endangered species in **2006** Red list is due to habitat loss and over exploitation for meat .

Fragmentation:- It is the process of reduction of habitat into smaller and scattered areas, few birds can reproduce only in dense forests.

Indiscriminate hunting:- For various uses of animals like food , musk, tusk, horn, fur, etc. extinction of dodo bird extensively hilled due to its beautiful feathers . Extinction of cheetah forest mammal of India (last died in Delhi Zoo in 1994)

According to reports of WPST , more than 60 tigers have been poached in different parts of the country during 94-95 . Some Rhino poaching occurred around in Kazi ranga park .

1. **Introduction of exotic (alien) species)**:- is threatening the existence of native species-e.g. American Cockroach (*Periplanta americana*) is threatening the existence native cockroach *Blatta orientalis*.

2. **Over exploitation of natural resources**:- over fishing. Mechanical catching of animal species etc. Is a serious threat to the valid life.

3. **High ways**: The construction of high ways through high biodiversity areas/wild life habitats is also a serious threat to the life of several important organisms.

4. **Environmental pollution**: - large scale use of synthetic compounds .release of radioactive chemicals, oil spills are polluting the rivers and reducing the species number.

Bio diversity Hot spots:- A biodiversity hot spots is a biogeographic region with a significant reservoir of biodiversity that is under threat for humans . The concept of biodiversity hot spots was originated by Norman- Myers in two articles in the environmental list in 1998 and 1990. A biodiversity hot spot posses two strict criteria.

1. It most contains at least 1500 species of vascular plants endemics and it has lost at least 70% of its primary vegetation. Around the world about 25 areas qualify under this definition. These sites support nearly 60% of the world plants, birds, mammals, reptiles and the amphibian's species with a very high share of endemic species.

Only a small percentage of the total land area within hot spots in now protected. Several international organization are working in many ways to conserve bio diversity hot spot. Critical ecosystem partnership fund (CEPF) is a global programme that provides funding and technical assistance to NGO and participation to protect earths richest regions and plant and animal diversity including hot spots. Some of the biodiversity hot spots of India are .

1. **The Western Ghats and Sri Lanka:** the eastern Himalayan Native of Indian Rhinoceros with 10000 species of plants of which 1/3rd are endemics also sharing by golden longor.

2. **Indo-Burma**.

Reasoned for bio diversity loss: in hot spots:-

1. **Habitat destruction.**

2. **Resource miss management**.

3. **Poaching.**

Endangered organisms:- These are those species or taxa whose number have been reduced to critical levels or whose natural habitats have been adversely affected so these are near extinction and may become extinct, if their casual factors continue operating . In India **WLP act 1902** provides four schedules categorizing the fauna of India based on their conservation status. **Schedule I** lists the rare and endangered species which are provided legal protection. It is considered that about 76 species of mammals, 70 species of birds and 3 species of amphibians 25 reptiles are endangered in India.

Plants:- *Berbers nilgheriesis* , *Coptis tacta* (Ranunculaceae)

Animal:- White wines wood duck, mile swan , tortoise, turtle. - Diagnostic X-ray exposures on pregnant women may cause the cancers in children.

Red data book is the document established by IUCN for documenting the rare and endangered species of plants, animals, fungi and also a few local species that exist within a state or country.

Importance:

1. Red data book is beneficial for providing detailed information for studies and researches.
2. It also helps in monitoring programs on rare and endangered species.
3. It helps in protecting the species that are on the verge of extinction.

- The red data book is maintained by the **International Union for Conservation of Nature**. This organization was founded in 1965 and works in the field of conservation of nature and sustainable use of natural resources.
- The headquarters of IUCN are located in **Gland, near Geneva**, in **Switzerland**.
- In 2008, the name of IUCN is changed to **WCU (World Conversation Union)**. It is responsible for necessary actions regarding better environmental management

Support international conversations, agencies and government to formulate new policies and laws for best practice.

- There are three colored pages in The Red data book — Red, Pink, and Green. The color-coded information sheets arranged by species:

Red - species that are endangered,
Amber - vulnerable,
White - rare,
Green - out of danger,
Grey - species that are endangered, vulnerable or rare but with insufficient information.

- The IUCN Red List is an important indicator of the health of the world's biodiversity. It is an influential tool to inform and catalyse action for biodiversity conservation and policy change, critical to protecting the natural resources that different life forms need to survive.

Threatened Species

Threatened species are the species which are susceptible to extinction in the near future.

- International Union for Conservation of Nature has divided the threatened species into three categories:

Depending upon the degree to which they are threatened

I. **Vulnerable,**

II. **Endangered**

III. **Critically endangered**

Critically Endangered Species

According to IUCN, critically endangered species are the wild species that are at the highest risk.

The name suggests that the number of these species have decreased, or it will decrease by 80% within three generations. It is thus considered to be facing an extremely high risk of extinction in the wild.

Endangered (EN) species

Endangered species is a population of organisms which are at risk of becoming extinct as they are either few in numbers, or threatened by changing the environment or by increased predation parameters.

Deforestation is also another factor for the decline in the number of these species as it may lead to a lack of food and/or water.

Vulnerable (VU) species

Vulnerable (VU) species are those that have been categorized by the IUCN, as susceptible to become endangered unless the circumstances threatening its survival and reproduction improve.

Hence, these species face a high risk of extinction in the wild.

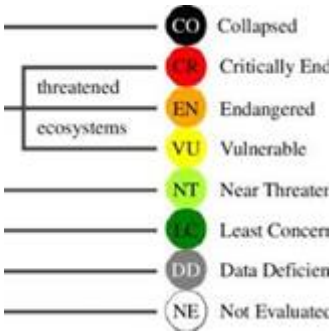
Extinct, Functionally Extinct and Extinct in the wild

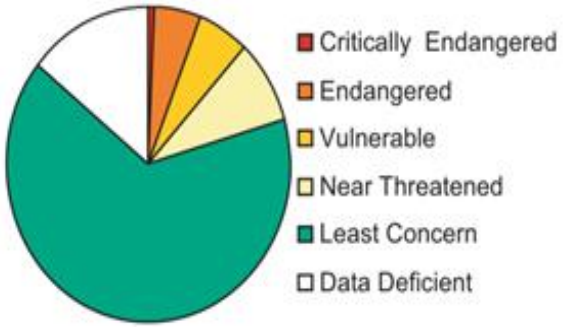
Extinct: ***A species is considered extinct when the last existing member of that species dies***. Extinction is when there are no surviving individuals that are able to reproduce and create a new generation.

Functionally Extinct: A species become **functionally extinct** when only a few of individuals survive, these individuals are unable to reproduce due to poor health, age, sparse distribution over a large range, or due to lack of individuals of both sexes (in sexually reproducing species).

Extinct in the wild: According to IUCN, Extinct in the Wild Species" (EW)are those species that are not known to have any living specimens in the wild, but are maintained only in zoos or other artificial environments.

Some of these species are functionally extinct; as they are no longer part of their natural habitat and it is unlikely the species will ever be re-established in the wild.





Difference between **Red List** and the Red Data Book:

The **red list** contains only the names of the endangered species, however, the Red Data Book contains the information about the species that are on the verge of extinction.

Green data book

The **green data** book is a small pocket-sized book containing environmental data of more than 200 economies. Agriculture, forestry, biodiversity, pollution, and sanitation are the key indicators.

Critically endangered species of India as per the Red Data List:

The critically endangered species of India are:

- Kashmir Stag
- River Dolphins
- Kondana rat
- Malabar Civet

In-situ biodiversity conservation:- In site conservation ,means outside conservation. Here the plant or animal species are protected In its natural habitat. This is carried out by two method.

1. Either by protecting or cleaning up the habitat itself.

2. By defending the species from predators and here stress is laid upon the protection of total ecosystem and this leads to the declaration of protected areas. For protecting such areas, legal or other effective strategies are used.

Protected parks in India:-

1. **National Parks**- A national park is an area which is strictly reserved for the betterment of wild life and where activities like forestry, gazing or cultivation are not permitted. In these parks even private ownership rights are not allowed.

   There are about 66 national parks in India (1988) spread over an area of 3398814 sq. km or nearly 1% of the countries geographical area. Some important national parks of India are Kazi ranga national park Assam, famous national park for one hundred Rhinoceros, Sunder banns west Bengal, desert national park Rajasthan.

Sanctuaries:- A sanctuary is a protected area which is reserved for the conservation of only animals and Human activities like harvesting of timber , collection of minor forest products or private ownership rights are allowed so long as they do not interfere with the well being of animals. There are about 392 sanctuaries in India which cover about 3.2% of countries geographical areas. Some important sanctuaries of our India are;-

1. Annamali sanctuary Tamil Nadu (958 sq. km).
2. Dachigam sanctuary (89 sq. km)
3. Periyor sanctuary Kerala (777 sq. km)

Biosphere reserves:- A biosphere reserve is a specified area in which multiple use of the land is permitted by dividing it into certain zones specified for a particular activity. Under **MAB** programme UNESCO established a number of biosphere reserve in the world and its concept was launched in 1975 by **UNESCO** for dealing with the conservation of ecosystem and the genetic material or resources containing there in.

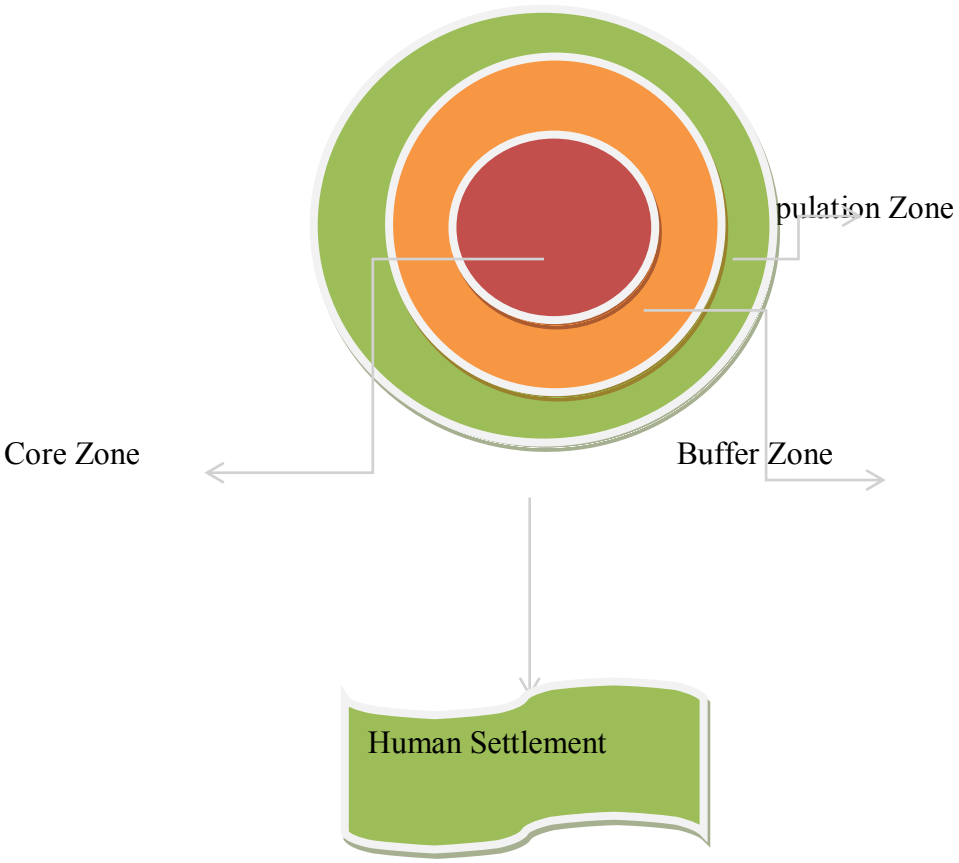
Biosphere reserves are basically divided into 3 zones.

1. **Core zone**: - it lies at centre where no Human activities are allowed. It is legally protected.
2. **Buffer zone**:- In this zone limited human interference is allowed . *It* surrounds core area.
3. **Transaction zone** (Manipulation zone):- In this zone multiple human activities are allowed but ecology is not permitted to be disturbed. It is the outer most part of the bio reserve. India has 14 bio sphere reserves. There are examples of natural

biomes with human as an integral part. 1. Niligiri (1986) Nanda Devi (1988), Panchmari Biosphere reserve.

Objectives:

1. To conserve bio diversity for present and future race use, the diversity and integrity of biotic communities of plants and animals with in natural ecosystem.
2. To provide areas for ecological and environmental research.
3. To provide facilities for Education and training.
4. To promote economic develop.



(Bio Sphere reserve)

Environmental pollution:- Un limited exploitation of nature by man disturbed delicate ecological balance between living and non living components , of the biosphere . it is very common to find warning at places as **air unfit for breathing** , **water unfit for drinking** . India today is one of the first 10 industrial countries of the world. But pollution is the necessary evil of all development. Due to the lock of development of culture of pollution, there has resulted a heavy back log of gaseous, liquid and solid pollution in our country.

Pollution:- pollution is an undesirable change in the physical, chemical or biological characteristics of air, water and soil that may harmfully effect the life or create a potential health hazard of any living organisms. Pollution is thus direct or indirect in any component of the biosphere that is harmfull to the living component and in particular undesirable for man affecting adversely the industrial processes, cultural and natural assets or general environment.

Pollutants:- Any substance which causes pollution is called pollutant. A pollutant may thus include any chemical or geochemical (dust, sediments) substances, biotic component or its products, physical factors (heat) that is released intentionally by man into the environment I such a manner that it may have adverse harmful or unpleasant affects.

Pollution may also be defined as any solid, liquid and gaseous substance present in such a concentration may be or tend to be injurious to the environment.

Environmental pollutants:-

1. Deposited matter:- soot, dust, girt ,etc.
2. Gases:- oxides of N, S, C and halogens.
3. Acid droplets as H_2SO_4, HNO_3 etc.
4. Metals.
5. Agro chemicals:- bio acids and pesticides.
6. Complex organic substances.

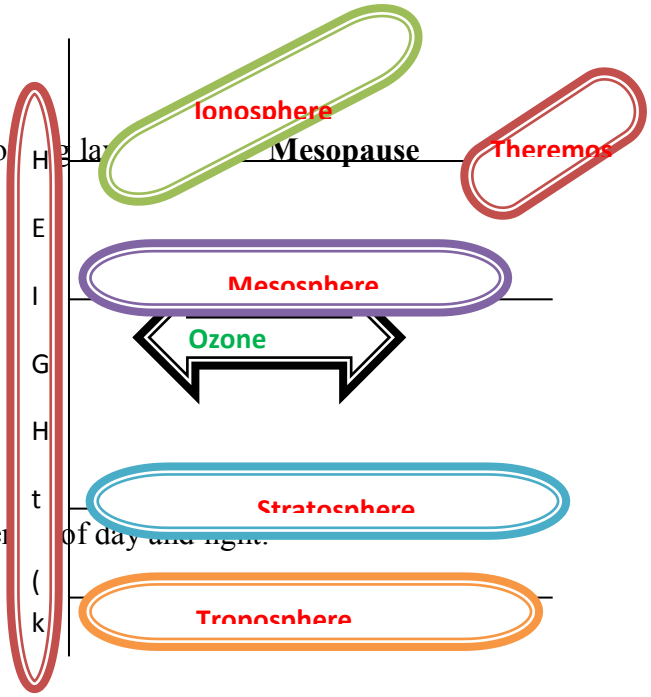
7. Solid wastes and noise.

Kinds of pollution:- on the basis of type of the environment being polluted, we may recognize air pollution , water pollution, soil pollution etc of the variety of pollutants, we recognize two basic type of pollutants.

1. **Non biodegradable pollutants**:- these are the ,materials and poisonous substances' like ammonium cams, mercuric salts. DDT etc that either do not degrade or degrade every slowly in nature.

2. **Biodegradable pollutants**:- they are domestic wastes that can be rapidly decomposed under natural conditions.

Air pollution:- The earths vertical extended atmosphere, a gaseous envelop is divided into follo[illegible] la[illegible] **Mesopause**



i. Troposphere 5km.

ii. Stratosphere up 45 km 90° c. 50

iii. Mesosphere (45 – 80 km) 80°c.

iv. Thermo sphere (80 km above).

It is the source of essential gases, maintains a narrow differ[illegible] of day and night.

Normal composition of clean air.

N_2 78% Ar 0.934%

O_2 20% CO_2 0.0314%

CH_4 0.002%, H_2 0.0005% & other gases.

Air pollution:- introduction of chemical , SPM, biological materials within the atmosphere making the living organisms discomfort particularly humans or cause damage to the natural environment.

Sources:- Air pollution results from gaseous emissions from mainly industry, thermal power stations , automobile domestic combustion etc.

2. **Industrial chimmeny wastes**:- there are a number of industries which are sources of pollution.. Petroleum refineries are the major sources of gaseous pollutants. The chief gases released are **SO2** and **NOx**. Mathura based petroleum refinery is posing great threat Taj Mahal in Agra and other monuments in Fatipur sikiri complex's. Cement factories emit plenty of dust which is potential health hazard. Also stone crushers emit **SPM** and in such areas SPM are more than five times the industrial safety limits. A study was made by CPCB in Najafgarh Road. This area has 32 air polluting units which emit every month 75.3 tones of SO2 and 794. 5 tones SPM.

3. **Thermal power stations**:- there are a number of thermal power stations and super thermal power stations in country. The coal consumption in thermal plants is several million tons and the chief pollutants released are fly ash, SO2 and hydro carbons. The three thermal power stations at the indraprata state rajgat and badarpur in Delhi are the main sources of air pollution. Budarpur the largest consumes daily about **10000** tons of coal.

4. **Automobiles**:- the toxic vehicular exhausts are a source of considerable air pollution, next only to thermal power plants. In all the major cities of the country about 800-1000 tons of pollutants are being emitted in to the air daily of which 50% comes from automobiles exhausts. In micro cities vehicular exhausts accounts for 70% of all Co, 50% of all hydro carbons 30-40% of all oxides and 30% of all SPM. It is estimated that a car (without cleaning) on burning 1000 liters of fuel emits 350kg of Co, 0-6 kg SO2 , 0.1 kg lead, 1.5 kg SPM.

    i. **Dust fall measurement**:- it is a simplest means of evaluating air quality . Here sampling period of dust settlement in buckets' is 30 days and results are reported as dust settled (sq. mts.) 30 days.

    ii. **High value sampler**:- here the sampler operates like a vacuum cleaner by simply forcing a large quantity of air through a flitter. sampling period is generally 24 hrs. And over 70000 cubic feet of air is sacked through the fitter during the time.

    Effects:- exposure to air pollution is associated with numerous effects on human health including pulmonary, cardiac, vascular and neurological. Effects

can be acute or chronic. Some chronic diseases may be long capacity and longer cancer due to long term exposure to air pollutants.

Control:- The main sources of pollution are .

1. Motor vehicles.
2. Industries their chimney wastes.
3. Fossil fuels (coal based thermal power plants. Steps are to be taken to control pollution as source as well as after the release of pollutants in the atmosphere.

Source correction:- this is the easiest solution to air pollution problem where we stop the guilty . **(a) Source Correction Methods:**

Industries make a major contribution towards causing air pollution. Formation of pollutants can be prevented and their emission can be minimised at the source itself.

By carefully investigating the early stages of design and development in industrial processes e.g., those methods which have minimum air pollution potential can be selected to accomplish air-pollution control at source itself.

These source correction methods are:

(i) Substitution of raw materials:

If the use of a particular raw material results in air pollution, then it should be substituted by another purer grade raw material which reduces the formation of pollutants. Thus,

(a) Low sulphur fuel which has less pollution potential can be used as an alternative to high Sulphur fuels, and,

(b) Comparatively more refined liquid petroleum gas (LPG) or liquefied natural gas (LNG) can be used instead of traditional high contaminant fuels such as coal.

(ii) Process Modification:

The existing process may be changed by using modified techniques to control emission at source. For example,

(a) If coal is washed before pulverization, then fly-ash emissions are considerably reduced.

(b) If air intake of boiler furnace is adjusted, then excess Fly-ash emissions at power plants can be reduced.

(iii) Modification of Existing Equipment:

Air pollution can be considerably minimised by making suitable modifications in the existing equipment:

(a) For example, smoke, carbon-monoxide and fumes can be reduced if open hearth furnaces are replaced with controlled basic oxygen furnaces or electric furnaces.

(b) In petroleum refineries, loss of hydrocarbon vapours from storage tanks due to evaporation, temperature changes or displacement during filling etc. can be reduced by designing the storage tanks with floating roof covers.

(c) Pressurising the storage tanks in the above case can also give similar results.

(iv) Maintenance of Equipment:

An appreciable amount of pollution is caused due to poor maintenance of the equipment which includes the leakage around ducts, pipes, valves and pumps etc. Emission of pollutants due to negligence can be minimised by a routine checkup of the seals and gaskets.

(b) Pollution Control Equipment:

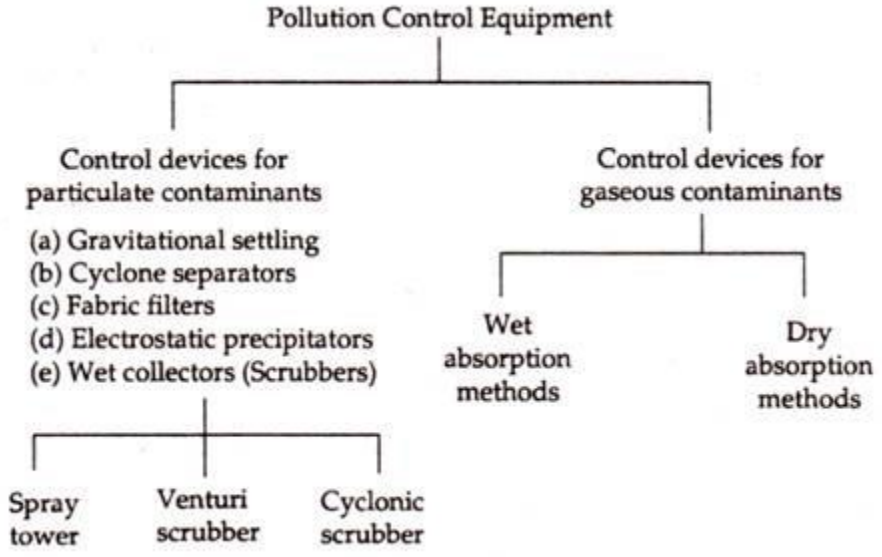
Sometimes pollution control at source is not possible by preventing the emission of pollutants. Then it becomes necessary to install pollution control equipment to remove the gaseous pollutants from the main gas stream.

The pollutants are present in high concentration at the source and as their distance from the source increases they become diluted by diffusing with environmental air.

Pollution control equipment's are generally classified into two types:

(a) Control devices for particulate contaminants.

(b) Control devices for gaseous contaminants.



In the present book only the control devices for particulate contaminants are dealt with.

Control Devices for Particulate Contaminants:

(1) Cyclone Separators (Reverse flow Cyclone):

Instead of gravitational force, centrifugal force is utilized by cyclone separators, to separate the particulate matter from the polluted gas. Centrifugal force, several times greater than gravitational force, can be generated by a spinning gas stream and this quality makes cyclone separators more effective in removing much smaller particulates than can possibly be removed by gravitational settling chambers.

A simple cyclone separator (Fig 5.2) consists of a cylinder with a conical base. A tangential inlet discharging near the top and an outlet for discharging the particulates is present at the base of the cone.

Mechanism of Action:

The dust laden gas enters tangentially, receives a rotating motion and generates a centrifugal force due to which the particulates are thrown to the cyclone walls as the gas spirals upwards inside the cone (i.e. flow reverses to form an inner vortex which leaves flow through the outlet). The particulates slide down the .walls of the cone and are discharged from the outlet.

(2) Fabric Filters (Baghouse Filters):

In a fabric filter system, a stream of the polluted gas is made to pass through a fabric that filters out the particulate pollutant and allows the clear gas to pass through. The particulate matter is left in the form of a thin dust mat on the insides of the bag. This dust mat acts as a filtering medium for further removal of particulates increasing the efficiency of the filter bag to sieve more sub micron particles (0.5 μm).

A typical filter (Fig 5.3) is a tubular bag which is closed at the upper end and has a hopper attached at the lower end to collect the particles when they are dislodged from the fabric. Many such bags are hung in a baghouse. For efficient filtration and a longer life the filter bags must be cleaned occasionally by a mechanical shaker to prevent too many particulate layers from building up on the inside surfaces of the bag.

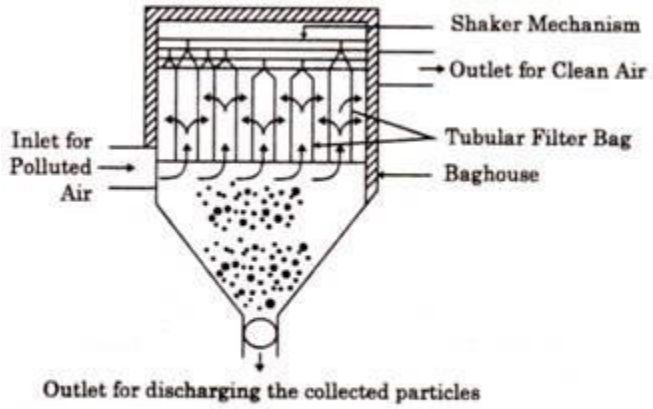


Fig. 5.3 Fabric Filter (Baghouse Filter)

(3) Electrostatic Precipitators:

The electrostatic precipitator (Fig. 5.4) works on the principle of electrostatic precipitation i.e. electrically charged particulates present in the polluted gas are separated from the gas stream under the influence of the electrical field.

A typical wire and pipe precipitator consists of:

(a) A positively charged collecting surface (grounded).

(b) A high voltage (50 KV) discharge electrode wire.

(c) Insulator to suspend the electrode wire from the top.

(d) A weight at the bottom of the electrode wire to keep the wire in position.

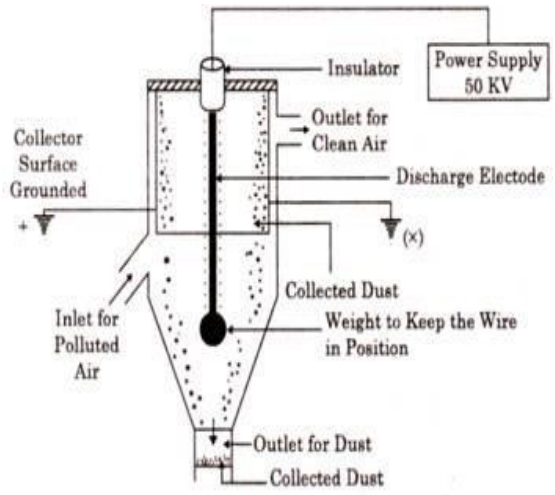


Fig. 5.4 Electrostatic Precipitator

Mechanism of Action:

The polluted gas enters from the bottom, flows upwards (i.e. between the high voltage wire and grounded collecting surface). The high voltage in the wire ionises the gas. The negative ions migrate towards the grounded surface and pass on their negative charge to the dust particles also. Then these negatively charged dust particles are electrostatically drawn towards the positively charged collector surface, where they finally get deposited.

The collecting surface is rapped or vibrated to periodically remove the collected dust-particles so that the thickness of the dust layer deposited does not exceed 6 mm, otherwise the electrical attraction becomes weak and efficiency of the electrostatic precipitator gets reduced.

As the electrostatic precipitation has 99 + percent efficiency and can be operated at high temperatures (600°C) and pressure at less power requirement, therefore, it is economical and simple to operate compared to other devices.

(4) Wet Collectors (Scrubbers):

In wet collectors or scrubbers, the particulate contaminants are removed from the polluted gas stream by incorporating the particulates into liquid droplets.

Common wet scrubbers are:

(i) Spray Tower

(ii) Venturi Scrubber

(iii) Cyclone Scrubber

(i) Spray Tower:

Water is introduced into a spray tower (Fig. 5.5.) by means of a spray nozzle (i.e. there is downward flow of water). As the polluted gas flows upwards, the particulates (size exceeding 10 μm) present collide with the water droplets being sprayed downward from the spray nozzles. Under the influence of gravitational force, the liquid droplets containing the particulates settle to the bottom of the spray tower.

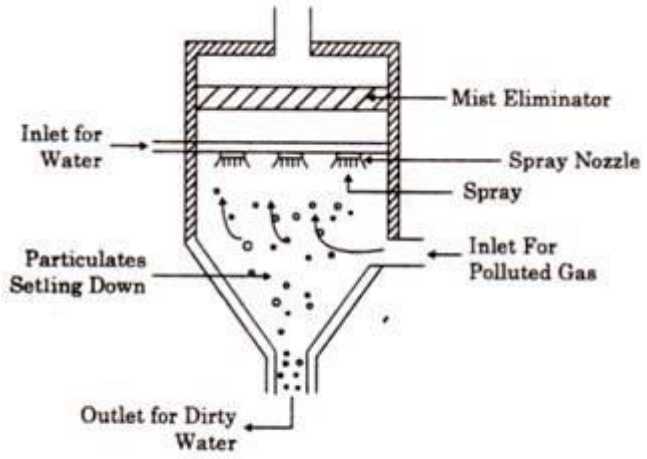


Fig. 5.5. Spray Tower

(iii) Cyclone Scrubber:

The dry cyclone chamber can be converted into a wet cyclone scrubber by inserting high pressure spray nozzles at various places within the dry chamber (Fig. 5.7).

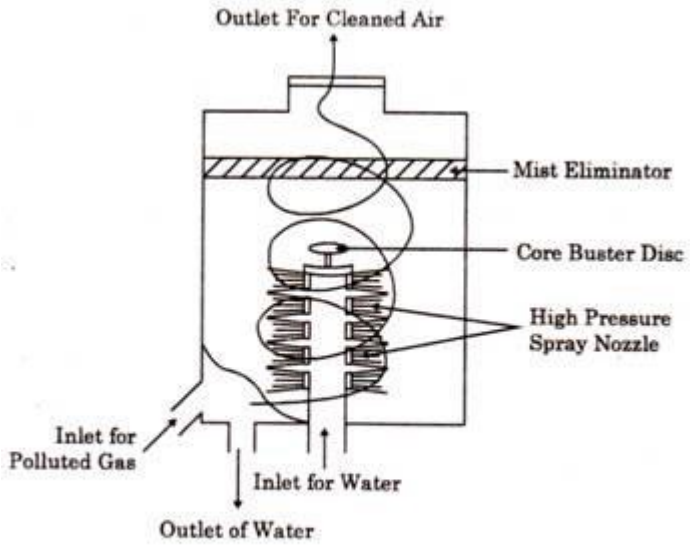


Fig. 5.7. Cyclone Scrubber

The high pressure spray nozzles generate a fine spray that intercepts the small particles in the polluted gas. The centrifugal force throws these particles towards the wall from where they are drained downwards to the bottom of the scrubber.

(c) Diffusion of Pollutants in Air:

Dilution of the contaminants in the atmosphere is another approach to the control of air pollution. If the pollution source releases only a small quantity of the contaminants then pollution is not noticeable as these pollutants easily diffuse into the atmosphere but if the quantity of air contaminants is beyond the limited capacity of the environment to absorb the contaminants then pollution is caused.

However, dilution of the contaminants in the atmosphere can be accomplished through the use of tall stacks which penetrate the upper atmospheric layers and disperse the contaminants so that the ground level pollution is greatly reduced. The height of the stacks is usually kept 2 to $2^1/_2$ times the height of nearby structures.

Dilution of pollutants in air depend on atmospheric temperature, speed and direction of the wind. The disadvantage of the method is that it is a short term contact measure which in reality brings about highly undesirable long range effects.

This is so because dilution only dilutes the contaminants to levels at which their harmful effects are less noticeable near their original source whereas at a considerable distance from the source these very contaminants eventually come down in some form or another.

(d) Vegetation:

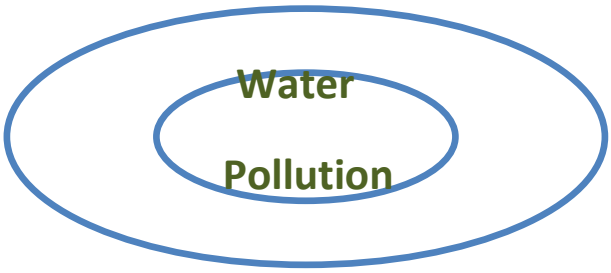
Plants contribute towards controlling air-pollution by utilizing carbon dioxide and releasing oxygen in the process of photosynthesis. This purifies the air (removal of gaseous pollutant—CO_2) for the respiration of men and animals.

Gaseous pollutants like carbon monoxide are fixed by some plants, namely, Coleus Blumeri, Ficus variegata and Phaseolus Vulgaris. Species of Pinus, Quercus, Pyrus, Juniperus and Vitis de-pollute the air by metabolising nitrogen oxides. Plenty of trees should be planted especially around those areas which are declared as high-risk areas of pollution.

(e) Zoning:

This method of controlling air pollution can be adopted at the planning stages of the city. Zoning advocates setting aside of separate areas for industries so that they are far removed from the residential areas. The heavy industries should not be located too close to each other.

New industries, as far as possible, should be established away from larger cities (this will also keep a check on increasing concentration of urban population in a few larger cities only) and the locational decisions of large industries should be guided by regional planning. The industrial estate of Bangalore is divided into three zones namely light, medium and large industries. In Bangalore and Delhi very large industries are not permitted.



Water pollution is defined as the addition of some substance (organic, inorganic, biological, and radiological) or factor (e.g., heat) which degrades the quality of water so that it either becomes health hazard or unfit for use.

Sources of Water Pollution and Effect of Water Pollutants:

1. Domestic Wastes and Sewage:

Raw sewage contaminates water with pathogens. Microorganisms causing degradation of sewage take up most of the oxygen present dissolved in water.

Sewage produces foul-odour and makes the water brownish and oily. Organic waste gives rise to scum and sludge that makes the water unfit for recreational and industrial use.

It induces the growth of some algal blooms that add to the depletion of oxygen, addition of more organic matter and fouling of water. Modern day detergents degrade very slowly. They, therefore, accumulate and render the water unfit for human and animal use. The phosphates present in detergents further stimulate algal growth that add to the organic loading of water.

2. Surface Run-Off:

The pollutants present on the surface of land and fertilizers added to the soils" are washed down into water reservoirs and water courses during rains. This flow of fertilizer rich water into streams and lakes gives rise to eutrophication.

3. Industrial Effluents:

They are industrial wastes which are allowed to pass into water bodies. The important toxic chemicals presents in them are:

(i) Mercury:

It is released during combustion of coal, smelting of metallic ores, chloralkali, paper and paint industries. Mercury is persistent. In water it gets changed into water soluble dimethyl form [(CH,) 2Hg)] and enters the food chain accompanied by biological or ecological amplification. Human beings feeding on poisoned animals and fishes develop a crippling deformity called minamata disease.

(ii) Lead:

The sources of lead pollution are smelters, battery, industry, paint, chemical and pesticide industries, automobiles' exhausts, etc. It is mutagenic and causes anaemia, headache, and bluish lines round the gums.

(iii) Cadmium:

It shows biological amplification and accumulates inside kidneys, liver, pancreas and spleen. It causes renal damage, emphysema, hypertension, testicular necrosis and damage to placenta.

(iv) Other metals: Copper, zinc, nickel, titanium, etc. cause toxaemia and change in enzyme functioning.

(v) Liquid Effluents: Several types of liquid effluents containing toxic chemicals, acids and bases, are added to the rivers and other water bodies. They kill fish and other aquatic life besides being toxic to human beings. Some examples of large scale effluent addition into rivers are Yamuna (near Okhla, Delhi), Gomti (near Lucknow), Ganga (near Kanpur) and Hoogli (near Calcutta).

4. Thermal Pollution:

Many industrial processes are causing thermal pollution leading to higher temperatures. These industries do not contaminate the water supply, but use a lot of water for cooling purposes and return this water to the stream at a higher temperature, which affect the biotic components in the aquatic habitat. Warmer water holds less oxygen (14 ppm at 0°C, 1 ppm at 20°C) and hence its Biological Oxygen Demand (BOD) increases. Green algae are replaced by less desirable blue-green algae. Trout eggs fail to hatch while Salmon does not spawn at higher temperature.

5. Marine Pollution:

Oceanic pollution is caused by ship-generated discharges of oil and petroleum products, noxious liquids, packaged dangerous goods, sewage, garbage etc. Migrating birds caught m the oil slicks losing their power of flight due to closer interlocking of barbules of the feathers, is common enough. Employment of detergents to clean the oil slicks has been found to be harmful to marine life.

Eutrophication:

Any lake or sheet of fresh water, to begin with is oligotrophic, supporting a minimum of life forms. Therefore, its productivity would be minimal. But in times it comes to be occupied by immigrant life forms, which on death and decay would make further immigration possible.

The lake is then said to have reached a mesotrophic level. Finally, it comes to be occupied by a rich flora and fauna when it is said to have reached the eutrophic level i.e., when its productivity had reached its maximum. In nature, this would take place through thousands of years but with industrialisation and other forms of human activity, this process of eutrophication, as it is called is achieved into a few decades.

Degree of Water Impurity:

Water pollution by organic wastes is measured in terms of bio-chemical oxygen demand (BOD). BOD is defined as the amount of oxygen required by microorganisms to stabilize decomposable organic matter in waste under aerobic condition. It is oxygen required in milligrams for five days to metabolize waste present in one litre of water at 20°C. A weak organic waste will have BOD below 1500 mg/litre, medium between 1500—1400 mg/litre while a strong waste above it. Since BOD is limited to organic wastes, it is not a reliable method of measuring water pollution. Another slightly better mode is COD or chemical oxygen demand. It measures all oxygen consuming pollutant materials present in water.

Chemical Oxygen Demand (COD):

It is an indicator of water or effluent quality which measures oxygen demand by chemical (as distinct from biological) means using potassium dichromate as the oxidizing agent. Oxidation takes 2 hours and the method is thus much quicker than a 5-day BOD assessment. The BOD: COD ratio is fairly constant for a given effluent.

Control of Water Pollution:

Water pollution can be controlled to a large extent on the principle, "the solution to pollution is dilution."

The various methods for the control of water pollution are discussed below:

1. The sewage pollutants are subject to chemical treatment to change them into non-toxic substances or make them less toxic.

2. Water pollution due to organic insecticides can be reduced by the use of very specific and less stable chemicals in the manufacture of insecticides.

3. Oxidation ponds can be useful in removing low level of radioactive wastes.

4. Thermal pollution can be reduced by employing techniques—through cooling, cooling ponds, evaporative or wet cooling towers and dry cooling towers. The purpose is that the waters in the rivers and streams should not get hot.

5. Domestic and industrial wastes should be stored in large but shallow ponds for some days. Due to the sun-light and the organic nutrients present in the waste there will be mass scale growth of those bacteria which will digest the harmful waste matter.

6. Polluted water can be reclaimed by proper sewage treatment plants and the same water can be reused in factories and even irrigation. Such a treated water being rich in phosphorus, potassium and nitrogen can make good fertilizer.

7. Suitable strict legislation should be enacted to make it obligatory for the industries to treat the waste water before being discharged into rivers or seas.

8. Water hyacinth popularly known as Kaloli and Jalkumbhi, can purify water polluted by biological and chemical wastes. It can also filter out heavy metals like cadmium, mercury, lead and nickel as well as other toxic substances found in industrial waste waters.

Green House Effect

Green house effect: Since CO_2 is confined exclusively to the troposphere, its higher concentration may act as a series pollutant. Under normal conditions the temperature at the surface of the earth is maintained by the energy balance of the sun rays that strike the earths and heat that is radiated back into space. However, when there is an increase in CO_2 concentration, the thick layer of this gas prevents the heat from being re-radiated out. Thus thick CO_2 layer thus functions like the glass panels of greenhouse or the glass windows of a motar car, allowing the sunlight to filter through but preventing the heat from being re-radiated in outer space. This is called **Greenhouse Effect**. Thus most of the heat is absorbed

by CO_2 layer and water vapors' in the atmosphere, which adds to the heat that is already present. The net result is the heating up of earths atmosphere. The increasing CO_2 levels tend to warm the air in the lower layers of atmosphere on a global scale. Nearly 100 yrs ago the CO_2 level was 275 ppm. Today it is 350 ppm, and by the year 2035 it is expected to reach 450 ppm, so imagine the earths temperature.CO_2 increases the earths temperature by 50% while CFCs are responsible for another 20% increase

According to some estimates CO_2 in the air may have risen by 25% since the middle of the 19th century, it may even be doubled by 2030 A.D. doubling the CO_2 level will increase the global mean temperature (15 C) by 2 degrees. There are also other gases which contribute to the green house effect. These are SO_2, NOx, CFCs discharged by the industry and agriculture.

Some analysts believe that the changes in earths mean temperature will be apparent by 2050, when the temperature would increase by 1.5 to 4.5 C. Changes will be the least in the tropics and the most at the poles.. According to U.S. Scientist, **George Woodwell**, India's annual monsoon rains may even cease altogether. A rise in the sea level of 50-100 cm caused by ocean warming will flood lands in Bangladesh and W.B. due to greenhouse effect there will be more hurricanes and cyclones and within next 25yrs there will be rise in sea level by 1.5-3.5 meters and in Bangladesh alone 1.5 million people will have to move away or drown.

Global Warming

Global warming: If the causes of green house effect remain unchecked, it could alter temperatures, rainfall and sea level of the earths. The UNEP choosed a slogan called global warming to alert the people on world environment day, June 5 1989. So global warming is a phenomena which results freom the emissions of green houyse gases likeCFCs, CH_4, NOx and SO_2 etc ,thus alters earths temperature, and hydrological cycle and its effects are global.

Predicting the consequences of global warming is one of the most difficult tasks for the world's climate researchers. This is because the natural processes that cause rain, hail and

snow storms, increases in sea level and other expected effects of global warming are dependent on many different factors. It is also difficult to predict the size of the emissions of greenhouse gases in the coming decades, as this is determined to a great extent by political decisions and technological breakthroughs.

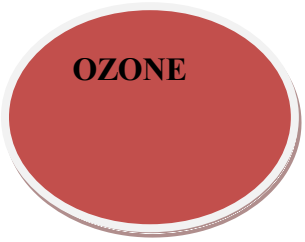
The effects that can be predicted include:

1.more drought and more flooding

2 less ice and snow

3 more extreme weather incidents

4 rising sea level



Ozone is a **secondary air pollutant,** when present in the **troposphere,** when present in **streptosphere,**it becomes very **harmful** to the human beings. It is formed by the **oxides of nitrogen molecules** reacting with **oxygen molecules. NO_2 breaks in to NO and O in presence of sunlight.**

$$NO_2 \longrightarrow NO+O$$

This O then combines with O_2 molecules and form O_3 and energy *M*

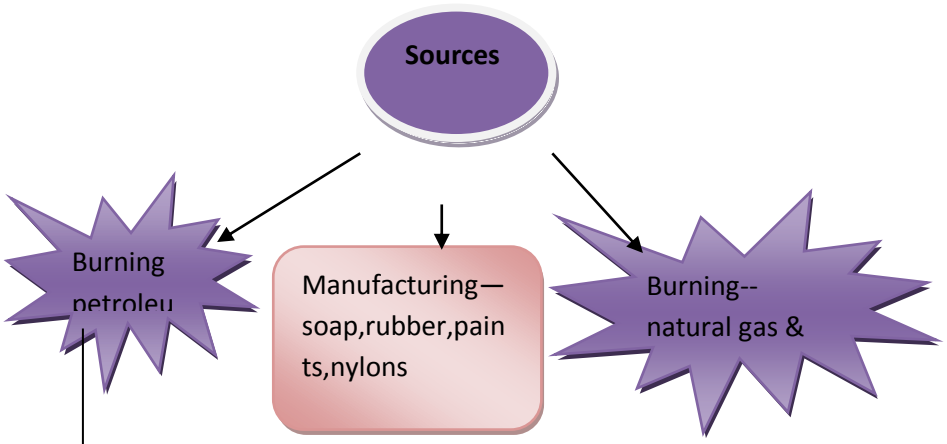
$$O + O_2 \longrightarrow O_3 + M$$

This O_3 combines with NO and give NO_2 and O_2

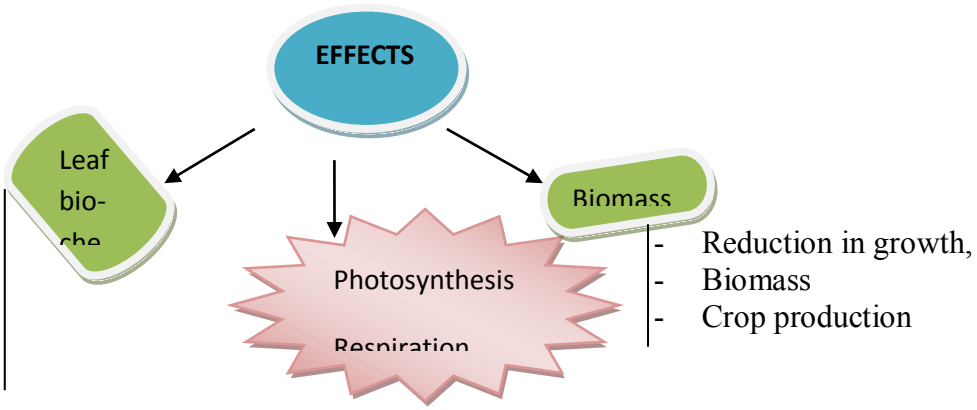
$$NO + O_3 \longrightarrow NO_2 + O_2$$

$$NO_2 + O_2 \rightleftharpoons NO + O_3$$

This NO_2 reacts with incomplete unburnt unsaturated hydrocarbons and acts as secondary pollutant and increases concentration of O_3.



- Automobiles, diesel, rail engines, aeroplanes,
- Process of petroleum refining



- Reduction in growth,
- Biomass
- Crop production

- Due to high oxidixing potentionality ozone reacts with the cell wall constituents and release free-radicles thus alter leaf bio-chemistry.
- -Reduction in rate of photosynthesis
- Respiration

➢ Ozone is a gaseous envelop in the stratosphere which protects us from the harmful UV radiations from the sun. its molecular formula is O_3. Inspite of being in such a

small proportion (o.2-0.7ppm), it plays a major role in the climatology and biology of our earth. Its depletion by human activities may have serious implications and this becomes a subject of much concern over last few years. On the other hand ozone is also formed in the atmosphere through chemical reactions involving certain pollutants (SO_2, NO_2, Aldehydes) on absorption of UV radiations. Ozone when present in troposphere acts as a destroyer as well as protector in stratosphere.

Ozone: the destroyer: ozone layer in stratosphere (8-16 km) has two properties.

Firstly, it absorbs UV radiations and thus protects all life on earth from harmful effects of radiations. Secondly, by absorbing the UV radiation the ozone layer heats up the stratosphere, causing temperature inversion. It limits the vertical mixing of pollutants thereby causing the dispersal of pollutants near earth's surface which upon absorbing light and UV radiations forms the ozone near earth's surface. Ozone and other pollutants such bas PAN and hydrogen peroxide are formed by light dependent reactions between NO_2 and hydrocarbons. Ozone is also formed by NO_2 under UV radiations called photochemical smog.

Increase in ozone conc. near earth's surface reduces crop yield significantly. In plants it damages leaves, decreases yield and quality of the product. In humans it causes irritation of eyes, nose asthma and lung diseases.

Ozone - the protector: ozone protects us from harmful UV radiations from the sun it filters out all the radiations' below 3000 A and keeps the temperature warm on earth necessary with the life sustaining processes.

Ozone depletion: thinning of ozone means ozone depletion. The first global conference on the depletion of ozone layer was held **in Vienna (Austria)** in **1985**, the year scientists discovered hole in south pole, as big as united states. Some pollutants enter in our stratosphere and remain there for years until they react with ozone and convert it to other products. Major pollutants responsible for ozone depletion are CFCs, Nox from fertilizers and hydrocarbons. CFCs are widely used as coolants in A.C's and refrigerators, cleaning solvents, fire extinguishers. The supersonic aircrafts are causing major disturbances in stratosphere ozone levels. CFC's are known to deplete ozone by 14%at the current emission and NOx by 3.5%

Depletion of ozone has serious effects. One percent reduction in ozone increases UV radiations on earth by 2% cancer is the best established threat to man. Nearly 6000 people die every year in USA.. Plant crop yield is reduced by b20-50%

So **Montréal protocol 1987** was signed by European countries which called for a 50% reduction in the use of CFC's by 1998 reducing to the level of 1986.

Solid waste pollution

The solid waste includes glass containers as bottles, crokeries, plastic containers, polythene and other packing materials that are used and then thrown as garbage. These pile up at public places and cause obstruction in daily life. Besides these there are also other used things like automobile spares, machines ,cycle parts etc that are thrown as junk .the wastes from building material, dead animal skeletons and heaps of crop residues also contribute to solid wastes.

Solid wastes are causing much problem in developed countries as USA and other European countries. In India also several million tons of solid waste is dumped along highways and other cities as Delhi, Madras, Jaipur etc. on an average two million tons of solid waste is generated in class Ist cities per year where as in Class 2nd cities about 0.25 million tons per year. To solve these problems technologies have been developed to recycle most of the solid wastes e,g paper cans, newspapers and metallic parts and rubber etc.

Solid waste management; solid waste can be disposed to land or oceans. Solid wastes can also be recovered and reprocessed, a procedure popularly known as recycling. Before disposal or recovery, however, the waste must be collected. All these I,e collection, disposal and or recovery form a part of solid waste management system.

(i) **Collection**; the common method of collection is by covered trucks. Much time is spent in loading and its transport to dumping or recovery site. Many new devices and methods have been proposed to cut collection costs. these include

(A) **Garbage grinders** reduce the amount of garbage in refuse .If such grinders are used, this cuts the frequency of collection in communities

(b**) Pneumatic pipes** receive the refuse from the park and deliver the waste to a central processing plant.

(c)**Compacters** in the kitchen also reduce cost collection

(d) **Transfer stations** are applicable to large communities .It involves several stations scattered around a city to which collection trucks bring the waste .The drive to the nearly station is fairly short. At transfer stations bulldozers cram this refuse into large vans which in turn take the material to the final disposal

(II) **DISPOSAL**: The dump is the most inexpensive and most popular means of solid waste disposal .One of the most acceptable means of disposal is sanitary landfills.

Sanitary landfills differ markedly, as these are engineered operations, designed and operated according to acceptable standards .The basic principle of Land Fill operation is to deposit the refuse, compact it with bulldozers and cover the material with at least six inches of dirt at the conclusion of each days operation and a final cover of two feet soil when the area is full. Finally the area be utilized for public use or private use after the operation is complete.

(III). **RESOURCE RECOVERY**: Also known as recycling, this is theoretically very appealing. After the material has been collected from consumers it must b cleaned sold to an industry, transported, remanufactured and sold once again to consumer's .The two basic reasons for a cycling are conservation of resources and reduction in volume of refuse to be disposed. Some of the common materials suggested as recyclable are paper, metals, glass, and organics

Radioactive Waste

Radioactive Waste: Radioactive waste is waste that contains radioactive material. Radioactive waste is usually a by-product of nuclear power generation and other applications of nuclear fission or nuclear technology, such as research and medicine. Radioactive waste is hazardous to most forms of life and the environment, and is regulated by government agencies in order to protect human health and the environment.

Radioactive Waste Management: The management and disposal of radioactive waste is called radioactive waste management.

For radioactive waste, this means isolating or diluting it such that the rate or concentration of any radionuclide's returned to the biosphere is harmless. To achieve this, practically all radioactive waste is contained and managed, with some clearly needing deep and permanent burial. From nuclear power generation, unlike all other forms of thermal electricity generation, all waste is regulated – none is allowed to cause harmful pollution.

All parts of the nuclear fuel cycle produce some radioactive waste and the cost of managing and disposing of this is part of the electricity cost (***i.e.*** it is internalised and paid for by the electricity consumers). Nuclear power is characterised by the very large amount of energy produced from a very small amount of fuel, and the amount of waste produced during this process is also relatively small. However, much of the waste produced is radioactive and therefore must be carefully managed as hazardous material.

Radioactive waste is not unique to the nuclear fuel cycle. Radioactive materials are used extensively in medicine, agriculture, research, manufacturing, non-destructive testing, and minerals exploration. Unlike other hazardous industrial materials, however, the level of hazard of all radioactive waste – its radioactivity – diminishes with time. All toxic wastes need to be dealt with safely – not just radioactive waste – and in countries with nuclear power, radioactive wastes comprise a very small proportion of total industrial hazardous waste generated.

Types of radioactive waste

Radioactive waste includes any material that is either intrinsically radioactive, or has been contaminated by radioactivity, and that is deemed to have no further use. Government policy dictates whether certain materials – such as spent nuclear fuel and plutonium – are categorised as waste.

Every radionuclide has a half-life – the time taken for half of its atoms to decay, and thus for it to lose half of its radioactivity. Radionuclides with long half-lives tend to be alpha and beta emitters – making their handling easier – while those with short half-lives tend to emit the more penetrating gamma rays. Eventually all radioactive waste decays into non-radioactive elements. The more radioactive an isotope is, the faster it decays. Radioactive waste is

typically classified as either low-level (LLW), intermediate-level (ILW), or high-level (HLW), dependent, primarily, on its level of radioactivity:

LOW-LEVEL WASTE

Low-level waste (LLW) has a radioactive content not exceeding four giga-becquerels per tonne (GBq/t) of alpha activity or 12 GBq/t beta-gamma activity. LLW does not require shielding during handling and transport, and is suitable for disposal in near surface facilities.

LLW is generated from hospitals and industry, as well as the nuclear fuel cycle. It comprises paper, rags, tools, clothing, filters, ***etc.***, which contain small amounts of mostly short-lived radioactivity. To reduce its volume, LLW is often compacted or incinerated before disposal. LLW comprises some 90% of the volume but only 1% of the radioactivity of all radioactive waste.

INTERMEDIATE-LEVEL WASTE

Intermediate-level waste (ILW) is more radioactive than LLW, but the heat it generates (<2 kW/m^3) is not sufficient to be taken into account in the design or selection of storage and disposal facilities. Due to its higher levels of radioactivity, ILW requires some shielding.

ILW typically comprises resins, chemical sludges, and metal fuel cladding, as well as contaminated materials from reactor decommissioning. Smaller items and any non-solids may be solidified in concrete or bitumen for disposal. It makes up some 7% of the volume and has 4% of the radioactivity of all radioactive waste.

HIGH-LEVEL WASTE

High-level waste (HLW) is sufficiently radioactive for its decay heat (>2kW/m^3) to increase its temperature, and the temperature of its surroundings, significantly. As a result, HLW requires cooling and shielding.

HLW arises from the 'burning' of uranium fuel in a nuclear reactor. HLW contains the fission products and transuranic elements generated in the reactor core. HLW accounts for just 3% of the volume, but 95% of the total radioactivity of produced waste. There are two distinct kinds of HLW:

- Used fuel itself.

- Separated waste from reprocessing the used fuel.

HLW has both long-lived and short-lived components, depending on the length of time it will take for the radioactivity of particular radionuclides to decrease to levels that are considered non-hazardous for people and the surrounding environment. If generally short-lived fission products can be separated from long-lived actinides, this distinction becomes important in management and disposal of HLW.

HLW is the focus of significant attention regarding nuclear power, and is managed accordingly.

VERY LOW-LEVEL WASTE

Exempt waste & very low-level waste (VLLW) contains radioactive materials at a level which is not considered harmful to people or the surrounding environment. It consists mainly of demolished material (such as concrete, plaster, bricks, metal, valves, piping, ***etc.***) produced during rehabilitation or dismantling operations on nuclear industrial sites. Other industries, such as food processing, chemical, steel, ***etc.***, also produce VLLW as a result of the concentration of natural radioactivity present in certain minerals used in their manufacturing processes (see also information page on Naturally-Occurring Radioactive Materials). The waste is therefore disposed of with domestic refuse, although countries such as France are currently developing facilities to store VLLW in specifically designed VLLW disposal facilities.

Waste Management

The steps employed in radioactive waste management depend on the nature of the radioactive waste being managed. There are a series of basic steps that commonly take place:

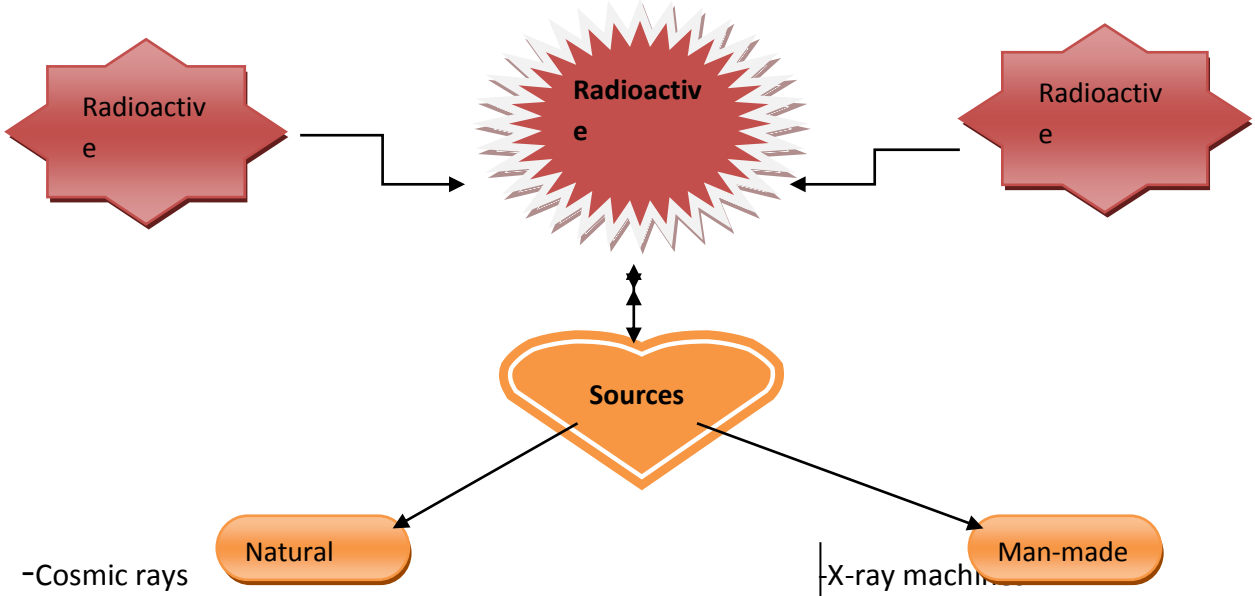
Treatment involves operations intended to change waste streams' characteristics to improve safety or economy. Treatment techniques may involve compaction to reduce volume, filtration or ion exchange to remove radionuclide content, or precipitation to induce changes in composition.

Conditioning is undertaken to change waste into a form that is suitable for safe handling, transportation, storage, and disposal. This step typically involves the immobilisation of waste in containers. Liquid LLW and ILW are typically solidified in cement, whilst HLW is calcined/dried then vitrified in a glass matrix. Immobilised waste will be placed in a container

suitable for its characteristics. (For more information, see information paper on Storage and Disposal of Radioactive Wastes).

Storage of waste may take place at any stage during the management process. Storage involves maintaining the waste in a manner such that it is retrievable, whilst ensuring it is isolated from the external environment. Waste may be stored to make the next stage of management easier (for example, by allowing its natural radioactivity to decay). Storage facilities are commonly onsite at the power plant, but may be also be separate from the facility where it was produced.

Disposal of waste takes place when there is no further foreseeable use for it, and in the case of HLW, when radioactivity has decayed to relatively low levels after about 40-50 years.



-Cosmic rays

-Environment (rocks,water,air)

-Living organisms

-Radio-nucleotides (Rd,U,Th)

-Isotopes (K,C- soil,rock,air,water)

-Marine sediments

-Radiations from atmosphere

-Radioactive gases (Thoron,Radon)

-Radioactive substances in body tissues(U,Th,K,C-isotopes)

-X-ray machines

-Nuclear tests

-Nuclear reactor wastes

- Research labs (industrial and medicinal)

-Miscellaneous (TV, watch dials)

- Thermal plants

Somatic

Direct effects on cell & tissue

-Higher exposures may kill animals quickly like(gonads).

-Low doses destroy immune system egg/embryo.

-Leads to eye-cataracts, leukemia, tumors,

-Cardio-vascular disorders, pre-mature aging and reduce life spain.

Genetic

Brought about by man-made radiations

-Cheifly damages dividingcells

-mutational/ lethal effects on

-Effects rate of mutation.

- Diagnostic X-ray exposures on pregnant women may cause the cancers in children.

botane.patho@gmail.com