GYMNOSPERMS, PLANT ANATOMY, ECOLOGY & BIOTECHNOLOGY

B.Sc. Botany, Second Year, Paper – II

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FOREWORD

Since its establishment in 1976, Acharya Nagarjuna University has been forging ahead in the path of progress and dynamism, offering a variety of courses and research contributions. I am extremely happy that by gaining a B++ (80-85) grade from the NAAC in the year 2003, the University has achieved recognition as one of the front rank universities in the country. At present Acharya Nagarjuna University is offering educational opportunities at the UG, PG levels apart from research degrees to students from about 300 affiliated colleges spread over the three districts of Guntur, Krishna and Prakasam.

The University has also started the Centre for Distance Education with the aim to bring higher education within reach of all. The Centre will be a great help to those who cannot join in colleges, those who cannot afford the exorbitant fees as regular students, and even housewives desirous of pursuing higher studies. With the goal of bringing education to the doorstep of all such people, Acharya Nagarjuna University has started offering B.A. and B.Com courses at the Degree level and M.A., M.Com., M.Sc, M.B.A. and LL.M. courses at the PG level from the academic year 2003-2004 onwards.

To facilitate easier understanding by students studying through the distance mode, these self-instruction materials have been prepared by eminent and experienced teachers. The lessons have been drafted with great care and expertise within the stipulated time by these teachers. Constructive ideas and scholarly suggestions are welcome from students and teachers involved respectively. Such ideas will be incorporated for the greater efficacy of this distance mode of education. For clarification of doubts and feedback, weekly classes and contact classes will be arranged at the UG and PG levels respectively.

It is my aim that students getting higher education through the Centre for Distance Education should improve their qualification, have better employment opportunities and in turn facilitate the country's progress. It is my fond desire that in the years to come, the Centre for Distance Education will grow from strength to strength in the form of new courses and by catering to larger number of people. My congratulations to all the Directors, Academic coordinators, Editors and Lesson - writers of the Centre who have helped in these endeavours.

K. Vijayamma Rao

Prof. V. Balamohandas

Vice - Chancellor,
Acharya Nagarjuna University

B.Sc. BOTANY II YEAR

Paper - II (Theory)

(Gymnosperms, Plant Anatomy, Ecology & Biotechnology)

UNIT - 1

(Gymnosperms)

1. General features of Gymnosperms and their classification.
2. Geological time scale; Fossils and Fossilization.
3. General account of Pteridospermales and Bennetitales.
4. Morphology of vegetative and reproductive parts; Anatomy of root, stem and leaf; Reproduction and life cycle of Pinus and Gneum.

UNIT - 2

(Plant Anatomy)

5. Tissues and Tissue systems.
6. Shoot System: Shoot apical meristem and its histological organisation; various theories of shoot apical meristems; Normal secondary growth; Characterization of growth rings; Sapwood and Heart wood; Periderm; Abnormal secondary growth (Boerhaavia; Dracena).
7. Leaf; Internal structure of dicot, monocot and xerophytic leaves.
8. Root System: Root apical meristem and its histological organisation; Recent theories of root apical meristem; Normal secondary growth; Abnormal secondary growth (Tinospora, Beta vulgaris).

UNIT - 3

(Plant Ecology)

9. Plants and Environment: Atmosphere (gaseous composition), Water (properties of water cycle), Light (global radiation, Photosynthetically active radiation), Temperature, Soil (development, soil profile, physico - chemical properties) and Biota.
10. Population ecology: Growth curves, Ecotypes, Ecads.
11. Community ecology: Community characteristics, Frequency, Density, Cover, Life forms, Biological spectrum and Ecological succession - Hydrosere, Xerosere.
12. Vegetation types of India: Forests and Grasslands.

UNIT - 4

(Biotechnology)

13. Functional definition, r DNA technology, Somatic hybridization, Cybrids, Application of biotechnology in agriculture, medicine and human welfare.
14. Basic concepts of plant tissue culture; Methods of culture of shoot tip, anther, ovary and embryo and their importance.
15. General account of vegetative propagation, grafting and economic aspects.

Part - II BOTANY PRACTICAL SYLLABUS

Practical - II (Gymnosperms, Anatomy, Ecology & Biotechnology)

I. GYMNOSPERMS:

a. Observation, identification, notes and drawings of reproductive parts of Pinus and Gnetum; Pteridospermales and Bennettitales.

b. Preparation and staining of thin section of stem & needle of Pinus.

II. ECOLOGY:

a. Analysis of Soil P^H by strip method.

b. Quadrat method.

c. Frequency of herbs.

d. To determine moisture content and water holding capacity of different soils.

e. To estimate dust holding capacity of leaves of different species.

f. To study the vegetation structure through profile diagram.

g. To measure the dissolved oxygen content in polluted and unpolluted water samples.

h. Observation of Museum specimens and permanent slides of hydrophytes and xerophytes.

III. Anatomy:

a. Study of cytohistological zonation in the L.S. of shoot tip and root tip.

b. Preparation, staining, observation, drawing, identification of:

i. Cucurbita stem (primary)

ii. Bignomia and Boerhaavia stems (Secondary)

iii. Tinospora root

iv. Casuarina phylloclade

v. Nerium leaf

vi. Nymphaea petiole

IV. Biotechnology:

a. Evaluation of toxicity of biopesticiedes / bioinsecticides (Neem, Vitex, Annone, Tagetus, Lantana).

B.Sc. DEGREE EXAMINATION, MAY 2007

(Examination at the end of Second Year)

Part II - Botany

Paper II - BOTANY

(GYMNOSPERMS, PLANT ANATOMY ECOLOGY AND BIO-TECHNOLOGY)

Time : Three hours Maximum : 100 marks

SECTION A (5 x 15 = 75 marks)

Answer All questions

1. (a) Give a brief account on the general features and classification of gymnosperms.

విృత బీజాల యొక్క సాధారణ లక్షణములను తెలిపి వర్గీకరణ గురించి వివరింపుము.

Or

(b) Explain the anatomy of root.

వేరు అంతర్నిర్మాణము గురించి వివరింపుము.

2. (a) Describe the tissues and tissue systems in Plants.

మొక్కల కణజాలాలు మరియు కణజాల వ్యవస్థల గురించి వివరింపుము.

Or

(b) Explain abnormal secondary growth in roots of Tinospora and Beta Vulgaris.

టీనోస్పోరా మరియు బీటా వల్గారీస్‌ల యొక్క వేరు అసంగత ద్వితీయ వృద్ధిని గూర్చి వ్రాయుము.

3. (a) Write an essay on vegetation types of India.

భారతదేశంలో గల వివిధ రకాల శాఖీయ వర్ధనాలను గురించి వ్రాయుము.

Or

(b) What is Soil Profile? Explain its physico-chemical properties and biota.

మృత్తికావ్యవస్థ గురించి వ్రాయుము. దాని యొక్క భౌతిక రసాయనిక ధర్మాలను గురించి వ్రాయుము.

4. a. Discuss the applications of biotechnology in Agriculture.

వ్యవసాయంలో జీవసాంకేతిక శాస్త్రము యొక్క అనువర్తనాలను వివరింపుము.

Or

(b) Explain embryo culture and its importance

పిండ వర్ధనం మరియు దాని యొక్క ప్రాముఖ్యతను గురించి వివరింపుము.

5 (a) Write an essay on Cybrids.

సైబ్రిడ్‌ల గురించి ఒక వ్యాసం వ్రాయండి.

Or

(b) Explain vegetative propagation and its importance.

శాఖీయ వర్ధనమును గూర్చి వ్రాసి దాని యొక్క ప్రాముఖ్యతను వివరింపుము.

SECTION B - (5 X 5 = 25 marks)

Answer any FIVE of the following

Each question carries 5 marks.

6. Fossils

   శిలాజాలు

7. Sapwood

   రసదారువు

8. Theories of shoot apical meristem.

   వేరు యొక్క అగ్ర విభాజ్య కణజాల సిద్ధాంతాలు.

9. Green house effect

   హరిత గృహ ప్రభావము

10. Ecotypes

    ఆవరణ వ్యవస్థలు

11. Hydrosphere.

    జలావరణ వ్యవస్థ

12. Ozone

    ఓజోన్

13. Cloning vector

    క్లోనింగ్ వాహకాలు

14. Electrofution.

    విద్యుత్ ప్రేరిత సంయోగము

15. Somatic hybrids.

    శాఖీయ సంకరాలు

CONTENTS

Lesson - 1

GYMNOSPERMS GENERAL FEATURES AND CLASSIFICATION

CONTENTS

1.1 OBJECTIVES OF THE LESSON

After reading this part you will know

- Vegetative and reproductive features of Gymnosperms, Similarities or differences with angiosperms and the classifications of Gymnosperms proposed by Chamberlain (1917), C.A Arnold (1948), D.D pant (1957) and K.R Sporne (1965) separately.

1.2 INTRODUCTION

The seed bearing plants are called "Spermatophyta". They are divided into "Gymnospermae" (Gymnosperms) and "Angiospermae" (Angiosperms). The seeds are exposed and present on megasporophylls in Gymnosperms where as they are embedded in the ovary i.e fruit in Angiosperms. The word "Gymnosperms" (Gymnos = naked; sperm= seed) was coined by Theophrastus (300BC).

The Gymnosperms were dominant vegetation, throughout the late Palaeozoic and Mesozoic eras and steadily declined thereafter. Their fossil record also reveals that they were dominant during the past geological ages. Certain primitive Gymnosperms like Cycadofilicales, Cycadeoidales and Corditales are now extinct. Cycadofilicales (Pteridosperms) are focused as an evidence for the origin of Gymnosperms from an ancient stock of fern-like individuals. They appeared during Carboniferous period.

Cycadeoidales which flourished during the early Mesozoic era show striking similarities with modern cycads. The Coniferales constitute the most conspicuous order of living Gymnosperms and include the familiar Conifers that became very important in world's economy. Cycadales, Ginkgoales, Coniferales, Taxales and Gnetales are the five orders where modern Gymnosperms.

The living conifers include 52 genera and are widely distributed throughout northern and southern hemispheres of the globe. *Sequoia gigantea* that lives for 4000 years and attains a height of about 100 meters. Gnetales are represented by three living genera *Gnetum, Ephedra and Welwitschia.* Living Gymnosperms are spread over 70 genera and 725 species (Bold, 1963). About 16 genera and 53 species (Raizada and Sahni, 1960) are reported in India.

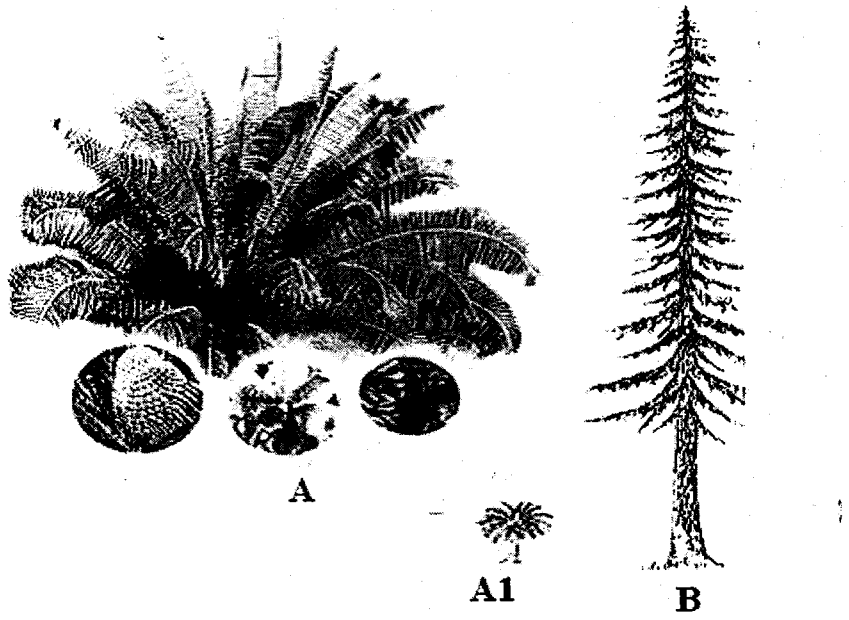


Plate 1.1 Change in habit & Size in Cycadophyta & Coniferophyta A& A1- Cycas, B-Pinus.

1.3 GENERAL FEATURES

These can be divided into two types. **1) Vegetative features.**
2) Reproductive features.

1.3.1 VEGETATIVE FEATURES

These are mainly perennial trees or shrubs. These evergreen plants grow in desert environment or less water available environs. Generally conifers are conical or pyramid

shaped plants The tallest plants in the world belong to this group. *Sequoia semipervirens* which grows in California mountains of North America attains a height of 140 meters. *Taxodium mexicanum*_ (Mexican Cyprus) had a girth of 17 meters. Plants which belong to 'Cycadales' show palm habit. These are short and slow growing.

At the tip of the unbranched stem, a crown of big pinnate compound leaves are present. (Plate:1.1A1). Rarely stems are branched. Plants which belong to 'Conifers' are conical or pyramid shaped trees (Plate:1.1B) Racemose branching is present. Plants which belong to the 'Gnetales' resembles dicotyledonous plants. Generally these are small trees or shrubs (*Ephedra sps.*). Mostly woody climbers. Root system is well developed from primary root. In *Pinus* ectomicorhiza is present. In *Cycas* coralloid roots, an "Algal zone" is present in the cortex, internally.

Stems of Gymnosperms are woody, straight and posses well developed xylem Branches are of two types.

1. **Branches of unlimited growth :-** They are also known as "long shoots".
2. **Branches of limited growth :-** They are also known as "short shoots".

Generally the plants are evergreen. But *Taxodium* and *Larix*_ are deciduous. Leaves are of two types **1. Scale Leaves 2.Green Leaves** (Plate1.2)

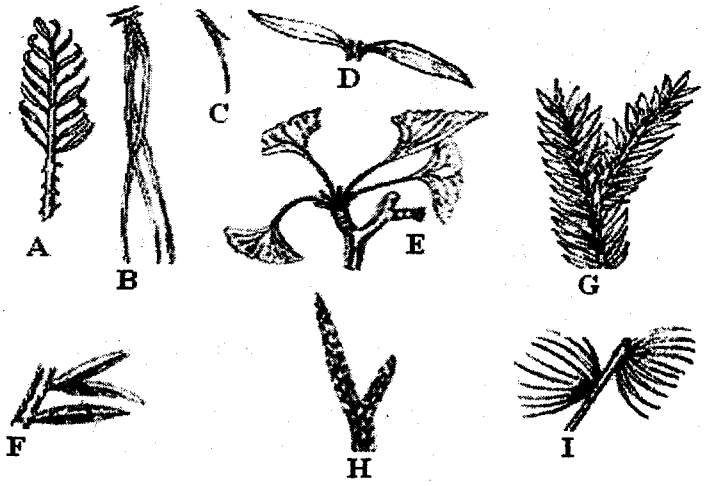


Plate 1.2 Vegetative leaves in Gymnosperms

A. Green leaf in *Cycas;*
B. Needle like leaves in *Pinus;*
C. *Picea* leaf.
D. *Arucaria* leaves;
E. *Ginkgo* leaves on short shoot;
F. *Taxus baccata* leaves;
G. *Sequioa semipervirens* leaves;
H. *Curpressus semipervirens* leaves;
I. *Cedrus deodara* needle like leaves.

The green leaves are of two types **a) Simple Green Leaves b)Compound Green Leaves** Simple green Leaves are linear, needle like. Compound green leaves resemble fern leaves in shape and phyllotaxy (Cycads, Cycadeoids).

There is variation in leaf arrangement. They exhibit Spiral arrangement (*Podocarpus, Taxus.)*, Opposite Decussate (*Ephedra, Gnetum*) and Whorled Arrangement (Cupressaceae), Venation in leaves is of three types, namely Reticulate venation (*Gnetum*), Parallel venation (*Welwitschia*) and Dichotomous venation (*Ginkgo).*

Young leaves show circinate vernation i.e., young leaves are twisted like a watch spring. This is very common in fern leaves and is a primitive character present in Gymnosperms.

Internal Structure of Vegetative Parts

Roots are well developed from primary root. Protoxylem is exarch in position. Based on their number roots are 'Diarch, 'Triach or Tetrach.

The stems of Gymnosperms resemble Dicot plants in their internal structure. A centralised parenchymatous pith/medulla, is surrounded by a few **conjoint, collateral and open** vascular bundles. This is known as "Eustele". A strip of cambium divides xylem and phloem in a vascular bundle, hence they are open. The xylem in stems shows endarch arrangement. Medullary rays are present. Secondary growth is common in stems of Gymnosperms. During secondary growth secondary xylem and secondary phloem are formed, abundantly. Annual rings are seen easily and clearly in secondary growth occured stems. Secondary xylem in Gymnosperms is of two types.

1. **Manoxylic - Cycadopsida**
2. **Picnoxylic - Coniferopsida**
   Gnetopsida.

In manoxylic variety, the tracheids are loosely arranged, their cell walls are thin, large sized and many medullary rays made up of parenchyma are present. So the wood is not hard and economical value is also less. In picnoxylic xylem, tracheids are compactly arranged, cell walls thick, small sized and a few thin medullary rays are present. This wood is hard and economically important. The xylem of Gymnosperms consists of tracheids and xylem parenchyma. Only, Gnetales possess 'Vessels', in other Gymnosperms they are absent. Bordered pits with torus are common in Gymnosperm tracheids. The borered pits on the radial walls of Gymnosperm xylem is of two types

1. **Araucarian Bordered Pits.**
2. **Abietinian Bordered Pits.**

In the araucarian variety, they are arranged in two or more rows, angularly, one after another in an orderly fashion. They are also alternate and circular in outline. In abietinian variety they are arranged in one or two rows only and are separately arranged and circular. If present in two rows, they are oppositely arranged. Resin ducts are common in the secondary xylem of many Conifer's (eg: *Pinus, Picea*). One variety of resin ducts are longitudinally arranged in xylem and are scattered. Second variety arranged transversely, spindle shaped and embedded in xylem.

In secondary phloem sieve tubes, phloem parenchyma and phloem fibres are present. Companion cells are absent in Gymnosperm phloem. But in Gnetales albuminous cells are

present instead of companion cells. Pallisade tissue is present in mesophyl. But in some it is absent. Pegged parenchyma is present in *Pinus.* Vascular bundles in the leaves of cycads are diploxylic. Both centripetal and centrifugal xylem is present in this. In others conjoint and collateral vascular bundles are present. In many leaves transfusion tissue is present. Resin ducts are present in conifers (*Pinus, Cedrus*). Leaves are hypostomatic i.e., stomata are present only in the lower epidermis. Basing on their formation they are of two types.

1. Haplocheilic Variety 2. Syndetocheilic Variety

Haplocheilic stomata are present in pteridospermales, Cycadales, Corditales, Ginkgoales and Coniferals. In this guard cells and subsidary calls have different origin. Syndetochielic stomata are seen in cycadeoidea and in some *Gnetum* species. In this both guard cells and subsidiary cells arise from the same mother cell.

1.3.2 REPRODUCTIVE FEATURES

Gymnosperms are generally monoecious. Some are rarely diocious (*Pinus*). Microspores are called pollengrains. They are produced in the microsporangia which are formed on the microsporophyll. In the female cone, megasporophylls are present. Megaspores are formed in the megasporangia (ovules), which are formed on the megasporophylls. In some fossil orders like Pteridospermales and in some genera like *Cycas, Ginkgo* and *Taxus,* there is no female cone formation. Prof B. Sahni (1920) divided Gymnosperms in to two large groups and used the terms mentioned below.

1. **Phyllospermae:-** The seeds are being inserted on the modified leaves.
2. **Stachyospermae:-** These are more or less microphyllous plants with seeds inserted on them (Plate 1.3).

MICROSPOROPHYLLS (STAMENS):

These are flat, broad and leaf like (*Cycas*) or umberella like (*Taxus*) with microsporangia at the bottom, in clusters. The formation of microsporangia is of "Eusporangiate" type i.e formation of microsporangia (Anther sacs) from a group of hypodermal cells. In the microsporangia many "Microspore Mother Cells" (MMC) are formed. They undergo, a reduction division and each produce four haploid microspores (Pollen grains).

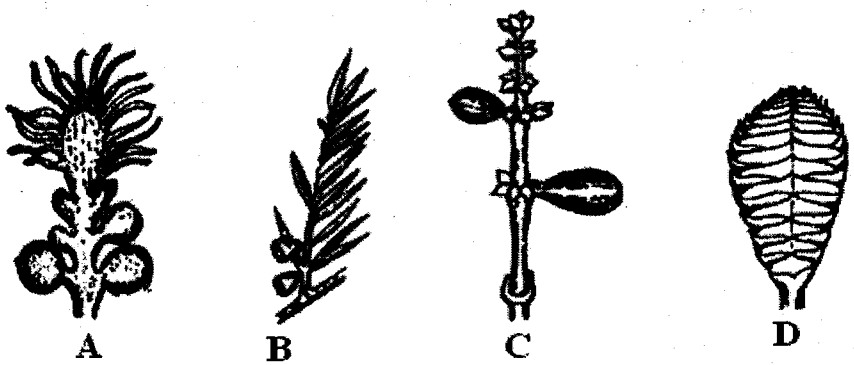
A B

***Plate 1.3 A. Pinus* Cones *B. Pinus* Tree**

There is a furrow like germpore in the pollen grain. Some pollen grains are winged. The number of wings many be 1-3. But in many, there are no wings to pollengrains. The wings are present in Conifers and some fossil Gymnosperms. The pollen grains are produced in more numbers (Pinaceae).

MEGASPOROPHYLLS (Plate 1.4):

In the female cones, megasporophylls are present. Megaspores are produced in the megasporangia (ovules) which are present on the megasporophylls. But in *Cycas, Ginkgo* and *Taxus* there are no female cones. Pteridospermales (Fossil Gymnosperms) do not posses male & female cones. The "Megasporangia" (ovules) in Gymnosperms are not covered by carpels (Unlike Angiosperms). They are naked. Ovules are orthotropous and possess three layered integument.



A B C D

Plate 1.4 Female cones of Gymnosperms

A. *Cycas revoluta* megasporphyll with ovules; B. *Taxus baccata* female Cones C. *Gnetum ula* female Cones; D. *Welwitschia* female Cone.

In the integument outer and inner layers are soft and are called sarcotesta. Middle layer is hard and sclerenchymatic. It is called sclerotesta. Except at the micropyle, the nucellar tissue is covered by integument. But in some Gymnosperms, the integument is having an attachment at the base of nucellar tissue, remaining part is free from integument. The vascular supply is present in the integument. In some vascular supply is absent. The nucellar tissue below the micropyle disintegrates and forms pollen chamber. Pollen grains reach this chamber after pollination.

During the pollination period, the nucellar tissue in the ovule produces sweet, mucilagenous fluid. This liquid forms a 'Pollination drop' at the tip of the micropyle of the ovule, because of its exudation. The air helps in the dispersal of pollen grains. Such pollen grains, which are travelled in the air caught in the pollination drop of the ovule. After it's drying, the pollen grains reache the pollen chamber through the micropylar canal.

MALE AND FEMALE GAMETOPHYTES DEVELOPMENT

Pollengrains mature before their liberation. Hence they are considered as "Partially grown Male Gametophytes". The nucleus in the pollengrain divides and produces one (Cycadales), two (*Pinus, Ginkgo*), or more prothallial cells and one antheredial cell. But in Taxaceae, Cupressaceae and Taxodiaceae, there are no prothallial cells. The antheredial cell undergoes a mitotic division and produces a small generative cell and a big tube cell. Generative cell undergoes another division and produces a small stalk cell or sterile cell and a big spermatogenous cell. This spermatogenous cell produces two non-motile male gametes due to normal cell division. But in Cycadales and Ginkgoales multiciliate motile male gametes are formed.

The partially developed pollen grain after reaching the pollen chamber only shows the above developments. After these developments in the pollen grain, pollen tube emerges out through germinal pore. Pollen tubes act as haustoria in Cycadales but in other they are only useful in transporting the non-motile male gametes, towards female gametes.

The megaspore mother cell present at the micropylar region undergoes reduction division and produce megaspore tetrads. They are arranged in one column. Out of the four, three which are at the top degenerate and the lower one develops. This fertile megaspore, after several divisions produces female gametophyte. In the megaspore development, several free nuclear divisions will occur and later membranes are formed and cellularization starts. This cellular female gametophyte acts as "endosperm" in Gymnosperms.

The formation of endosperm in Gymnosperms occurs before fertilization and it is in haploid condition. On contrary in angiosperms the endosperm formation occurs only after fertilization and it is triploid

Near the apex of female gametophyte some of the hypodermal cells form archegonia. The archegonia in Gymnosperms are primitive. Each archegonium is flask shaped with a short neck and a ventre. In the ventre there is an egg cell and a ventral canal cell are present. In *Gretum* and *Welwitschia* there is no formation of archegonia. Fertilization in gymnosperms is through siphonogamy. Male gametes reach the archegonia through pollen tubes.

EMBRYOGENY

In embryogeny, the zygote undergoes many free nuclear divisions. Free nuclear divisions in zygote is a Gymnosperm character. The number of free nuclei are specific to every genus. These may be 30-1000. After this, wall formation occurs and pro-embryo is formed. At the tip of the micropylar end of ovule in the embryo a suspensor is differentiated and it pushes the embryo in to the endosperm tissue as and when it is required. Because of this, embryo draws its nourishment from the endosperm. Embryo in Gymnosperms is embedded in the endosperm and is orthotropous. Primary plumule in Gymnosperm embryo is developed towards the chalazal end of the ovule. This type of embryo is called 'Endosporic embryo' (Plate 1:5). There are two (*Cycas*) or many (*Pinus*) cotyledons in embryo.

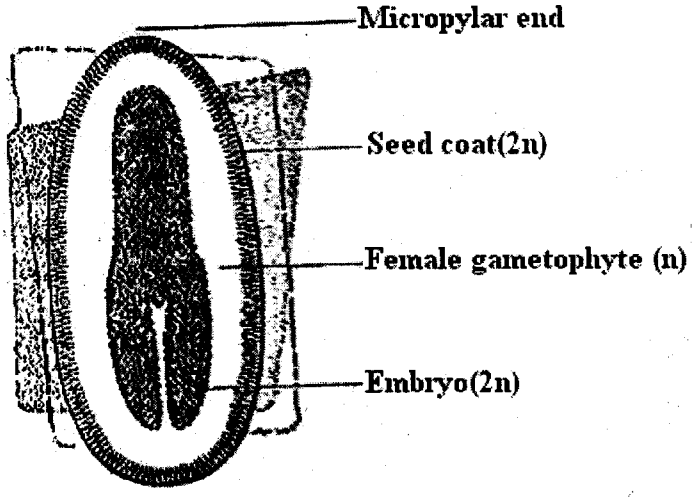
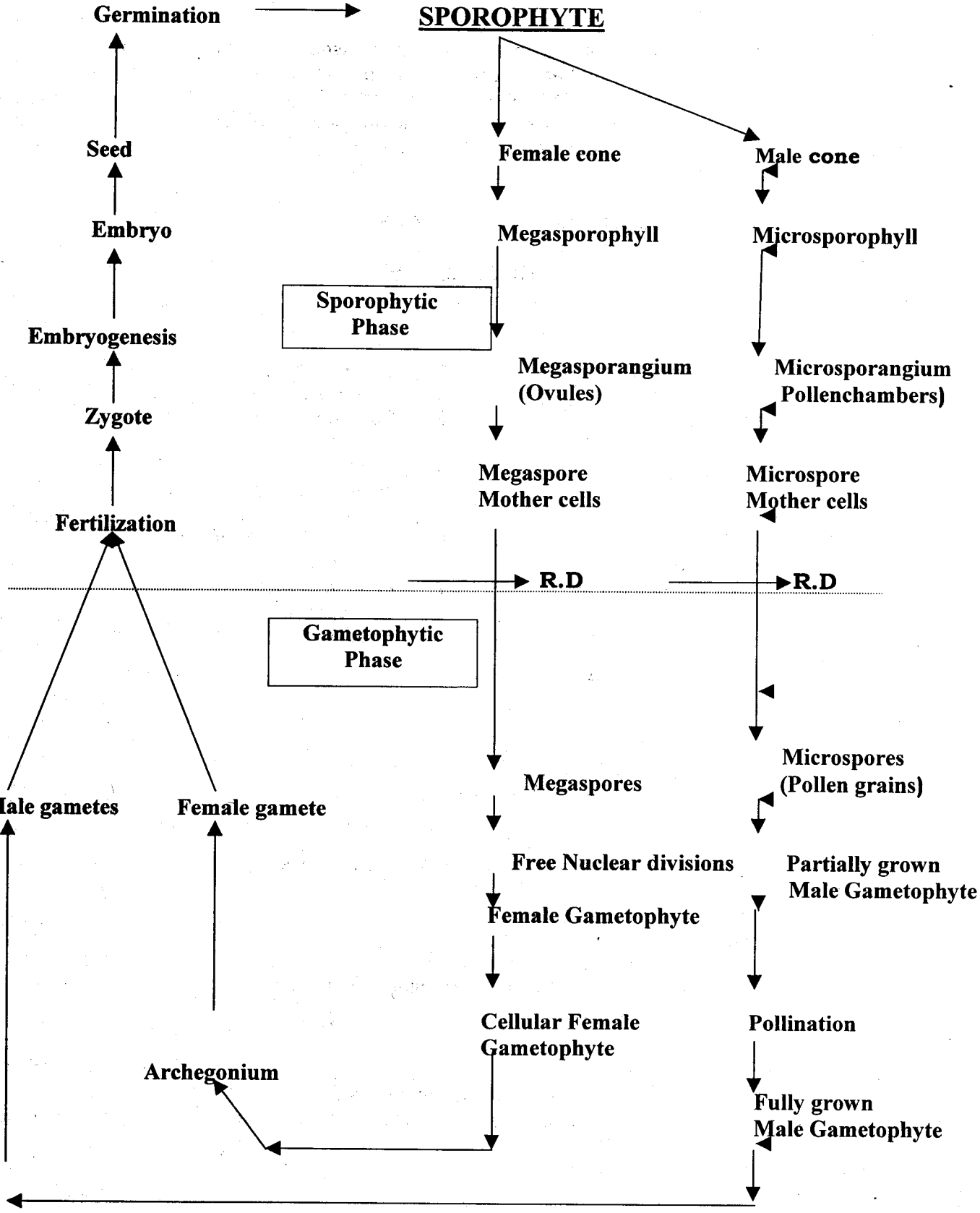


Fig : 1.5: Sturcture of a Gymnospermous type of seed in longisectional view.

Presence of 'polyembryony' is a common character among Gymnosperms. It may be simple polyembryony or cleavage polyembryony. But in one ovule only one embryo develops, all the other embryos will disintegrate due to lack of food and space. A fully grown embryo shows a primary radicle, primary plumule, hypocotyl and two or many cotyledons.

Due to several post fertilization changes ovule develops in to seed. Integument develops in to seed coat with three layers. The outer sarcotesta becomes juicy, middle sclerotesta is hard and shell like and the inner sarcotesta remain as a thin layer. After some period of rest the seed germinates and produces a seedling. The germination is epigeal.

THE LIFE –CYCLE OF THE GYMNOSPERM PLANT IS GIVEN BELOW



1.4. DISTRIBUTION

The number of living Gymnosperms in India decreases as we proceed from eastern to western Himalayas. The total number of living Gymnosperms in the world is approximately 70 genera and 725 species. Raizada and sahni(1960) reported 16 genera and 53 species of living Gymnosperms from India. The Gymnosperms are mainly dwellers of temperate regions. They are poorly distributed in the plains of India. Only Himalayas region is the best suited area for the growth of Gymnosperms in India.

Comparison of Angiosperms and Gymnosperms :-

DIFFERENCES

S.No.	Angiosperms	S.No.	Gymnosperms
1.	Annuals, Biennials or Perennials.	1.	Generally Perennials.
2.	Xylem possess vessels	2.	Xylem is devoid of vessels.
3.	Phloem consists of companion cells.	3.	Companion cells are absent.
4.	Flowers are formed.	4.	' Strobili ' or 'Cones' are formed as reproductive structures.
5.	Flowers are bisexual.	5.	Cones are unisexual.
6.	'Double fertilisation and 'Triple fusion' occur during fertilization	6.	Double fertilisation and 'Triple fusion' Do not occur in Gymnosperms..
7.	Endosperm is the product of	7.	Female gametophyte acts as
8.	fertilization. Endosperm is generally triploid in nature.	8.	endosperm. Endosperm is monoploid In nature.

Similarities

1. Roots are exarch and diarch to polyarch.
2. Secondary growth occurs in both the groups.
3. Plants are heterosporous.
4. Stems are eustelic.
5. Secondary growth in both results in manoxylic or pycnoxylic wood.
6. Both are endosporous plants.
7. Gametophytes are reduced in both.
8. Megasporangium is never shed in both.
9. Megasporangium forms the seed in both the groups.
10. In both the groups, nucellus is covered by integument and forms the structure called 'ovule'.

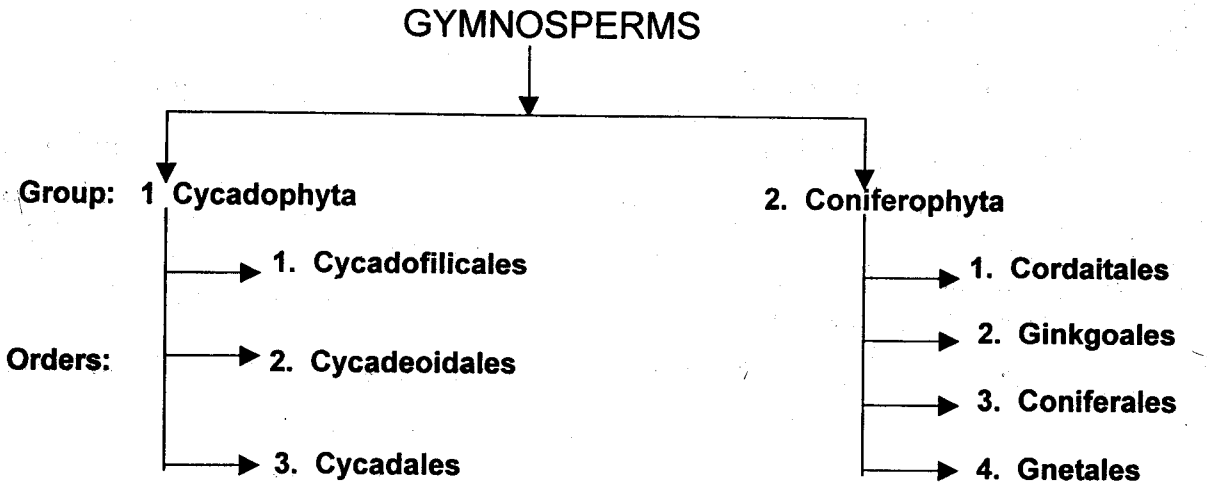
1.5 CLASSIFICATION

The relative ranks assigned to the different orders and their subdivision is , however, a debatable subject. Robert Brown (1827) recognized Gymnosperms as a distinct group. Eichler

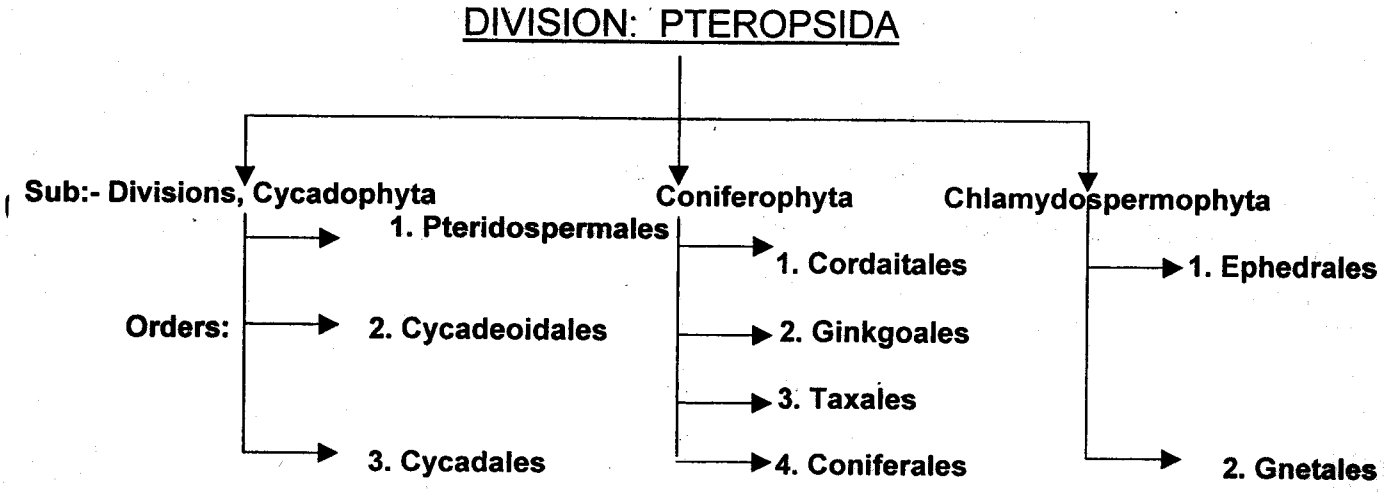
(1889) treated Gymnosperms as a separate order. Coulter and Chamberlain (1917) in their " Morphology of Gymnosperms" divided Gymnosperms in to the following orders.

1. Cycadofilicales 2. Bennettitales 3. Cycadales 4. Cordaitales, 5. Ginkgoales 6. Gnetales.

Chamberlain (1934) divided Gymnosperms in to two major groups basing on habit, shape of leaf lamina, structure of xylem and structure of ovule.



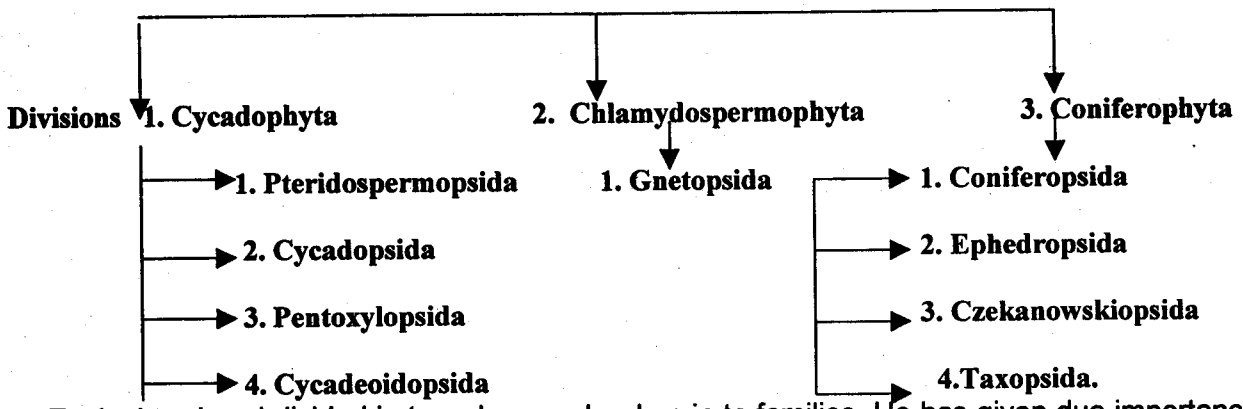
C.A Arnald (1948) included all Gymnosperms in one group i.e. "Pteropsida". He has not used the term " Gymnospermae". Pteropsida is divided in to 3 divisions.



In this classification Arnald has not followed the international code of botanical nomenclature. Since the Gymnosperms are not monophyletic in origin he has not used the term " Gymnospermae". According to him, Gymnosperms are polyphyletic in origin. This view was later supported by many.

D.D. pant (1957) divided the group Gymnosperms in to 3 divisions. Divisions are further subdivided in to classes.

Gymnosperms



Each class is subdivided in to orders, and orders in to families. He has given due importance to fossil Gymnosperms along with living Gymnosperms.

Sporne (1965) divided Gymnosperms in to three classes.

Class 1 ⟶ Cycadopsida (In this 4 orders and 13 families are present)

Order ---1:- Pteridospermales

Family:- Lyginopteridaceae

Ex:- *Lyginopteris.*

Order ---2:- Bennittitales

Family:- Williamsoniaceae

Ex:- *Williamsonia*

Order ---3:- Penetaxylales

Order ---4 :- Cycadales.

Class 2 ⟶ Coniferopsida (In this 4 orders and 15 families are present)

Order ---1:- Chorditales

Order ---2:- Coniferales

Family:- Pinaceae

Ex:- *Pinus.*

Order ---3:- Taxales

Order ---4 :- Ginkgoales.

Class 3 ⟶ Gnetopsida (In this 1 order 3 families are present)

Order ---1:- Gnetales

Family:- Gnetaceae

Ex:- *Gnetum.*

Cycadopsida Characters:

1. Manoxylic wood in stems. 2. Long, fern -like compound leaves .
3. Actinomorphic ovules.

Coniferopsida Characters:

1. Fan-like or needle-like simple green leaves. 2. Zygomorphic ovules.

Gnetopsida Characters:

1. Picnoxylic wood. 2. Presence of vessels. 3. Flowers with perianth.
4. Regular flowers or cones.

1.6. SUMMARY

The seeds are exposed and present on megsporophylls in Gymnosperms. These are evergreen, perennial trees. Abundant secondary growth occurs in there plants. Tracheids are the main conducting tissue in these plants. Companion cells are absent in phloem tissue. Cones or flowers are unisexual. Megasporophylls or microsporophylls are compactly arranged in to cones or they may be present freely. Only one germ pore, on the pollen and that it is like a strait furrow. Male gametes are generally non -motile but in some they are multiciliate and motile. Pollination agent is air. Fertilization in Gymnosperms is through siphonogamy. The ovule has only one integument and it has three layers. The endosporic embryo is embedded in the endosperm of the seed. Endosperm is formed before fertilization.

1.7 Technical Terms

Gymnosperms : The seeds are naked and present on megasporophylls

Circinateernation : Young leaves are twisted like a watch spring.

Eustele : Many conjoint, collateral, and open vascular bundles are arranged like a ring around the parenchymatic pith.

Monoecious : Presence of only male or female reproductive organs on the plant.

Simple Polyembryony :More than one archegonium is present in one seed, and the female gametes in them are fertilized and produce more embryos.

Cleavage Polyembryony: From the same zygote many embryonic cells are formed and each develops into an embryo.

Endosporic embryogeny: The shoot end of the embryo is directed away from the micropylar side.

1.8 MODEL QUESTIONS

I. Answer each question in 30 lines.

1. Write the general features of Gymnosperms ?
2. Write the classification of Gymnosperms ?
3. With a line sketch explain the Life – History of a Gymnosperm ?

II. Answer each question in 10 lines.

1. Angiospermic Characters present in Gymnosperms
2. Write the classification of Sporne ?
3. Write the anatomical features of Gymnosperms ?

1.9 SUGGESTED READINGS.

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Lesson - 2

GEOLOGICAL TIME SCALE; FOSSILS AND FOSSILIZATION

2.1 OBJECTIVES

After reading this part.

You will know the geological eras, periods and their age in geological time scale. The relative distribution of Gymnosperms and other plants in these geological eras & periods. The difference between "Organ genus" and "Form genus" and their definitions. The differences between different types of fossils i.e., impressions, casts and petrifactions and their identification.

STRUCTURE

2.1 INTRODUCTION

The total life span of the earth from the time of its origin is called geological time. The discovery of radioactivity served as a tool in estimation of geological time. The origin of life before prehistoric periods was estimated to be 2000 million years. The entire geological age was divided in to 3 eras. **1. Palaeozoic era 2. Mesozoic era 3. Cenozoic or Cainozoic**

era. Cainozoic era is the recent period, in which the highly evolved land plants like Angiosperms are dominant. The Gymnosperms are an ancient group of plants dating back to the Devonian period of palaeozoic era (Fig 2.1 and Table 2.1). They were predominant over the earth's surface during Jurrasic and Cretaceous periods of Mesozoic era (Table 2.1). However by the end of the era, they were gradually replaced by Angiosperms (Magnoliophyta). Several of the primitive Gymnosperms (Cycadofilicales, Bennettitales and Cordaitales) became extinct now. They are known only as fossils. A large number of Cordaitales and seed ferns existed during Carboniferous period. The Ginkgophytes made their appearance in Permian period and were widespread by the Triassic period. In the Tertiary, the conifers declined in diversity, giving room to primitive angiosperms. But during this time Gnetales flourished. The most convincing evidence of plant evolution comes from the fossil plant record which is helpful in the study of past plant life. The ways and means by which fossils are formed and preserved are discussed here.

2.2 GEOLOGICAL TIME SCALE (TABLE 2.1)

The entire geological age was divided in to basically 3 eras.

1. Palaeozoic era, 2. Mesozoic era, 3. Cainozoic era or Caenozoic era

The eras are further divided in to periods and periods in to epochs. The eras, periods and epochs (of the geological time) are arranged in an orderly manner. This arrangement is called '*Geological time scale'.*

2.2.1 PALAEOZOIC ERA

The first and most primitive period of life is named as Palaeozoic era. It consisits of 7 periods. They are Cambrian, Ordovician, Silurian, Devonian, Carboniferrous, Permian and lower Triassic periods (Table2.1). Before Cambrian, there is a period called Pre –cambrian. In this Pre-cambrian period fungi and bacteria are reported to have occured 2000 million years ago. In the Cambrican period of Palaeozoic era, very primitive groups of plants like marine algae, representing thallophyta were supposed to be exiting and there is some evidence of land plants too. In Ordovician and lower Silurian periods of Palaeozoic era some advanced marine algae were supposed to have existed in water. Next in the upper Silurian and lower Devonian periods of Paleozoic era herbaceous marsh plants like *Psilophytum* and *Zosterophyllum* and some small shrubs made their appearance. In the upper Devonian and lower Carboniferous periods the early Gymnosperms appeared. Conifers and Bennettitales are the Gymnosperms present in the upper Permian and lower Triassic periods or at the late Palaeozoic era of geological age.

Primitive groups of fossil Gymnosperms like Pteridospermales along with early gymnosperms like Cycadales also appeared. Among Cycadales, *Cycas* is the only genus still persistant without any change. *Cycas* hence has been called

" Living fossil".

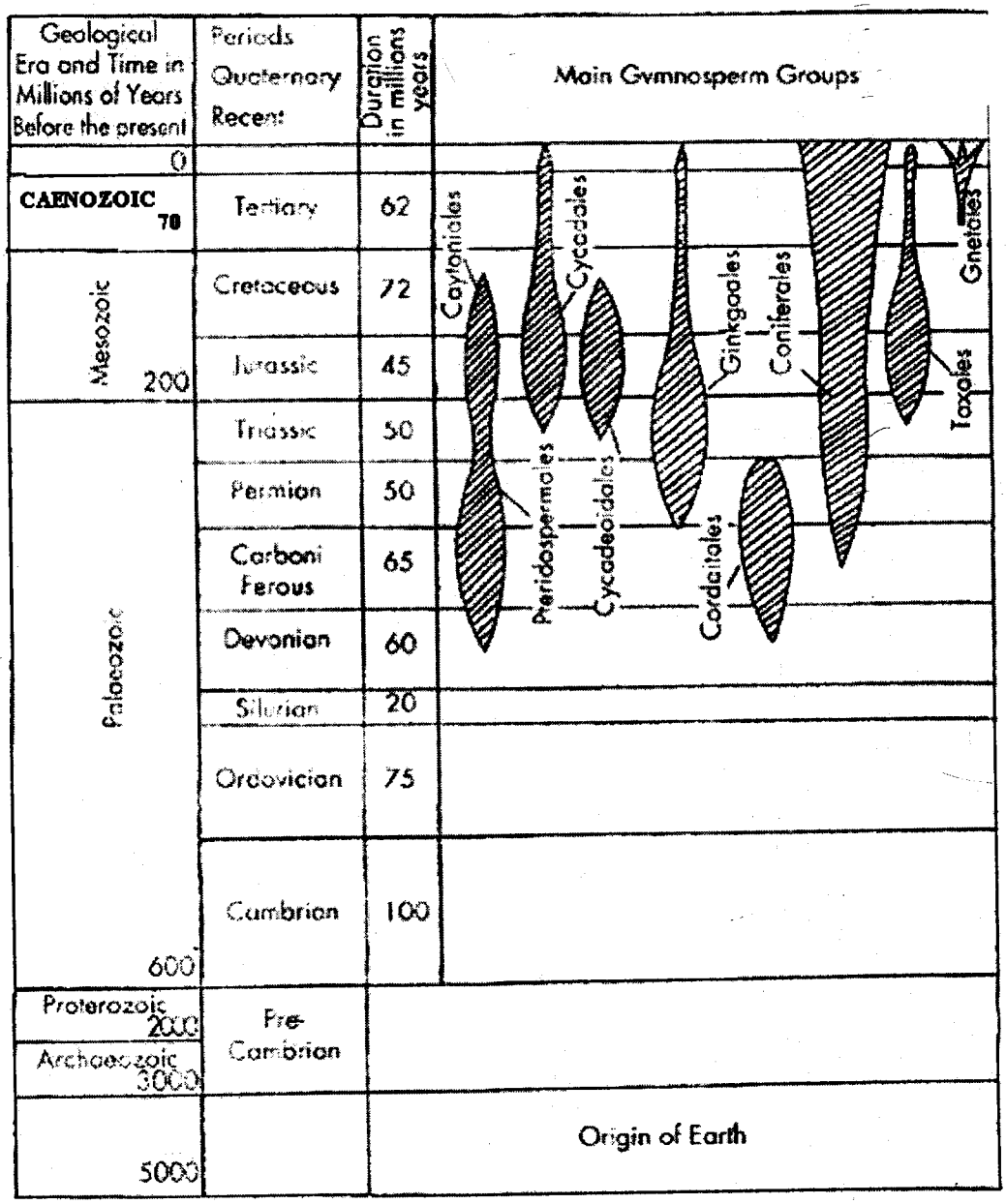
Table:2.1 **GEOLOGICAL PERIODS**

ERA	PERIOD	AGE (MILLION YEARS)	TYPE OF VEGETATION
CENOZOIC	Quaternary	1	Modern
	Upper tertiary, Pliocene. Miocene.	10 20	Modern
MESOZOIC	Lower Tertiary, Oligocene. Eocene. Upper Cretaceous.	35 50 75	Modern, With tropical Plants in Europe
	Lower Cretaceous Upper Jurassic	100 300	Gymnosperms dominant (Conifers and Bennettiales)
	Lower Jurassic (Liassic) Upper Triassic (Rhaetic)	140 160	Luxuriant forests of Gymnosperms & Ferns.
PALAEOZOIC	Lower Triassic (Bunter) Upper Permian	180 190	Sparse desert flora with Gymnosperms (Conifers & Bennettitales)
	Lower Permian upper carboniferous (Coal Measures)	 200	Tall swamp forest with early Gymnosperms, Tree Lycopods, Calamities and ferns.
	Lower Carboniferous Upper Devonian	250 260	Early Gymnosperms, Large Tree Lycopods and Ferns.
	Middle Devonian	275	*Rhynia* Vegetation in marshy localities
	Lower Devonian Upper Silurian	 300	Herbaceous marsh plants (*Psilophyton & Zosterophyllum)* and some Small shrubs.
	Silurian	350	Marine algae
	Ordovician	425	Marine algae
	Cambrian	500	Marine algae, but some Evidence of land plants too.
PRECAMBRIAN		4500 ?	Fungi and bacteria reported to have occured 2000 million years ago.

Table 2.2. Geological Table for Gymnosperms (Cycadophyta)

2.3.2 MESOZOIC ERA (TABLE 2.2)

- This second era present in between Palaeozoic era and Cainozoic era with Triassic, Jurassic, eretaceous and lower tertiary periods.



In the lower Jurassic and upper Triassic periods luxuriant forests of gymnosperms and ferns were present. In the upper Jurassic and lower Cretaceous periods Gymnosperms were dominant. Among them Conifers and Bennittitales are present. In the Tertiary (Oligocene and Eocene) period along with Gymnosperms modern tropical plants also made their appearance in Europe. As a whole Mesozoic era can be called the *'Age of Gymnosperms'*.

2.3.3 CAINOZOIC ERA (CAENOZOIC ERA)

This era consists upper Tertiary and Quaternary periods. In this era the Conifers declined in diversity giving room to Angiosperms along with Gymnosperms. The vegetation in this era is called modern vegetation.

2.4 FOSSILS AND FOSSILIZATION

2.4.1 FOSSILS

The word fossil came form the latin verb " Fodere" which means to dig. Fossils are the remains of plants and animals that have been preserved in rock or preserved traces left by any organism, while it was alive. In short it can be defined as a record of former life. The study of fossils is useful academically as well as economically. The most convincing evidence of plant evolution comes from the fossil plant record which is helpful in the study of plant life. Palaeobotany is the study of fossil plants i.e. the plants now entirely extinct and existed in the past. Some fossils are associated with petroleum, coal and other ecomically important products because they are confined to definite strata of earth's crust. Certain fossils are present just above the coal mines. Fossils also help us in determining the climate of ancient times in different regions.

The fossils are studied as fragments or bits. The well preserved fossils are collected as bits. They are classified under "*Form Genera*". The naming of such genera is by suffixing the plant part from which it came. For example Phyllum means leaf, *stigmaria* means rhizome, deydron is the tree trunk, xylum is the woody part, *strobilus* means cone. The reconstruction of the whole plant becomes possible only if all the bits of such fossils are available. Some times only some parts are available and others are not. The naming of plant fossils occurs in a specialized manner. Depending upon the fossil available it may be recognized as "*Organ genus*" or" *Form Genus*". If the botanical affinities of the available fossil is known, then it is called Organ genus. If the botanical affinities are not known, basing on the shape, it is considered as form genus. Continuous search and research of roots, stems, leaves and reproductive organs of past plant life, provided morphological and anatomical characters of fossil Gymnosperms. Their study and analysis enriched the fossil Gymnosperm science undoubtedly.

2.4.2. FOSSILIZATION

Rivers flowing down the mountains bring pieces of rocks and large quantity of sand. This sand settles down at the bottom of the water. Along with this sand, plant parts and animals which get in to that, would have a chance of being preserved. This sand gets compressed and sedimentary rocks are formed at the bottom of water. Fossil plants are found in sedimentary rocks that originated from fresh water or brackish water or occasionally of marine bed. Coal balls retain fragments of roots, stem, petioles and seeds. Volcanic ashes also retains plant fossils. During fossilization plant parts get deposited on the site under which they grow. Some times they are carried by rivers and deposition takes place in estuaries (eg: Indian Gondwana coal deposits). During fossilization protoplasmic contents disappear. The cutinized tissues, hard wood and sclerenchyma resist the decay. Due to pressure of

sedimentary rocks fossils are highly compressed. This compression reduces the vacant spaces in side the cells.

2.4.3 TYPES OF FOSSILIZATION

There are several varieties of plant fossils. Among them 1. Impressions, 2. Compressions 3. Casts or Incrustations 4. Petrifactions are important. Along with them Mummifications and Ambers are also present.

The fossil plants provide most valuable information, about morphology, anatomy and old carbon compounds of those past lived plants. Impressions provide only external morphological details. Compressions provide the past era plants in a compressed form (eg spores, pollengrains, cuticular layers and leaf parts). If the whole plant is available as it was, it is called mummification (eg Diatoms and Desmids). In the moulds type fossils the carbon compound is not available. These provide only tridimentional morphological view of the part plants. Plant parts which are very soft will become mould type of fossils.

In nature, the whole plant availability as a fossil, is very rare. The reason for this is, before fossilization some plant parts degenerate, because of bacterial or viral action in the soil. Even if the whole plant becomes a fossil, the changes that occurs in the earths crust, where the fossils are available, like earth quakes, faults and organogenesis, these whole fossil plants will break in to pieces, and they will separate. Among these separated pieces, only some of the parts are available to us during excavations. The different parts of the plants, collected from different regions and in different sedimentary rocks collection and naming is a difficault task for the palaeobotanists. A brief account our different fossil types is given here.

2.4.4. IMPRESSIONS

The surrounding mud gets deposited on the surface of the plant parts or gets in to the cavity and their impressions are left. These impressions are of two types. May be the surface markings of plant material. May be the markings of the internal cavities in the plant material. In some fossils even details of the epidermal hairs, structure of stomata and details of the venation are seen clearly. But in this type of fossils, carbon material of the past plants is not available. If a leaf fall on semistiff clay, first its organic matter decays and it leaves the imprints of its form and viens (Fig : 2.1D).

2.4.5 COMPRESSIONS

Most common kind of preservation is compression. This type of fossil is formed as a result of great vertical pressure of the sediments. The orgaric matter of the plant is preserved with the impression of the plant (Fig 2.1E). The study of coal as this section or by maceration reveals the presence of spores, pollen grains, cuticles, parts of leaf, wood fragments (Fig 2.1A), bark fragments and resins. These include familiar carbonizations, around 250 millions of years ago in the Carboniferous period. The compression reveals fossils out line but not its thickness.

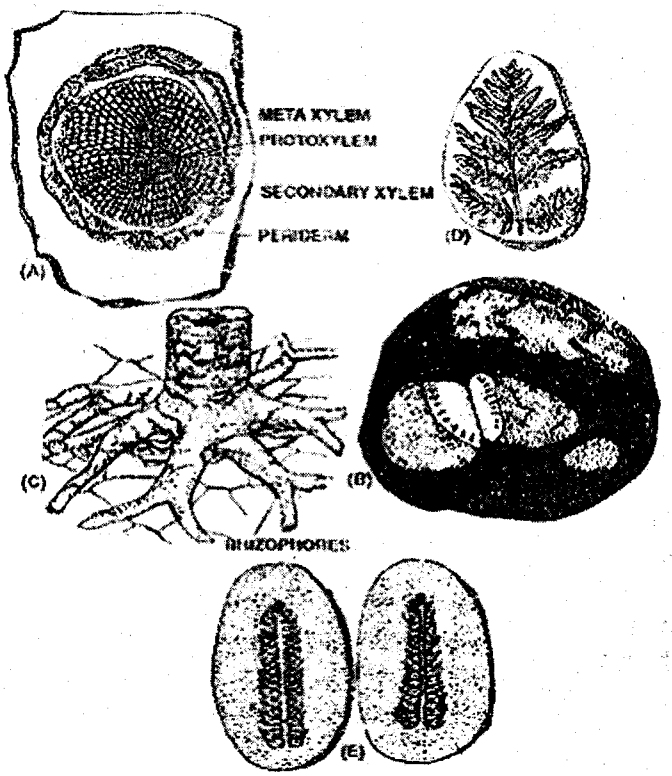


Fig :2.1 (A) Section of coal ball showing stems (B) Section of coal ball showing petrification of stem of Sphaenophyllum (C) Cast of Stigmaria (D) Impression of Neuropteris leaf (E) Compression of Lepidostrobus in a clay nodule.

2.4.6 CASTS OR INCRUSTATIONS.

Casts are also most common type of fossils. A cast does not contain original plant but gives an idea about the external structure. Firstly, the covering of sand or mud on the parts of the plant occurs and in course of time the plant material in side rots leaving a hollow structure. This cavity gets filled with some solid material or rock forming material in course of time, the inside as well as outside plant material solidify in to a stone. From this stone the external parts peel off leaving an exact cast of the original plant (Fig 2.1C).

2.4.7 PETRIFACTION

These fossils preserve the external form, internal structure and some times substances of the original plant. This is the only method of transformation of the organic tissue in to stone. These fossils are very rare. The infiltration is followed by precipitation, so that calcium carbonate, magnesium carbonate, silica, iron sulphide etc., are formed. The original organic material may get destroyed but the cell wall compounds may remain and make the whole structure stone like (Fig : 2.1B). Some times delicate parts remain intact so that the external morphology and internal structure is clearly preserved.

2.4.8 MUMMIFICATION

The total plant part, just like in algae and fungi, is available as fossil. This is known as mummification. Occasionally the tissues in leathery leaves or though fruits, are retained in a mummified condition.

2.4.9. AMBERS (PSEUDO-FOSSILS)

Diatomaceous earth formed by skeletons of billions of diatoms, depositing on the sea bed; different types of algal limestones; graphite used in the lead pencils and even petroleum comes under this category. Besides the insects and small animals, the above plant fragments are well preserved and are ranked as fossils because any object connected with ancient organisms is considered as a fossil. But in strict sense they are to be called pseudo-fossils. 'Amber' is the resinous extraction of coniferous trees. It becomes hardened and accumulated over long periods. Some times this encloses a flower or insect.

2.5 Summary

The total life span of the earth from the time of its origin is called geological time. The discovery of radioactivity served as a tool in estimation of geological time. The geological time is divided into major divisions called *"Eras"*. The eras into *"Periods"* and the periods into *"Epochs"*. The eras, periods and epochs of the geological time if arranged in an orderly manner that arrangement is called *"Geological time scale"*. The oldest is named as *"Palaeozoic era"*. The Mesozoic era is called *" The Age of Gymnosperms"*. Cainozoic era is the recent period where the highly evolved land plants like angiosperms are dominant.

The word 'Fossil' is derived from the latin verb "Fodere" which means to dig. Fossil is a record of former life. "*Palaeobotany*" is the study of plants existed in the past and now entirely extinct. In nature the whole plant is not available as fossil. Only parts of the plants, at different regions are available due to break down of plant parts. The fossil are of various types ie., The most convincing evidence of plant evolution comes form the fossil plant record. Continuous study and analysis of roots, stems, leaves and reproductive organs of past plant life enriched the fossil gymnosperm science. The naming of plant fossils occurs in a specialized manner. If the botanical affinities of the available fossil plant is known then it is called *'organ genus'* if not known, then that fossil is known as *'form genus'*.

1. Impressions 2. Compressions 3. Casts 4. Petrifactions.
5. Mummifications 6. Ambers (Pseudo-fossils).

The Gymnosperms, is an ancient group dates back to Devonian period. They were predominant over the earths surface during Jurassic and Cretaceous periods of Mesozoic era by the end of Mesozoic era they were gradually replaced by angiosperms. Many Gymnospermous genera are available as fossils only. (eg Pteridospermales, Corditales, Bennittitales). Among fossil plants Gymnosperms have a separate status and recognition.

2.6 Technical Terms

- **Fossil** : A record of former life.
- **Palaeobotany** : The study of plants existed in the past and now entirely extinct and became fossils.

- **Organ genus** : The botanical affinities of the available fossil is known it is organ genus.
- **Form genus** : The botanical affinities of the available fossil is not known it is form genus.
- **Geological time** : The total life span of the earth from the time of its origin is called geological time.
- **Geological time scale** : The geological time is divided in to eras, periods and epochs. They are arranged in an orderly manner and this arrangement is called geological time scale.

2.7 MODEL QUESTIONS

I Answer in 30 Lines

1. Give an account of fossils and fossilization methods?
2. What are fossils ? Describe different types of fossils?
3. What is geological age? Give details of Geological time scale ?

II Answer in 10 lines

1. Types of fossils
2. Mesozoic era
3. Diagrammatic representation of Geological time scale

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Lesson -3

GENERAL CHARACTERS OF PTERIDOSPERMALES AND CYCADEOIDALES

3.1 OBJECTIVES

After reading this lesson you will know
General characters of Pteridospermales, classification and similarities with ferns and Cycadophytes. Charecteristic features of Lyginopteridaceae. Cycadeoidales general characters, classification and affinities. Fossils which belongs to family: Cycadeoidaceae, order: Bennittitales.

STRUCTURE

3.2 INTRODUCTION

The Gymnosperms had an elaborate and extensive fossil history. Among the fossil plant groups Gymnosperms had a distinction. Many Gymnosperms are available as fossils only (Pteridospermales, Corditales and Bennettitales). Pteriospermales, came into existence during the upper Devonian and lived through Carboniferous and Permian period. They reached their climax in the Mesozoic era. The fossils of this period consists fern like-leaves and seeds that

were unprotected. Hence they were named as *"Pteridosperms"*. *"Pteridospermae"* is the name coined by Oliver & Scott (1903). In the Gondwana rocks of India, these Pteridosperms were identified. The members of Cycadeoidales(Bennettitales) have been collected from various places in the world from upper Jurassic period to upper cretaceous periods of Mesozoic era. Even though *"Bennettitales"* is the name already they had, Arnold (1948) favoured the name *"Cycadeoidales"* to this fossil group. In the external features Cycadeoidales group resembles cycads.

3.3 PTERIDOSPERMALES

The Eur-American coal reserves are formed from the Pteriospermales, remnants of tree plants once lived. They have grown luxuriantly in the marshy soils in those days. The Pteriospermales resemble *"ferns"* on one side and *"Cycads"* on the other side. Pteridospermales are most primitive plants among spermatophyta. The fossil remains of Pteridospermales are available incompletely and shows diversity in availability.

3.3.1 General Characters of Pteridospermales

It is difficult to define this group, due to incomplete fossil remains and more diversity in the available fossils. The fossils of Pteridospermales are available in both Palaeozoic and Mesozoic eras. More data is available on Palaeozoic era fossils. The stems of these plants are slender, erect and weak. The leaves are large, Pinnately compound and frond – like.

Thick cuticle is present on leaves and leaf traces are large, consisting of a single mesarch strand or several strands. Primary xylem is mesarch or rarely exarch. The stem is monostelic or some-times polystelic. The stele is a medullated protostele or siphonostele. Secondary growth is present in stems. Secondary wood is manoxylic. The tracheids bear multiseriate bordered pits on their radial walls. Secondary phloem is formed in small amounts. Plants are heterosporous and heterothallic. Microsporangia are free or grouped in to synangia. The microsporangia have no annulus.

The pollen grains are monolete or trilete as in Lyginopteridaceae. Pollen grains do not develop pollen tubes. So it is presumed that the male gametes are motile and are directly discharged in to the pollen chamber. Male gametophyte is multicellular. The ovules/seeds are borne directly on an unmodified or modified frond called *"Megasporophyll"*. They are organized into cones or inflorescences.

The ovules/seeds possess close resemblances with the ovules of modern cycads. The ovules lack annulus and are surrounded by " *Cupule*-like" structures. The ovules have integument either free are fused with the nucellus. A dlistinct micropyle and pollen chamber are present, in ovules. The megaspore is surrounded by a thick wall. The seeds are radially symmetrical.

3.3.2 Affinities of Pteridospermales.

The Pteridospermales resemble *"Fern Plants"* of *"Pteridophta"* on one side and *"Gymnosperms"* on the other side.

The have the following Similarities with ferns (Pteridophyta)

Pinnately compound large leaves. Dichotomously branched lateral viens. Polystelic condition in the stems of some genera. The xylem is mesarch, rarely exarch. The leaf traces are also mesarch and may be single or many. Like the pteridophyte spores the microspores are trilete. Motile and multiflagellate spermatozoids. Like those of many heterosporous ferns, the gametophytes are endosporic. Presence of archegonia in the ovules. The production of synangia on pinnules. A thick wall surrounds the megaspore.

The Pteridospermales and cycads resemble each other in the following aspects. The leaf traces are mesarch. Absence of female cones in all living cycads. Fond – like nature of pollenbearing structures. Megasporophylls are foliar in nature. Female cones are absent in living cycads. Endosporic and reduced gametophytes. The ovules are with single integument. This integument consists of three layers. The ovules posses pollen chamber and archegonia. Annulus is absent in microsporangia. The megaspore has a thick wall. Bierhost (1971) opined that Bennittitales. had their origin from pteridospermous stock and they have evolved parallel to cycads.

3.3.3 Classification of Pteridospermales

Sporne (1974) recognized seven families in the Pteridospermales order. Out of them three families belong to Palaeozoic era and the remaining four belongs to Mesozoic era.

PALAEOZOIC PTERIDOSPERMALES

1. Lyginopteridaceae. 2. Medullosaceae. 3. Calamopityaceae.

MESOZOIC PTERIDOSPERMALES

1. Glossopteridaceae 2. Peltaspermataceae
3. Corystospermaceae 4. Caytoniaceae.

The details about family, Lyginopteridaceae enriches the knowledge about Pteridospermales.

3.3.4 Family: Lyginopteridaceae.

This family belong to Palaeozoic Pteridospermales. The plants of this family are available as fossils only. The fossil plant parts such as stems, leaves, microsporangia and ovules are available like impressions, compressions and petrifactions only. These fossil plant parts were discovered in different parts of the world and described under different genera

Stems	-----	**Lyginopteris, Heterangium.**
Leaves	-----	**Sphenopteris.**
Ovules	-----	**Lagenostoma, Spherostoma.**
Pollen –Bearing organs	-----	**Crossotheca, Telangium.**

Fig. 3.1 *Lyginopteris oldhamia* branch.

1. Young leaf showing circinate vernation; 2. Big fern like compound leaf. 3. Stem; 4. Microsporophyll.

STEMS

Lyginopteris oldhamia was described in detail among all the plants. There is sufficient information about the seeds of Lyginopteris but on the pollen bearing organs there is no authentic information. *"Calymmatotheca hoeninghausi"* is the another name to *Lyginopteris oldhamia.* These plants are abundantly available in the coal mines of lower carboniferous age of England and America. *Lyginopteris* habit resembles a straggler or woody liane (Fig:3.1).

The stem of *Lyginopteris* is slender and weak with a diameter of 3-5 inches. Leaves which are fern frond-like and big in size are arranged spirally on the stem and with a length of half meter. Adventious root system is present. On all the plant parts (Except on roots) glandular hairs are formed. Due to disintegration of the cells, on the tip of these glandular hairs they exude resin or mucilage.

The leaves of *Lyginopteris,* if found alone, they are called " *Sphenopteris hoeninghausi".* They are big in size. In shape, they resemble fern leaves and grows up to half meter length. The leaves are spirally arranged on stems. On the rachis of the leaf there are several glandular hairs present. The pinnae arrangement on the leaf rachis is feather like. Leaflets are present on pinnae. Young leaves shows circinate vernation.

The lower and upper epidermis are present on the leaflets of *Lyginopteris* . Thick cuticle is present on the epidermis. These leaves were collected as fossil impressions because of this cuticle on leaves. In the mesophyll these is differentiation into palisade and spongy parenchyma. Stomata are present only in the lower epidermis.

ANATOMY OF STEM

The transection of *Lyginopteris oldhamia stem* is circular in outline (Fig 3.2). It shows 3 regions **Epidermis, Cortex and Stele.** Young stem shows a broad cortex covered by epidermis. Cortex is divided in to outer cortex and inner cortex.

The outer cortex consists of radially elongated fibrous strands, forming a network. Parenchymatous cells are present in between these strands. This network of anastomosing fibres is called *dictyoxylon* cortex and is characteristic of *Lyginopteris* stem. The dictyoxylon cortex cells appear as black streaks and resemble *"Roman letters"*. This makes identification very easy. The inner cortex is many layered and made up of parenchymatous cells. Below the inner cortex, pericycle with sclerotic cells is present as a first layer of stele.

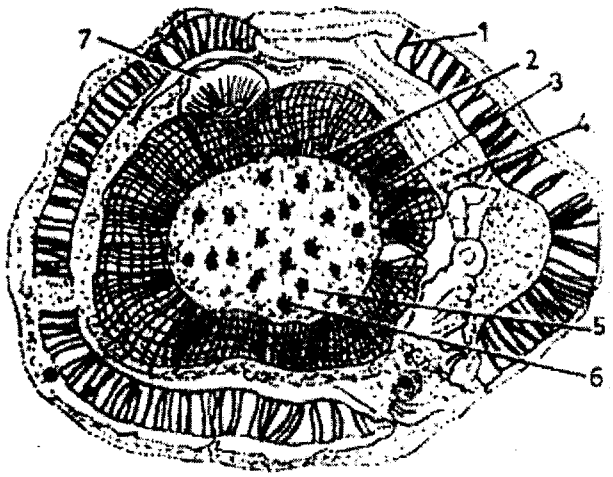

Fig. 3.2. ***Lyginopterisoldhamia*** **Stem T.S Showing secondary growth**
1. Outer cortex (Dictyoxy lon cortex); 2. Primary Xylem
3. Secondary Xylem; 4. Inner cortex; 5. Pith
6. Sclerotic cell groups; 7. Leaf trace.

The stele is described as eustele because. 5-8, Conjoint, collateral, open and mesarch vascular bundles are arranged in a ring around the central pith. Meristematic tissue is present in-between xylem and phloem in the vascular bundle. *"Sclerotic nests"* which has thick walled cells, are also present in the pith.

Secondary growth in the stems is very common. The production of secondary xylem during secondary growth is limited. The secondary xylem is manoxylic type. In the xylem, tracheids and multilayered parenchymatic *"Xylem rays"* are present. The scalariform tracheids in the xylem show multiseriate bordered pits on their walls.

The leaf trace arises by tangential division of the stem bundle. This strand traverse through the secondary xylem and it divides in to two, as it passes the inner cortex. At the base of the petiole, the leaf trace bundles reunite to form a V- Shaped trace. It traverses through the

petiole as a single trace, and then branches in to two, and each branch enters in to a forking rachis. The vascular bundle in the petiole is V-Shaped with the concavity facing upwards.

REPRODUCTIVE STRUCTUURES:

OVULES (SEEDS)

The ovules (seeds) are covered by a protective covering called "*Cupule*". The cupule is covered with capitate glands, typical of those found on the stems and leaves. These features confirm that these ovules/seeds belongs to *Lyginopteris* plants (Fig: 3-3A). Scattered seeds are described as *Laginostoma Lomaxi (Fig 3.3B).* Oliver & Scot (1904) first described these *Lyginopteris* seeds that are small i.e. 5.5 mm. X 4.2 mm. These ovules are orthotropous and barrel shaped.

The integument is fused with the nucellus except at the apical region, where it is divided in to nine lobes (Fig: 3.3A). A single vascular bundle enters into the ovule at the base and divides in to nine strands, each one enters in to the nine lobes of the integument.

- *Laginostoma lomaxi* ovular integument consisits of two layers. The outer layer is stony, where as the inner one is soft with parenchymatic cells.

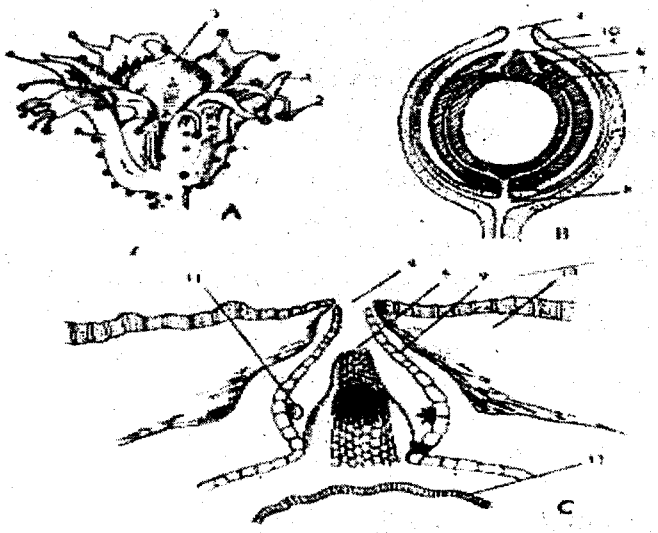

Fig. 3.3 *Lagenostoma lomaxi* seed.

A. Seed with glandular haired cupule; B. L.S of seed; C. Upper part in seed L.S.

1. Flower like cupule. 2. Glandular hairs; 3. Seed; 4. Micropyle. 5. Lagenostome. 6. Pollen chamber;7. Female Gametophyte; 8. Conducting Tissue. 9. Necellar Sclerotic layer; 10. Integument; 11.Pollen grain; 12. Female Gametophyte upper layer.

At the apical region of the ovule, where the integument and nucellar tissue are not fused, a beak like structure is formed because of the bulging of the nucellar tissue. This is called as " *Laginostome*" (Fig 3.3C). Around this a circular pollen chamber is formed (3.3C). Long (1944) observed archegonia in the female gametophyte tissue, in the ovules of *Laginostoma ovoides.* It is understood that in *Lyginopteris* ovules, pollen grains after reaching

the circular pollen chamber, germinate to produce male gametes just like in modern cycads and participate in fertilization.

POLLEN –BEARING STRUCTURES

The male reproductive organs microsporangia of *Lyginopteris* is not known with certainty. The form genus *Crossotheca* is generally regarded as the Pollen bearing organ of this species.

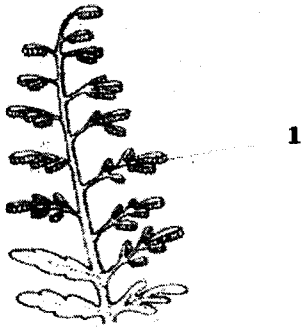

Fig .3.4 *Crossotheca hoeninghausii* Pollen bearing organs

1. Hanging male sporangia.

Arnold (1947) opined that *"Telangium"* may be microsporangia bearing structure in *"Lyginopteris"* plant. The microsporangia bearing structures along with *Lyginopteris* stems were not found till today. So, *Crossotheca* whether belongs to *Lyginopteris* plant or not, is not confirmed so far. But along with *Crossotheca, Lyginopteris* stems were available separately (Fig 3.4). *Crossotheca hoeninghausii* (Fig3.4) is a pinnately branched frond–like structure bearing microsporangia. The fertile frond terminated in a number of lateral branchlets which appeared like a miniature hair brushes. Each brush has a small ovate plate at the end about 2 mm. length. About 6 bilocular sporangia with several microspores are attached at one face of the plate. Many species of *Crossotheca* are available with *Sphenopteris* leaves as fossils.

3.4 CYCADEOIDALES (BENNETITALES)

These plants belonged to, Jurassic and cretaceous periods of Merozoic era. They are wide spread on earth during this period and showed their dominance on the earth. This order was named as *"Bennetitales"* in recognition of a British scientist J.J. Bennett. But Arnold (1948) suggested that *"Cycadeoidales"* is the correct name to this order. Hence these two names are used as synonyms to recognize this order.

The fossils of this order were collected from Western Europe, France, America, Poland, Austria and Germany. These fossils have been also collected from Bihar, Gujarat and Andhra Pradesh are the states in India and also in the delta region of Krishna and Godavari. In 1825, from Great Britain the fossil stem *"Bucklandia"* has been identified. In 1932, Prof. Birbal Sahni of India identified and reconstituted the most important fossil plant *Williamsonia sewardiana*.

Most of the fossils collected during Mesozoic era belonged to cycadeoidales. Hence the Mesozoic era can be called *" Age of cycadeoidales"* in stead of *"Age of Cycads"*. Thomas and Bankraft (1913) after observing the leaves of Cycadeoidales and Cycads found that there are two types of stomata in these fossils. Haplocheilic Stomata in which the guard cells and subsidiary cells originate from different initials. (eg Cycadales) and Syndetocheilic Stomata where the guard cells and subsidiary cells originate from the same initial cell. (eg Cycadeoidales).

3.4.1 GENERAL CHARACTERS OF CYCADEOIDALES

Stems are strait, woody and cylindrical. Branching is very rare. Plants are perennial trees. On the stem there are persistent rhomboidal leaf bases and in between them ramental hairs are present. At the tip of the stem pinnately compound leaves are present just like a crown. Leaves in *Williamsoniella* are simple. Venation is parallel or simply diverging type. Stomata are syndetocheilic type.

"Eustele" type of conducting tissue is present in stems. In this a broad pith, encircling this pith, vascular bundles are arranged in a ring. Vascular bundles are conjoint , collateral, open and endarch. Secondary Xylem is manoxylic type. No annual rings in the stem. *"Flowers"* are the reproductive structures. The flowers may be unisexual (*Williamsonia*) or bisexual (*Cycadeoidea*). These flowers resemble Ranalian flower of angiosperms, superficially. In this flower bracts with ramenta like hairs are arranged in spiral manner. The microsporophylls resemble long pinnate compound leaves. They are present, at the base of the thalamus in a circular manner. 8-20 microsporophylls are present in a whorl. The fertile pinnae (which are present on the rachis) have two rows of bean shaped *Pollen Capsules or Synangia* in sub – lateral position. Each synangaum contain variable number of pollen sachs. Each pollen sac produces many pollen grains Pollen grains are monocolpate.

Conical shaped thalamus, upon which stalked ovules, in between them inter- ovular scales are present in female flower. The inter-ovular scales had a widened distal end and forms a protective plate around female flowers. Ovules are orthotropous. There is a three layered integument around nucellus in the ovule. The nucellar tissue stretches and forms a pollen chamber at the micropylar end. Seeds contain dicotyledonous embryo. Seeds are not endospermic.

3.4.2 AFFINITIES OF CYCADEOIDALES:

" Cycadeoidales" have resemblances with ferns, Pteridospermales, Cycadales and Angiosperms.

A. RESEMBLANCES WITH FERNS

Pinnately compound leaves just like fern leaves. Leaf traces directly enter in to petiole. Microsporangia are like a capsule, as in *"Maratiaceae"*. Ramenta like hairs are present which are brown in colour.

B. RESEMBLANCES WITH PTERIDOSPERMS:

Long, pinnately compound sporophylls. Mesarch vascular bundles in leaf traces. Manoxylic type of xylem. According to Sporne (1965), there is no evidence that Bennetitales

have originated from Pteridospermales. However, most of the scientists are of the opinion that both Bennetitales and Pteridospermales originated from a common ancestor *"Progymnospermopsida"* stalk in two parallel lines.

C. RESEMBLANCES WITH CYCADALES:

Stems are branched or unbranched. Crown like arrangement of leaves on the distal part of the stem. Pith is broad.

D. RESEMBLANCES WITH ANGIOSPERMS:

Wieland (1906) and Arber and Parkin (1907) opined that bisexual flowers in Cycadeoidales resemble angiospermic flowers. According to them the angiosperms are originated from Bennetitales. The flowers of Bennetitales and magnolian flower of Angiosperms shows the following resemblances.

Perianth like bracts. Long and conical shaped thalamus. The arrangement of reproductive parts. Spirally arranged ovules.

Basing on the above resemblances it is proposed that Angiosperms are evolved from Bennetitales. But the flowers of Bennititales and Angiosperms shows the following differences.

1. Microsporophylls possess many pollen capsules. These pollen capsules are not similar to Angiosperm stamen.
2. The seeds in Bennetitales are gymonospermous. But in *Magnolia* ovules are in a closed ovary which is formed by the fusion of carpel.
3. Inter-ovular scales are not present in Angiosperms but are present in Bennetitales.
4. The *" Fruit wall"* in Bennetitales is formed because of the union of broadened tips of inter-ovular scales. But in the Angiosperms it is formed from the ovary wall.

Basing on these differences there is no relationship between Bennetitales and Angiosperms. Sporne (1974) recognized 3 families in the Cycadeoidales order.

1. Williamsoniaceae 2. Wielandiellaceae. 3. Cycadeoidaceae.

3.4.3 FAMILY: CYCADEOIDACEAE

" Cycadeoidea" is the only genus present in this family. *" Bennetitis"* is the name given to this genus in European countries. From the rocks of Jurassic and Cretaceous periods (Mesozoic era) *cycadeoidea* fossils were identified in several Countries.

EXTERNAL MORPHOLOGY

Stems are short and stout (Fig.3.5). Stems are spherical or conical, with branched or unbranched trunk having diameter of 60 cm. The stem is covered with an armour of persistent leaf bases with multicellular hairs or ramenta in between them.

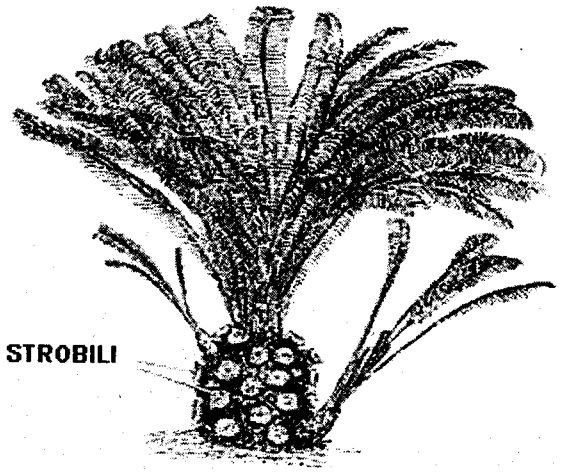

Fig : 3.5 Cycadeoidea dacotensis

The trunk is up to 1 meter height and terminated in a crown of large pinnate compound leaves. The leaves are about 2-3 meters long. The leaflets are xerophytic and had parallel veins. However the attachment between the trunk and leaves is not found so far.

STEM ANATOMY:

Cycadeoidea stem is circular in outline in transverse section. Numerous leaf bases and multicellular ramenta are present on this tem. Epidermis is not clear. Parenchymatous, broad cortex is present. It is traversed by many mucilage canals and leaf traces. Conjoint, collateral, open and endarch vascular bundles are arranged in a ring in the stele (Fig : 3.6). The leaf trace is single at the place of its origin and is passed straight in to the leaf with out forming the girdling traces. The 'C' shaped leaf trace splits in to a number of mesarch strands as it passed through the cortex. They are horse-shoe shaped. Pith is broad and encircled by primary xylem. This is surrounded by secondary xylem out side. Secondary xylem is manoxylic type.

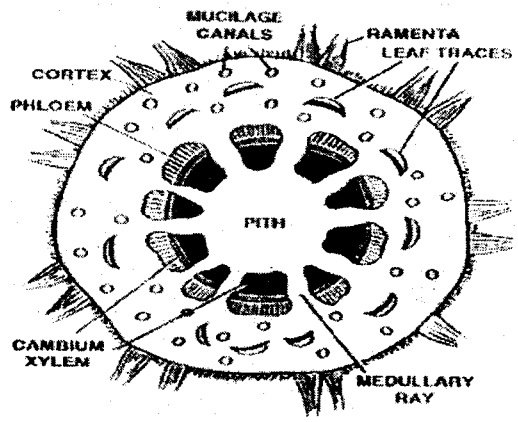

Fig :3.6 T.S Cycadeoidea stem

Uniseriate or biseriate medullary rays are present. Secondary phloem is present at the out side of the secondary xylem.

ANATOMY OF LEAF

A vertical section of the pinnule shows clear upper epidermis and lower epidermis (Fig: 3.7). Epidermis is uniseriate but the cells are thick walled. The mesophyll which is present in between the upper and lower epidermis is differentiated in to palisade and spongy parenchyma. Bundle sheath is present around the vascular bundles. Xylem is towards the upper epidermis and phloem is towards the lower epidermis in the vascular bundle.

REPRODUCTIVE PARTS

The reproductive parts in *Cycadeoidea* are termed as howers. Flowers are shortly pedicellate, bisexual but in some unrisexual (*Cycadeoidea wielandii).* All the floral buds are found to be at the same stage of development. It presumed that all floral buds opened simultaneously and the plant flowered only once in its lifespan (Monocarpic).

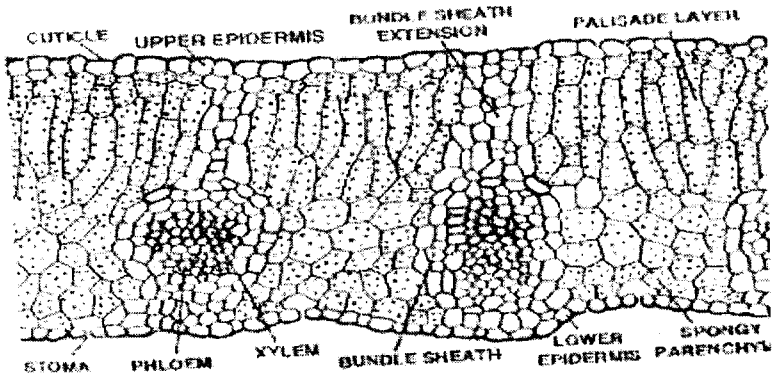

FIG :3.7 Cycadeoidea Pinnule T.S.

The microsporophylls 8-20 are arranged circularly on thalamus (Fig: 3.8). These are similar to pinnate compound leaves which are long. At the base they are united. Wieland (1906-1916) opined that microsporophyll is equivalent to a stamen. In the early stages of development these microsporophylls are closed. Each microsporophyll/stamen is 10-12 cm long and more like a foliage leaf having a central rachis with numberous pinnae, alternating each other. The pinnae were sterile at the base and the extreme tip. The fertile pinnae have two rows of bean shaped pollen capsules or synangia in sub – lateral position (Fig:3.8). Delevoryas (1963) believed that the microsporophyll have never expanded. Mostly self pollination will occur in these flowers (Crepet, 1972 and 74). Female part of the flower consists of a conical or spherical receptacle which bears hundreds of stalked ovules and inter-seminal scales. They were at right angles to the surface.

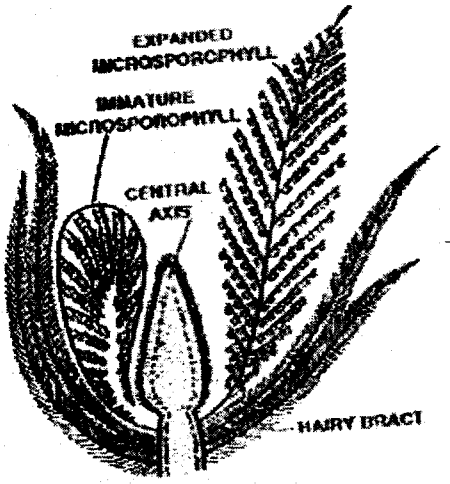

Fig: 3.8 *Cycadeoidea* bisexual flower

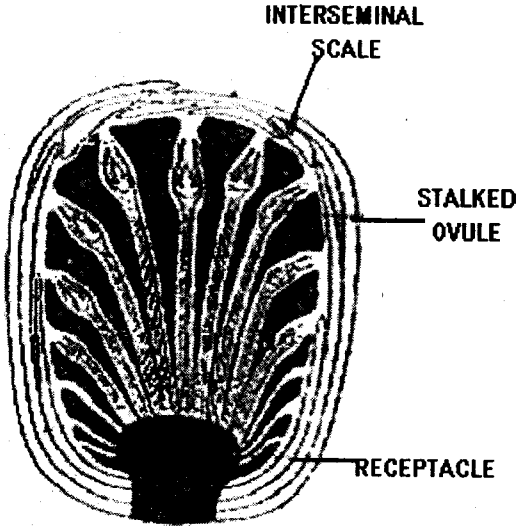

Fig :3.9 Cycadeoidea gynoecium V.S

Each ovule has an integument, which surrounds nucellar tissue. Integument stretches long and produces micropyle. The integument is having three layers. The middle one is hard and the inner and outer layers are soft and parechymatous. There is a nucellar beak and also a pollen chamber. Ovules and interseminal scales unite and produce a fruit like structure (Fig. 3.9). A thick seed coat is present around the seed. The embryo is with two cotyledons.

3.5 SUMMARY

The fossil history of gymnosperms is great and long. This part deals with fossil Gymnosperms like Pteridospermales *(Lyginopteris)* and Cycadeoidales *(Cycadeoidea).* The stem of *Lyginopteris* is having mesarch xylem anatomically. The stele is *"Eustele"* type. The outer cortex is dictyoxylon type i.e. specialized reticulate arrangement of thick walled cells are present. The leaves of *Lyginopteris* if collected separately they are called *Sphenopteri hoeninghausii.* They are very big compound leaves. The margins are lobed and shows dichotomous venation. Seeds are known as *Lagenostoma.* They are small and formed in a cupule. They had a specialized pollen chamber.

Pollen bearing organs are known as *Crossotheca.* Many microsporangia are arranged at the bottom of the pinnately branched lateral branches of a rachis of *Crossotheca.* The stem of *Cycadeoidales* is short and stout, conical or spherical in shape. Stems are unbranched and covered with rhomboidal, persistent leaf bases just like a protective sheath on the stem. Big, compound leaves are arranged like a crown at the tip of the stem. Conjoint, collateral, open and endarch vascular bundles are arranged in the form of a ring, around the large pith. Secondary growth is common in stems. Secondary xylem is manoxylic type. Out side the xylem, secondary phloem is present. In the transaction of leaf upper and lower epidermis are present. In the mesophyll there is a distinction of spongy and palisade parenchyma.

The flowers of Cycadeoidea are generally bisexual but in some monoecious. Solitary flowers are formed in the leaf axils. Bracts with ramenta like hairs, surround the flowers, like perianth. In each flower, 8-20 microsporophylls are arranged in a ring on the thalamus. Two rows of synangia or pollen capsules are arranged in lower region of these microsporophylls. On the receptacle, there are several stalked ovules alternately arranged with inter –seminal scales. An integument surrounding the nucellar tissue is present in the ovule. Seeds are oval in shape, covered with thick seed coat, and an embryo with two cotyledons inside the seed are present.

3.6 TECHNICAL TERMS

- Eustele : Around the broad pith conjoint, collateral, open and endarch vascular bundles are arranged like a ring.
- Dictyoxylon Cortex: Cortical cells are stretched lengthwise and these fibers shows reticulate plate like arrangement.
- Lagenostome: At the base of the micropyle where the integument and nucellar tissue are not united, the nucellar tissue bulges like a beak in *Lyginopteris.* This is called lagenostome.
- Syndetocheilic stomata: Both the guard cells and subsidiary cells formed from the same mother cell.
- Haplocheilic stomata: Only guard cells arise from the same mother cell.

3.7 MODEL QUESTIONS :

I. Answer each question in 30 lines.

1. Describe the vegetative and reproductive characters of *Lyginopteris* ?
2. Write in detail about *"Cycadeoidea"* ?

II. Answer each question in 10 lines.

1. Write about Lagenostoma in short ?
2. Write about Crossotheca in detail ?
3. Write about Bennittitales ?
4. Write about Pteridospermales ?
5. Write about the reproductive parts of Cycadeoidea ?

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- Dr.P.V.RAMA RAO.

Lesson - 4

PINUS

4.1. OBJECTIVES

After reading this part you will know

The external morphology and internal structure of sporophyte of *pinus*. Also the reproduction and economic importance of *Pinus* are dealt in detail.

STRUCTURE

4.2. INTRODUCTION

The genus *Pinus* belong to the order coniferales of the class coniferopsida. Bierhorst (1971) recognized 6 families in the **order : Coniferales.** Pinaceae is the fourth family. Plants of this order are mostly trees or shrubs. These are pyramidal or conical in shape. They have unbranched trunk up to some height in the trees. *Sequoia sempervirens*(Taxodiaceae) grows up to 100 metres height. There are 52 genera and 566 species are present in Coniferales.

Some conifers have an exceptionally long life. In the 'National Forest of California' (USA) there is a tree of *Pinus aristata* which is more than 4600 years old.

Coniferous forests are largest, continuous and dense in the world. Conifer leaves are mostly needle like and evergreen. They are not deciduous. In *"Araucaria"* the leaves may persist and remain green for as long as 15 years. Anatomically transfusion tissue is present in leaves which is associated with lateral conduction. The roots of conifers have ectotrophic mycorrhizal association. Mycorrhiza is a symbiotic association between the root cells and fungi. In Araucariaceae, the mycorrihizae are endotrophic, while the roots of Podocarpaceae develop root nodules.

Pinaceae is one of the importance farnities of order coniferales. The family includes about 18 genera and 200 species. Plant parts shows spiral arrangement. The microsporophyll contains two pollen sacs. The ovuliferous scale and bract scale in megasporophyll are free or they may be united at base. Two ovules are present on each ovuliferous scale. Generally they have winged seeds. Polyembryony is present. The exact origin of Pinaceae is not known. But the family certainly appeared by early Cretaceous period. The genus *Pinus* is represented by over one hundred speices.

4.3 Pinus Distribution and habit

Pinus species are widely distributed in the temperate and cold regions of Northern part of globe. About 6 species of *Pinus* have been recorded from different parts of India. Generally they grow in hill slopes and in plains. Pinus plants are perennial and ever green trees. Generally they grow as huge trees. Some plants are shrubs. (eg *Pinus pumilla*).

4.4 External Morphology

4.4.1. Stem :

Young pine tress are pyramidal in shape with horizontal branches at regular intervals. The symmetry is lost, as the tree matures and the crown becomes round or spreading. Generally they grow as huge trees. They attain a height of 70-200 feet. They trunk had a diameter of 10-12 feet. Main stem grows strait and woody. It possess bark on trunk and rough in nature. The apical bud is active and shows growth throughout the life of the plant. The branches at the bottom are long and they are comparatively short as it proceeds to the top. So it gives pyramid shape to the plant (Fig 1.1B). The plant looses its symmetry because of the death of the old branches or falling of branches due to wind pressure.

4.4.2 Branches :

There are two types of branches produced in *Pinus, viz.,* The branches of unlimited growth (long shorts and limited growth (dwarf shorts). The long shoots occur on the main stem as lateral buds in the axils of scale leaves. The dwarf shoot develops on a long shoot in the axis of a scale leaf (Fig.4.1).

4.4.3 Leaves :

Two types of leaves are produced in *Pinus*. They are 1). brown colored scale leaves and 2). vegetative, green needle-like leaves. Scale leaves are produced on both the branches. After the growth of the branches they will fall off.

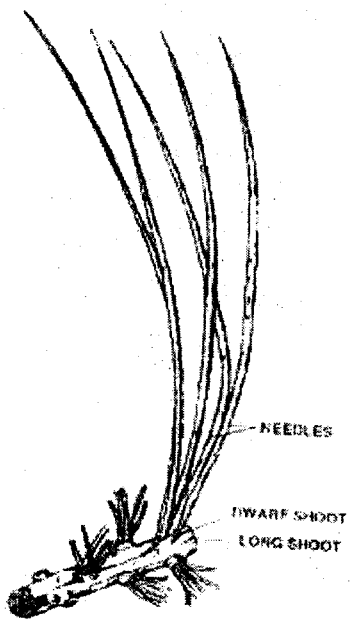


Fig : 4.1 Vegetative branches of *Pinus*

Green leaves are needle-like (Fig 4.1). They are produced in the axils of scale leaves on dwarf shoots only. The green needle-like leaves remain on the plant, for many years. These leaves shed only along with dwarf branches. The green needle-like leaves are intact on the plant for many years, so the plants are evergreen. The number of needle-like leaves on dwarf branches vary in different species.

The number of needle like leaves is one in *Pinus monophylla* two in *Pinus sylvestris* three in *Pinus roxburghii* four in *Pinus quadrifolia* and five in *Pinus wallichiana*

4.4.4. ROOT :

There is a primary tap root with large number of laterals (Long roots). Some of these roots branch dichotomously and form corolloid masses with ectotrophic mycorrhizae. These are called mycorrhizal roots. The *Pinus* plant grows on hill slopes where the soft soil is less and rocks come in the way of roots. So, the primary tap root exists for certain period and laterals roots outgrow the tap root and gives stability to the *Pinus* plant. The ectotrophic mycorrhiza, is useful in the absorption of nitrogen, from soil to the root.

4.5 Internal Anatomy:

4.5.1. Stem Anatomy:

The primary structure or *Pinus* stem resembles dicot stem anatomically. The transection shows three distinct regions, i.e.,

1. Epidermis 2. Cortex 3. Stele.

Epidermis is single layered. The cells in this layer are not regularly arranged. Cuticle is present on the epidermis. Stomata are present here and there. Below the epidermis, there is a multilayered cortex. The outer cortex which is present below the epidermis is sclerenchymatous. Inner cortex is parenchymatous. Endodermis is single layered and present at the end of the cortex. The cells are having casparian thickenings on their radial walls. Resin canals in the cortex is a special character. There is an epithelial layer around the resin duct. They are filled with turpentine. This turpentine is useful in healing the wound, if occurs on the stem.

The stele is surrounded by a pericycle that consists of several layers of parenchyma. There are 6-8, conjoint, collateral, open, endarch vascular bundles are arranged in a ring (Fig: 4.2). The stele is *"Eustele"*. The primary xylem contains of circular or spiral thickenings. In the metazylem tracheids, bordered pits are present on their radial walls and tangential walls.

- There are no vessels in the xylem but xylem parenchyma is present.

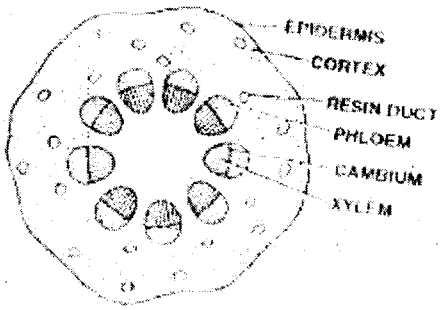


Fig : 4.2 T.S. of ***Pinus*** Young stem (Diagrammatic)

In the centre of the stem enclosed within the ring of vascular bundles there is broad parenchymatous pith. It also contains tannin filled cells. Medullary rays are uriseriate or multiseriate and the latter contain resin ducts.

SECONDARY GROWTH

The secondary growth in *Pinus* stem is similar to dicotyledonous stem, there by secondary tissue increases and the diameter of the stem also increases. "*Cambial ring*" is formed by the union of fascicular cambium (Present in the vascular bundle) and interfascicular cambium (formed in between the vascular bundles during secondary growth). These meristematic cells in the cambial ring produces secondary xylem towards inside and secondary phloem towards outside. Secondary xylem is picnoxylic type. In the secondary xylem distinct

"Annual rings" are formed (Fig.4.3). The autumn wood and spring wood together from the annual ring. The tracheids in the autumn wood the are narrow with thick walls and the lumen is also small. In the spring wood tracheids are broad with thin walls and the lumen is also broad. The age of the plant can be determind by counting the annual rings in its wood.

In the secondary xylem there are tracheids, xylem rays and resin canals but there is no xylem parenchyma. On the radial walls of tracheids, bordered pits are formed. Their arrangement is uniseriate or biseriate. In the uniseriate arrangement, circular bordered pits are formed individually. In the biseriate arrangement bordered pits are formed oppositely. This arrangement is known as *"Abietinian pitting"*. In the *"Auracarian pitting"* the bordered pits are multiseriate and alternate in arrangement. In this the bordered pits are arranged closely and their margins are compressed, they look like "A*ngular*". In many *Pinus* species, in between the uniseriate and biseriate bordered pits which are present on radial walls, there are rod-like thickenings, horizontally. They are called *"Crassulae"*. These crassulae are rod like thickenings formed on the secondary walls of tracheids.

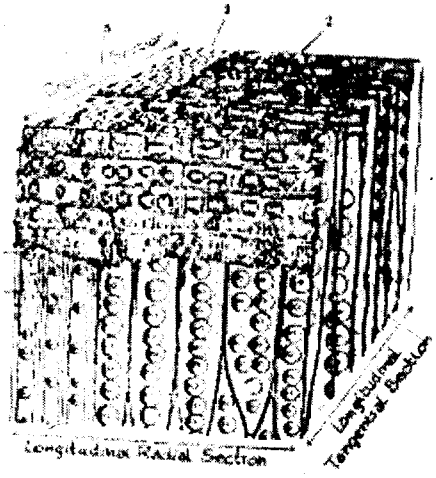


Fig :4.3 Three dimentional view of *PINUS* Xylem (Xylem T.S., T.L.S & R.L.S Can be seen).

1. Autumn Wood; 2. Spring Wood; 3. Xylem ray; 4. Bordered pit 5. Xylem tracheid.

In the pinus wood, xylem rays (Fig:4.3) are of two types namely, short and uniseriate xylem rays and long, spindle shaped multiseriate xylem rays. Resin canals are present in the multiseriate rays.

1. The uniseriate rays with a single row of cells are also called *"Linear vascular rays"*.

2. The ays which include a resin canal become multiseriate and fusiform. Such rays are called *"fusiform vascular rays"*.

The rays vary in size from 2-12 cells in height. In the xylem rays of *Pinus* ray tracheids are also formed at the margins. In *Pinus* wood resin canals are present. This is a charasteritic

feature of *Pinus* wood. Resin canals are also two types. In the secondary phloem there are sieve cells, phloem parenchyma and phloem fibres. Phloem rays are also present. Secondary growth also occur out side the stele i.e in the cortex. This starts with the formation of phellogen in the cortex, which produces secondary cortex towards in side and cork to out side.

4.5.2 Anatomy of Root :

Anatomy of *Pinus* root resembles dicotyledonous root. There are three regions in its transverse section i.e., **1. Piliferous layer 2. Cortex 3. Stele.**

1. Piliferous layer : This is made up of single row of parenchymatous cells. This is also known as epidermis. Root hairs are formed from this layer. The root hairs are less in number in *Pinus* root. In some they are completely absent. Instead of these root hairs ectotrophic mycorrhiza is present and helpful in assimilation.

2. Cortex : Parenchymatous, multilayered cortex is present below the piliferous layer. At the end of cortex, endodermis is present. Casparian thickenings are present on the radial walls of endodermal cells.

3. Stele:- Multilayered layered pericyele is present below the endodermis. Vascular bundles are radial, separate, diarch to tetrarch. Xylem is exarch in arrangement. Proto xylem shows 'Y' shaped arrangement. On the two free arms of 'Y' proto xylem is present and in the mid region a resin canal is present. Xylem consists of tracheids only. No vessels are present. Phloem alternates with xylem sieve tubes and phloem parenchyma are present in the phloem. The companion cells are absent in the phloem.

Parenchymatic cells are present in pith. They contain starch. Primary xylem rays are formed opposite to proto xylem (Fig:4.4). Secondary growth in root is similar to dicotyledonous root. Secondary phloem in less quantity and secondary xylem in large quantity are formed during secondary growth.

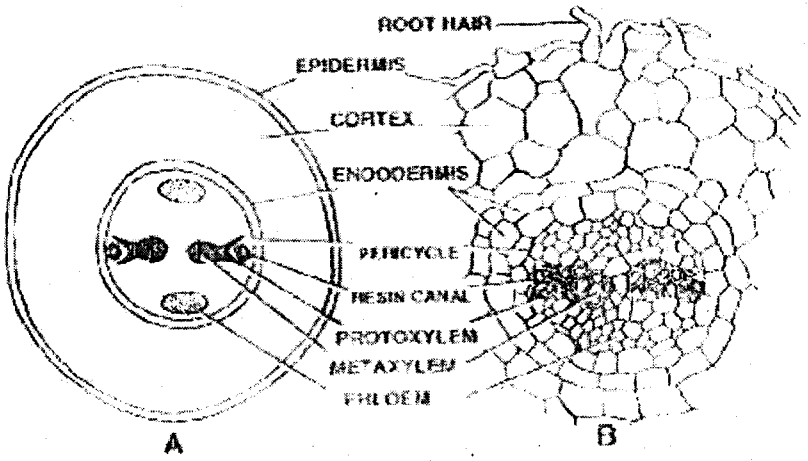


Fig :4.4 *Pinus* – Young Root

(A) Diagrammatic representation of transverse section
(B) Sector enlarged

4.5.3 LEAF (NEEDLE) ANATOMY

Needle–like green leaves are half-circular, triangular or spherical in transection in the *Pinus* species. Anatomically all the species have the same cellular structure (Fig: 4.5). There are three regions in the transection of needle – like green leaf, Viz.,

1. Epidermis 2. Mesophyll 3. Stele.

1. Epiderms :- Epidermis is single layered. Outer walls of these epidermal cells are thick and they are covered with thick mucilage. Stomata are present in pits. They open in to a substomatal cavity. They show haplocheilic development.

2. Mesophyll:- There is no differentiation in to palisade and spongy parenchyma in the mesophyll. There are three regions in this mesophyll:

a) Hypodermis b) Parenchymatic cell zone c) Endodermis.

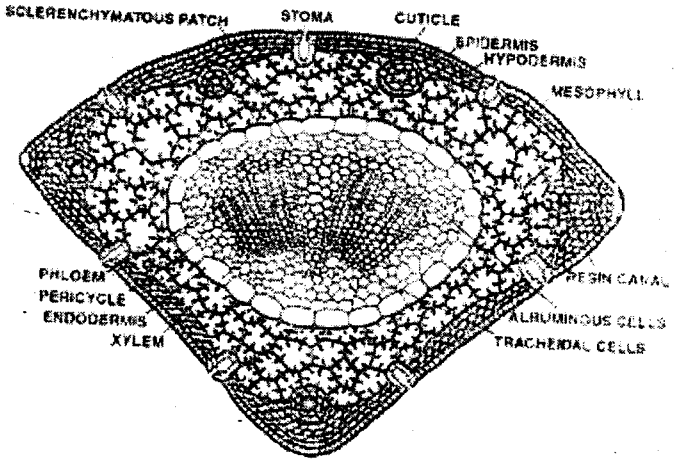


Fig :4.5. *PINUS* NEEDLE T.S.

a) Hypodermis is made up of sclerenctymatic cells. At the corners this is 4-5 layered and in other places it is 1-2 layered. Sclerenchyma is absent below the stomata (Fig:4.5).

b) Parenchymatous Cell zone : Below the hypodermis many layered parenchyma is present. These parenchymatous cells possess abundant chloroplasts and starch grains. Hence they are known as chlorenchymatous cells. The walls of the mesophyll parenchymatous cells give rise to many peg–like infoldings which increase the photosynthetic area of these cells. These are known as peg–like invaginations, arms, flanges or simply folds. They increase the internal photosynthetic area and

compensate the reduction in the leaf surface. Pegged parenchyma is a distinct feature in *Pinus* needle. In the parenchymatic cell zone 2-3 resin canals are present.

c). Endodermis: A single layer of parenchymatous cells at the end of cortex constitutes the endodermis. These cells are filled with starch and no casparian thickenings.

3. Stele: This is divided in to three zones, namely

a) Pericycle. b) Transfusion tissue. c) Vascular bundles.

a) Pericycle : Below the endodermis, multilayered pericycle is present. It consists of parenchymatous cells along with transfusion tissue.

b) Transfusion tissue : There are two types of cells in the transfusion tissue.
(i) Albuminous cells and (ii) Tracheidal cells.

Albuminous cell are bigger in size and occur above the phloem of vascular bundles. They are parenchymatous but are filled with proteins and starch grains. Tracheidal cells are tracheid-like which occur close to the xylem of vascular bundles. These cells possess bordered pits and help in conduction of water and nutrients to mesophyll. Sclerenchymatous cells forms the T- shaped girdle above the two vascular bundles.

b) Vascular Bundles: The number of vascular bundles vary in different species of *Pinus*. These vascular bundles are collateral, open and endarch.

Xerophytic Characters of *Pinus* Needle : The green leaves are needle-like and this shape reduces the transpiration rate. The outer walls of epidermal cells are thick and covered by thick cuticle. Sclerenchymatous hypodermis, sunken stomata, peg-like invaginations of the mesophyll parenchyma, and presence of peculiar transfusion tissue are also xerophytic features.

4.6 REPRODUCTION

Pinus tree is monoecious. Both male and female cones are formed on different branches of the same plant.

4.6.1 MALE CONE : At, end of the long shoots, male cones 15-140 are formed in clusters. Each male cone is formed in the axil of scale leaf where the dwarf shoot is expected. Male cone is oval in shape with 3-4 cm in length and 0.64 cm in diameter. At the base of the cone, there is an involucre, consisting of a number of small bracts. The microsporophylls are borne spirally on a central axis (Fig. 4.6A). Each microsporophyll is equivalent to a stamen and male cone is equivalent to male flower of angiosperms, as per many scientists. But only two microsporangia are present as against the four in angiosperm stamen. The scales situated at the base of the cone are sterile. The release of pollen grains occur in between April – June months. After the release of pollen grains male cones are dried and shed.

MICROSPOROPHYLL: On the central axis of male cone many microsporophylls are arranged spirally. Each microsporophyll is triangular in outline and consists of a short stalk and leaf – like expanded structure. Two pollen sacs or microsporangia are present on the lower side of the

sporophyll (Fig: 4.6D). Each sporangium is filled with numerous winged microspores (Fig 4.6 C&E).

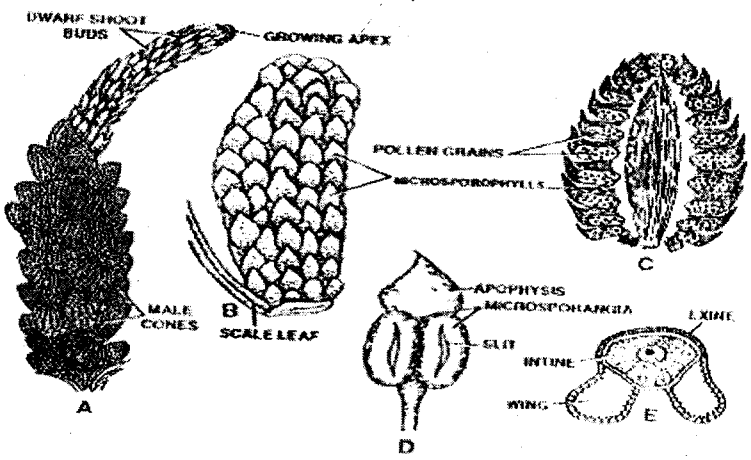


Fig:4.6: Male cone – (A)
A. A group of male cones; B. A single male cone; C. Longitudinal section of male cone; D. Microsporophyll; E. Pollen grain.

The pollen gains are light yellow in colour and the microspores are liberated in such a large number that pine forests appear yellow at the time of dehiscence. This is called "*Shower of Sulphur*".

Microsporangium Development : The development of microsporangium is of *'Eusporangiate'* type. A group of hypodermal cells divide periclinally to from the outer *"Parietal cells"* and inner *"Archesporial cells"*. The parietal cells divide and produce a four layered wall of the microsporangium. The inner archesporial cells divide to from *"Sporogenous cells"* which ultimately differentiate into *"Microspore mother cells"*. The microspore mother cells under go meiosis to form tetrads. These tetrads develop in to microspores (pollen grains). Each pollen grain has three layered wall.

They are exine, sexine and intine. The sexine, which is the middle layer froms two air bladders or wings on two sides of the pollen grain. These are helpful in the dispersal of pollen grains. *Pinus* pollen grains are heteropolar. i.e., the distal surface and proximal surface are not alike. On the distal surface of the pollen grain a scar like germ pore is formed and this is called as *"sulcus"*. The pollen grains develop partially before their dispersal. They contain four celled male gametophyte, by the time they shed from microsporangium.

4.6.2. FEMALE CONE : Female cone arise from the axils of the scale leaves in the place of long shoots. 1-4 female cones arise as a group. Young female cones are compact, small in size (Fig.4.7A&B). They take about three years to mature (Fig.4.7C). They are stalked and reddish green in colour. They are protected by an involucre of bract scales. It consists of a central axis, upon which 20-80 megasporophylls are spirally arranged (Fig.4.7A&B). At the base of the female cone there are some sterile megasporophyll are present.

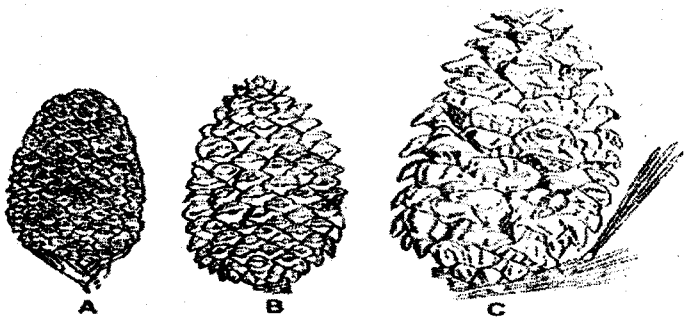


Fig. 4.7. Female cones

A &B Young Cones. (C) Old Cone.

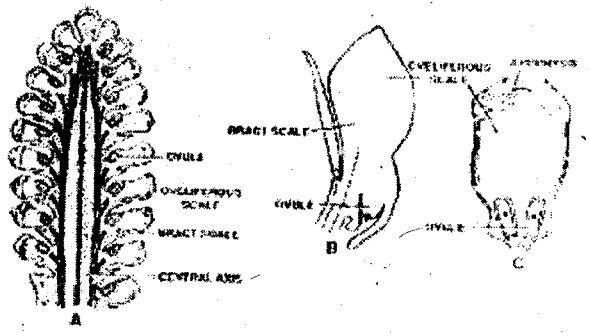


Fig.4.8. Female cone

(A) Longitudinal Section; (B) Megasporophyll; (C) Ovuliferous scale

Each mesasporophylls is a compound structure. Paired scales are present ineach megasporophyll (Fig.4.8B) .Bract Scale: This is the lower scale. It is directly attached to the central axis of the cone. It is like a thin and dried layer. These are spirally arranged on the main axis of the cone. One vascular bundle also enters into this scale. In this vascular bundle, xylem is present towards the adaxial surface. Hence, the bract scale is considered as a modified leaf (Fig .4.8 A&B).

Oveliferous Scale: In the axil of each bract scale, there is one big ovuliferous scale (Fig.4.8C). This is hard, triangular and united at the base with the bract scale. The tip of the ovuliferous scale is sterile and bend towards back. This part is called *"Apophysis"*. On the adaxial surface of the ovuliferous scale, at the base, two ovules are arranged. Their micropyles are directed towards the axis of the cone.

The surface of the cone is marked by rhomboidal areas each with a small central conical point. This pint is known as *"Umbo"*. The rhomboidal areas are the outlines of the broad sterile apophyses of the ovuliferous scales and the tips of the apophysis is the umbo. In the aged female cone, the megasporophylls are wide open form the central axis to fecilitate pollination (Fig 4.7C).

4.6.3. Morphlogy of the Megasporophyll : The morphological nature of the paired scales has long been a debated question. There are several views expressed by different scientists. This is due to, two different questions which come in the way of understanding the nature of these paired scales. i.e., whether the *Pinus* female cone is simple or compound structure (an in Flore science)

ROBERT BROWN (1827) opined that the ovuliferous scale is an open carpel, arising in the axil of the bract scale. But bract scale is leaf like. And in its axil, no leaf is formed. Hence the pinion is not correct morphologically.

BRAUN (1847) explained that the ovuliferous scale is formed by the fusion of the first two leaves of an axillary shoot. According to him bract scale is equivalent to green leaf.

Based on the formation of two green leaved dwarf branches, in some special cones of *Pinus*. Oyersted (1864) upheld the view that the ovuliferous scales is equivalent to two leaved axis. According to SACHS & EICHLER (1868) bract scale is equivalent to sporophyll. Ovuliferous scale is equivalent to ligule. Basing on this, he has drawn the following similarities are attributed between angiosperm female flower and *Pinus* female cone.

FLORIN (1951, 54) consider that each ovuliferous scale is a modified dwarf shoot. Such special branches are present on the central axis of the female cone. Hence the female cone of *Pinus* is a compound structure, according to Florin. **Morphologically, *Pinus* female cones are equivalent to angiosperm inflorescence. This view was accepted by many.**

4.6.4. OVULE : Each ovule of *Pinus* consists of a central mass of tissue called nucellus. This is covered by an integument. The integument is not united with the nucellus at the top of the ovule. This forms the micropyle of the ovule. The integument extends into a long tube beyond the nucellus , to from micropylar canal (Fig 4.9).

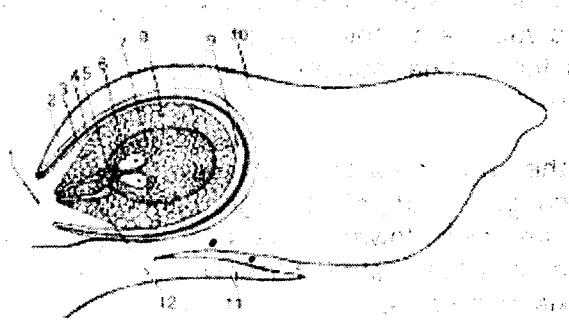


FIG .4.9. *PINUS* OVULE L.S
SHOWING MATURED FEMALE GAMETANGIA

1. **Micropyle**
2. **Outer Sarcotesta**
3. **Middle Sclerotesta**
4. **Inner Sarcotesta**
5. **Female Gametangium Ventre**
6. **Female Gametangium.**
7. **Female Gametophyte**
8. **Nucellus**
9. **Chalazal end**
10. **Ovuliferous Scale**
11. **Bract Scale**
12. **Pollen tube.**

The inner and the outer layers of the integument forms the *sarcotesta* of the ovule (Fig. 4.9). The middle layer forms the *"sclerotesta"* of the ovule. Superficial cells of the nucellus metamorphosed into archesporial initials. These cells divided periclinally to form outer parietal cells and inner megaspore mother cells. The megaspore mother cell undergoes a reduction division and produce 4 haploid megaspores, which are arranged in linear fashion. Among them the upper 3 megaspores degenerate and the fourth functional megaspore produces female gametophyte. In a matured female gametophyte, 1-5 archegonia are present (Fig. 4.9).

4.6.5 POLLINATION : There is enormous production of pollen grains in *Pinus*. Pollination is anemophilous. Pollen grains are light yellow in colour. Pollen grains are microscopic structures. They are 75x40 microns diameter and also possess two lateral wrings, on their distal surface. The floating pollen grains are caught in the gelatinous *"Pollination Drop"* which is formed at the microphyle of the ovule. These pollen grains enter micropylar canal and subsequently drawn in to 'pollen chamber' due to the drying of the pollination drop. After the completion of pollination the ovuliferous scales come nearer to the central axis and the cones are intact. Before this the hypodermis cells grow and close the micropyle.

4.6.6. Male gametophyte development : Initially the division in the pollen grain nucleus results in the formation of a small *"prothallial cell"* and a big cell (Fig .4.10). This big cell divides and produce the second *"prothallial cell"* and *"antheredial cell"*. This antheredial cell divides and produces a *"generative cell"* and a *"tube cell"*. At this four celled stage the pollen grains liberate from the microsporangium.

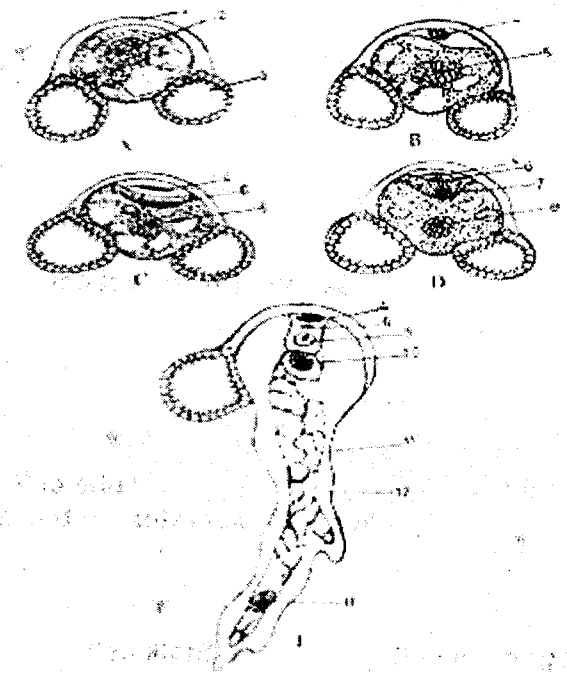


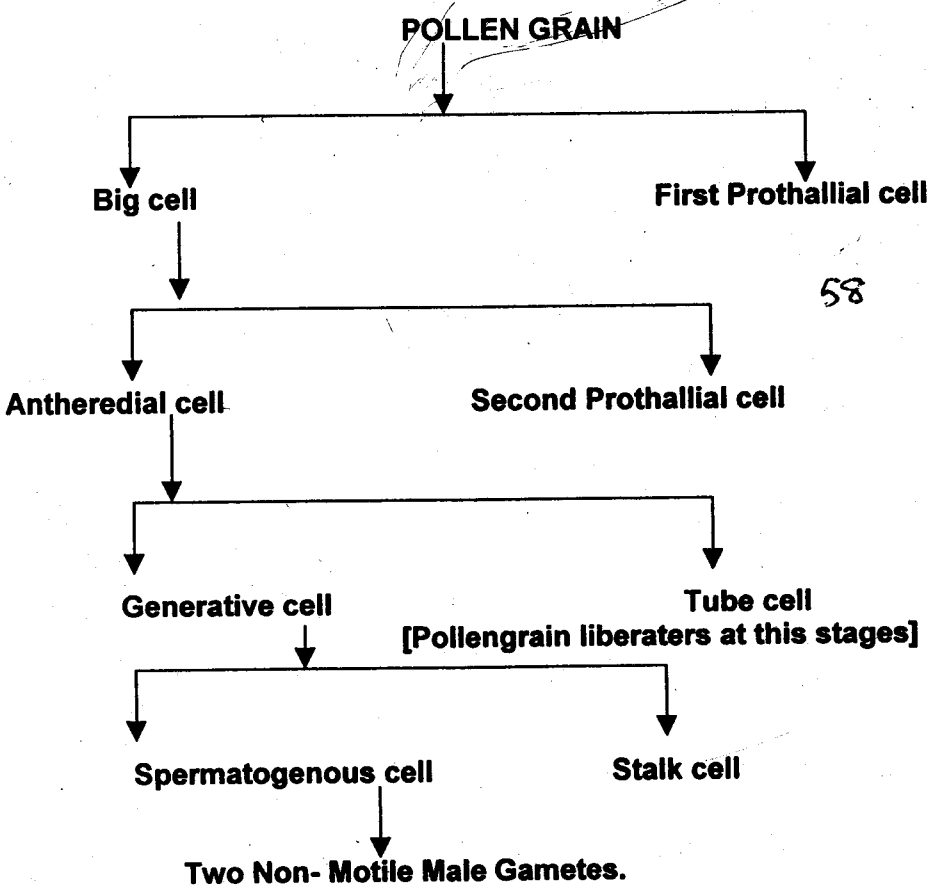
Fig .4.10. *PINUS* Male Gametophyte Stages.

1. **Exine**
2. **Nucleus**
3. **Air Chamber**
4. **Prothallial Cell**
5. **Antheredial Cell**
6. **Second Prothallial Cell**
7. **Generative Cell**
8. **Tube Cell**
9. **Stalk Cell**
10. **Microspore Mother Cell**
11. **Cytoplasm**
12. **Pollentube.**

The pollen grains that are at this stage of male gametophyte development are trapped in the pollination drop at the micropyle of the ovule and due to the drying of the pollination drop, pollen grains reache the pollen chamber and germinate. By this time, the prothallial cells are degenerated. Pollen tube emerges out from the germ pore of the pollen grain, due to the activity of the tube cell. At this stage, pollen tubes growth is retarded temporarily and only after 9 months, the male gametophyte's growth restarts in the pollen grain.

During this, the tube cell first enters in to the pollen tube and later the generative cell. The generative cell divides to produce a *"stalk cell"* and a *"spermatogenous cell"*. The spermatogenous cell divides and produces two *" non-motile male gametes"* . These developments start, one week before fertilization. During the same one week, pollen tube grows and reaches the archegonia traversing the nucellar tissue.

MALE GAMETOPHYTE DEVELOPMENT



4.6.7 Female Gametophyte Development : The functional megaspore, at the chalazal end of the ovule develops in to female gametophyte. Nutrition to this megaspore is from the nucellar tissue of the ovule. Megaspore nucleus undergoes many free nuclear divisions and produces several nuclei. These nuclei are arranged towards the wall of the megaspore. This results in the formation of a big vacuole in the centre of the megaspore. Later walls develop in between the nuclei, to produce *"Cellular female gametophyte"*. This cellular female gametophyte is called as *"Endosperm"*. As this endosperm is formed before fertilization, it is haploid in nature.

The superficial cells in the female gametophyte, which are very nearer to the micropyle act as *"archegonial intials"*. That develop into archegonia. The nucleus in the archegonial initial cell divides and produces a small *"Primary neck cell"* and a big central cell. From the primary neck cell, archegonial neck is formed. There are no neck canal cells in *Pinus*. The division of central cell results in the formation of *"ventral canal cell"* and a large *"egg cell"*. The archegonia are sunken (Fig 4.9).

4.6.8. Fertilization : Before fertilization, pollen tube shows growth and reaches the Egg cell through the archegonial neck. The tip of the pollen tube splits and releases the tube cell and the two non-motile male gametes in to the ventre of the archegonium. One of the male gametes unites with the egg cell (syngamy). With this the fertilization is completed. The remaining cells soon degenerate. There is a gap of one year in between pollination and fertilization.

4.6.9. Embryogenesis : Fertilised egg is known as *"Zygote"*. This is the first cell in the sporophytic stage. Zygote develops and produces the *"Embryo"*. There are two stages in the development of zygote to embryo, viz., Pro-embryo stage and Secondary Embryo.

Pro-embryo stage : (Fig. 4.11 A-F) : Initially free nuclear divisions occur in the zygote. In two divisions 4 nuclei are formed. These four nuclei reaches the bottom of the zygote and by another division produces 8 nuclei. These 8 nuclei are arranged in two tiers each having 4 nuclei. Later wall layers are formed around these nuclei (Fig 4.11). But the cells in the upper tier will have no wall on their upper region. So the nuclei are floating, freely in the cytoplasm of the zygote in the upper tier. Afterwards the 8 cells undergoes another division and produces 16 cells. The embryo which is having 16 cells is known as "*Pro-embryo*" (Fig.4.11F). The 16 cells are arranged in 4 tiers of four cell each. The four tiers of cells in the pro-embryo are named serially from above as. They are Embryonal tier (Fig.4.11G) and Suspensor tier Rosette tier Open tier.

The four cells in the open tier, had no role to play in the embryo formation. The suspensor tier cells, elongate and produce primary and secondary suspensors and push the embryo cells in to the endosperm, to draw nourishment to the developing embryo. Pro-embryo in *Pinus* is symmetrical.

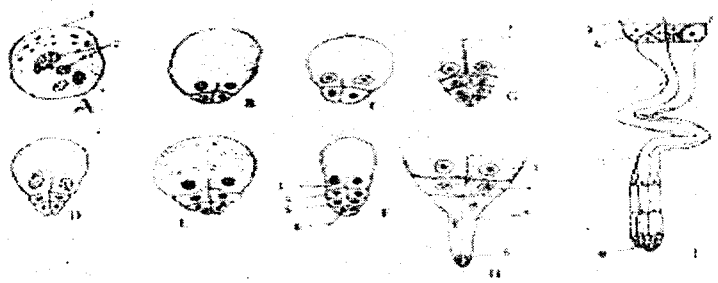


Fig: 4.11: Embryogenesis in ***PINUS***
A-F Pro-embryo Stage and G-I Secondary embryo stage.

1. **Zygote Wall**
2. **Free Nuclei in Zygote.**
3. **Open tier.**
4. **Rosette tier**
5. **Suspensor tier**
6. **Embryonal tier**
7. **Secondary suspensor**
8. **Embryo**

SECONDARY EMBRYO STAGE.

In the embryonal tier, several divisions occur and a multi cellular embryo is formed. At the same time, the suspensor tier cell divide and produce secondary suspensors (Fig.4.11 G-I). These secondary suspensor cells which are intact up to this stage dissociate and along with them the embryo cells are also dissociate to produce four separate embryos. These embryos are formed because of separation of suspensor cells. Hence this is known as *"cleavage polyembryony"*.

There are 3-4 archegonia in the each ovule. The egg cells in different archegonia may get fertilized and produce four embryos (due to fertilization in all the four archegonia). This is *"simple polyembryony"*. Some times the cells in the rosette tier may divide and produce more embryos. This is known as *"rosette polyembryony"*. However only one embryo matures and all others degenerate. In a fully grown mature embryo a central axis is present upon which slender, yellow coloured, 10 cotyledons are arranged in a ring. On this axis *"primary root"* is towards micropyle, below the cotyledons. *"hypocotyl"*, is adjacent to a small epicotyl are present. Thread like suspensor cells are arranged in a bundle and attached at the primary root tip.

4.6.10 Seed : After fertilization seeds are developed from ovules. Seed coat is formed from integument. The outer soft sarcotesta disappears and the middle sclerotesta will become *"testa"*. The inner sarcotesta developing into *"tegmen"*. (Fig.4.12-B). The left over endosperm at the micropyle will form a thin layer of *"Perisperm"*. In the seed there are 3-10 cotyledons are present. So seed is *"Polycotyledonous"*. Seed has a single wing in *Pinus* (Fig .4.12-A).

Seed dispersal : Ovuliferous scales in the female cone, wide open from central axis during the seed dispersal. This opening also releases a breakage sound. At this stage, winged seeds are released from cone and travel to long distances along with air.

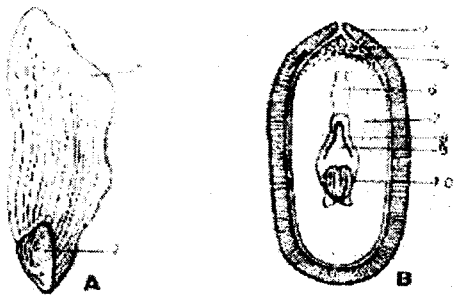


Fig .4.12 *PINUS* seed.

A. Winged Seed.
B. Longitudinal section of seed

1. Wing	**6. Suspensor**
2. Seed	**7. Nucellus**
3. Seed coat	**8. Primary radicle.**
4. Inner seed coat	**9. Hypocotyl**
5. Perisperm	**10. Cotyledons.**

Seed Germination : The germination of the seed is *"epigeal"*. In the germinating seed, the seed coat breaks open and the primary radicle emerges out and grown into the soil, in the form of primary root. Then epicotyl comes out of the seed and also brings out 3-10 green cotyledons along with it. Later from epicotyl region vegetative parts are formed. This is known as *"young sporophyte"* (Fig 4.13). This gradually develops in to *"adult sporophyte"*. The life history of *Pinus* plant is depicted in (Fig.4.14).

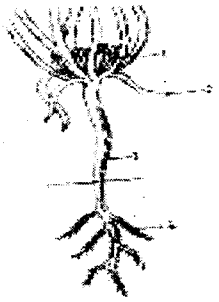


Fig : 4.13 *PINUS* Young Sporophyte

1. Primary Leaves. 2. Cotyledon 3. Hypocotyl 4. Primary Root.

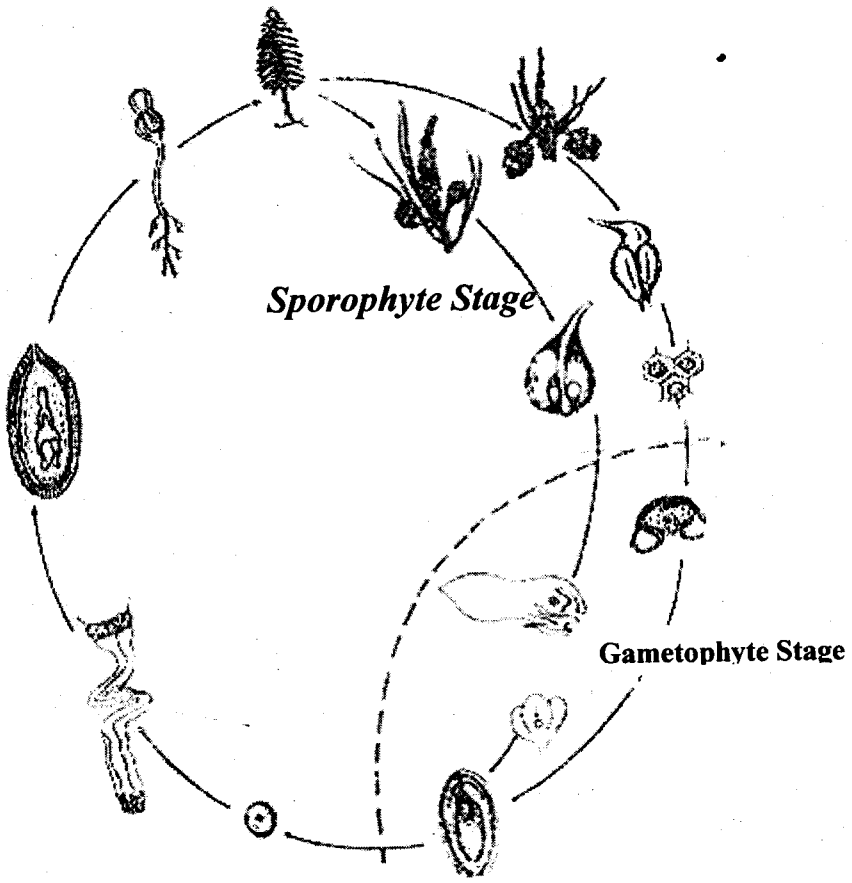


Fig .4.14. Life – Cycle in *Pinus*

4.7. ECONOMIC IMPORTANCE OF PINUS :

The stems of *Pinus* yield quality wood. Some Pinus species produce wood pulp. *Pinus sylvestris, Pinus wallacliana, Pinus roxburghii and Pinus insularis* yield economically important resin and turpentine. They are used in paint industries, varnish industries & medicine industries. Seeds of *Pinus gerardiana* are edible and known as *"Chilgoza"* in India. The xylem of young plant parts of *Pinus mercucii* is used in the preparation of craft paper. The long trunks of *Pinus* plants are also used as telegraph poles.

4.8 SUMMARY

Pinus is widely distributed genus, in the northern globe. The branches in *Pinus* are of two types.(Long shoots and Dwarf shoots) and also leaves (brownish scale leaves and green needle–like leaves). Scale leaves are present on both the branches. But needle-like green leaves are present only on dwarf shoots. The internal anatomy of *Pinus* stem is similar to dicotyledonous stem. The wood is picnoxylic with resin ducts in stem. The of needle–like green leaves mesophyll contain, angular chlorenchymatous mcsophyll cells and their inner wall

produces peg-like invaginations in to the cell lumen. This increases the photosynthetic occurring surface area in the leaf.

In the *Pinus* leaf there is transfusion tissue with albuminous cells and tracheidal cells and it is useful in lateral conduction. Anatomically the root is similar to dicotyledonous root. Ectotrophic mycorrhizae is present on the roots of *Pinus*. This helps in the absorption of nutrients from the soil. *Pinus* is a monoecious plant. Both the male and female cones are present on the same plant, on different branches. Male cones are small and produced in the axils of the scale leaves Microsporophylls are spirally arranged on the cone axis. Two pollen sachs with numerous winged pollen grains are present on the adaxial surface of the microsporophyll.

Female cones are bigger in size and produced in the place of long shoots. Each female cone consists of many spirally arranged megasporophylls subtending bract scales. Thin, dried bract scale on the central axis of the cone and a triangular ovuliferous scale with two ovules at bottom, in the axil of bract scale is present. Archegonia (2-3) are formed on the female gametophyte of the ovule.

The pollen grains reache the pollen chamber of the ovule where they germinate and release sends the non- motile male gametes (haploid) nearer to egg. The egg and male gamete unite (syngamy) and produces a zygote. The zygote (diploid) is the result of fertilization and it is the first cell in the sporophytic plant. It zygote undergoes divisions and produces a 16 celled pro-embryo. The lower tier in the pro-embryo divides and produces the embryo. From the pro-embryo, mature embryo is formed. The embryo is polycotyledonous with 3-10 green cotyledons. Seed dispersal is due to air and the germination is epigeal.

4.9 TECHNICAL TERMS

- **Annual ring** : Spring wood and autumn wood unitedly called annual ring .
- **Abietinian pits** : If bordered pits are in two rows on the radial walls of tracheids, they are placed opposite to each other. Such pits are called abietinian pits.
- **Araucarian pits** : The multiseriate bordered pits on the radial wall of tracheids are placed alternately.
- **Crassulae** : On the inner side of the tracheidal wall there are rod like thickenings. They are transversely arranged in between the bordered pits. These are called crassulae.
- **Piliferous layer** : Epidermis on the root with root hairs and with out cuticle.
- **Transfusion tissue** : Albuminous cells and tracheidal cells are present in pericycle of *Pinus* leaf responsible for lateral transport
- **Adaxial surface** : The plane of a leaf which is nearer to the axis.

- **Abaxial surface** : The plane of the leaf which is away from the axis.
- **Anemophily** : The pollination taking place with the help of air is anemophily
- **Syngamy** : The union of male and female gamete(egg) nuclei.
- **Pro-embryo** : Embryo with 16 cells is pro-embryo.
- **Polyembryony** : Presence of more than one embryo in the seed.
- **Normal polyembryony** : More than one egg fertilized to produce, more than one embryo will be
- **Cleavage polyembryony** : Polyembryony is due to the separation of suspensor cells along with embryo
- **Polycoty ledonous** : Presence of more than two cotyledons in the embryo.

4.10 MODEL QUESTIONS

I. **Answer each question in 30 lines.**

1. Describe the vegetative characters of *Pinus* and add a note on the distribution of *Pinus* in India
2. Write the anatomy of the needle–like green leaf with a neat labeled diagram and mention the xerophytic characters present in that
3. Write the anatomy of the young *Pinus* stem with a neat labeled diagram
4. Describe the structure of *Pinus* male cone and microsporophyll
5. Give an account of Write the structure of *Pinus* female cone and the megasporophyll
6. Describe the embryo development in *Pinus*

II **Answer each question in 10 lines.**

1. Economic importance of *Pinus*
2. Ovuliferous scale in *Pinus*.
3. Seed development and dispersal in *Pinus*.
4. The structure of ovule in *Pinus*.
5. Dwarf shoot in *Pinus*.
6. Male gametophyte development in *Pinus*.

4.11 SUGGESTED READINGS :

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- Dr. P.V.RAMA RAO

Lesson - 5

GNETUM

OBJECTIVES

By the end of this lesson you will be able

* describe the external feature of the sporophyte of *Gnetum*.
* draw the internal structure and label the parts of the primary, secondary stem and anomalous secondary growth in stem and also the leaf and root.
* distinguish between the structure of the male and female strobilus off *Gnetum*.
* describe different interpretations of different scientists on the three envelopes of *Gnetum* ovule
* differentiate the developmental stages of male gametophyte from a female gametophyte.
* describe the pollination mechanism and fertilization process in *Gnetum*.
* describe the development of embryo & endosperm of *Gnetum* and
* list of the various uses of *Gnetum* plant.

STRUCTURE

UNIT - III
LESSON-11

PLANTS AND ENVIRONMENT

11.1 OBJECTIVE:

It is aimed to learn about the relation between plants and environment which is known as ecology. Ecology is considered important for its multifarious application for human welfare. It is helpful for understanding of our surroundings and how they interact with each other in an ecosystem.

11.2 STRUCTURE

11.2.1 INTRODUCTION

The various environmental factors affect the plant and animal life and inturn they change the environment. The field of ecology is concerned with the relationship between the living organisms and the environment in which they live. The word "ecology" is coined by a German ecologist Reitre in 1885. This term was derived from two greek words. " Oikos" means house and " logos" means study of , and the term ecology means study of living organisms in their natural habitats or homes. Many ecologists defined ecology in their own terms. Odum (1963) defined ecology as the study of structure and function of nature. According to J.Kreb (1972),

ecology is the scientific study of the interaction that determines the abundance and distribution of organisms.

Ecology has many sub divisions like autecology, synecology, habitat ecology, population ecology, community ecology, ecosystem ecology, etc. The basic concepts of ecology are two. 1. structural concepts which include materials, living organisms, energy, etc. and 2. functional concepts which include metabolic and catabolic reactions, flow of energy, material cycling, etc. The earth along living organisms and environment is called BIOSPHERE. This include all things which react upon with each other resulting into continuous changes. These changes are in an orderly manner assuring the survival and gradual evolution of the organisms. The different components of biosphere are -1.Lithosphere, 2.Hydrosphere and 3.Atmosphere. They can be well understood by studying the atmosphere, water, light, temperature, soil and biota.

11.2.2 ATMOSPHERE:

This is the outermost gaseous composition of biosphere surrounding the earth. The atmosphere influences the climate and life on the earth. Sutcliffe(1966) divided atmosphere into four zones. Smith (1974) divided atmosphere into five zones.

1. **Troposphere:** This is the immediate layer above the earth's crust and is about 8 km at poles and 20 km at the equator. This is referred as air and it contains about 78% Nitrogen, 20% Oxygen, 0.03% Carbondioxide, 0.93%Argon and traces of Helium, Hydrogen, Sulphurdioxide, Ammonia, Methane, water vapour, etc. Cloud formation, lightning, thundering, storms take place in this zone. Air temperature gradually decreases with height at the rate of 6.4 degree celcies / km. The upper limit of this zone gradually merges in the next zone and is called tropopause.

2. **Stratosphere:** This is about 30 km in the thickness. The gases present in stratosphere and troposphere are almost similar except in the presence of Ozone. The temperature increases upto 90 celcies due to ozone formation under the influence of UV rays of sun light. This ozone forms ozonosphere and it absorbs UV and Infrared rays of sun and saves the life on earth acting as a blanket.

3. **Mesosphere:** The third layer from earth is mesosphere and is about 40-80 km thick. It contains little gaseous mass, no water vapours and temperatures are nearly -80Oc.

4. **Heterosphere:** This is extending from 80-300 km from the earth's surface. The gases are in atomic state. The lower portion is called thermosphere containing nitrogen and oxygen in equal quantities. Upper portion contains hydrogen and helium. Between mesosphere and thermosphere the layer is called ionosphere.

5. **Exosphere:** It exists beyond 300km and extends up to 40,000km from the earth's surface. Here hydrogen is the major element. The temperatures are from 200^0F.-

10,000^{0}F. The lower layer of exosphere contains a layer of helium. This helium checks the entry of UV rays of sunlight to the earth's surface. See Fig 1.

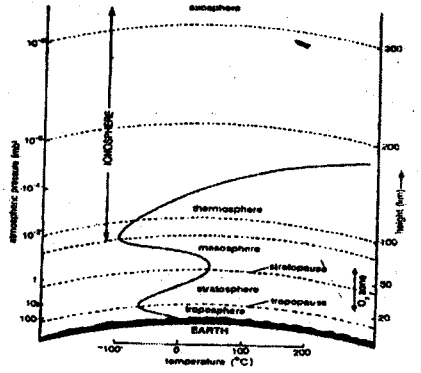


Fig. 1. Atmosphere showing zonal differentiation

11.2.3 HYDROSPHERE:

All the water of this earth is considered as hydrosphere. Water is very important factor for living organisms. Protoplasm, the very basis of life is made of mostly water. Along with other factors water determines and regulates the structure and arrangements of plant communities. Nearly 70% of the earth's surface area is covered with water, in the form of seas, lakes, rivers, etc. Water is also present below the earth's surface as ground water or water table. Water is also found in the form of ice on south and north poles and on the tops of high mountains.

In nature water is found as a) atmospheric moisture, b) precipitation and c) soil moisture.

a) ATMOSPHERIC MOISTURE: It is of two forms-1) invisible vapour form and 2) visible cloud or fog form. The vapour present in air is called atmospheric humidity and it affects the rate of transpiration. The total amount of invisible water vapour actually present in the atmosphere at a particular temperature is called absolute humidity. The ratio between **absolute humidity** of the air to the amount of vapour actually required to saturate it at the same temperature is called **relative humidity (RH).** Humidity is measured by **psychrometer** and continuous record by **hygrometer**. Low atmospheric humidity increases water loss through transpiration and thus affects growth. The roots of most land plants do not grow into the soils devoid of water. Humidity in the atmosphere forms the source of water for dessicated masses, lichens and tropical archids, epiphytes, etc. For microbes and fungi moisture plays an important role in the germination of spores and subsequent stages in life cycle.

b) PRECIPITATION: This is the chief source of water for life on the earth. This is available to the plants and animals by rainfall. Precipitation results as a consequence of cooling and condensation of water vapour at high altitudes. The low temperatures at high altitudes cool the air and it loses its vapour holding capacity and gets saturated. Further decrease in temperature

result water vapours into liquid form and are pulled down by gravity. Precipitation occurs as drizzles, rain, snow, dew, frost, hail, storm sleet, etc. Epiphytes and lithophytes depend mainly on atmospheric water vapour humidity and dew.

The amount and distribution of rain fall influence the vegetation in a region. Eg. Forests, deserts, etc. Heavy rainfall throughout the year results into evergreen forests. If rainfall is confined to a few months only. It results in the deciduous forests. The regions with scanty rainfall change into deserts. Rains in India are caused by monsoons.

The monsoon seasons are :

1) The seasons of north west monsoon which are:
   i) cool season from mid December to February. and
   ii) hot dry season from March to mid June.

2) the seasons of the south east monsoon are –
   iii) wet season from mid June to mid September. and
   iv) season of retreating monsoon from mid September to mid December.

On the basis of annual rainfall, the country is divided into 4 climatic zones.

1. Wet zone with very heavy rainfall of more than 200cm.- Western Ghats, Meghalaya, Bengal, part of Uttar Pradesh.
2. Intermediate zone with heavy rain fall of 100-200cm. - West Bengal, Bihar, Eastern Madhya Pradesh Andhra Pradesh part of Western Ghats, Eastern Tamilnadu.
3. Dry zone with moderate rain fall of 50-100cm.- Punjab, UP, Delhi, Maharashtra , Karnataka, etc.
4. Arid zone with scanty rainfall of below 50cm. - Part of Punjab, Rajasthan, Gujart, etc.

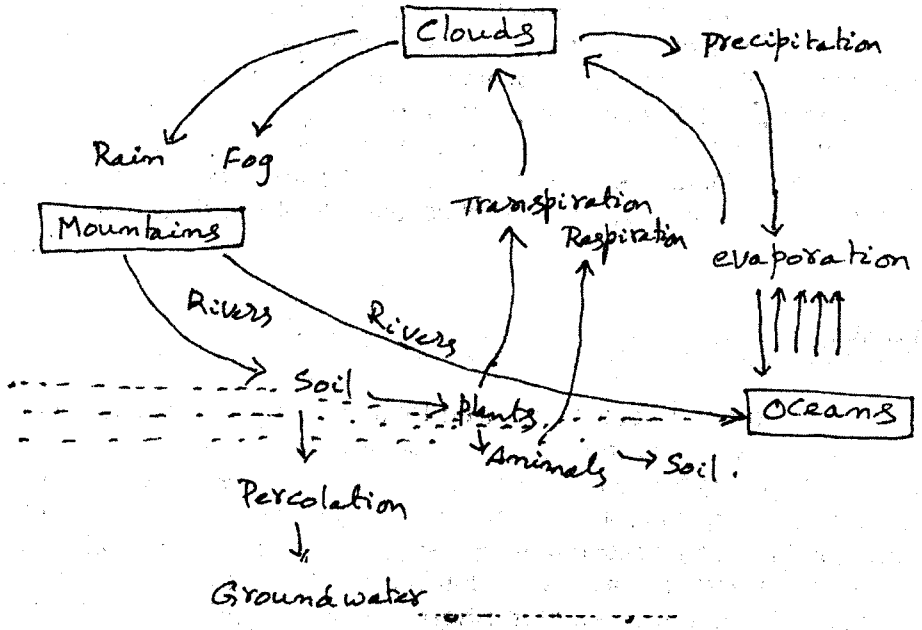
c) **SOIL MOISTURE:** This is the terrestrial water. Soil water is classified into four types.

i) Hygroscopic water- present as a thin film around soil particles.
ii) Capillary water- present in the pores of soil, held against gravitational pull.
iii) Gravitational water-present at lower areas of soil by gravitational force.
iv) Combined water- present in the form of chemical bonds.

Depending on the requirements and availability of water in the habitats the plants are classified as different communities. They are 1. Hydrophytes- the plants grow in abundant water areas.2.Xerophytes- plants adapted to grow in scanty water conditions, and 3. Mesophytes- plants grow in moderate or sufficient supply of water conditions.

WATER CYCLE : As water forms most essential inorganic nutrient for living organisms it is necessary to have the knowledge of water cycle. A continuous cyclic flow of water is maintained through precipitation, evaporation and transpiration, etc. The cycle is referred as hydrological cycle. The water is continuously evaporated by the heat of the sun and reaches the atmosphere. These water vapors condense at higher altitudes of the atmosphere and form clouds. These clouds are moved by winds and pass over land where further cooling takes place and precipitate to form rain, dew, frost, snow, etc. Water thus reached earth's surface in this way runs into streams and rivers and ultimately return to the ocean. During rainfall some of water percolates into the deeper layers of the soil. The water in the soil is utilized by the plants and is returned to the atmosphere through transpiration. Animals drink water and part of it is returned to the atmosphere through evaporation and excretion. Thus there is always a continuous circulation of water from earth to atmosphere and vice-versa is maintained in nature. This is called hydrological cycle or water cycle.

It is estimated that 80,000 cubic miles of water from the oceans and 15000 cubic miles from lakes and land surface evaporate annually. About 24,000 cubic miles of this water fall on land in the form of precipitation. The distribution of rain fall through the year is one of the leading features of climate. It is closely related with the distribution of great vegetational zones in the world. See Fig. 2.



11.2.4 LIGHT

Light is the most important factor in determining the growth of plants and the development of vegetation. The sun is the only source of light energy for our planet earth. Light is reaching earth in the form of electromagnetic waves which range from 2900A to 50,000A in their wavelengths (290 $m\mu$ to 5000 $m\mu$ or nanometers). (10 A^{o} =1 $m\mu$ = 1nm= 10^{-9} meters). The visible light has the wavelengths from 390 750mu. When it passes through a prism it exhibits

VIBGYOR with seven different colours. The red and blue lights are used by the plants (700-626 $m\mu$ red colour and 490-435 $m\mu$-blue colour). The light with wavelength less than 390mu is ultraviolet and more than 700mu is infrared. Light is measured in terms of candle power. The amount of light received from a standard candle at one foot distance is called foot candle and at one meter distance is called meter candle or lux. The intensity of light is measured by photometer or lux meter. Secchidisc is used to measure the distance in water up to which the light penetrates, which indicates the turbidity of water. The light intensity is varying from place to place. It is influenced by few factors like a) atmosphere,b) suspended particles, c) water vapor, d) vegetation and e) topographic conditions.

Atmospheric gases like Nitrogen and Oxygen absorb and disperse light of shorter wavelengths. The intensity of light is greater in dry areas. Solid particles, gases, vapours in atmosphere have screening effect. The intensity of light is reduced in water medium. Submerged plants receive little light than the floating forms. The foliose of trees also affects the light intensity. The tallest plants receive more light than the underlying shrubs and herbs. Diurnal variations also are common. The light intensity is maximum at midday time and minimum in mornings and evenings. In summer intensity is very high and in rainy and winter seasons it is low. At equatorial line the intensity of light is more and prevails for 12 hrs. Light directly or indirectly affects the plants and their growth in many ways. Light is essential for photosynthesis. Chlorophyll and other pigments use light energy in preparing food molecules during photosynthesis. Light also effects the formation of chlorophylls and pigments and also their arrangement, number, distribution, etc. It has effect on the leaf structure and function. It also affects the rate of transpiration, stomatal movement, and respiration. Based on the light receiving period by the plants(photo period) the plants are called long day plants, short day plants and day neutral plants. Light effects the germination of seeds, plant movement etc. On the basis of light requirement plants are classified into 1) **heliophytes** or **photophilous,** 2) **sciophytes** or photophobic. Again they are classified as obligate and facultative types in each.

11.2.5 TEMPERATURE:

The measurement of degree of hotness is called temperature. The radiant solar energy is converted in to heat energy and it is measured in calories. Temperature is measured by thermometers. Thermograph records the temperature on graph and the recorded graph is called thermogram. Temperature varies indifferent environments. Temperatures of ponds are usually ranging from 0^0C to 36^0C and temperatures in deserts vary -70^0C to 80^0C. The land plants live in a wide range of temperatures and are called **eurythermic** plants. The aquatic plants grow in a narrow range of temperatures and are called **stenothermic** plants.

EFFECTS OF TEMPERATURE ON VEGETATION :

Temperature plays a significant role in determining the distribution of plants on this planet. At equator temperature is maximum and it gradually decreases as one moves to the poles. This variation brings about the variation in vegetation both in structure and composition.

The ever green forests are located at the equator and tropical regions, where the temperatures are high. The coniferous forests are in the temperate region where the temperatures are low. Very few and small plants are seen at the polar regions where temperatures are extremely low. Such type of variation is also prevailing with the changes in altitudes too. The lower regions are rich with trees and gradually when one moves to the top of the mountain the vegetation is changing to less and small with sparse distribution. On the basis of vegetation, distinct zones like tropical, subtropical, temperate and alpine are recognized.

HEAT TOLERING CAPACITY OF PLANTS:

Depending upon the capacity of heat tolerance, the plants are categorized into-

1. **Megatherms:** Plants tolerating high temperatures throughout the year. Found in tropical rain forests.

2. **Mesotherms:** Plants tolerating high temperatures during summer and lower temperatures during winter. Found in temperate deciduous forests.

3. **Microthrms:** Plants tolerating lower temperatures throughout the year. Found at the higher altitudes.

4. **Hekistotherms:** Plants that tolerate extreme low temperatures throughout their life. Found in alpine vegetation and polar regions.

EFFECT OF TEMPERATURE ON PLANTS:

The effects of temperature on plants are significant in distribution of the plants. Some of them are given here.

1) Effect on metabolism: The metabolic reactions are carried out by certain enzymes which are influenced by temperature. Most of the reactions are carried out between 20^0C-30^0C. The reactions decrease below 10^0 C and above 30^0 C because of the inactivity of the enzymes.

2) Effect on seed germination: The suitable temperatures for germination of seeds varies from 20^0C-27^0C for most of the plants. The dormancy of seeds is broken by exposing them to low temperatures and it is called stratification.

3) Absorption of water is also influenced by temperature. It is reduced at low temperatures as viscosity of water increases.

4) Mineral absorption: The absorption of essential minerals is influenced by temperatures, as it requires energy which ultimately depends up on the rate of the metabolic activities like photosynthesis, respiration, transpiration, etc.

5) Transpiration: Transpiration increases with increase in temperatures.

6) Respiration: Variations in the temperatures alter the enzyme activity during respiration. Generally the rate of respiration increases with the increase in temperature up to the optimum limit.

7) Vernalisation: Induction of flowering by cold treatment is called vernalization. Many cereals bianuals and perennial trees require certain period of lower temperature to flower.

THRMOPERIODISM: The plants are adjusted to the diurnal changes in the temperature for leafing, flowering, fruiting and abscission of leaves in plants etc. The regulation of these phonological events to periodic thermal changes is called thermoperiodism.

INJURIES DUE TO EXTREME TEMPERATURES:

Extreme low or high temperatures injure plants. To overcome such odd conditions plants develop adaptations. To overcome injuries by high temperatures plants show some symptoms like leaf chlorosis, stem leisions, leaf scorch, thin lamina rapid rate of transpiration, vertical orientation of leaf, shining surfaces with wax, etc., thick cuticle, corky layers, sclerenchyma, dry seeds and spores with low water content, etc. To over come injuries by lower temperatures plants become dormant with lower biological activity. See. Fig. 3

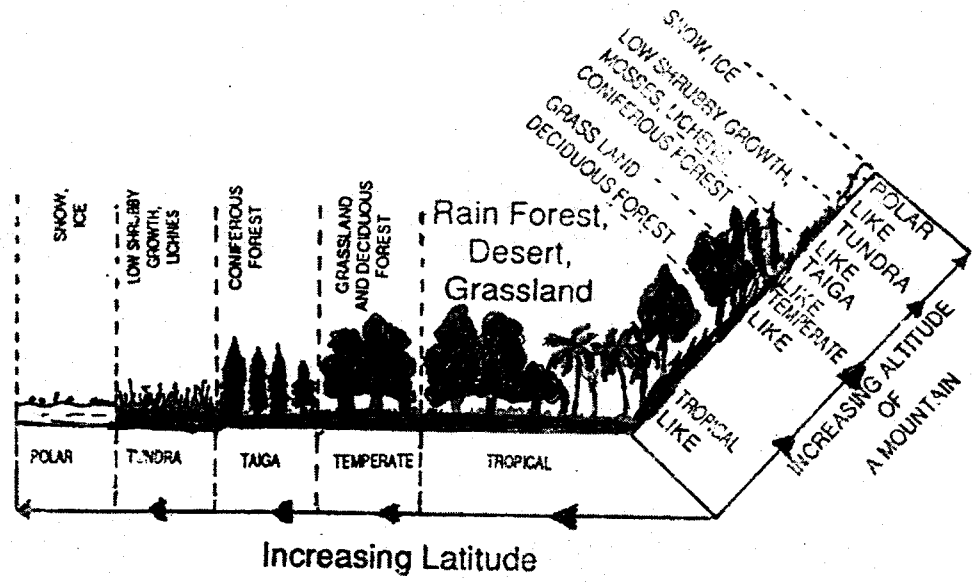


Fig. 3. The sequences of zones of vegetation on latitudes and altitudes

11.2.6 SOIL:

Soil forms the basis for living organisms and is called lithosphere. It is considered as an important ecological factor as the living organisms depend on soil for the nutrients, water supply and to live on. The various components of soil interact and relate with each other and form soil complex. Soil is defined as the weathered rock of the earth's crust mixed with organic materials, nutrients, organisms, gases, etc. Soil has mineral matter (40%), organic matter(10%), soil solution(25%), and air(25%).

DEVELOPMENT (FORMATION) OF SOIL:

Rock from which soil is formed is called parent rock or parent material. The process of breaking up of the parent material into smaller particles is called weathering. Weathering may be due to physical, chemical or biological agents. The rocks break into mineral particles like sand, silt, clay, etc.

i) **Physical weathering:** Action of water, temperature effects, gravitational force, movement of glaciers, wind, etc. cause physical weathering. These phenomena result into the breaking of the rocks by cooling, drying, moving, pulling, etc.

ii) **Chemical weathering:** Hydration, hydrolysis, reduction, oxidation, carbonation, chelation, etc. cause chemical weathering of rocks.

iii) **Biological weathering:** Micro organisms, lichens, roots of higher plants, etc. cause biological weathering. These living organisms are able to bring changes in rock constitution by secreting some acids, extracting nutrients, widening and separating crevices, absorbing and retaining water, accumulating organic material as humus, etc.

SOIL PROFILE:

The vertical section of soil showing many layers super imposed one on the other, is called soil profile. A typical soil profile shows the following layers.

HORIZON A:

This is the top most layer of the soil that bears the vegetation and is also called top soil. It appears dark in colour and rich with organic matter and is biologically active. This layer is further divided into five layers. They are in this order-

Aoo: This is the top most layer consisting of many things like fallen leaves, litter, twigs, etc.

Ao : This layer consists partly decomposed organic matter. The decomposing organs lose their identity.

A1: This layer is rich with mineral matter and high quantities of organic matter.

A2: This layer contains lesser nutrients and light coloured one.

A3: This is the transitional zone between horizon A and horizon B. But it is more like horizon A.

HORIZON B: This layer is made of light brown particles which are fine, dense and compactly arranged. This is having more clay and serves as reservoir of water. This layer shows maximum accumulation of silicate clay, mineral, and organic matter.

HORIZON C: It is having large rocks which are incompletely weathered.

HORIZON D: This is composed of unweathered parent rocks and is also called regolith region and is without biological activity. Soil profile can be well understood by Fig. 4.

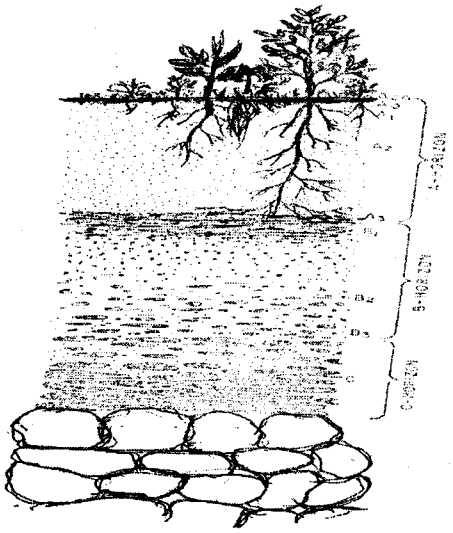


Fig. 4. Soil profile or a section of soil various zones

PHYSICO-CHEMICAL PROPERTIES OF SOIL

All the physico- chemical properties of soil can be under various heads and some of them are given here.

1. **Texture (mineral particles):** Texture of the soil plays an important role in determining the vegetation of any place. Soil texture is determined by the size and relative proportion of the mineral particles of the soil. The criterion for soil particle size is given here.

   Stones------10 mm-100 mm
   Coarse gravel------10 mm-5 mm
   Fine gravel------5 mm-2 mm
   Coarse sand------2 mm-0.2 mm
   fine sand------0.2-0.02 mm
   Silt------0.02 mm-0.002 mm
   Clay------less than 0.002 mm.

   Silt and clay help in holding water where as other types give physical support to the plants.

2. **Soil organic matter (humus):** It gives fertility to the soil and increases water holding capacity. Organic matter contains two components – 1. Minor constituents and 2. Major constituents. The minor constituent include carbohydrates, lignin compounds, fats, protiens, aminoacids, alkanes, etc. The major constituents include resins, celluloses,

lignins, etc. If these components under go decomposition they produce dark amorphous substance and it is called humus. This process is called humification and is brought out by soil organisms.

3. **Soil water:** The source of soil water is rain fall. Water is required for metabolic activities and it is the medium for absorption of nutrients.The process of dew composition of organic matter depends upon the availability of water. Part of rain water is lost as runaway and part of rain water is retained by soil, in different forms like gravitational water which is percolated and forms water table (ground water), hygroscopic water present on the surface of the soil particles, capillary water held between the soil particles by capillary force and combined water which is found as hydrated oxides of aluminium, iron, silicon, etc. The total amount of water present in the soil is called hollard. Of this the amount of water absorbed by plants is called **cressard** and remaining amount not used by plants is considered as **echard**.

4. **Soil solution:** The organic and inorganic materials present in the soil in the form of solution. Carbonates, sulphates, nitrates, chlorides, salts of Mg, Na, K, etc. are found in dissolved state. The soils rich with nutrients are called **eutrophic** and with less nutrients are called **oligotrophic** soils.

5. **Soil acidity(pH):** This is expression in pH values Many plants live well in slightly acidic soils. Where the soil pH values are 4.4-5.8. Slight alkalinity is favourable for grasses and legumes.

6. **Soil atmosphere:** In between the soil particles the space is filled with air. This air is consisting mostly CO_2 released by soil microorganisms. Oxygen of soil air also important for the microbial activity during humification and respiration of soil organisms like earthworms and other soil fauna.

7. **Soil temperature:** At 20^0-30^0C the metabolic activities including water absorption are optimum. Soil is heated by sunrays. It effects the growth of the roots of higher plants and microbial activity. At low temperatures the viscosity of water is higher and plants cannot absorb and utilize this water. Plants growing in high temperatures, show stunted and rosette structure.

8. **Soil organisms:** Many types of organisms live inside the soil. They include bacteria, protozoa, nematodes, insects, annelids, earthworms, molluscs, higher plants and vertebrates. Some activities of soil organisms include decomposition of organic matter, nitrozen fixation, soil aeration, release of hormones, toxic substances, etc.

11.2.7 BIOTA (BIOTIC FACTORS) :

Biota includes all the organisms that live in the ecosystem. They depend on and influence each other and bring changes in the ecosystem. This dependence and all these influences and resulting changes as a whole is considered as biological interrelationship. According to McDugal (1918) the interrelationship is of two types-1. Disjunctive symbiosis (associated organisms are not in constant contact). This is again considered as social and nutritive types.2. Conjunctive symbiosis (associated throughout their life time). This is also again considered as social and nutritive types. Odum (1971) considered the relationships among living organisms as positive interaction and negative interaction. We considered these biotic relationships as 1.effect of plants, 2. effect of animals and 3. effect of man.

1. **Effect of plants:** There are many relationships- 1. Mutualism-Both the associated organisms get benefit. Eg. Lichens, symbiotic nitrogen fixing organisms, mycorrhizal association, etc. 2. Commensalism- In this relationship only one organism gets benefit. Eg. Epiphytes, lianes,etc. 3. Amensalism (allelopathy)- One organism do not allow the other one to grow nearby. It secretes some antibiotics, or toxic substances. Eg. Bacteria producing antibiotics. 4. Saprophytism- It involves the digestion of dead organic matter by heterotrophes which are called natural scavengers. They bring recycling of materials. 5.- Parasitism- One organism derives nutrients from the other with the help of haustoria. Eg. Fungi, bacteria, etc. 6. Predation- In this relationship one organism feeds the other organism. Eg. Insectivorous plants. 7. Competition- There will be always competition for sunlight, nutrients, etc. between the associated plants. The plants may be of same kind (intraspecific) or of different kinds(interspecific) eg. Weeds, crop plants.

2. **Effect of animals:** Animals and plants cannot live without influencing eachother. Some of the effects are given here. 1. Grazing and brousing- Animals depend on plants for their food directly or indirectly in food chain. Grazing refers eating herbs completely by the animals. Brousing refers eating the leaves of shrubs and trees. 2. Myrmecophily- Some plants provide shelter to ants. They also secret some food substances for the ants. The ants act as body guards of the plants against any disturbing agents like other animals. 3. Zoophily- Insects, birds, bats, squirrels, etc. help plants in pollination for the production of seed. These animals take the edible pollen and nectar. In this process the pollengrains get transferred from one flower to the other as these animals visit many flowers. 4. Zoochory- Some animals like monkeys, bats, birds, etc. help the plants in disseminating the seeds, fruits, spores, etc. They help the plants in migration and dispersal.

3. **Effect of man:** Man is the most important biotic factor regulating other living organisms in many ways. His actions have changed the face of the earth. Indiscriminate felling of trees, use of natural resources, killing of animals, etc. resulted into depletion of resources and extinction of many plants and animals. Over population, destruction of forests, construction activities, increase in cultivable land, forest fires, etc. all lead to the deterioration of ecosystem including plants and animals, and also environment at various levels.

11.3 SUMMARY

Different environmental factors interact with each other and influence plant and animal life on this earth. Study of living organisms in their natural habitats is called ecology. Planet earth with its living organisms and environment together is called Biosphere. All these interact with each other and lead to the development of organisms as well as ecosystem. This leads to evolution too. There are basically three components of biosphere. They are atmosphere, hydrosphere and lithosphere.

Atmosphere is the outermost gaseous composition from the earth's crust. It is divided into troposphere, stratosphere, mesosphere, heterosphere and exosphere.The ozone layer is present between troposphere and stratosphere absorbing UV light which can damage life on this earth. The water content of this earth is called hydrosphere. This water is present in gaseous vapour, liquid water and solid ice. Water covers almost 70% of the earth's surface and it is continuously moving from one place to the other. Water evaporates from the seas and form clouds, after reaching the heights it cools and precipitates and reaches earth in the form of rain. Part of rain percolates into the ground and part runs off as streams, rivers, etc. and reaches sea. The percolated water is used by plants and reaches atmosphere by transpiration. Animals use part of water and it reaches earth by excretion. On the basis of rain fall our country is divided into wet zone, intermediate zone, dry zone and arid zone. On the basis of availability of water the plants are classified into hydrophytes, mesophytes and xerophytes.

Light and temperature are important factors in determining the development of plants and vegetation. They effect the structure and function of plants including metabolic activities. Soil forms the basis for the organisms, to live on. It is formed by weathering of rocks by physical, biological and chemical means. Soil profile and soil composition effect and influence the plant's growth and life cycle in various ways. All the relationships as effects of plants, effects of animals and effects of man are studied.

11.4 TECHNICAL TERMS: Ecology, ecosystem, atmosphere, hydrosphere, lithosphere, biota.

11.5 MODEL QUESTIONS:

Essay type questions:

1. Describe the role of biotic factors on plant community.
2. Write about the atmospheric factor.
3. Explain how the temperature factor influence the structure of the vegetation.
4. Write about the water cycle.

Short answer type questions:

1. Biosphere
2. Light factor
3. Soil profile
4. Hydrosphere

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- Dr. M. Vijaya Koteswari

LESSON-12

POPULATION ECOLOGY

12.1 OBJECTIVE :

The objective of this lesson is to know about population ecology. We learn how the plant populations are studied and try to understand their distribution, growth patterns, phytosociology, etc.

12.2 STRUCTURE

12.2.1 INTRODUCTION :

The Species is the smallest taxonomic unit in the classification.It refers to a group of similar individuals among which breeding takes place. A group of organisms of the same species growing in a place is called population (the individuals may exchange genetic information). They occupy a particular space and function as a part of a biotic community, which is defined as an assemblage of populations that function as an integrated unit. A population has characters like density, natality (birth rate), mortality(death rate), age distribution, biotic potential, dispersion and growth form. These are expressed as statistical functions of the group but not at individual level. The populations also posses genetic characters related to their ecology (adaptiveness, reproductive fitness **(Darwinian)**, and persistence (probability of leaving descendents over long periods of time). Populations undergo seasonal changes and cyclic changes which can be described as population fluctuations and cyclic oscillations.

12.2.2 CHARACTERISTICS OF POPULATIONS :

Each population has certain characters like density, mortality, natality, age distribution, population regulation etc.

1. **POPULATION DENSITY:** All the organisms in a community constitute the population size. The number of individuals in a given area is called population density. It varies with the time. The individuals may migrate, die, in a population. Population density is of two types. 1. Crude density-it is calculated by taking the number of individuals and the total area, and 2.Specific density or ecological density it is the area, the individuals of a population occupy actually.

$$\text{Density (D)} = \frac{\text{no. of individuals(n)x time(t)}}{\text{area (a)}}$$

2. **NATALITY:** The newly born and added individuals in a population in a given time and given area is called Natality. This is also called birth rate in human beings. Increase in population is due to vegetative, asexual reproduction and budding, binary fission, formation of spores and seeds, laying eggs or giving birth to new off springs, etc.

$$\text{Natality rate (b)} = \frac{\text{Birth rate in unit time}}{\text{Average individuals in a population}}$$

$$b = \frac{dNn}{Ndt}$$

b = birth rate
d = change in values
N = initial population
n = population after unit time
t = unit time

3. **MORTALITY:** The incidence of number of deaths in a population is called mortality. It is of two types. 1. Realized mortality or related to ecological factors. The actual deaths occurred in a population are considered for realized mortality. 2. Minimum or specific mortality. The minimum deaths in a population in favorable conditions are called minimum or specific mortality. Even in favorable conditions as the ageing occurs to all individuals, the individuals become old and die.

$$\text{Death rate} = \frac{\text{No. of deaths in a unit time}}{\text{No. of individuals on average.}}$$

The ratio of natality and mortality is called vital index.

$$\text{Vital index} = \frac{\text{Birth rate}}{\text{Death rate}} \times 100$$

4. **SURVIVORSHIP CURVES:** The individuals in a population, depending on age are divided into three age groups. 1. Group of individuals before reproductive age. 2. Group of individuals in reproductive age and 3. Group of individuals in post reproductive age.

The individuals remaining in a group after the death of some individuals of different ages in a population are called survivors. The capacity to withstand the unfavorable conditions and survive is called survivorship. If they are shown in a graph, the resulting curves are called survivorship curves. Different species show different survivorship curves. They are identified as three types.

1. **Convex type:** In this the incidence of death generally occurs in old age. E.g. many crop plants and many annual plants. The plants die in old age after reproduction and seed setting.

2. **Concave type:** In this type more deaths occur in juvenile or young age. The individuals deprive of food or suffer with disease and do not get medicines. E.g. the animals in hilly regions or desert areas.

3. **Normal type:** These curves are generally in the form of straight lines. In these populations death rates are almost equal in all age groups. It can be clearly seen in vertebrates and plants. With the help of survivorship curves we can guess the reasons for the causes of the deaths in a population. If we identify the reasons for deaths we can control the loss of individuals in a population and regulate it.

5. **AGE DISTRIBUTION:** The rates of births and deaths depend on the age distribution in a population. If the young and old individuals are more in number in a population then that is called young population. If the old and middle age individuals are more in a population it is called stable population. In humans all the three stages prevail for same length of time where as in plants and animals the old age prevails for a long time. In any declining population the old age individuals are more in number.

6. **AGE PYRAMIDS:** The three age groups of a population are shown as a graph and is called age pyramid. The young individuals at the base, middle aged at the centre and old age

individuals at the top should be taken for the preparation of the graph .The shape of these pyramids indicate, increase, stability and decrease of the population. The triangular pyramid indicates more number of young individuals in the population. e.g. bacteria, yeasts, etc. In a stable population the diagram of pyramid appears in a bell shape. All the three age groups of populations are more or less same in number. In a declining population the older individuals or, individuals of post reproductive age are more in number and the pyramid appears in urn shape. See Fig. 5.

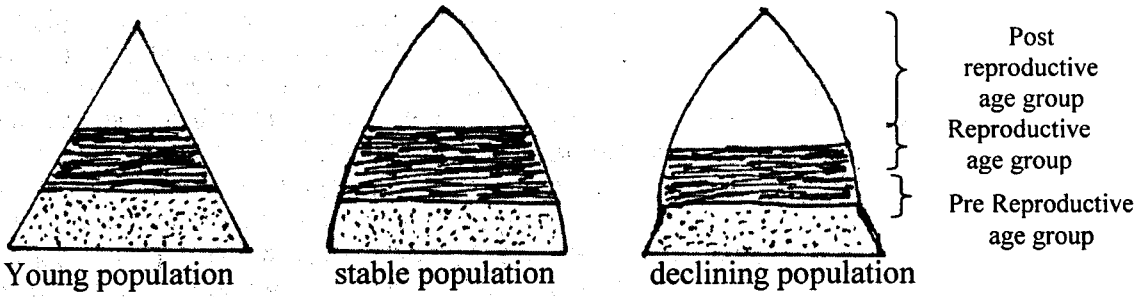

Fig. 5. Age Pyramids

7. POPULATION GROWTH AND GROWTH CURVES:

Population growth is an intrinsic character of a population.The growth rate can be caliculated by the following equation.

$$\text{Growth rate} = \frac{\text{Number of births(b) - Number of deaths(d)}}{\text{Average population of a unit time}}$$

Growth or depletion of a population is controlled by many factors which act systematically. The reproductive potential and environmental resistance together determine the population growth of a species. The maximum rate of reproduction of the organisms in an ecosystem in favourable conditions is called biotic potential.

Biotic potential =birth rate(b) - death rate(d)

The biotic potential depends on age groups of the population, sex ratio, etc. The environmental factors which tend to decrease the population growth rate is called environmental resistance. This includes food supply, availability of sun light, nutrients, heat, space and also competition and adaptation of the individuals. The difference between biotic potential and environmental resistance (er) is called the carrying capacity of the ecosystem. That can be written as shown here.

Carrying capacity = biotic potential _ environmental resistance.

The line diagrams representing the growth or depletion in a population in a given time are called growth curves. For this we have to take time on x- axis and number of individuals in a given area on y- axis. Usually the growth curves are of two types. They are 1. 'S' shaped or sigmoidal growth curves and 2. 'J' shaped growth curves.

1. **S-shaped or sigmoidal growth curves:** This indicates the growth of the population in a new area or ecosystem. Initially the growth of a population is slow. After the acclimatization to the new environs the population attains stability. This is called Lag stage or positive acceleration phase. Later the population increases very fast. This is called log phase. In this phase the birth rate is far higher than the death rate. After some time the birth rate declines and death rate increases. Because of this, the population growth rate decreases. This is called negative acceleration phase. After this stage, later the population comes to an equilibrium, indicating zero population growth. Now the rate of births almost equals to the rate of deaths in the population. The density of the population acts as environmental resistance and helps in decreasing population growth. This state of equilibrium where the population shows zero growth is called its **carrying capacity**. After reaching carrying capacity or zero growth in the population we see the decline in population growth rate. This type of population growth is density dependent gradient, because growth rate is dependent on number of individuals in the population. See Fig. 6.

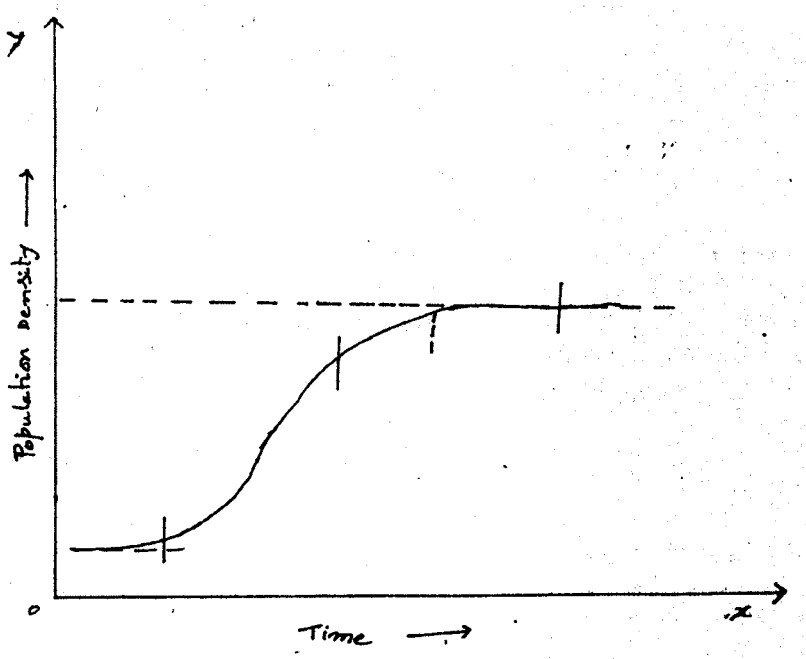

Fig. 6. Graph showing 'S' shaped growth curve

Many Types of organisms both in laboratories and external environments show this type of 'S' shaped or sigmoid growth curves. eg. Many types of micro- organisms, plants, animals, etc. We can calculate the growth rate of population which show sigmoidal growth curves by the following equation.

Population increase $\frac{dN}{dt} = rN\left(\frac{K-N}{K}\right)$

N = initial number of individuals of a population in an ecosystem.
K = carrying capacity.
r = biotic potential.
t = time interval
d = change in values.

The phytoplankton in ponds and seas show this type of growth rate. Bacteria or yeast grown on culture media show this type of growth curves.

2. **'J' shaped growth curves:** In A new environment, after the initial lag stage the population increases fast and tremendously. This line diagram appears as the letter 'J'. This increase suddenly is stopped by the environmental resistance. This is called 'crash'. In this type of growth of a population does not depend on the density of the population i.e., density independent. This is because population grows until it reaches crash , does not depend on population density. Sudden changes in the environment like, seasonal changes or end of reproductive phase or breeding season,or death of the supporting organisms lead to crash.eg. ephids grow very fast and show 'J'shaped growth curve when temperatures are low, but when the temperatures turn high they suddenly die and decrease in number and population density. Another example to this type of population growth curves is annuals. They grow fast and reach reproductive stage, form seeds and all of them suddenly die. Algae grow in rainy season and disappear when the ponds dry. This type of 'J'shaped growth curves can be seen in many populations. See. Fig. 7. This 'J'type of growth rate can be calculated by the following equation.

$$\frac{dN}{dt} = rN$$

N = number of individuals in a given time
t = time interval
r = biotic potential
d = change in values

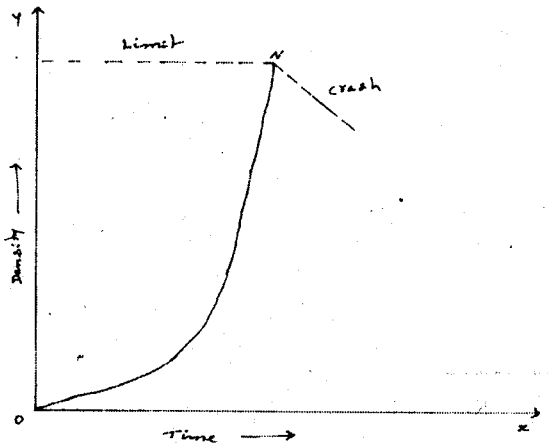

Fig. 7. Graph showing 'J' shaped growth curve

In unisexual organisms we have to consider the females as n, and biotic potential as r of every organism.

8. **POPULATION FLUCTUATIONS:** Populations show three types of fluctuations. 1. Seasonal,2. Annual and 3. Irruptive. The fluctuations are due to 1. Changes in the physical environment. 2. Interactions in a population. And 3. Both the reasons.

9. **POPULATION DISPERSAL:** The living organisms move out or in to the population through reproductive structures and this is called population dispersal. The agents which help in the dispersal are wind, water and animals.

12.2.3 POPULATION STRUCURE:

It shows three characters. 1. Dispersion and 2. Aggregation and 3.Isolation and territory.

1. **DISPERSION:** In this we generally study the dispersion of the population and mutual interaction of the individuals. Dispersion indicates how far the individuals of a population are present. Random type of dispersion is observed in the places where environmental factors are unchanging. E.g. plant depending on water or wind for seed dispersal. In the uniform dispersion the individuals occupy specific or equal distances. e.g. crop plants archards. In clumped dispersion the individuals are grouped or clustered. e.g. vegetatively reproducing plants.

2. **AGGREGATION:** Living organisms growing in groups is called aggregation. The reasons for aggregation are changes in micro climate, changes in seasons, reproduction and social attraction. e.g. grass plants, bees, etc. Aggregation leads to intraspecific competition and at the same time as a group they can face the unfavorable conditions like cyclones or enemies and save themselves. See. Fig.8

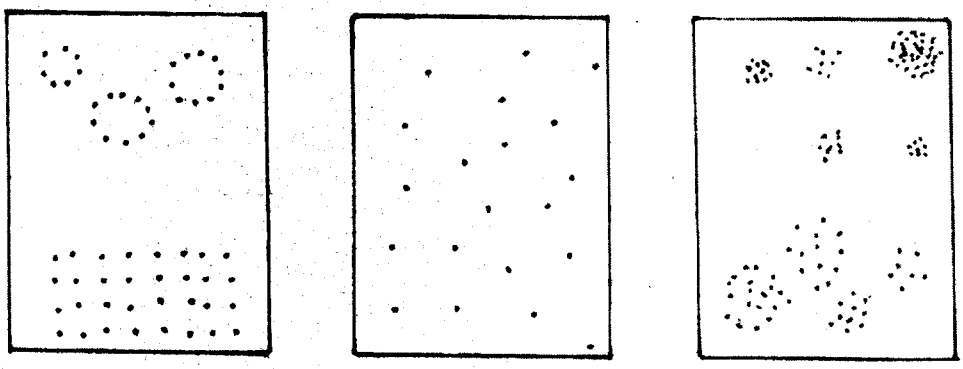

Fig. 8. Population Dispersion

3. **ISOLATION AND TERRITORY:** Some animals restrict themselves to a limited area with its companions and off springs. This area is called its territory. This is helpful in dividing utilizing the area for procuring food and for reproductive activities.

12.2.3 ECADS:

Ecads are morphologically different forms of a species. They are also called as ecophenes, habit forms,etc. Mostly the variations are environmentally induced. The plants differ in appearance, size, number of leaves, flowers, growth habit, length of stem, pigmentation, etc. But they are genetically similar. These characters may be altered if the environmental conditions are changed. Thus these variations or differences are not genetically fixed and these changes are reversible and temporary.If different ecads are transplanted in similar environmental conditions these differences disappear and all plants of the population appear similarly. eg. Euphorbia hirta. The plants growing in grassland showed prostrate and profusely branching. The plants growing on the foot path are compact with small leaves. When these two types are grown in same habitat the differences were disappeared.

12.2.4 ECOTYPES :

They are also called ecological races. The variations seen among the populations are genetically fixed and permanent. They can not be reversed. These variations are distinct and the plants are interfertile and are in one taxonomic species.If they are grown in same habitat their differences persist and will not change. Eg. Lindenbergia polyantha, Euphorbia thymifolia. There are different types of ecotypes. Few of them are given here.

1. **Climatic ecotypes:** This type of ecotypes have developed with climatic factors like light, temperature, water etc. Turesson(1930) recorded climatic ecotypes in Leontondon autumnalis.
2. **Edaphic ecotypes:** Edaphic ecotypes develop with the differences in soil factors. They include soil moisture, excess or deficiency of nutrients. eg. Euphorbia thymifolia recorded by Ramrkrishna (1961)
3. **Climatoedaphic ecotypes:** Both climatic and edaphic factors influence the plant populations and the climato edaphic ecotypes may be developed. eg. Cenchrus ciliaris reported by Pandey and Jayan (1970)
4. **Physiological ecotypes:** Physiological changes like photoperiod, nutrient uptake, water absorption,etc. are the causes for the formation of physiological ecotypes. Olmsted (1944) recorded physiological ecotypes in Bontelona curtipendulla.
5. **Altitudinal and latitudinal ecotypes:** This type of ecotypes develop with the change in altitude and latitude. Generally they are observed in gymnosperms. Eg. Pinus

Formation of ecotypes indicates the capacity of adaptability of a species to various factors. They can be grown in wider areas which are environmentally different. They can lead to the evolution of new plant species.

12.3 SUMMARY :

In this chapter population ecology we study about the characteristics and structure of the population, ecads and ecotypes. Species is the smallest unit in classification. The group of similar individuals, genetically identical, living in a given area are called population. They show certain characters like natality, mortality, density, age distribution, etc. The seasonal are considered as fluctuations. Different statistical measures are considered to study the populations. The populations show two types of growth curves, where in 'S' shaped curve the populations show steady point after growing fast for some period and 'J' shaped curve the populations abruptly stop growth suddenly. In the population structure we studied about the dispersion, aggregation and isolation and territory.

Plants with different morphological characters are called ecads and ecotypes. Ecads when grown in similar conditions do not retain their differences, and they are environmentally induced. Ecotypes are the pSlants when grown in similar conditions they retain characters and they are genetically fixed.

12.4 TECHNICAL TERMS :

Population, population ecology, growth rate, mortality rate, growth curves, ecads and ecotypes.

12.5 MODEL QUESTIONS:

Essay type questions:

1. What is population, describe the characters of population.
2. write an essay on population strcture
3. Explain the characters of population. 4. Explain population growth curves.

Short answer type questions:

1. Population density
2. Surviversh ip curves.
3. Age pyramids
4. Population structure
5. Birth rate and death rate.
6. Population dispersion.

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LESSON-13

COMMUNITY ECOLOGY

13.1 OBJECTIVE:

The objective of this lesson is to study about the community ecology. We learn about the characters of plant communities, life forms, biological spectrum etc. in this chapter.

13.2 STRUCTURE

13.3 INTRODUCTION:

A group of individuals which are genetically identical and breed, belong to a species. A number of individuals of a species growing in an ecosystem is called **population**. Many such populations sharing the same ecosystem is called **community**. All the types of populations living in a given ecosystem are collectively referred as biotic community. We can see many types of plants, animals, microorganisms living together in a forest. Then all of them are referred to forest community. It is very difficult to study all the plants and animals in an ecosystem like forest. So it is conveniently divided into plant community and animal community. Biotic community refers to both plant and animal communities. Community ecology refers to the study of the ecosystem and collective actions and reactions of biotic communities in the given ecosystem.

13.4 BIOTIC COMMUNITY:

Biotic community has certain characters. They are-

1. **Size of the community-** It ranges from few meters tofew hundreds of kilometers with or without specific boundaries.Area between two zones where one gradually merges with the other is called transitional zone or ecotone.

2. **Specific abundance and diversity-**The number of populations in a community shows diversity. Some ecosystems may have few or one type of populations. We see only one species in a grassland ecosystem and some ecosystems like tropical forest may have many types of populations in a community. Depending upon the number of populations the species richness may be considered. The ratio between number of species and total number of organisms is called species diversity. This depends on many factors like environment and community stability.

3. **Community dominance:** In a community all species may not have equal opportunities, one or few may dominate the others. Size, number, structure, agility, rate of reproduction etc. may influence in dominating others.

4. **Coexistance :** Any organism can not live alone. It coexists with other organisms in its lifetime for various things. Exploitation refers the coexistence where one gets benefit and the other loses. Mutualism refers coexistence where both the organisms get benefit.

5. **Community structure:** In a community the organisms show a definite spatial differentiation giving the community a shape and structure ,etc.We can consider this into a) vertical stratification or differentiation, b) horizontal stratification and c) temporal differentiation.

6. **Succession:** The communities are replaced by one after the other over a period of time in an ecosystem. This phenomenon is called ecological succession or ecosystem development. Finally a stable community lasts for a long time and is called climax community and the replaced communities are called seral communities.

13.5 CHARACTERS OF PLANT COMMUNITIES

Any plant community broadly shows two types of characters.1) analytical characters and 2)synthetic characters. Analytical characters are again of two types- quantitative characters and qualitative characters. The qualitative characters include physiognomy, phenology, stratification, sociability, vitality, vigor, life forms. The quantitative characters include density, frequency, cover, etc. The synthetic characters include presence, fidelity, dominance, etc.

FREQUENCY: Frequency can be defined as the homogeneity in distribution of a species in the communities. It indicates the availability and presence, adequacy of a species in the given area. Its value depends on the size of the sampled area and at the same time the size, structure and distribution of the individuals of the population. Raunkier (1934) identified five frequency classes as shown below.

Class A---------- 0—20 %
Class B----------21---40 %
Class C----------41---60 %
Class D----------61---80 %
Class E----------81---100 %

Raunkier proposed 'law of frequency' after his extensive study on various frequency ratios. According to his law of frequency the various classes are in the A >B>C>D<E order. In a community of homogenous distribution of species are in the following ratio. They are A=53, B=14, C=9, D=8 and E=16. If this is shown in a diagrammatic way it shows 'J' shaped curve. If species belonging to A class are less and E class are more, then we can say that the distribution in that community is heterogenous. See.Fig. 9.

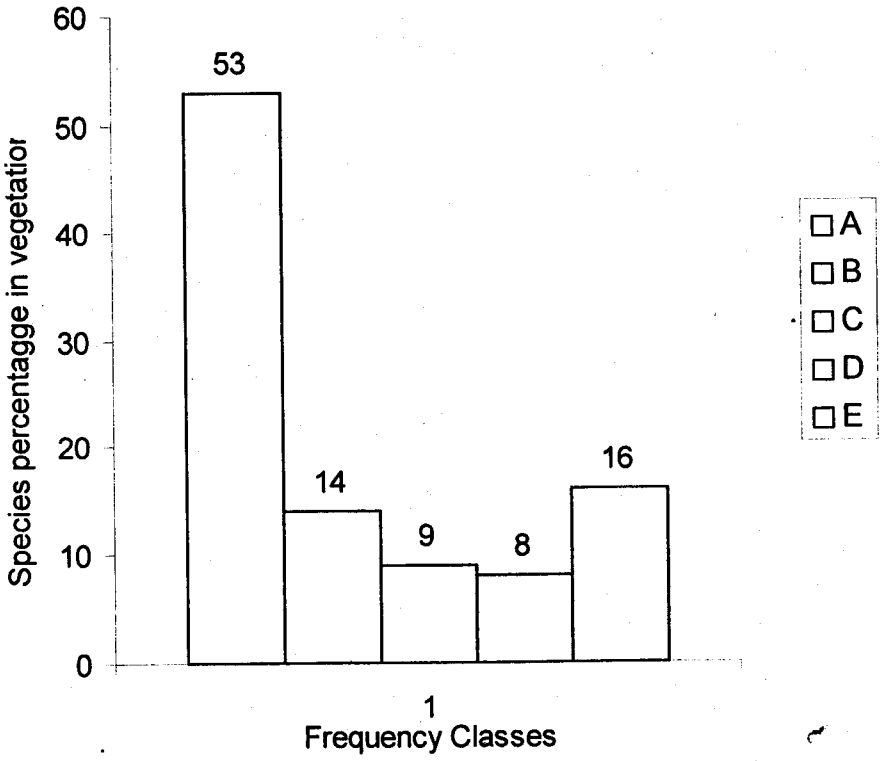

Fig. 9. Frequency Distribution Graph

DENSITY: Density can be defined as the number of organisms living in a unit area. Generally we use unit area for terrestrial organisms and unit volume for aquatic organisms. E.g. 100

coconut trees in an acre, 2 fish for cubic meter. Density can be calculated by the following equation

$$\text{Density (d)} = \frac{Number\ of\ living\ organisms\ (n)}{Area / Volume (a)}$$

Density of organisms depends on various ecological factors like available light, annual rain fall, edaphic factors, climatic and topographic factors. Density indicates the number of organisms in the community present in that area.

COVER AND BASAL AREA: The area on the surface occupied by the total plant including its branches and leaves is called cover. It can be denoted in two ways. 1.) basal area and 2.)canopy cover. The area of transverse section of a stem at 2.5cm. height from the ground is called basal area. In trees the height is considered to take at 1.5 meters. This area indicates the area of soil used by this plant to penetrate. The branches and leaves occupy certainly some area and forms shade. This area is considered as canopy cover. This is measured by tapes and calculated by the following equation.

$b = \pi r^2$ b= canopy area

r=radius diagram

We can calculate cover (coverage) by taking the canopy cover of all the individuals devided by total area of the habitat. The organisms showing higher values of coverage dominate. In the field we follow the given scale to determine the coverage of a species. See. Fig. 10.

Class—1------occupying less than 1% area

Class—2------occupying 1/20—1/4 of the area

Class—3------occupying 1/4—1/2 of the area

Class-- 4------occupying1/2—3/4 of the area

Class—5------occupying3/4—whole of the area.

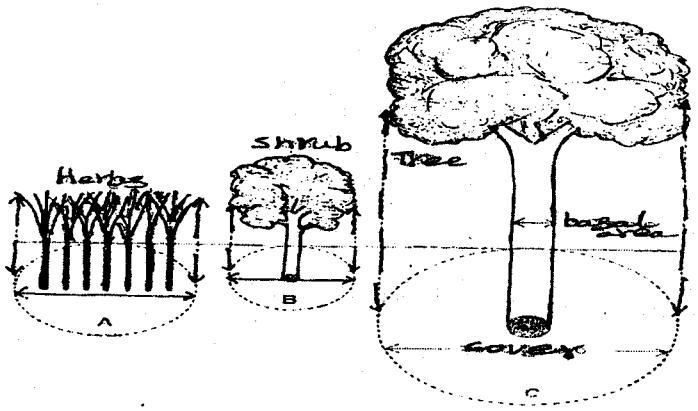

Fig. 10. Diagram showing cover and basal area of the different plants in an area

13.6 LIFE FORMS:

Many phytosociologists tried to classify the plant communities by taking some characters into consideration. Some of them are

1. Growth form—tree, shrub, herb, etc.
2. Habitat—edaphic factors
3. Environmental factors—temperature, rain fall, humidity, etc.

Of all, the classification proposed by Raunkier (1934) received support from many. Raunkier classified the plants on the basis of the plant adaptations to the unfavorable conditions. He defined life forms as the adaptations to the environmental conditions. Species abundance depend on its adaptations to unfavorable conditions. Generally the plants reproduce vegetatively and sexually, producing tubers, seeds, rhizomes, etc. which can survive the unfavorable conditions. He recognized 5 types of plants on the basis of the position of the perennating buds and the way the plants protects them. They are-

1.Phanerophytes (Ph): The plants are woody. They grow as shrubs and trees. Some scientists consider epiphytes as phanerophytes.They produce the perennating buds at least at the height of 30 cm, which are naked. Sometimes the buds are covered with scales. They are found in tropical and sub-tropical forests. Their growth is less in temperate regions. Again they are divided into four sub-classes based on height, protection of the buds, etc. They are given here under.

1) Megaphaenerophytes------ growing more than 30 meters.
2) Mesophaenerophytes------ growing 8- 30 meters
3) Microphaenerophytes------ growing 2-8 meters
4) Nanophaenerophytes------ growing less than 2 meters

2. Chamaephytes (Ch) : They are small in size. They include shrubs and prostrately growing plants. The reproductive(perennating) buds may be formed in the height of at least 25 cm. These types of plants are mostly seen in colder temperate regions.

3. Hemicryptophytes(H): This class includes grasses and herbs.The perennating buds form near the ground and are covered by fog, litter, etc. e.g. biannuals and perennials growing in colder regions.

4. Cryptophytes or geophytes (Cr) In this class the plants produce the perennating reproductive buds under the soil layers or in water and are protected. Many aquatic plants and plants producing rhizomes, corms, bulbs, etc. They are called geophytes since they produce under ground buds. In favorable conditions they develop into aerial shoot systems and complete their life cycle.

5.Therophytes (Th) : In this type the plants do not produce any special structures to survive in unfavorable conditions. They overcome the odd conditions by being in the form of seed. E.g. annuals growing in dry arid conditions. See. Fig. 11.

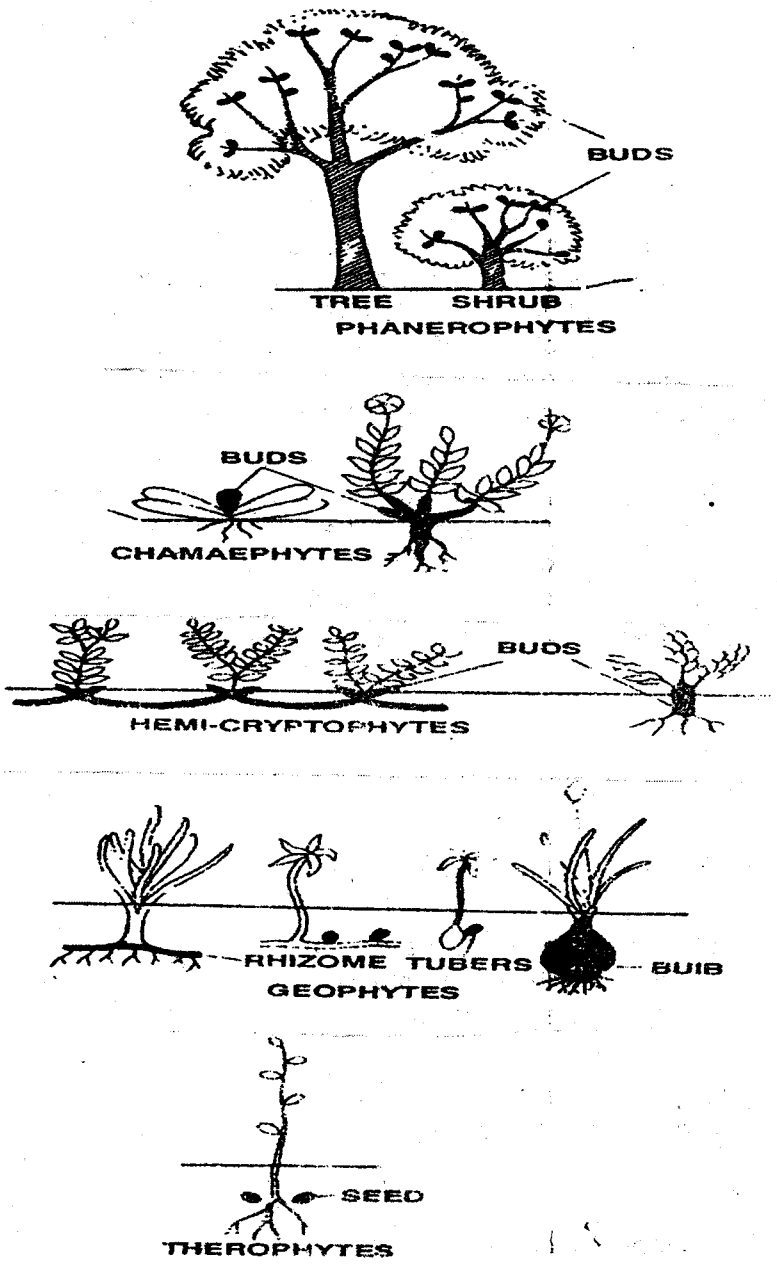

Fig. 11. Raunkiers life gorms

Depending upon the size of the leaves which indicate climatic conditions Raunkier identified six types of leaves. The leaf in each class is nine times greater than its previous class.

Community type	**leaf size (mm2)**
Leptophyll	25
Nanophyll	225
Microphyll	2025
Mesophyll	18225
Macrophyll	164025
Megaphyll	>164025

13.7 BIOLOGICAL SPECTRUM:

The percentage composition of different life forms in a plant community is called biological spectrum or phytoclimatic spectrum. As these life forms belong to specific ecosystem, the biological spectrum of the community acts as an indicator. If the plants are chemiphytes, we can understand the climatic factors of that region are cold.If there are many therophytes then we can say that the climateprevailing is dry.If the biological spectrum of different ecosystems are similar then we can understand that the climates are similar. In 1934 Raunkier prepared one biological spectrum of phanerogamic flora and calculated that different life forms are presenting the following ratio.

Phanerophytes—46%

Chemiphytes—9%

Hemicryptophytes—26%

Cryptophytes—6%

Therophytes—13%

If we compare our data of any community with this and find similar then we understand the climatic factors of that region. But it not always so, because biotic factors including mans activities come in the way. See. Fig. 12 & 13.

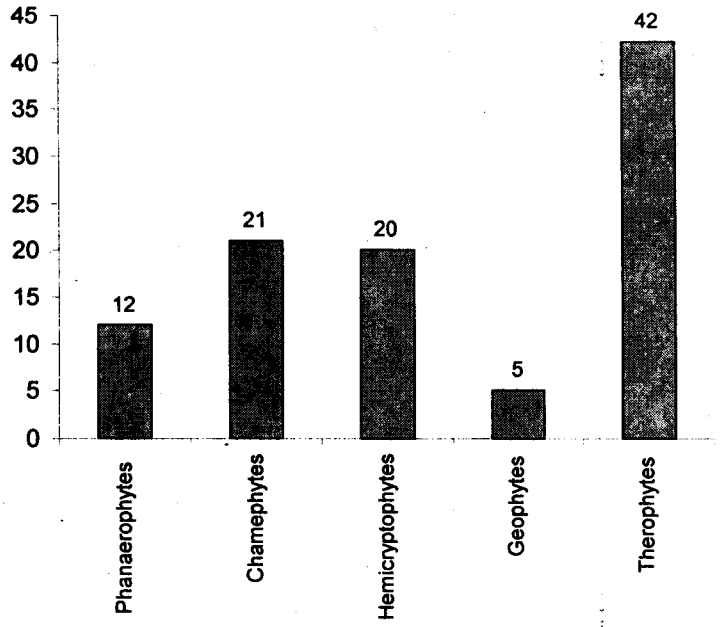

Fig. 12. Biological spectrum in a desert

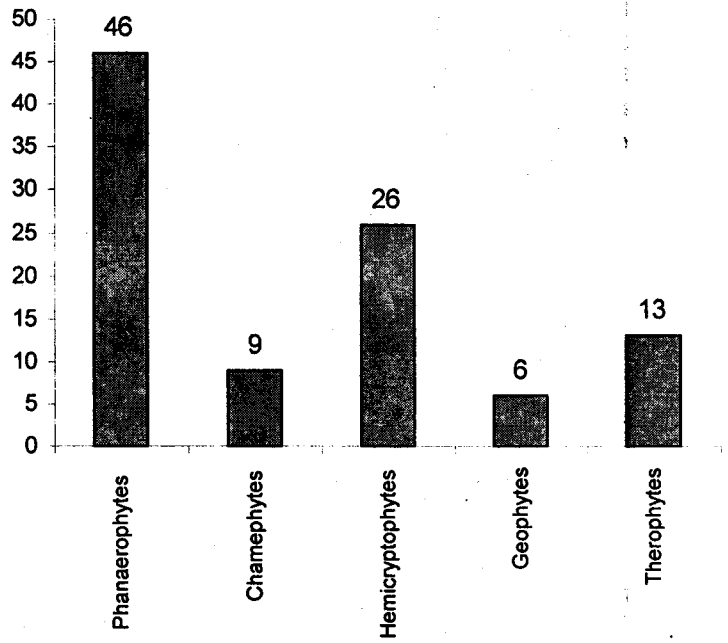

Fig. 13. Biological spectrum of flowering plants

13.8 SUMMARY:

Similar individuals belong to a species and as a group forms population. Such different populations growing in an area are called community. Community ecology is about the study of populations which influence each other and their phytosociological behaviour, life forms, etc. The biotic community has certain characters like size, species abundance and diversity, community dominance, coexistence, community structure, etc. The plant communities do exhibit analytical and synthetic characters. Various life forms like phanerophytes, chamaephytes, hemicryptophyts, cryptophytes and therophytes were identified in plants by Raunkier. The biological spectrum shows the percentage composition of different life forms.

13.9 TECHNICAL TERMS:

Community ecology, biotic community, life forms, biological spectrum.

13.10 MODEL QUESTIONS:

Essay type questions:

1. Define community and write about its characters.
2. How we study plant communities

Short answer type questions:

1. Quadrats.
2. Raunkier life forms
3. Strtification4. Biological spectrum.

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LESSON – 14

ECOLOGICAL SUCCESSION – HYDROSERE, XEROSERE

14.1 OBJECTIVE

It is aimed to learn about plant succession. The objective is to learn the development of ecosystems which are controlled by chiefly the environment. This is helpful to understand better the relationship of living organisms and their surroundings in which they live.

STURUCTURE

14.2 STRUCTURE:

14.2.1 INTRODUCTION :

The consequence of biological regulation in the community as a whole is the phenomenon known as ecological succession. It is also known as ecosystem development. Communities are made up of populations, which interact in many ways and influence their development over time. Ecological succession involves an orderly process of community changes which are directional and predictable. A sequence of temporary communities replace one another in a given site, bringing changes in the physical environment. This changed environment is not suitable for the present communities. Therefore it paves way for new communities. This determines the pattern of succession. The transitional series of communities

which develop in a given area are called sere or seral stages. Ultimately one community becomes stable and is called the climax community. Ecological succession is therefore community controlled although the environment determines the succesional pattern. Therefore we can define succession as successive colonization of the same area by different plant communities over a period of time. The term succession is first used by Hault (1885). In India studies on ecological succession have been made by R. Misra, Ranganathan, Champion, Puri, Pandey, etc.

14.2.2 PROGRESS OF SUCCESSION :

According to Odum 1971 succession has features like: 1. It involves an orderly change in the communities, 2. Its direction is predictable unless disturbed by outside factors. 3. It results from the modification of the environment by the community. 4. It results finally in the stability of successful community and is called climax.

14.2.3 KINDS OF SUCCESSION : There are two types of succession

1. **Primary Succession :** It starts always on bare area where it is previous unoccupied by living organisms. E.g. recently cooled lava flow, newly exposed sand dune, etc.

2. **Secondary Succession:** It starts on the site where it is previously supporting inhabited community which is cleared by denudation. Here the pace of succession is very fast because the area is fertile and favourable to living organisms especially plants. E.g. deforested regions, abandoned crop fields, etc.

Depending on the type of habitat the primary and secondary succession is of the following types.

a) **Halosere** –succession begins on a salty soil or in saline water.
b) **Hydrosere** – succession starts in aquatic bodies like pond, lake, river, stream, etc.
c) **Xerosere** – it starts in dry habitats like sandy areas (psammosere) or rock surfaces (lithosere).
d) **Autogenic succession:** The outside forces are the chief agents of the modifications in the environment.
e) **Autotrophic Succession:** This is initially dominated by the autotrophic organisms i.e., green plants.
f) **Heterotrophic succession:** In this early dominance is observed by heterotrophs like fungi, bacteria and animals, etc.

14.2.4 CAUSES OF SUCCESSION:

We know that succession is caused by many factors. 1. Climatic factors 2. Topographic factors and 3. Biotic Factors.

1. Climatic Causes – Wind, drought, lighting, storms, etc. kill and eliminate the previous existing life forms.

2. Topographic causes – erosion and deposition of soil by various agents like wind, water, gravity, etc. lead to topographic changes. These changes produce primary bare areas.

3. Biotic causes – they include destruction of habitats by man, overgrazing by animals and diseases caused by bacteria, fungi, etc.

14.2.5 PROCESS OF SUCCESSION :

The process of succession involves many steps in sequence. They are

1. **Nudation :** The development of a bare area without any form of life is called nudation. It is caused by many factors like topography, climate and biota.

2. **Invasion:** Organisms which grow in the neighbouring areas of the newly formed bare area invade into and grow. It involves migration, ecesis and aggregation. Migration is arrival of seeds, spores, etc. by wind, water, animals, etc. into the new bare area. This determines the type of plants which grow initially. The first arrivals are called pioneers or pioneer colonizers and the process of invasion is called colonization. Ecesis is the establishment of the pioneers. It has the process like germination, growth and reproduction. Ecesis is controlled by climatic, edaphic and biotic factors. Seed dormancy, germination, establishment of seedlings also play important role. Aggregation is establishment of a species in an area after ecesis. The species reproduce and multiply in number. This results into the increase in number of a species in the colony and is called aggregation.

3. **Competition:** If the number of a species in a colony increases it results into competition for space, sunlight, water, nutrients etc. The competition may be among the individuals of the same species (intra specific) or among the individuals of the different species (inter specific). In the competition for various things the weaker species are eliminated and stronger ones are retained.

4. **Reaction:** This is considered as important biotic factor because it brings change in the habitat. The above ground or aerial environment is altered by plants in few ways. Some of them are

1. The roots bind the soil and check the soil erosion.
2. The roots which penetrate into rock crevices cause weathering of rocks leading to formation of soil.
3. The plants protect the ground from rainfall and waterflows.
4. They check the wind velocity
5. They give food to the heterotrophic organisms playing important role in food chain.
6. After their death the plants enrich the soil with nutrients by the formation of humus.

All these changes make the habitat less favourable to the existing colonizers and favourable to the new invaders. Again the process repeats and other new invaders occupy the habitat. Thus a series of invasions occur in the same area and a sequence of plant communities appear in that area. The whole sequence of communities that replace one another in the given area is called sere. The various communities which constitute the sere are called seral communities or seral stages or developmental stages.

4. **Stabilisation :** This is also climax. After some seral stages the succession results into stabilization of the vegetation develop into climax community. This climax community is stable and maintains a complete harmony with environment factors of that area. The conditions of environment are almost remain unchanged. In this condition the number of individuals increase but the number of species decrease. The soil is fully occupied by plants and the seral communities are largely affected by soil and other factors. Since the climax is mainly controlled by the climatic factors it is also called climatic climax by Clements 91916).

14.2.6 THEORIES OF CLIMAX CONCEPT:

The climax community is the ultimate among the seral stages. It may take hundreds or thousands of years to reach climax in any area. Climax community is self maintaining and slow changing community and area and time. Previously it is believed that the climax communities do not undergo changes and is called ageing. The community may be aged due to storms, diseases and other biotic and abiotic factors. Another stable community may develop on the margins of the climax community and is called proclimax. Sometimes some areas of higher life form is found within a climax and is called post-climax e.g. the plant community present along a stream in a grass land climate constitutes the post climax stage. If the climax is in localized regions with plants of lower order than climax, the strip of formation is called as sub-climax or pre-climax. There are two opinions regarding the number of climax communities in a given climatic region – they are.

1. MONOCLIMAX THEORY:

Frederick Clements (1916) put forwarded this theory. According to him there develops only one true climatic climax in that region. This concept is generalized as mono-climax theory because the climax formed is controlled by the climate only. In 1928 Clements considered the

climax formation as an adult Organism. It is a fully developed community of a region and all other communities are in the different stages of development and are called sub ordinate communities. His theory of mono-climax has been strongly criticized by many ecologists. Coweles, disagreed and stated that equilibrium state is never reached and succession is in fact a variable approaching another variable rather than a constant. Sometimes it is also not correct to consider the climate as the sole governing factor to reach climax. It is also common to see different types of communities according to the topography, soil and other factors.

2. POLYCLIMAX THEORY:

Tansley (1935) put forwarded this theory. According to this theory climax reflects not only the climatic factors but also other factors of the environmental complex like edaphic, biotic, etc. The climaxes are of several kinds which are different from the climatic climax of the area. They can be called as edaphic climax influenced by soil, biotic climax controlled by biotic components, anthropogenic climax effected by man, grazing climax showing grazing effects, likewise zootic climax by animals, topographic climax by topographic factors, fire climax brought out by repeated fires. Other ecologists like Whittaker, Braun, etc supported this polyclimax theory and its concept. According to Daubenmire (1968) if we accept monoclimax theory then it means that we are not considering other environmental factors primarily.

14.3 HYDROSERE

Succession is the natural process by which the same locality becomes successively colonized by different communities of plants. In other words it is the orderly process in which different forms of plant communities occupy a particular area before the development of climax vegetation. Before reaching the climax stage the same area is occupied by a series of different plant communities constituting the seral stages and the whole sequence of communities is called **sere**. The succession which begins in an aquatic environment is called as **hydrach** and the different stages of this succession are collectively called **hydrosere.** Hydrosere can be best studied in a fresh water pond or a lake. Succession is quite conspicuous in standing water of moderate depth. The different seral stages in a hydrosere are as follows.

1. **Phtoplakton stage:** This is the first and pioneer stage of hydrosere. The autotrophic plankton forms like diatoms, green flagellates, blue-green algae, bacteria are the pioneers to colonise the pond. They multiply rapidly and constitute the food for zooplanktons which are heterotrophic. The death and decomposition of plankton produces organic matter. This organic matter mixes up with clay and silt lying at the bottom of water reservoir, which is brought about by rainwater or by wave action of pond water. It forms soft mud at the bottom of the pond. This new habitat, which is a little shallower and where light penetration occur, becomes suitable for the growth of rooted hydrophytes.

2. **Rooted submerged stage:** Because of the death and decay and decomposition of phytoplanktons the habitat is more suitable for the growth of rooted submerged hydrophytes. Plants like *Vallisnaria, Ceratophyllum, Utricularia, Potamogeton, Hydrilla, Myriophyllum,* etc. are some rooted hydrophytes which are seen in second seral stage of hydrosere.They are rooted in the mud and the rest of the plant grows submerged giving

some resistance to the water currents. This results in to the deposition of silt and sand because these plants obstruct the water currents. Further the death and decay of these plants forms the substratum. Thus the pond becomes shallower. This new condition invites the floating leaved plants. The submerged plants are forced to shift to the deeper areas of the pond.

3. **Rooted floating stage:** By this time the pond is 5-10 feet deep. The rooted floating plants colonise this pond with their rhizomes. They have large leaves floating on the water surface and rooted at the bottom of the pond. Plants like Nymphea, Nelumbo, Trapa, Monocharia, etc. are some plants of this category. Some free floating hydrophytes like *Azolla, Lemna, Pistia, Eichornia, Wolffia, Salvinia* also are present along with these rooted floating plants. This is possible because many nutrients are present in these waters. These plants cover the water surface cut off sun light to the submerged plants. The death and decay of these plants in course of time build up the substratum. This soil building process continues and in a few years the pond becomes very shallow making the habitat more unsuitable for the growth and survival of the floating plants.
4. **Reed-swamp stage:** This stage is also called as amphibious stage. The water in the pond is 3 to 5 feet. The plants of this seral stage have larger and much branched rhizomes. The shoots stand erect due to the presence of mechanical tissues. Plants like *Rumex, Pontoderia, Phragmitis, Sagittaria, Typha*, etc. are few to mention These plants cut off light for the floating plants making the area unfavorable for them. The plants transpire large quantities of water, leading to further reduction in the water level. They contribute more organic matter which decays slowly and this habitat becomes unfavorable for the amphibious species.
5. **Sedge-meadow stage:** This stage is characterized by marshy soils. Now the waters are 1-2 feet. Plants like *Juncus, Carex, Polygonum, Eleocharis*, etc. colonise this area. They form a mat like vegetation towards the centre of the pond. Herbs like *Mentha, Caltha, Polygonum, Galium, Iris,* etc. are also found with these sedges. Due to high rates of transpiration the mud is exposed to air. As a result ammonia, sulphides, etc get oxidized to nitrtes and sulphates. This leads to mesic conditions of the area and the marshy vegetation is gradually replaced by species of another community.

6. **Wood land stage:** By the time the marshy vegetation disappears the soil becomes drier for most of the time in the year. Then terrestrial plants like *Salix, Cephalanthus, Cornus*, etc. invade in to this area. Trees like *Alnus, Populus, Cassia, Terminalia*, etc. also inhabit this area. Because of the presence of shrubs and trees the water levels still decrease. The humus added by these plants enrich the soil with nutrients. Many microorganisms grow in this soil. Further new tree species grow in that area.

7. **Forest stage or climax stage:** Now the area is invaded by many tree species. These plants do not allow the growth of previous types of plants. Now the nature of the climax is controlled by the climatic factors of this region. In tropical regions where rain fall is heavy and dense, tropical rain forests develop. In temperate regions mixed forests with *Quercus, Ulmus, Abies, Taxus, Spruce,* etc. In regions of moderate rain fall, deciduous forests or monsoon forests develop. See. Fig. 14

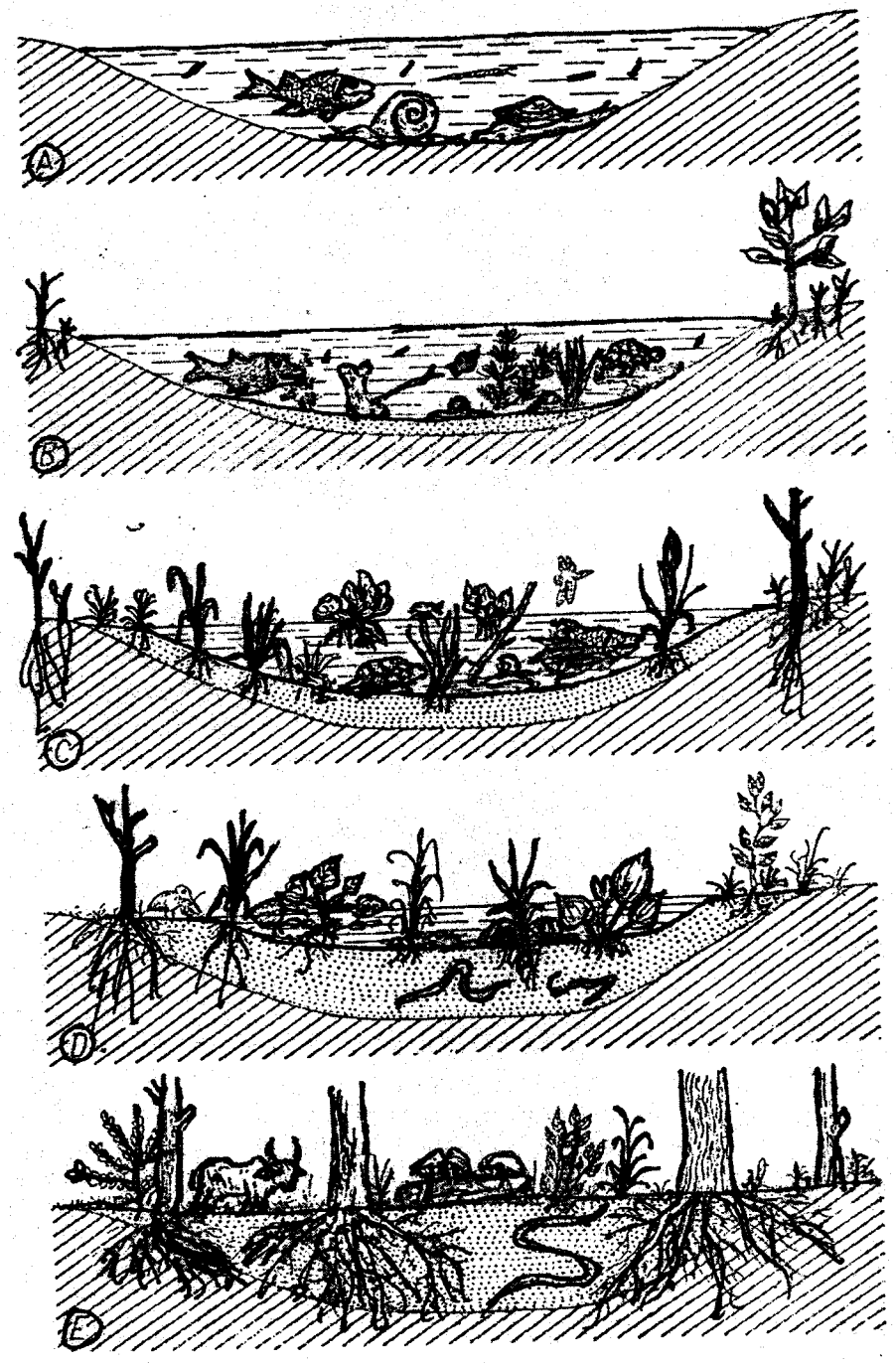

Fig. 14: Hydroseric succession, stages A to E showing formation of different communities at the same place. Gradual silking of pond bed reduces the depth and finally a terrestrial community has formed.

14.4 LITHOSERE OR XEROSERE

Succession stages start on bare rocks, sand dunes, etc. which faces extreme deficiency of water is called xerosere. Xerosere on bare rocks is called lithosere. Crustose lichens are the pioneers to colonize such exteme water scarce areas. After a series of seral stages the succession finally results into a forest constituting the climax community.

The different stages in the xerosere are given hereunder.

1. **Crustose lichen stage:** Generally on bare rocks water is deficient and that area is not favourable to the growth of the plants due to lack of nutrients. The crustose lichens inhabit this area and become pioneers of xerosere. *Rhizocarpon, Rinodina, Lecanora*, etc. are some crustose lichens growing in this area. They produce carbonic acid which causes weathering of rocks. The dead organic matter of these lichens mixes with dust particles of the weathered rock. This thin layer of soil invites foliose type of lichens to grow on.

2. **Foliose lichen stage:** They inhabit the rock after the crustose lichens. Plants like *Parmelia, Dermatocarpon, Umbilicaria,* etc. grow in this stage. These plants have large leaf like thalli and cover the crustose lichens. These crustose lichens die and accumulate on rocks. The foliose lichens can absorb more water from the rocksurface where the debri of pioneers absorb and store water during rains. The rocks also are weathered by the acids secreted by living as well as decaying plants. This results into the formation of thin layer of soil on the rocks, pavinig way for the growth of masses. Further the rocks form crevices and become rough.

3. **Moss stage:** The rocks with rough surfaces and crevices favours the growth of some xerophytic mosses. *Bryum, Barbula, Funaria, Poltrichum*, etc. are some mosses inhabiting the rough rocks. Spores of these mosses are brought over by wind currents. They germinate and grow in rainy season and they compete with the lichens for water and nutrients. These mosses have strong capacity to withstand desiccation. By their death and decay they accumulate humus and mineral nutrients. This is favorable for the growth of higher plants.

4. **Herb stage:** The annual herbs grow after the mosses. They are replaced by xerophytic herbs. The early herbs are *Poa, Aristida, Eleusine, Justicia, Tridax,* etc. Forms like ***Adiantum, Actinopteris, Asplenium, Chielanthus*** also accompany these grasses and herbs. The roots of these plants further corrode the rock, when they die they increase the humus content. The tall herbs cover the soil surface and create moist conditions. Evaporation rate is reduced. The heterotrophs like fungi, bacteria appear in large numbers in the soil. They help in recycling the minerals. Gradually these changes result into mesic conditions with decreasing xeric qualities.

5. **Shrub stage:** The increased accumulation of soil allows shrubs to grow. These shrubs invade this area by seeds or rhizomes, etc. Initially the xerophyic shrubs inhabit this area. They love sunlight and change the habitat, wind movements, and evaporation of water are further retarded. Their roots penetrate deep and take part in further breaking of rocks. This results into increase in the soil mass. Further the soil is enriched with nutrients by decay of twigs, leaves, and dead bodies. More water is retained in the humus. This lead to the growth of larger shrubs like *Zizyphus, Capparis,Fragaria, Rubus,* etc.

6. **Climax forest:** The first trees to inhabit this area are xeric and show stunted growth. Later the area becomes favorable for mesic trees. Thus the vegetation finally becomes mesophytic climax community. See. Fig. 15

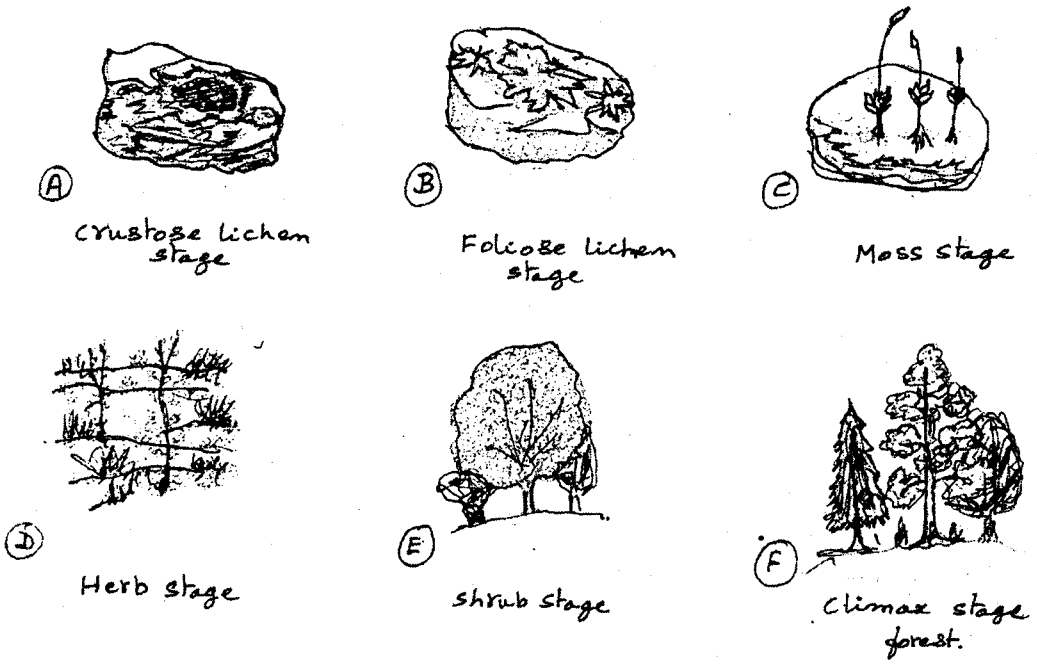

Fig. 15 : Xerosere succession showing all the different stages

14.5 SUMMARY

Succession is the natural process by which the same area becomes successively inhabited and colonized by different communities of plants. Before reaching the climax stage the same area is occupied by a series of different plant communities constituting the seral stages and the whole sequence of communities is called a sere. The process of succession is controlled by environment and finally reaches a stable climax stage. There are many kinds of succession depending upon the habitat nature, different forces of the environment or initial inhabitants. The causes of succession are mainly climatic, topographic and biotic. The process of succession involves nudation, invasion, competition, reaction and stabilization. There are two theories regarding climax concept. They are monoclimax theory and polyclimax theory. The succession which starts in aquatic body is called hydrosere. The different seral stages involved in hydrosere are phytoplankton stage, rooted submerged stage, rooted floating stage, reed swamp stage, sedge meadow stage, woodland stage and climax stage. Succession stages started on bare rocks is called xerosere. Crustose lichen stage, foliose lichen stage, moss stage, herb stage, shrub stage and climax stage are the different seral stages of xerosere.

14.6 TECHNICAL TERMS

Succession,communities, environment, autogenic, allogenic, autotrophic, heterotrophic, monoclimax, polyclimax hydrosere, xerosere, lithosere, climax.

14.7 MODEL QUESTIONS

Essay type questions:

1. Describe the various stages in succession.
2. Explain succession in a pond.
3. Write about lithosere.

Short answer type questions:

1. Climax theory.
2. Reed-swamp stage.
3. Sedge- meadow stage.

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- Dr. M. Vijaya Koteswari

Lesson 16

RECOMBINANT DNA TECHNOLOGY, SOMATIC HYBRIDS AND CYBRIDS

16.0 OBJECTIVES

* Students can understand the steps and procedure, involved in r-DNA technology.
* Pupil comprehend the differences between somatic hybrids and somatic cybrids.

STRUCTURE

16.1 INTRODUCTION

Recombinant DNA is a hybrid DNA formed by joining of at least two different fragments of DNA. It is abbreviated as r-DNA, also known as chimeric DNA. The organism containing r-DNA is called a Recombinant. The production of recombinant DNAs is said to be recombinant DNA technology, often called as Genetic Engineering.

16.2 REASONS FOR THE CONSTRUCTION OF RECOMBINANT DNA

- To protect the foreign DNA from nuclease enzymes of the recipient cells.
- To make the foreign DNA replicate along with the vector DNA.
- To have genetic markers helping us for the selection of recombinants.
- To enhance the expression of the desired foreign DNA or gene in the recipient cells.

3. STEPS IN THE CONSTRUCTION OF rDNA

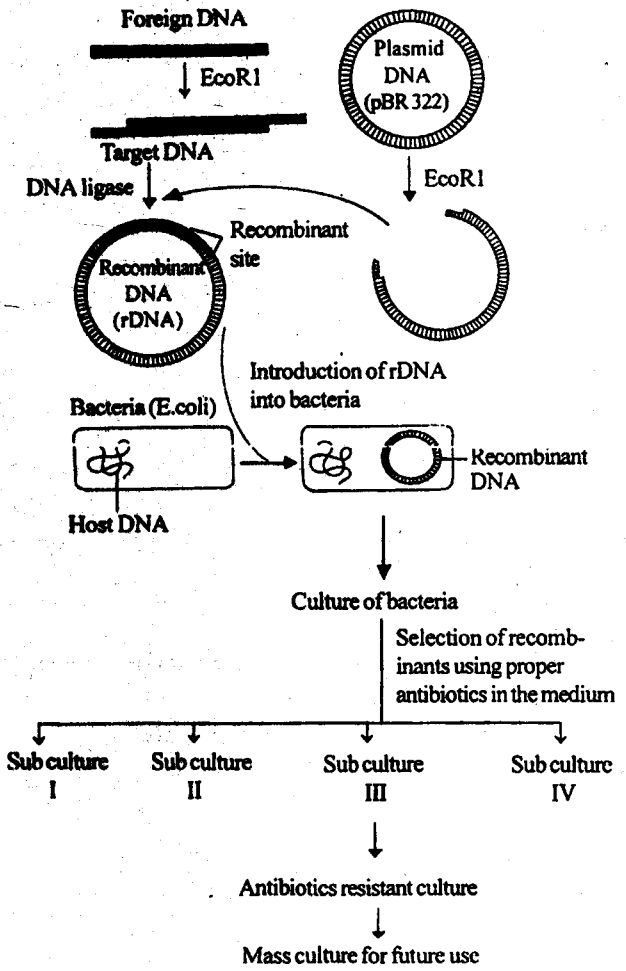

Fig –1 : An outline of genetic engineering methodology

Recombinant DNA can be constructed by the following steps

(i) Isolation of desired DNA (gene).
(ii) Isolation of Plasmid.
(iii) Insertion of desired gene into the plasmid DNA.

16.3.1 Isolation of Desired DNA (gene)

This is the first essential step in Genetic Engineering. Different methods are employed for the isolation of DNA from different groups of living organisms. Few of them are as follows:

(A) From Natural Source: Various steps are involved in isolation of DNA from cells. They are

(i) Cells are treated with a solution containing 0.14M Sodium chloride and 0.02M Sodium citrate.

(ii) Cells are homogenised well to break the cell walls.
(iii) The homogenate is centrifuged at 300rpm for about ten minutes to remove cell debris.
(iv) Precipitate is then treated with Sodium chloride solution to separate the DNA from histone protein. These histonic proteins are removed by centrifugation.
(v) The resulting (or) supernatant solution is treated with Ethanol drop by drop and stirred well with a glass rod. Stirring is continued until a white fibrous precipitate is formed around the glass rod.
(vi) Precipitate is then dissolved in Ethanol-Sodium chloride solution (or) washed with TE Buffer (Tris EDTA Buffer). The resulting solution contains chemically pure DNA's. These DNA's are stored in a cool place for further use.

(B) From mRNA: Many eukaryotic genes are made from mRNA's. Human insulin gene can also be obtained from mRNA. The synthesis of desired gene from mRNA involves the following steps.

(i) Firstly the mRNA is isolated from a relevant tissue. For e.g., The cells of islets of langerhans of pancreas are rich in insulin mRNA.

(ii) mRNA is then exposed to the enzyme reverse transcriptase. This enzyme synthesizes a single stranded DNA copy using mRNA as the template. The DNA synthesized on mRNA is called complementary DNA (C-DNA).

(iii) The RNA-DNA hybrid is subsequently treated with an alkali, which destroys the RNA strand, leaving the C-DNA strand intact.

(iv) A hair-pin loop present at the 3′ end of the C-NA strand acts as a primer for the synthesis of the second strand. For this, the enzyme DNA polymerase is added to single stranded C-DNA.

(v) Single strand hair-pin is then removed by treating with an enzyme S1 endonuclease.

16.3.2 ISOLATION OF PLASMID

Plasmids are commonly used as cloning vehicles. Plasmids are the extra chromosomal, circular, double stranded DNA molecules found naturally in many prokaryotes and in a few eukaryotes such as *yeast, Saccharomyces cerevisiae.*

Salient Features of Plasmid DNA's:

(a) Plasmid DNA is a circular, double stranded molecule.

(b) It is inherited from organism to organism without the influence of chromosomal DNA.

(c) Replication of plasmid DNA is independent of the replication of chromosomal DNA.

(d) Number of copies of plasmid DNA's present in each cell is constant for many generations.

(e) Plasmids can be single copy plasmids i.e., one plasmid DNA per cell (or) multi copy plasmids i.e., 10-20 plamid genomes per cell.

(f) Plasmid DNAs may carry very important genes like genes for antibiotic resistance, toxin production, antibody production and genes for nitrogen fixation. These genes confer to the phenotypic traits of plasmids.

(g) Plasmids propagate the DNA fragment linked to them by *in vitro* ligation.

(h) Plasmids have ability to clone reasonably large pieces of DNA say about 50Kb pairs.

(i) Plasmids can be used as cloning vectors or vehicles.

The isolation and separation of plasmids from the bacterial cell involves the following steps.

(a) Bacterial culture is treated with chloramphenicol which inhibits protein synthesis resulting a rapid increase in the number of plasmid DNAs in the bacterial cell.

(b) Then the cultured bacterial cells are treated with lysozyme and EDTA. These chemicals degrade the complex polysaccharides of bacterial cell wall.

(c) The wall-less cells are then lysed by treating them with Sodium Lauryl Sarcinate solution (SLS).

(d) The cell lysate is centrifused to remove the larger and smaller debris from the cell lysate. After centrifugation, the supernatant solution contains plasmid DNAs along with some fragments of chromosomal DNA.

(e) This supernatant solution is again centrifused with Cesium Chloride to remove chromosomal DNA fragments from the plasmid DNAs. The plasmid DNAs thus obtained are purified for further use.

16.3.3 INSERTION OF DESIRED GENE INTO THE PLASMID DNA

After the desired gene and plasmid are prepared, the desired gene is inserted into the plasmid. It involves the following steps:

(a) The plasmid DNA is cut with the help of restriction endonuclease enzymes. These enzymes function as chemical knives in Genetic Engineering. These enzymes are called restriction endonucleases because they cleave DNA with in the molecule at restriction sites.

Type II restriction endonucleases recognize and break DNA at a specific site. The enzymes make two types of breaks, for *e.g.*, the enzyme $EcoR_1$ cuts the DNA between G & A as indicated by arrows in the sequence. The staggered ends with complementary bases are called sticky ends (or) cohesive ends, because they can combine with similar sequences produced by the same enzyme on a suitable vector DNA.

5′ --- G AATTC → 3′

3′ ← C TTAAG --- 5′

↑

Other enzymes, such as Sma I derived from *Serratia marcescens* produce blunt ended DNA fragments, cutting between C & G.

5′ --- CCC | GGG → 3′

3′ ← GGG ↓ CCC -- 5′

(b) The desired foreign gene DNA is also cut with the help of the same restriction enzymes. As a result, the foreign DNA with identical ends is formed. Its ends are also identical with the ends of plasmid DNA.

(c) The broken ends of plasmid and foreign DNAs are mixed together. The sticky ends of foreign DNA and the plasmid DNA are linked together by complementary base pairing. This process is called ligation. As a result, the chimeric plasmid DNA is formed.

(d) The enzyme DNA ligase is added to seal the nick present in between the plasmid DNA and foreign DNA. For blunt end ligation, the enzyme T_4 DNA ligase is used. The chimeric plasmid DNA containing the desired foreign gene is called recombinant DNA (or) rDNA.

16.3.3 INTRODUCTION OF r-DNA INTO BACTERIAL CELL

After the construction of rDNA, it is introduced into a host cell. The most commonly used host in rDNA technology is *E. coli*. Under natural conditions the recipient cells are unable to uptake the chimeric plasmid DNAs. For this the bacterial cell is treated with $CaCl_2$ which renders the cell membrane permeable to plasmid DNA. The transformed bacterial cell is then grown on nutrient agar plates under optimum conditions for the development of colonies. As each colony is the progeny of a single cell, all cells will have the same genetic make up. This is called a clone. As per the entry of plasmid DNA into the bacterial cell, it should be protected by histone proteins of the recipient cells against the degrading activity of cellular endonucleases and exonucleases.

The rDNA gets integrated into the genome of *E. coli*. The *E. coli* now gets the property of the desired gene. The process is called **Transformation**. The transformation leads to the genetic manipulation of bacterial cells for synthesizing the product of the desired gene. For *e.g.*, if the inserted gene is insulin gene, the *E. coli* begins to synthesize insulin which can be harvested from the medium.

16.3.4 SELECTION OF CELLS CONTAINING RDNA

The cultured cells may contain transformed cells as well as the normal cells. The transformed cells are selected by two methods namely biochemical testing and colony hybridization.

1. Biochemical selection: Plasmid DNA possess some sequences to confer resistance for the organism against certain drugs like ampicillin, chloramphenicol, tetracycline etc. These sequences are used as genetic markers to detect the transformed recipient cells.

For example: PBR 322 plasmid possess 2 sequences to confer resistance to organism against drugs. First sequence gives resistance against tetracycline and the other sequence gives resistance against ampicillin. If the gene is cloned at the tetracycline resistance sequence, the plasmid fails to give resistance to the organism against tetracycline. So, these cells do not form colonies in an agar medium containing tetracycline.

2. Colony hybridization: In this technique, radio active probes containing complementary sequence of cloned gene are prepared. These radio active probes are used to establish hybridization on the nitrocellulose filter paper. The cloned genes forms complementary base pairing with radio active probe. As a result hybrid DNAs are formed. The presence of hybrid DNAs indicate the clone containing the transformed bacterial cells.

16.4 APPLICATIONS OF r-DNA TECHNOLOGY

Recombinant DNAs have the following applications:

- Recombinant DNA technology is used in the synthesis of vaccines against diseases such as hepatitis B virus, Malaria, Foot and Mouth disease of cattle.
- Recombinant DNAs are used in biology-based industries to produce amino acids, organic acids, alcoholic beverages, etc.
- Transformed bacterial cells are also used in manufacture of steroid hormones.
- Genetically engineered microorganisms are used in production of biogas.
- Recombinant DNAs are used to make transgenic plants, transgenic animals and genetically engineered micro organisms (GEMO).
- Human growth hormone and Human insulin hormone are produced by *E. coli* containing rDNA.
- Rat insulin is also produced by genetically engineered *E. coli* cells.
- Colonbacilli, having rDNA can produce human L-interferon which acts against viruses.
- r-DNAs are also used in DNA sequencing in chain termination method.
- Site directed mutagenesis can be bought about by rDNA technology.
- Details of various diseases such as Down's syndrome, coronary artery disease, orthosclerosis, cytogenetic abnormalities, inborn errors of metabolism were revealed by rDNA studies.
- Recombinant DNA technology is also used in animal husbandry.

16.4 SOMATIC HYBRIDS AND CYBRIDS

Protoplasts are nothing but original plant cells which lack cell wall. They are prepared from plant cells by removing their cell wall. Protoplasts can be isolated by mechanical method or enzymatic digestion of plant cell wall. Takebe et al first employed the use of cellulase and maceroenzyme for the isolation of mesophyll protoplast of Tobacco. Isolation and culture of protoplasts are important steps in crop improvement.

Isolated protoplasts have a tendency of fusing with one another. As a result of this fusion, hybrids and cybrids are formed which are of very much important in crop improvement. The hybrid protoplasts are called somatic hybrids and the phenomenon is called as Somatic Hybridization. The somatic hybrid contains cytoplasm and nuclei of both the parent protoplasts.

In few instances, fusion takes place between the plastome (cytoplasm) of one protoplast and genome of other protoplast leading to the formation of cytoplasmic hybrids or cybrids. Cybrids when compared to hybrids possess a nuclear genome of one parent and cytoplasmic genes of both the parents. The fusion of protoplast resulting in the development of cybrids is known as Cybridization.

Cybrids can be obtained by using any one of the following methods.

(i) Fusion of normal protoplast from one parent with enucleated protoplasts of other parent.
(ii) Selective elimination of one of the nuclei from the heterokaryon.
(iii) Fusion of normal protoplast of one parent and protoplast, containing non-viable nucleus of the other.

16.6 SOMATIC HYBRIDIZATION

Somatic hybridization and fusion of protoplasts in plants are based on the following principles.

16.6.1 PROTOPLAST ISOLATION

Protoplasts can be isolated from a variety of plant tissues. The most convenient and suitable materials are leaf mesophyll and cells from liquid suspension culture. The important step of isolation includes the removal of cellulose cell wall without damaging the protoplast. Two methods are widely employed for the isolation of protoplasts. They are (i) Mechanical method (ii) Enzymatic method.

(i) Mechanical method: Mechanical isolation of protoplasts from higher plants was first introduced by Klercker in 1892 and is employed for the isolation of protoplasts from highly vacuolated cells of storage tissues such as onion bulbs, radish roots, beet root, cucumber mesocarp, etc.,

In this method, cell walls are mechanically removed with the help of knife, scissors, microscalpels, needles and forceps. For this, cell (or) tissue is kept in a hypertonic solution for a few minutes. This solution brings about exosmosis which leads to shrinkage of protoplast. As a result, the protoplast of the cell is physically separated from the cell wall. Now the cell wall of plasmolysed cell is broken with the help of micro scalpel and needle or with a knife under the microscope. The isolated protoplast is then transferred to isotonic liquid medium to prevent damage. But this method has the following disadvantages:

- Requires long time
- Only a small amount of protoplasts can be prepared.
- It must be repeated again and again to isolate viable protoplasts.

(ii) Enzymatic Method: Here the protoplasts are isolated by enzymatic digestion of plant cell wall. The cell wall degrading enzymes such as cellulase, hemicellulase, pectinase (or) macerozyme are used for digestion. These enzymes are available commercially and are isolated from Fungi.

The procedure involves the following steps.

- Healthy young leaves are collected from the upper parts of 7-8 week old plants and thoroughly washed with tap water.
- These leaves are surface sterilized with 10% ethanol and 2% sodium hypochlorate and washed with distilled water.
- The lower epidermal layer of the leaf is carefully peeled off with needles (or) scalpel and the mesophyll tissue is cut into small pieces.
- They are immersed in a hypertonic solution to plasmolyse the plant cells.
- Later, the leaf pieces are kept dipped in an enzyme mixture containing 0.5% macerozyme, 27% onazuka cellulase and 13% sorbital or mannitol. The enzyme mixture is then incubated at 25°C over a night.
- After incubation, the leaf pieces are gently pressed with fine forceps to liberate the protoplasts from the intact plant tissues. Then the mixture is filtered through a fine wire guaze to remove the cell wall debris from the enzyme mixture.
- The solution containing protoplasts is centrifuged for 5mts to get protoplasts at the bottom of the centrifuge tube.
- The protoplasts are again transferred to the enzyme mixture and incubated over a night. This process is repeated 2 (or) 3 times to produce pure protoplasts.
- The protoplasts are washed with a solution containing 13% sorbitol and 20% sucrose solution.
- It is centrifuged then the pure protoplasts float on the sucrose–sorbitol solution and the debris settle down at the bottom of the centrifuge tube.
- Protoplasts are carefully separated from the tube and stored for future use.
- In this method only one type of enzyme mixture is used, hence it is called single step enzymatic isolation.
- Two step enzymatic isolation involves two enzyme mixtures.

16.6.2 PROTOPLASTS FUSION

Protoplast fusion refers to the fusion of isolated protoplasts of two different plant species. It is also known as somatic cell hybridization (or) Somatic Hybridization. The product of protoplast fusion is called Hybrid protoplast. Plants, raised from hybrid protoplasts are known as somatic hybrids. Protoplast fusion helps to overcome sexual incompatibility between two closely related species (or) varieties. Protoplast fusion is a tool for gene manipulation in plants.

Protoplast fusion can be classified into two types. They are as follows:-

(a) Spontaneous fusion of protoplasts: During enzymatic isolation, protoplasts often fuse spontaneously and the phenomenon is called spontaneous fusion. When the cell walls are enzymatically degraded, the plasmo desmatal connections between adjacent cells enlarge. This enlargement of plasmodesmata allows the entry of organalles into neighbouring cells. As a result, hybrid protoplast is formed. The spontaneous fusion is strictly intraspecific and gives rise to homokaryons.

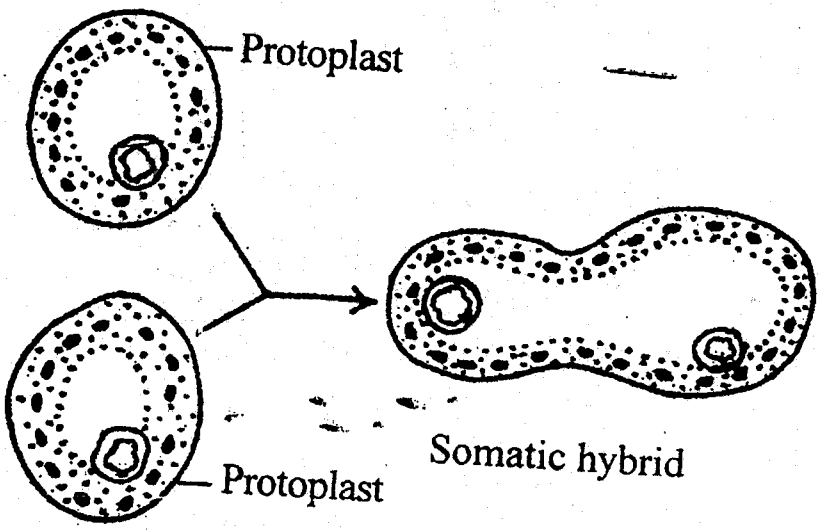

Fig. 2. Diagram showing protoplast fusion

(b) Induced fusion of protoplasts: Normally isolated protoplasts possess negative charge outside the plasma membrane. As they carry negative charge they do not fuse with each other and need some fusion inducing chemicals called **Fusogens** or mechanical pressure. It is again of 2 types.

(1) Chemical Method: Here some chemical substances called **fusogens** are used to induce fusion of protoplasts. Sodium nitrate, polyethylene glycol, calcium ions, polyvinylalcohol, lysozyme, artificial water, antibodies, dextran are the most commonly used fusogens. Inactivated Sendai virus is used to induce fusion in animals.

Mild electric stimulation is the recent technique employed for fusion of protoplasts called as Electrofusion. Zimmerman and Scheurich improved this method for large scale fusion of plant protoplasts.

(2) Mechanical Method: In this method, protoplasts of two different plant varieties are sucked in a micro pipett whose mouth is partially blocked and forced out quickly. The rapid flow of protoplast suspension through the partially sealed mouth create a mechanical force. This force induces adhesion of adjacent protoplasts tightly. As a result two protoplasts fuse together and form a hybrid.

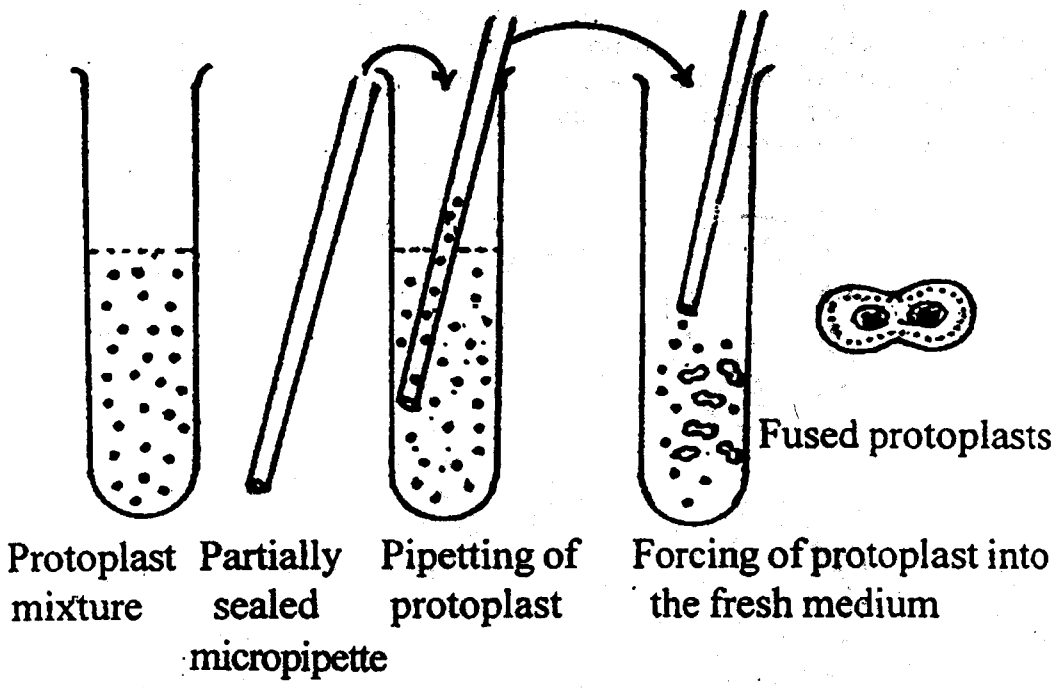

Fig. 3. Mechanical induction of protoplast fusion

16.6.3 Identification of Somatic Hybrids

After fusion treatments, the protoplast mixture contains a number of hybrid protoplasts and unfused parent protoplasts. The hybrid protoplast shows heterogenecity in its genome content and are useful for crop improvement. Chromosomes and the cell inclusions of one parent protoplast are gradually eliminated in some non viable protoplasts. As a result, the hybrid protoplast contains the nucleus of one parent protoplast and the cytoplasm of both protoplasts. Such somatic hybrids are called Cybrids. Thus, the somatic hybrid and cybrid differ by the presence of two nuclei and by the presence of a single nucleus respectively.

The preliminary identification of fusion product is done under a microscope. Identification is based on visible characters of protoplasts, like colour of protoplast, presence of chloroplasts and other pigments. In most of the hybridization experiments, green coloured and colourless protoplasts are used. This facilitater easy identification of fused protoplasts from unfused parental protoplasts. If both types of parental protoplasts look alike i.e., either colorless or pigmented, then the fusion product can be distinguished using nuclear staining technique. Recently, Patnaik (1982) used the dye fluorescein diacetate to detect the hybrid protoplasts.

17.7 POST FUSION EVENTS

The selected hybrid (or) cybrid protoplasts are transferred to a fresh culture medium. The nutrient medium generally contains nutrients similar to those required for callus and suspension cultures. Viable hybrid protoplasts in the culture medium regenerates a wall around

them and enter into mitotic cycle. Successive divisions result in the formation of a callus tissue. Complete hybrid (or) cybrid plants can be regenerated from callus tissue.

17.8 SUMMARY

Recombinant DNA is a hybrid DNA formed by joining of atleast two different fragments of DNA. The production of r-DNA is called Genetic Engineering. It contains 5 important steps they are (1) isolation of desired gene (2) isolation of desired vector (3) preparation of r-DNA (4) introduction of r-DNA into the suitable host and (5) selection of desired strains. Industrially so many valuable products such as insulin, growth hormone, interferans, amino acids, organic acids, steroid harmones, have been produced using this technology. Somatic hybridization is a process, carried between sexually incompatible strains. It a fusion or breeding between two somatic cells. Isolation of protoplast is the first step of somatic hybridization, followed by protoplast fusion and identification.

16.9 TECHNICAL TERMS

Genetic Engineering, Hybrid, Cybrid, plasmid, reverse transcription, Restriction endonucleases, Ligation, Colony hybridization, Protoplast isolation.

16.10 MODEL QUESTIONS

Essay type

(1) Write a detailed essay on r-DNA technology?
(2) What are hybrids, write the process of somatic hybridization?

Short notes

(1) Applications of r-DNA technology
(2) Somatic hybrids & cybrids
(3) Isolation of desired genes
(4) Vectors.

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- S. HARI KRISHNA RAMA PRASAD

Lesson 17

APPLICATIONS OF BIO-TECHNOLOGY

17.0 OBJECTIVES

(1) Students understand the significance of Bio-technology in the fields of Agriculture, Medicine and Industry.
(2) Students comprehend the importance of Bio-technology in the man's life.

STRUCTURE

17.1 INTRODUCTION

The term '**Biotechnology**' was coined by Carl Ericay in 1919. It consists all large scale productions carried out by using living organisms especially microorganisms. In the year 1978, European Federation of Biotechnology defined Biotechnology as "The integrated use of scientific and engineering principles in the processing of materials by using biological agents to produce valuable goods and services".

The common and widely used definition for Biotechnology is "Application of Engineering Principles in the field of biology to produce valuable products by using microorganisms".

In ancient times, biotechnological methods were employed in various fields such as agriculture, brewing industry, preparation of milk products etc., without knowing the scientific principles. Uses of biotechnology is well known for a long time in the process of fermentation with the help of microoragnisms, antibiotic production by certain microorganisms especially fungi and moulds.

Novel methods, research and development, advanced studies in microbiology and genetic engineering results opened new areas where microorganisms can be exploited for commercial production of industrially valuable products. Some of the important biotechnological products are alcoholic beverages, steroids, vaccines, antibiotics, vitamins, enzymes, pesticides, biofertilizers, food products, health products, aminoacids etc.

Biotechnological methods are employed in many fields. They are mainly differentiated into three important categories namely – Agricultural Biotechnology, Medical Biotechnology and Fermentation Technology.

17.1.1 AGRICULTURAL BIOTECHNOLOGY

Agricultural Biotechnology deals with the genetic improvement of crop plants to get high yield. Such genetically improved plants are disease resistant, resist to adverse climatic factors, and also herbicide resistant. This branch also explains the method of culturing microorganisms which can be used as Biofertilizers.

17.1.2 Biofertilizers

The carrier based microbial inoculants added to the soil to enrich the soil fertility are called biofertilizers. They are often known as Microbial fertilizers (or) Microbial inoculants. A Biofertilizer may contain nitrogen fixing microbes or phosphates olubilizing microbes or spores of VAM fungi. It is supplied to the soil either by seed treatment or by spreading it over the field during cultivation. Biofertilizer reduces the use of chemical fertilizers in agriculture and cost of production.

Nitrogen fixing Bacteria, phosphate solubilising bacteria, Spores of VAM fungi are the commonly used Biofertilizers.

17.1.3 Nitrogen fixing organisms

The nitrogen biofertiliser may have nitrogen fixing bacteria (or) Blue-green algae. Nitrogen fixing bacteria include Rhizobium, Azospirillum, Azatobacter, Azotococcus etc. Blue green algae such as Anabaena, Aulosira, Nostoc, Plectonema and Tolypothrix are used as nitrogen biofertilizers. Atmospheric air contains about 78% of nitrogen and is not readily available for plants. Nitrogen is the important macronutrient required for plant growth, but very few microorganisms namely nitrogen fixing organisms convert the molecular nitrogen into organic nitrogen. These nitrogen fixers are of two types -- symbiotic or non-symbiotic.

Symbiotic nitrogen fixers are Rhizobium, Anabaena etc. These organisms develop an association with the roots of higher plants and fix the atmospheric nitrogen in the soil. Examples of non-symbiotic nitrogen fixers are Azatobacter, Azotococcus, Klebsiella etc. They live freely in soil and fix nitrogen in the soil.

In nitrogen fixers, nif-genes play key role in fixing the atmospheric nitrogen. Now scientists are trying to transfer nif genes to non-nitrogen fixers using Recombinant DNA technology. Thus, *E. coli, Salmon ella, Typhimurium, Pseudomonas, Serratia marcenscens, Proteus mirabilis, Erwinia herbicola,* Yeasts have been genetically transformed to fix atmospheric nitrogen in soil. Cloning of nif-genes was first achieved by plasmid (PWK 120) mediated conjugation. Attempts are now being made to convert the genetic systems of cereals such that they fix atmospheric nitrogen in soil.

17.1.4 Phosphate Solubilizing Microorganisms / Phosphate Biofertilizers

Microbes which solubilise the bound phosphates and rock phosphates into simple soluble phosphates are called phosphate solubilizers, phosphate solubilizing bacteria (or) phosphate bacteria. They secrete organic acids such as propionic acid, formic acid, acetic acid, lactic acid, succinic acid and hydroxy acids to solubilize the bound phosphates in the soils. Phosphate is one of the most important macronutrient required for growth of plants. But plants absorb only in the form of soluble phosphate ions. As phosphate solubilizers solubilize the bound phosphates, the simple phosphates are readily available for the plants.

Phosphate biofertilizer can save 30-50 kg of super phosphate per hectare and increase the yield upto 200-500 kg/hectare.

Examples of important phosphate biofertilizers:

Bacteria	**Fungi**
Bacillus megaterium var *phosphaticum*	*Aspergillus awamorii*
Bacillus subtilis	*Aspergillus flavus*
Bacillus striata	*Aspergillus niger*
Bacillus pulvifaciens	*Aspergillus fumigatus*
Pseudomonas striata	*Pencillium digitatum*
Pseudomonas rathonis	
Pseudomonas liquifaciens	
Escherichia intermedia	

Phosphate biofertilizer is suited for all types of crops. They provide 20-30% of phosphate required for the crops.

17.1.5 Vesicular Arbuscular Mycorrhiza (VAM)

Vasicular arbuscular mycorrhizal fungi are a group of symbiotic, endotrophic mycorrhizal fungi found in roots of higher plants. They are included in the family Endogonaceae of

Zygomycetes. VAM fungi form a symbiotic association with the root system of higher plants, such as angiosperms, gymnosperms and pteridophytes, VAM fungi infects a plant root and forms vesicles and arbuscles in the root cortex, a permanent mantle of hyphae on the roots surface. They infect many plantation crops like rubber, coffee, tea, papaya, crop plants like rice, maize, potato, soyabean, cotton, tobacco, sugarcane, tomato, strawberry, citrus, avacado, pear, apples etc. VAM fungi play a very important role in plant growth. They increase the plants absorption capacity for zinc, phosphate, water, sulphur etc. VAM infected plants show increased rate of growth.

Apart from the VAM, sea weeds are also used as Biofertilizers. E.g. Sargassum, Laminaria, Gracillaria, Enteromorpha, Macrocystis, etc. They are used as green manure in the cultivation of wheat, potato, citrus, palm trees etc. These sea weeds possess nitrogen and potassium in their cells and hence they increase the quality and yield of plants. They also contain the elements like cobalt, manganese, boron, barium, iodine etc.

17.1.6 Biopesticides

Many microorganisms are known to be pathogenic to insects. These microbes are called as Entomopathogens. They include viruses, bacteria, fungi and protozoa. Biotechnology helps in production of these microbes in large number. Some commercial microbial insecticides are *Bacillus thuringiensis, B. sphaericus, Beauveria* spp., Nuclear polyhedrosis virus etc.

17.1.7 Tissue culture

The *in vitro* culture of plant cells or tissues in artificial medium is said to be plant tissue culture. It has many applications in crop improvement, preservation, breeding and in industries.

Tissue culture helps in rapid production of large number of identical clones *in vitro* within a short duration. This is called Micropropagation. Meristem culture helps in the production of pathogen free plants. Haploid plants can be raised from anther, pollen and ovule cultures. Haploids are used in plant breeding in order to improve the field and agricultural crops. Nearly 250 haploid species are established through anther culture method. e.g. Rice, Rye, Wheat, Potato, Tomato, Tobacco etc.

Somatic hybrids and cybrids are raised through tissue culture. These are nothing but the fusion products of protoplasts of two different strains (or) species. Somatic hybridization overcomes sexual incompatibility among plants and also used to raise new crop varieties.

Somaclonal variants are obtained through tissue culture. Generally, clones obtained through tissue culture show uniformity in their characters. However, a few clones show variations among the clones. They are called Somatic variants. The formation of variant clones from the cultured callus tissues is called somaclonal variation. Somactoral variation is used to develop new strains of plants with some noval character. Variations have been raised from plants like potato, cereals, tobacco, sugarcane etc.

17.1.8 Plant Protection

Biotechnology helps in protecting the plants from several environmental factors, diseases and pests through r-DNA technology by employing microorganisms.

Stress Tolerant Plants

Difficulty for the plants to survive is called stress. It is caused by extreme cold, heat, drought, etc. Efforts are being made to produce trangenic plants resistant to a variety of stresses. Stress tolerance is important for occurrence of a species in a wide range of climates. So far many cold tolerant, drought tolerant plants have been produced.

Herbicide resistant plants

Plants that can tolerate herbicides are called herbicide resistant plants. Herbicide resistance of main crops helps us for effective use of herbicides to control weeds. So transgenic plants with herbicide resistance have been developed by adopting genetic engineering. E.g. Glycophosphate resistance petunia, tomato, corn, etc.

Disease resistant plants

Trangenic viral resistant, fungal resistant and bacteria resistant plants are produced through genetic engineering.

17.2 MEDICAL BIOTECHNOLOGY

Medical Biotechnology deals with the production of immunologically active substances such as antibiotics, interferons, human growth harmones, vaccines, insulin, etc. These compounds play an essential role in diagnosis and treatment of some serious and dangerous diseases.

17.2.1 Interferons

Interferon is a protein produced by cells when they are infected by a virus for the first time and protects the cells against the second viral infection.

Isaacs and Lindermann first isolated interferons from the blood of patients suffering from viral attack in 1957. Interferons are very small protein molecules with the molecular weight of 20,000-34,000 daltons. They are sensitive to proteolytic enzyme such as Trypsine.

Inteferons are antiviral agents. They inhibit the invading viruses. Gillbert and Weismann, for the first time, produced genetically engineered human leukocytic inferferons in 1980. The US company Biogen started to manufacture the α-interferon in a large scale. The American Cancer Society made use of leucocytic interferons in the treatment of breast cancer γ-interferons are used in the treatment of renal carcinoma in man. β-interferons are used in the treatment of cancers of lymphnodes and lymphoma. Interferon A is used to treat hepatitis C.

17.2.2 Insulin

Insulin is a hormone secreted by the beta cells of the islets of langerhant of pancreas. It is essential for oxidation and utilization of blood sugar and for the maintenance of proper blood sugar level.

Inadequate secretion of insulin leads to hyperglycemia. This condition is said to be diabetics. The treatment of diabetics by injecting insulin is named insulin therapy. The antidiabetic role of insulin was first discovered by Sir F.G. Banting in 1922. In 1982, Eli Lilly (USA) started to produce human insulin from genetically engineered bacteria. Genentech Company in the USA chemically synthesized insulin gene. Gillbert and Villa-Komaroff in 1980 isolated insulin mRNA from beta cells of pancreas of rat and synthesized duplex complementary DNA. Rat insulin is also used to treat diabetics.

17.2.3 Monoclonal Antibodies

A single type of antibodies having the same antigenic determinant produced by a single hybridoma clone is called monoclonal antibody. The hybridoma is made by fusing a lymphocyte (B cell) with a myeloma cell. Presence of a single antigenic determinant is the useful feature of the monoclonal antibodies.

Monoclonal antibodies were first made by Milstein *et al.*, in 1973. They are used in the diagnosis and treatment of severe diseases. In genetic engineering, monoclonal antibody is used to screen recombinants. They are also used to determine the structure of cell membranes. Monoclonal antibodies are employed in serological classification of closely related bacteria, viruses and protozoans. They are used in RIA, ELISA and immunoflorescence assay in research to identify and detect some target products.

17.2.4 Vaccines

A suspension of killed or modified live virus or bacteium being injected into the body to stimulate immunity against the pathogen is called vaccine. The process of injecting a vaccine into the body is known as vaccination. Vaccinated body starts to produce antibodies against the antigen present in the vaccine. The antibodies act as a defence system and give protection to the body against the particular pathogen. This type of immunity is called Acquired immunity. Vaccines are used to prevent certain infectious diseases. E.g. Vaccines for hepatitis, herpes, measles, polio, tuberculosis, malaria, leprosy, rhematic fever etc.

Genetically engineered vaccine contains only a part of pathogen that stimulates the immunity. It contains one or a few antigens of the pathogen. As it is a subunit of pathogen, it is known at subunit vaccine. Subunit vaccines are otherwise calied Biopharmaceutical vaccines. Biotechnology has been used to raise vaccines in large quantities. Hepatitis-B virus was isolated and cloned in yeast cells for large scale production of vaccine.

3.0 Industrial Bio-technology

3.1 Fermentation Technology

Fermentation is an anaerobic breakdown of complex organic material by the action of anaerobic microorganisms or free enzymes. But now-a-days the word fermentation is not restricted to anaerobic break down of organic substances. It also denotes some other process in which the breakdown of organic substances taken place in the presence of some low amount of O_2. Fermentation results in a variety of products.

3.2 Amino acids

These are the building blocks of proteins. They are used in various industries.

Different kinds of organisms have the property of producing amino acids in the culture medium. This property of microbes is used in industries to produce amino acids.

Amino acid		Source organism
L – Glutamic acid, Glutamine		- *Corynebacterium glutamicum, Brevibacteium flavum, B. thiogenitalis, Microbacterium ammoniaphilum*
L – Lysine	-	*E. coli, Aerobacter aerogens, Aspergillus uster, Brevibacterium flavum.*
L – Threonine	-	*E. coli, Serratia marcescens, Candida guilliermondii, Corynebacterium aceto-acidophylum*
Phenylalanine	-	*Mutant of Brevibacterium lactofermentum*
L – Serine	-	*Corynebacterium glutinophilum*
L – Isoleucine	-	*Corynebacterium glutamicum (Mutants) Brevibacterium flavum*
L – leucine	-	*Brevibacterium lactofermentum*
L – valine	-	*Paracolobactrom coliforme, Aerobacter aerogens, Bacillus subtilis, Serratia marcescens.*
L – Arginine	-	*B. subtilis, Corynebacterium, glutamicum, Brevibacterium flavum, Serratia marcescens*
L – Histidine	-	Mutant of *Corynebacterium glutamicum*
L – Alanine	-	*Corynebacterium gelatinosum,*

		Brevibacterium monoflagellum *Micrococcus sodonensis*
Tyrosine	-	Mutant of *C. glutamicum*
Proline	-	Mutant of *Corynebacterium glutamicum*
L – cystine	-	*Pseudomonas*
Methionine	-	*Corynebacterium glutamicum*
Tryptophan	-	*Bacillus subtilis, Candida utilis, E. coli, Proteus rettgerii.*

17.3.3 Alcohols

Alcohols are produced during the fermentation carried by bacteria. The microbes produce ethanol, methanol and butanol in the medium. *Bacillus granulobacter* produces alcohols in the medium containing diluted corn-starch solution. It produces butanol, acetone and ethanol in the ratio 6 : 3 : 1. D.I. Wang used *Clostridium thermocellum* and *C. thermosaccharolyticum* for producing ethanol from cellulose and sugar.

Alchols are now used as fuels in many industries. They are called Biofuels.

17.3.4 Organic Acids

Different groups of organisms produce organic acids in the culture during their growth. Some important organic acids and corresponding process organisms are listed below:

Organic acid	**Process organism**
Aconitic acid	*Aspergillus itaconicus*
Gortic acid	*Penicillium charlesii*
Citric acid	*Aspergillus niger, P. citratum*
Formic acid	*Rhizopus, Mucor*
Glyceric acid	*Aspergillus niger*
Lactic acid	*Rhizopus, Mucor, Lactobacillus, Streptococcus lactis*
Malic acid	*A. fumaricus, A. niger*
Oxalic acid	*A. niger*
Propionic acid	*Propionibacterium jensenii, P. rubrum, Proteus mirabilis.*

17.3.5 Antibiotics

A wide class of bacteria and fungi produce antibiotics in the culture. They are used in the treatment of various pathogenic diseases. The different process organism and their products are listed below:

Name of Antibiotic	Process organism
Amphotericin-B	*Streptomyces nodosus*
Rifamycin	*Nocardia mediterrnea*
Tetracyclin	*Streptomyces rimosus*
Chloromycetin	*S. venezuelae*
Actinomycetin	*S. erythreus*
Erythromycin	*S. erythreus*
Carbomycin	*S. halsteidi*
Minocyclin	*S. aureofaciens*
Pencillin-G	*Penicillium crysogenum*
Novobiocin	*Streptomyces* sps.
Kanamycin	*Streptomyces kanamyceticus*

17.3.6 Alcoholic Beverages

A number of beverages like wine, vinegar, beer, brandy, wisky, rum are produced by fermentation. Production of beverages is purely a microbial process.

Beer is produced by the action of *Saccharomyces cerevisiae* on grain flour. Larger beer is produced by action of *Saccharomyces carlbergensis*. Wine is also produced by the action of *S. cerevisiae* Whisky & Brandy are produced by action of *S. cerevisiae* on marsh grain of corn, rye, malt and grape juice, apple, orange, peech. Rum is produced by action of *Clostridium saccharobutyricum* on Cane molasses (or) Beet molasses.

17.4. SUMMARY

Application of engineering principles in the field of biology to process the goods using microorganisms is called Biotechnology. It brings revolution in the biology especially in the field of agriculture, medicine and industry. In agricultural field, it brought alternative fertilisers named biofertilizers to increase soil fertility, bio-pesticides to control pest population and to increase yieldings.

Similarly it introduced tissue culture technique tc develop disease resistant and high yielding varieties. In medicine different medicines, interferors, insulin, monoclonal autobodies and vaccines have been developed to combat against viral, bacterial and other diseases. In

industries through fermentation technology, it has given different industrial products such as aminoacids, alcohols, organic acids, antibiotics and alcoholic beverages.

17. 5. TECHNICAL TERMS

Interferons, monoclonal antibodies, bio-fertilizers, bio-pesticides, vaccines, fermentation, phosphate solubilizing microorganisms, VAM etc.

17.6 MODEL QUESTIONS

Essay Type

(1) Write a detailed essay on applications of Biotechnology.

(2) Explain the role of bio-technology in the field of Agriculture and Medicine.

Short Notes

(1) Production of Mab's

(2) Interferons

(3) Bio-fertilisers

(4) Vaccines

(5) Plant protection

(6) Bio-pesticides

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Unit- IV
Lesson 18

PLANT TISSUE CULTURE – EMBRYO CULTURE, ANTHER CULTURE, MERISTEM CULTURE AND APPLICATIONS OF TISSUE CULTURE

18.1 OBJECTIVES

* Students learn about tissue culture and different steps involved in it.
* Students will know the significance of tissue culture by studying its application.

Structure

18.1 INTRODUCTION

Zygote (2x) is the first cell of plant life cycle. It is the resultant of fusion between male and female gametes. It further undergoes divisions and form many cells. These cells are differentiated into organs and ultimately plant body. If the entire plant body is developed from a single cell, why not each somatic cell of the developed plant can give a new plant. This, very idea motivated G. Haberlandt to develop "Tissue culture technique". But the cells which have totipotency nature can only give new plants when they are grown separately on culture medium. Totipotency means the genetic information required for the growth and development of an entire plant. The concept of totipotency was first put forward by Schleiden and Schwann (1838).

Development of a plant by culturing a single cell is called **single cell culture**, similarly culture of a tissue or an organ to give a plant is called **tissue culture** or **organ culture** respectively.

18.2 STEPS INVOLVED IN TISSUE CULTURE PRACTICE

Five important steps are involved in the tissue culture practice. They are

(i) Selection of explant
(ii) Preparation of Nutrient medium
(iii) Transfer of explant on the medium
(iv) Incubation of the culture and
(v) Acclimatization.

(i) Selection of explant

The cell or tissue or organ which is used as source material for getting a plant, is called 'explant'. Basing up on our need the explant is selected from healthy growing plant and it must be sterilized. For sterilization firstly explant is thoroughly washed with liquid detergent like teepol (5% v/v) and after 10 or 15 minutes it is washed with distilled water. Then it is surface sterilized with chemical disinfectant such as Bromine water (or) chlorine water (or) H_2O_2 (or) sodium hypo chloride in harmless concentrations. Soft explants can be sterilized with ethyl alcohol or iso propyl alcohol for just a few seconds.

(ii) Preparation of Nutrient Medium

All the essential nutrients, required for plant growth are taken as a single unit, called medium. Medium with only carbon source, nitrogen source and vitamins is called **Basal medium** and the medium, which is supplemented with growth harmones along with the above material is called **enriched medium**. Different media have been used by different scientists but a single medium is not useful for the entire process of tissue culture. For example basal medium is helpful upto callus formation (undifferentiated cell mass). For further growth enriched medium is needed. Some important media prepared by various scientists for tissue culture practices are – M.S. Medium (Murashige and Skoog), B5 Medium (Gamborg), White's medium, S.H. Medium (Schenk and Hilde brant). It clearly tells all the media are named after the discoverer. Generally an ideal medium contains macro nutrients, micro nutrients, carbon source, vitamins and amino acids. All these ingredients are measured in required concentrations and dissolved in distilled water and P^H must be adjusted to neutral. The total volume of medium would be one litre. This prepared medium is sterilized with the help of autoclave. Thermolabile substances in the medium can be sterilized separately by filter sterilization, means these materials are filtered through bacterial filters.

(iii) Transfer of explant on to the medium

The transfer of sterilized explant on to the nutrient medium is called *inoculation*. The inoculation must be carried in sterilized environment. Generally laminar air flow cabinet is used for this work, which keeps its working area as sterile as possible and controls contamination.

(iv) Incubation of the culture

After inoculation the culture medium with explant is incubated in culture room, where controlled conditions are provided. Under controlled conditions explant uses nutrients, divides and form 'callus tissue'.

(v) Acclimatization

The callus tissue is transferred to enriched medium, where callus differentiates into small plantlets. The plantlets are properly acclimatized, keeping them in green house, later they are transferred to real conditions.

18.3 ORGAN CULTURE

Culturing of a plant organ on nutrient medium to develop an entire plant is called organ culture. On the basis of the origin of the explant, organ culture is categorised into 2 types (1) Vegetative organ culture (2) Reproductive organ culture. In this chapter reproductive organs culture is discussed.

18.3.1 Embryo Culture

Embryo culture is defined as culturing of immature (or) mature embryos into plantlets in a nutrient medium. It is done in the plants in which fertilization takes place as usual but the fertilized egg fails to develop into mature embryo. This embryo culture method overcomes the non viability of seeds, hence called as Embryo rescue.

For the growth of immature embryos artificially in the laboratory, requires complex medium containing all inorganic salts, sucrose and hormones. Isolated embryo is placed on a filter paper bridge in a nutrient medium in a tube and incubated for a weeks. During incubation the embryo develops into a plantlet.

Uses of Embryo Culture: Embryo culture is used to raise plants from seeds having immature embryos during seed dispersal, *e.g.*: Banana, Orchids.

- ⇒ Also helps to overcome abortion of embryos at early stages.
- ⇒ Produces disease free plants.
- ⇒ Overcomes seed dormancy.
- ⇒ It is an ideal test for assessing the seed viability.

18.3.2 Anther Culture

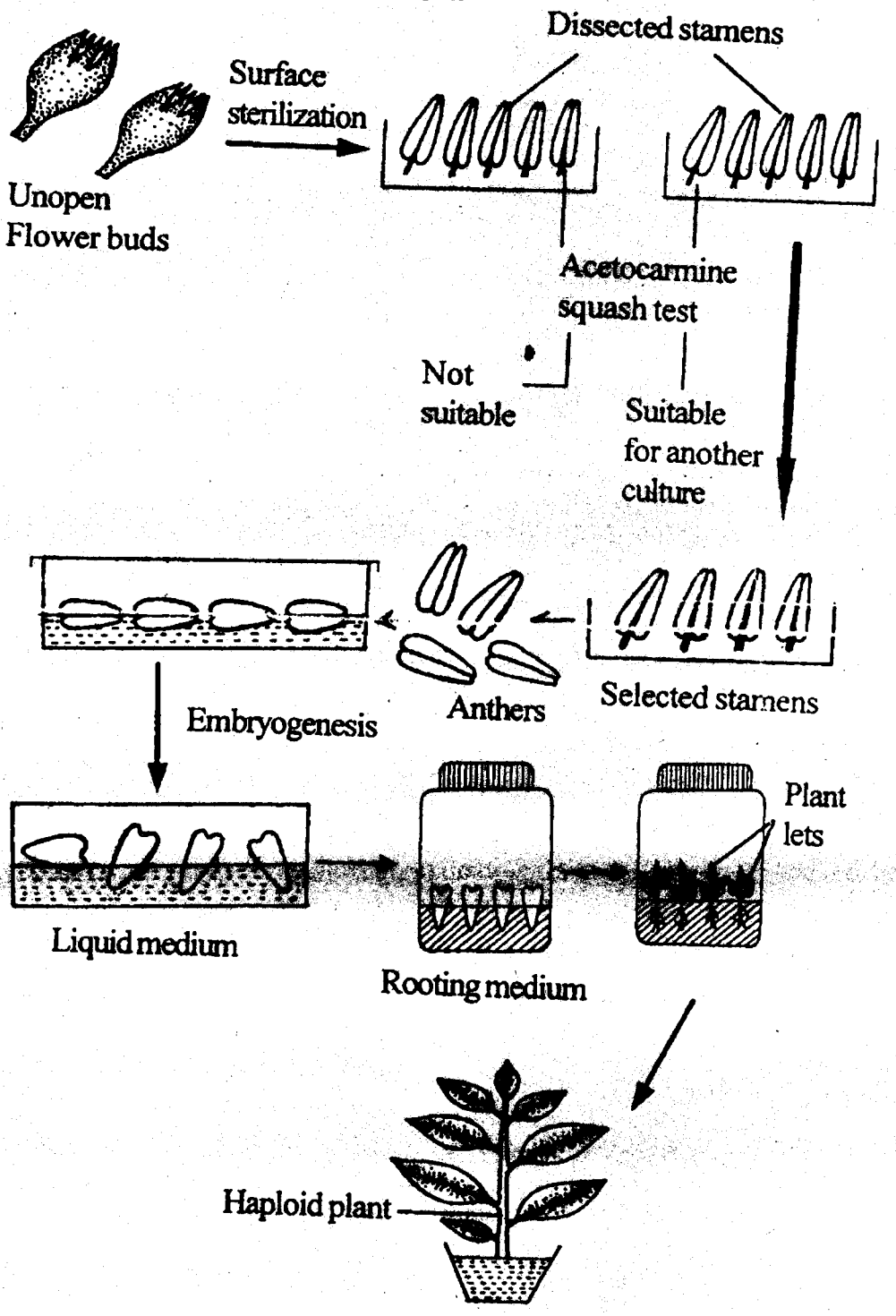

Fig. 1. Stages in anther culture

The culture of immature anthers into embryos is called Anther Culture. It was first done by Guha and Maheswari in Tobacco plants. Anther culture give raise to Haploid plants. Through anther culture about 250 species of dicot and monocot plants are regenerated. e.g.: Coconut, Rubber, Tobacco, Maize, Wheat etc.

Diploid cells are seen in young anthers. Diploid cells undergo meiosis to form haploid tetraspores. The haploid tetraspores get dedifferentiated into haploid parenchyma cells in nutrient medium. Thus a mass of parenchyma cells are produced in the anther. This cell mass becomes circular and forms a globular embryo which later develops into heart shaped embryo. Embryo attains the tarpedo shape finally. The stages in the development of embryoid from the anther looks like the development of zygotic embryo.

Procedure for Anther Culture

Unopened flower buds are collected from plants, then sterilized with tween 80 mercuric chloride solution and washed with distilled water. Anthers are taken from flower buds carefully and kept in petri dish. All the anthers are collected from flower buds and kept in separate petri dishes. Anthers just completed first meiotic division are selected by autocarmine, squash test. Selected anthers are aseptically transferred to a liquid or solid nutrient medium containing glutamine, L-serine and inositol and incubated at 25°C for about 15 days. During incubation anther grow into embryoids. Embryoids are then transferred to a rooting medium in the culture plates. 3000 lux illumination is given to the culture. Embryoids become plantlets into 4-5 weeks. Well developed haploid plantlets are planted in pots in a green house for proper acclimatization.

Uses of Anther Culture

⇒ Haploid plantlets are produced through anther culture.
⇒ These haploids contain reduced number of genes, hence used in the study of metabolic path ways.
⇒ Anther culture is of much use in *in vitro* mutagenesis for crop improvement.
⇒ The haploid embryoids are immersed in colchicine for 2 days and grown into plants in vitro. The resulting plants are diploid and fertile.

18.3.3 Meristem Culture

The growing tip of shoots is termed Meristem. It is often called as shoot apex. Shoot meristem has totipotency necessary to regenerate into a plantlet. Genetic make up of plantlet is similar to its explant source. Eg:- Sugarcane, Citrus, Cymbidium, etc.

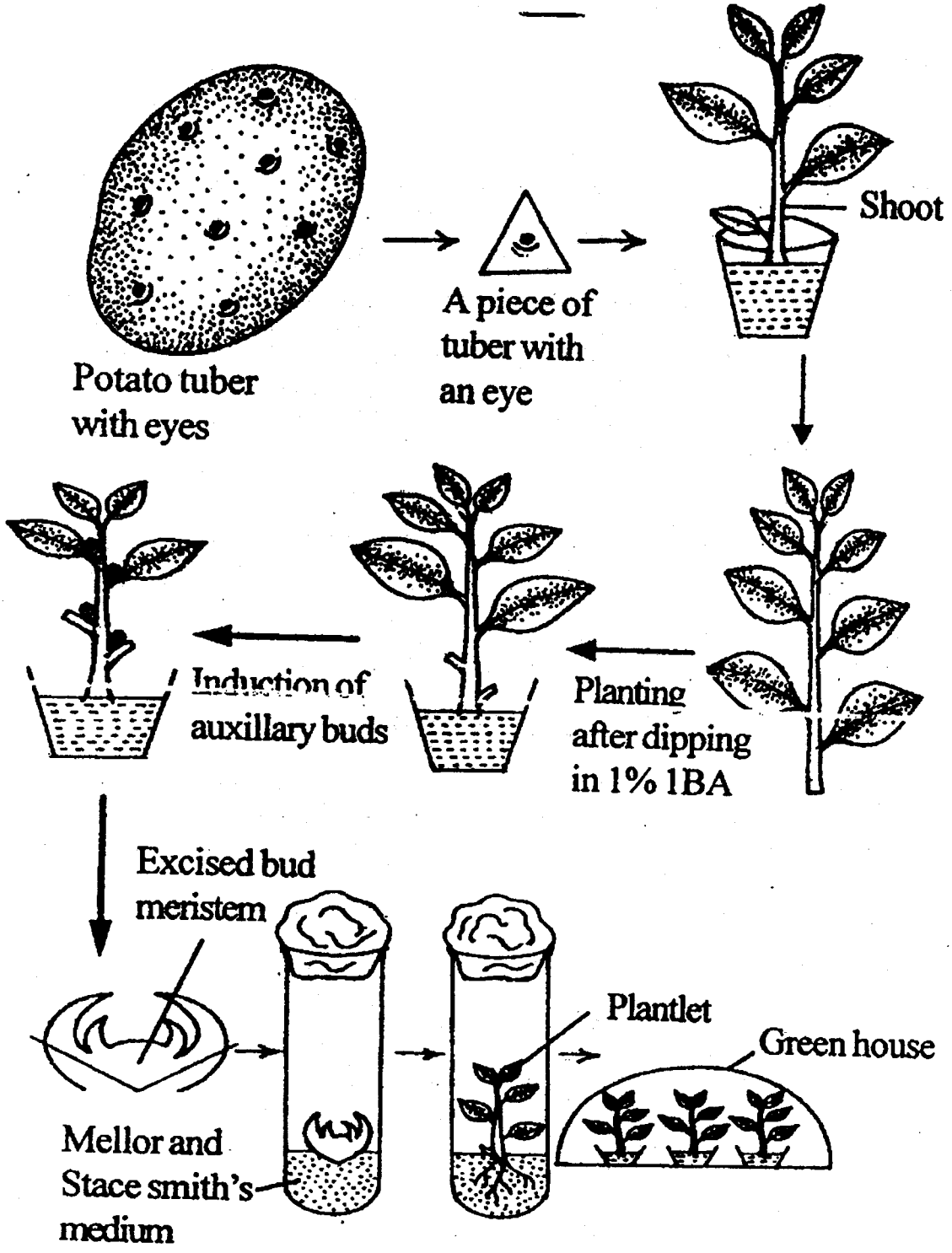

Fig. 2. Potato Meristem Culture

Steps involving in Meristem Culture

⇒ Potato tubers are taken and treated with cytokinin and gibberlic acid to induce the production of eyes. Each eye along with a portion of the tuber is excised and planted in a pot and allowed to grow for 8-10cms. A 6-8 mm long tip is cut and its two basal leases are removed. The cut end is dipped in 1% IBA to induce rooting and planted in a pot. Afterwards tip of the shoot is pinched out to promote the growth of auxilliary buds. Each auxilliary bud is excised and transferred to Mellor-Stace Smith's medium in a tube. Culture tubes are incubated at 25°C with little illumination of 4000 lux/day. The meristem i.e., auxilliary bud grows into a plantlet with shoots and roots with in few weeks. The well developed plantlets are then transferred to pots in a green house for proper acclimatization.

Uses of Meristem Culture

⇒ Disease free plants are produced. E.g.:- Virus free Potato, Sugarcane, Grapes, etc.,
⇒ Used in micro propagation of many crops. E.g.:- *Eucalyptus, Tectona grandis, Quercus*, etc.,
⇒ Used in germplasm storage of plants.
⇒ Meristem Culture is practised in woody plants in which root induction is difficult.

18.4 APPLICATION OF TISSUE CULTURE

The culture of excised plant cells or plant tissues in a synthetic culture medium under controlled aseptic conditions is known as Tissue Culture. It is also called *in vitro* culture. Plant tissue culture has many applications in crop improvement, preservation, breeding and industries.

Tissue Culture is employed in

⇒ Micropropagation
⇒ Elimination of pathogens from plant materials.
⇒ Germplasm storage
⇒ Production of somaclonal variants.
⇒ Embryo rescue.
⇒ Production of haploids.
⇒ Production of artificial seeds.
⇒ Production of secondary metabolites.
⇒ Production of somatic hybrids.
⇒ Production of Transgenic plants.

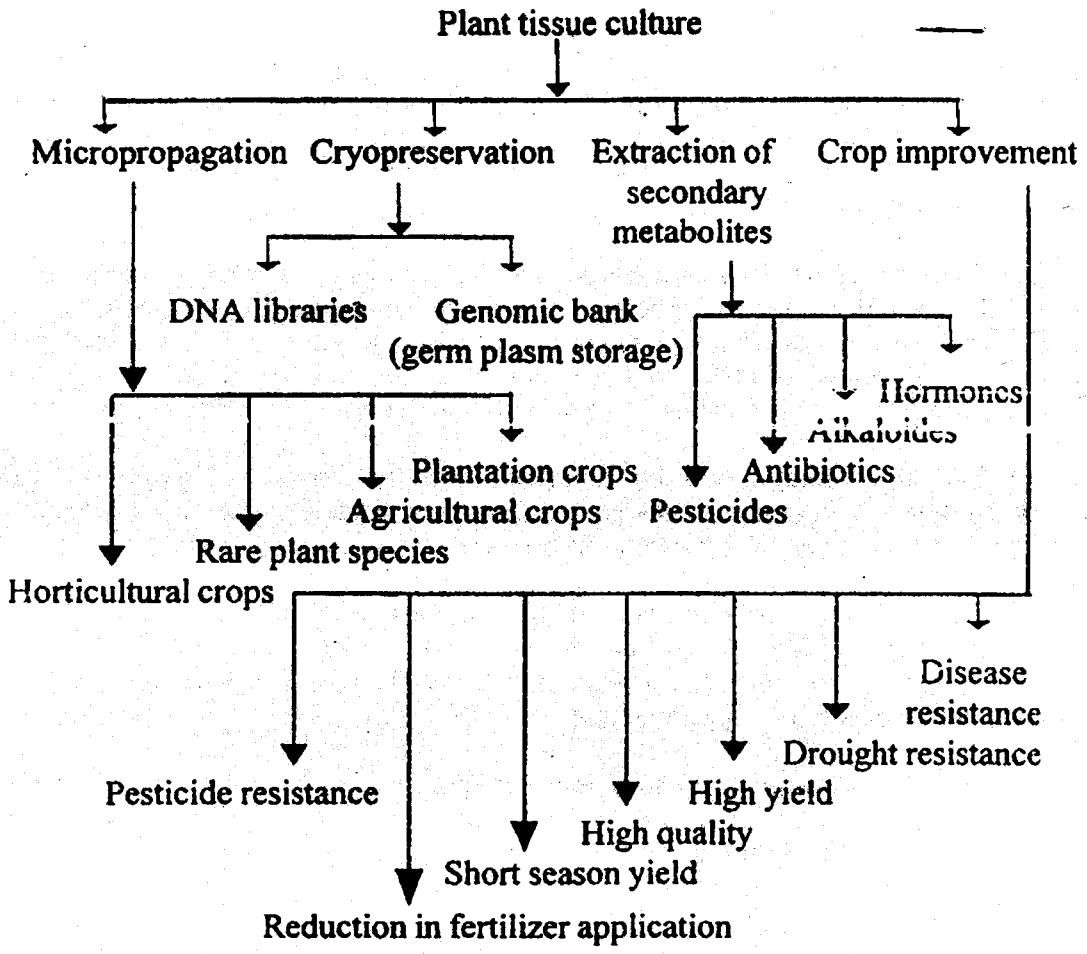

Fig. 3. Diagram showing the possible uses of plant tissue culture technology to the developing world

(a) Micropropagation: It is a process which involves rapid production of large number of identical clones in vitro within a short duration. This technique restores the original genetic make up of parent plant in the clones. Hence it is used to breed crop plants in large number Micropropagation is done in

- Plants in which seed production is difficult or impossible.
- Plants which fail to produce seeds with desired traits.
- Plants in which frequent changes take place due to cross pollination.

(b) Elimination of Pathogens: Plants free from pathogens can be raised from the existing plants through meristem culture. The resulting plant will be free from viruses, bacteria and mycoplasmas.

(c) Germplasm Storage: Germplasm is defined as the collection and storage of different plant species in a limited area. Rare plants and important plant species are stored in the form of

seeds or as growing plants. Invitro storage is the recent trend. The plant materials are stored in a minimal medium with low light intensity at a low temperature. This medium reduces growth rate of plant tissue but the total regeneration capacity of the plant is maintained in it. Large number of individuals are stored in a small area. Subculturing is done at regular intervals of one year. Sometimes meristematic tissues are stored in liquid nitrogen at -196°C. This is called as Cryopreservation.

(d) Production of Somaclonal Variants: Normally, plants (or) clones raised through tissue culture show similarity in their characters. But a few clones exhibit variations, such clones are called as somatic variants. They possess one or two characters which are not present in their parents. The formation of such variant clones from the cultured tissue is called somaclonal variation. These variants are formed due to mutation in some cells of callus tissue.

Somactonal variants can be obtained from the culture of explants like immature embryos, tip of inflorescences, stems, microspores, ovaries etc., by inducing mutations. Somactonal variation is used to develop new strains of plants with new characters.

(e) Embryo Rescue: In some plants, fertilization takes place normally, but the ovule fails to become an embryo. Such immature embryos are isolated from the immature fruits and cultured in tissue culture medium to develop into new plants. This method overcomes embryo immaturity hence called as embryo rescue.

(f) Production of Haploids: Haploid plants contain only one set of chromosomes. They are used in plant breeding to improve the field and agricultural crops. Haploids are produced from anther cultures, pollen cultures and ovule cultures. Haploid plants are used to identify recessive traits and recessive mutants of crops.

(g) Artificial Seeds: Artificial seeds were first prepared by Murashige in 1977. An artificial seed is a synthetic seed made up of somatic embryo surrounded by nutrient medium protected by a chemical membrane. Artificial seeds are smaller in size when compared to some natural seeds. Artificial seeds contain only the somatic embryos of a known plant strain. Nutrient medium which surrounds the embryo, nurishers the developing embryo. The protective chemical membrane is made up of sodium alginate (or) polyoxyethylene (or) polyacrylamide gel. Artificial seeds germinate as such as natural seeds. Viability is more and no dormancy is seen.

(h) Production of Secondary Metabolites: Plant cells in the bioreactor utilize the substrate material and convert it into valuable products. These valuable products are the secondary metabolites. These are also called as Defence Chemicals and they include alkaloids, phenols, terpenes, flavones and essential oils. These products are of very much importance in food industries, cosmetic industries, agriculture and pharmacy. As the plant cells are producing such products they are often called as biological factories.

(i) Production of Somatic Hybrids: The plant, which is raised as a fusion product of protoplasts of two different strains (or) species is called Somatic Hybrid. These hybrids are raised from sexually incompatible species. Somatic hybridization overcomes sexual incompatibility among plant species and also used to raise new crop varieties.

(j) Transgenic Plants: The plant whose original genetic makeup was altered is called Transgenic plant. Alteration of genetic makeup by introducing a noval gene is possible by genetic engineering. Herbicide resistance, insect resistance, degradable plastics, vaccination property, etc. can be developed in plants.

18.5 SUMMARY

Production of a plant by culturing a tissue on the artificial medium is called Tissue Culture. Totipotency is the pre-requisite for tissue culture practice. It is found in plant cell, hence this practice has been succeeded in plants. Selection and sterilisation of explant, preparation and sterilisation of medium, inoculation, incubation of culture and acclimatization are important steps, involved in tissue culture practice. Culturing of a plant organ on nutrient medium to develop an entire plant is called organ culture. It is classified into (a) Vegetative organ culture, and (b) Reproduction organ culture. Embryo culture is generally used to culture physiologically inactive embryos. Anther culture is performed to raise haploids and pure varieties. Culturing meristem is mainly with the aim of getting virus resistant varieties. Using tissue culture techniques many advantages have been achieved such as micropropagation, resistant varieties production, protection of endangered varieties, somatic hybridisation, transgenic plants etc.

18.6 TECHNICAL TERMS

Tissue Culture, Cell Culture, Organ Culture, Totipotency, Cryopreservation, Micropropagation, Somaclonal Variation, Explant, Basal medium.

18.7 MODEL QUESTIONS

Essay Type

(1) What is Organ Culture? Explain embryo, anther and meristem culture.
(2) Write a detailed essay on "applications of tissue culture".

Short Notes

(1) Preparation and Sterilization of medium.
(2) Embryo Culture.
(3) Anther Culture.
(4) Meristem Culture.

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Lesson - 19

VEGETATIVE PROPAGATION

OBJECTIVES

By the end of this lesson you will be able to

* describe advantages and disadvantages of vegetative propagation
* describe different methods of vegetative propagation
* describe the layering and grafting methods

STRUCTURE

19.1 INTRODUCTION

In flowering plants sexual reproduction is the most common method of reproduction. Some of the flowering plants are propagated by specialized structures formed by the plants in a natural way. This method of reproduction is similar to asexual reproduction where there is no involvement of male and female gametes and involves only one parent. New plants are formed mostly from vegetative parts of the plants like stem, root, leaf etc. The offspring will be similar to the parent plant. This method of propagation in plants is called VEGETATIVE PROPAGATION or VEGETATIVE REPRODUCTION. Vegetative propagation is useful to maintain the same genotype and phenotype of plant through generations without variation. Vegetative reproduction is a common method of reproduction in many

Lesson - 19

VEGETATIVE PROPAGATION

OBJECTIVES

By the end of this lesson you will be able to

* describe advantages and disadvantages of vegetative propagation

* describe different methods of vegetative propagation

* describe the layering and grafting methods

STRUCTURE

19.1 INTRODUCTION

In flowering plants sexual reproduction is the most common method of reproduction. Some of the flowering plants are propagated by specialized structures formed by the plants in a natural way. This method of reproduction is similar to asexual reproduction where there is no involvement of male and female gametes and involves only one parent. New plants are formed mostly from vegetative parts of the plants like stem, root, leaf etc. The offspring will be similar to the parent plant. This method of propagation in plants is called VEGETATIVE PROPAGATION or VEGETATIVE REPRODUCTION. Vegetative propagation is useful to maintain the same genotype and phenotype of plant through generations without variation. Vegetative reproduction is a common method of reproduction in many

plants. When a new variety of plant is developed through crop improvement technique, to retain the same characters of the parent in the offspring, vegetative propagation is preferred.

19.2 ADVANTAGES AND DISADVANTAGES

Advantages

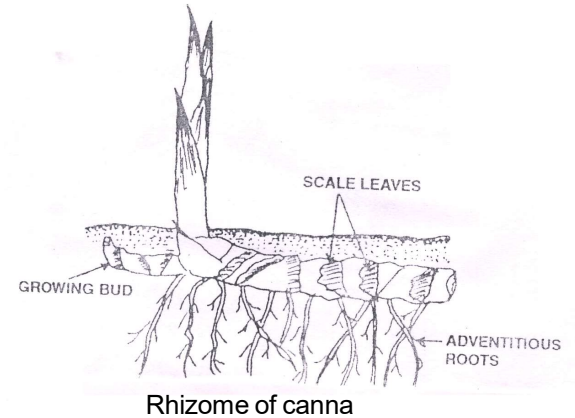
1) Some plants do not produce seeds. Then they are multiplied by vegetative propagation. e.g. ginger, turmeric, plantain etc. and ornamental plants like Jasmines, Hibiscus, Roses, Bougainvilleas etc.
2) Some field crops, many garden plants and fruit plants are normally propagated vegetatively through bulbs, rhizomes, corms, tubers, bulbs, stolons etc.
3) They are propagated by horticultural methods such as cuttings, budding, grafting.
4) It is an easy method of multiplying a large number of plants which are genetically similar.
5) Vegetatively propagated plants grow faster than seedling plants.
6) The vegetatively propagated plants are highly heterozygous and they do not breed true in sexual reproduction. But several plants can be produced from such a heterozygous parent plant by vegetative propagation. All the progeny thus produced will have the same genotype as that of the parent plant. So the characters of the parent plant are faithfully reproduced without any change unless mutations occur.
7) The hybrid vigour exhibited by the parent plant can be maintained in the progeny produced by vegetative propagation.
8) The performing potential of the vegetatively propagated sapling is known right from the time of planting.
9) Plants obtained by cutting or by layering flower earlier than seedling plants.
10) Grafting is of great importance in horticulture. Vegetatively propagated plants like mango, sapota start to bear fruits much earlier than seedling plants obtained by sexual reproduction.

Disadvantages

1) If the parent plant is infected with some disease, the progeny produced from it will have the same disease. e.g. Canker disease of citrus and bunchy top of banana.
2) Generally vegetatively propagated plants have shallow roots when compared to the deep rooted seedling trees. So vegetatively propagated plants cannot survive in dry conditions.
3) Vegetative propagation has restricted applications. It cannot be done in all plants.
4) New genes which are useful cannot be introduced into vegetatively propagated plants. So new varieties cannot be produced unless by mutation.

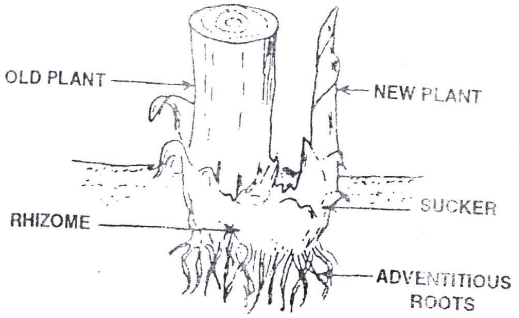
19.3 METHODS OF VEGETATIVE PROPAGATION

Various methods of vegetative propagation can be broadly classified as follows.

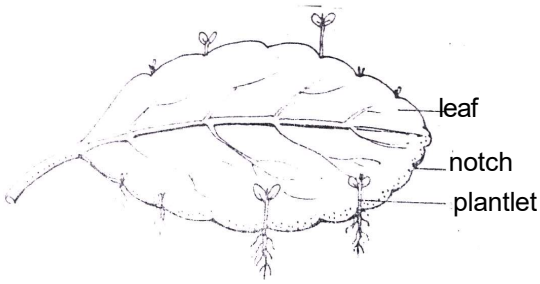


Rhizome of canna

Runner of Strawberry



Sucker of Banana



Leaf buds of Bryophyllum

Fig. 19.1

19.3.1 Propagation by specialized vegetative structures

Certain plant modifications are useful for vegetative propagation. These modified plant parts may be the stem, roots or leaves and are usually specialized for food storage. The plants showing these modifications constitute some vegetables and many others are valuable flowering ornamental plants.

Underground Storage Organs

The following storage organs are useful in vegetative propagation.

1) Tuberous roots of Dahlia, Sweet potato etc.
2) Bulbs of onion, Rhizomes of turmeric, ginger, amaryllus, garlic.
3) Tubers of potato.
4) Corms of Colocasia, Amorphophallus.

Runner

A runner is a long, slender branch which trails along the ground, and form a new plant at one of the nodes. The daughter plants, inturn, produce additional runners. Runner propagation is practiced in strawberry, dub grass (*Cyanodon dactylon), Oxalis corniculata, Hydrocotyl asiatica,* and *Lippia nodiflora.*

Offset

An Offset is a short, thickened stem of rosette – like appearance developing from the base of the main stem. *e.g. Pistia*. Offsets are separated from the mother plant by cutting them close to the main stem with a sharp knife. If the offset is well rooted, it can be potted or planted directly in the field. If the rooting is poor, the offset is first placed in the rooting medium (soil, sand, peat mixture).

Sucker

A sucker is an underground runner, which soon emerges to form a daughter plant, after striking roots. Suckers also develop from axillary buds at the nodes of underground runners (e.g. Pine apple). Plants developed from other under ground parts are also called suckers (roots of some shrubs and trees). Suckers are separated from the mother plant and are either planted in the nursery for rooting or in the field (Banana, Pine apple, Mentha).

Bulbils

These are produced on the branches of inflorescence after the fall of flowers. The bulbils are like plantlets. They grow into new plants when they fall on the soil. *e.g. Agave*.

Leaf buds

In *Bryophyllum*, epiphyllous buds occur in the notches of leaf margin. In *Kalanchoe*, they are at the leaf apex. In *Begonia*, they develop at the wounded or cut regions. In horticultural practices they are called bulbils. They fall down when mature. They can be separated from the leaf and planted. They are considered as plantlets as each one has root, stem and leaf.

19.3.2 PROPAGATION ON ITS OWN ROOT SYSTEM

19.3.2.1 Cuttings

This is a method of vegetative propagation in which a portion of any vegetative part like stem, leaf or root is separated from a parent plant and placed in suitable soil and environmental conditions. The cuttings form roots and shoots develop in to new plants. Ex: Rose, Sugar cane, Hibiscus. The method is simple. There is a high success rate, as adventitious roots will readily grow on many stems once they are separated from the parent plant.

When outside conditions are not suitable, plant cuttings can be grown with special equipment, or in green houses (with regulated temperature and humidity), in soils free from pathogenic organisms. Uniform distribution of water and nutrients help growth of the cuttings required for propagation. Herbaceous plants easily root than woody plants.

Propagation of plants by cutting uses either stem or root cuttings. In these methods before cutting a part of the plant for propagation, we should see that the plant has regions that can give rise to whole plant. Propagation by stem cuttings involves a piece of stem that is detached from the mother plant, which will have a few nodes. When this piece of stem is planted in suitable soil conditions, it grows into a new plant by developing roots. Cells in the cut portion will be active and produce new roots.

Stem cuttings

This is the most commonly adopted method of propagation of plants. In most cases, cuttings, are made from strong and mature stems. ex: *Hibiscus, Rosa*. These are called HARD WOOD CUTTINGS. In some plants the cuttings are prepared from semi hard stems and are called SEMI HARD WOOD CUTTINGS. Ex. *Clerodendron*, *Tecoma*. When cuttings, are taken from soft portion of the shoots as in *Dahilia*, *Geranium*, they are known as SOFT WOOD CUTTINGS. In all these, the cuttings are taken from healthy plants. A smooth and slanting cut is made below the basal node. In sugar cane, a small portion of the stem consisting of a node is used to produce new plants. For rooting, soft wood cuttings are usually maintained in materials like vermiculite, which provide more aeration. Also this type of materials can be sterilized to maintain the cuttings free from any pathogenic organisms.

Treatment with auxins reduces the time required for rooting. Auxins, like Indole-3-acetic acid (IAA), Indole butyric acid (IBA), Naphthalene acetic acid (NAA), are widely used in propagating stem cuttings.

Root cuttings

Plants that form adventitious buds on the roots can be propagated by root cuttings. The cut roots are kept in coarse sand for further development. eg: carrot, guava, apple, pear, cherry, plum.

Leaf cuttings

Certain plants with thick and fleshy leaves have the capacity to produce plantlets on their leaves. e.g. *Bryophyllum, Begonia, Peperomia, Sansevieria* etc. This is a simple and efficient method of vegetative propagation. It can occur in two ways : (a) by naturally growing foliar embryos or (b) by artificially induced plantlets from leaf cuttings. Leaves of *Pepromia* or *Sansevieria* are planted vertically in sand bed. They first produce root and then shoot. Leaves of *Begonia* and *Bryophyllum* are placed flat on the sand bed with petiole inserted into the sand to hold it in place.

19.3.2.2 Layering

Plants that are large and more difficult to propagate by cuttings can be made to root while still attached to the parent plant. Stems that form roots while still attached to the parent plants are called LAYERS. Propagating the plants in this method is known as LAYERING. In layering roots are induced. A layer is supported by the parent plant till it develops roots. Layers do not require close attention like watering, humidity, and temperature that cuttings require.

The common practice in layering is to injure the portion to be layered by notching, cutting, girdling. This is to reduce the downward movement of nutrients through phloem tissue. As a result, nutrients gather above the injured area and favour root formation by the layers. Downward movement of hormones is prevented in this type of injuries. This will help to develop roots. Two different types of layering are routinely practiced in horticulture.

Air layering

A healthy branch on a plant is selected. A girdle is made just below a node, by completely stripping of bark of 1 to 2.5 cm width, around the stem. The girdle portion is covered with moist moss plants. Then it is wrapped with a polythene sheet around the girdle. The paper is tied very tightly with a twine so that no moisture can escape from inside. This is left undisturbed till the roots appear from the layered area. Layered shoot is separated from the mother plant when healthy roots are visible through the polythene sheet. This air layering is practiced in crotons and climbing roses. A method very similar to air layering is carried out in plants where branches are hard or formed well above the surface of the soil. eg: pomegranate, guava, orange. This method is called **Gootee**. In this method, a healthy shoot is selected and a ring of bark is removed towards the base of the branch. The injured part is covered by moss plants and then covered by a polythene sheet and tied firmly. After some time, roots are formed from injured part. The branch is now cut below the bandage and planted separately.

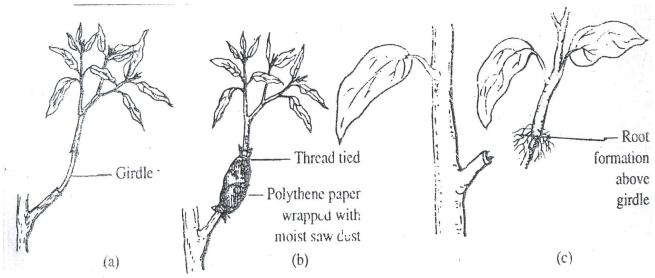


Fig. 19.2 Air layering

Ground layering

In rose, instead of planting a cutting, a branch nearer to the ground is bent down to touch the soil. On the basal side, under the node, a cut is made to form a tongue. The two cut portions are kept apart with the help of a small piece of wood. The tongue part is covered with soil. A stone is kept on the layered portion in position or covered with soil. Watering is done whenever required. After some time, the portion under the soil develops roots. The layered portion is separated from the parent plant and planted separately. eg: Jasmine, Rose, Grapevine, Ipomea etc.

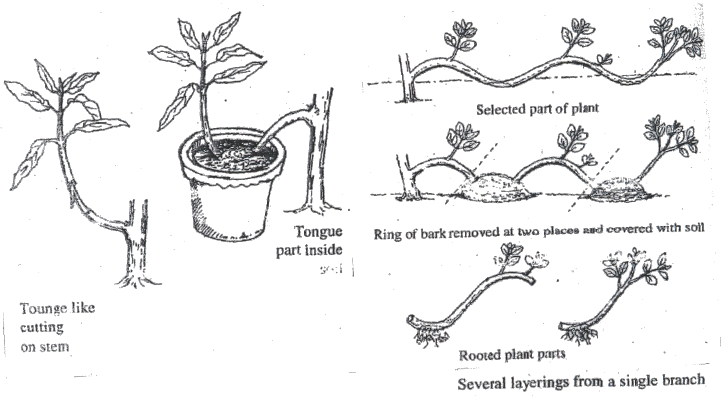


Fig. 19.3 Ground layering

19.3.3 Propagation on the Root system of other plants

A plant can be propagated by establishing it artificially on the root system of other plants through budding or grafting. This method requires two or more plants. Of these two plants, the one which supplies the root system is called the root – stock. The other which stands on the root stock and provides the shoot system is called the scion or graft.

19.3.3.1 Grafting

Grafting provides an opportunity to bring the characters of different plants into one plant. Grafting is a technique of inserting a part of one plant into another plant in such a way that the two will unite and continue their growth. The top part of new plant looses its own root system and unites with another plant that supports and supplies its nutritional requirements. The upper part of the union is known as **scion** or **graft** and the lower part is the **root stock** or **stock**. Grafting is practiced between closely related species. Each part of the plant retains its own characteristic features, as there will not be any interchange of genetic material. Thus, one mango trunk system can carry a number of different varieties of mangoes.

Grafting will be successful only when the union of the scion and root stock occurs through formation of new tissues. For successful union, vascular tissues of scion and stock should touch each other. There should be enough moisture and optimum temperature for formation of new tissues. Successful grafting cannot take place between unrelated plants. Plants in which grafting is done more routinely are mango, apple, guava etc.

19.3.3.1.1 Uses of grafting

Grafting helps to preserve and perpetuate varieties that cannot reproduce by vegetative method. Grafting changes less useful varieties to more desirable varieties. Adaptation to unfavourable environment can also be done by grafting. Different varieties can be grown on one plant as a novelty. It is economically feasible to produce plants through grafts for those that are difficult to propagate by cuttings. A high yielding commercial tree can be grafted to a hardy root stock, that is more resistant to disease, pests and drought.

19.3.3.1.2 Methods of Grafting

Atleast four methods of grafting are in practice. They are approach grafting, cleft grafting, tongue grafting and bud grafting. We will learn more details about these methods

Approach grafting

This is largely practiced for grafting sapota, mango and guava. In this method, both the stock and scion remain rooted. The stock is grown in a pot. A branch of scion of the same thickness as the stock is bent towards the stock and a small slice is cut off from its stem. A similar slice is cut off in the stock. The two cut surfaces of the stems are brought in contact and are held firmly in position by tape. The two stems get united in course of time. Then the top of the stock and the base of the scion are later cut off. The scion of a desired variety is allowed to grow on the root stock of different variety. This can be applied in all those plants where grafting is possible.

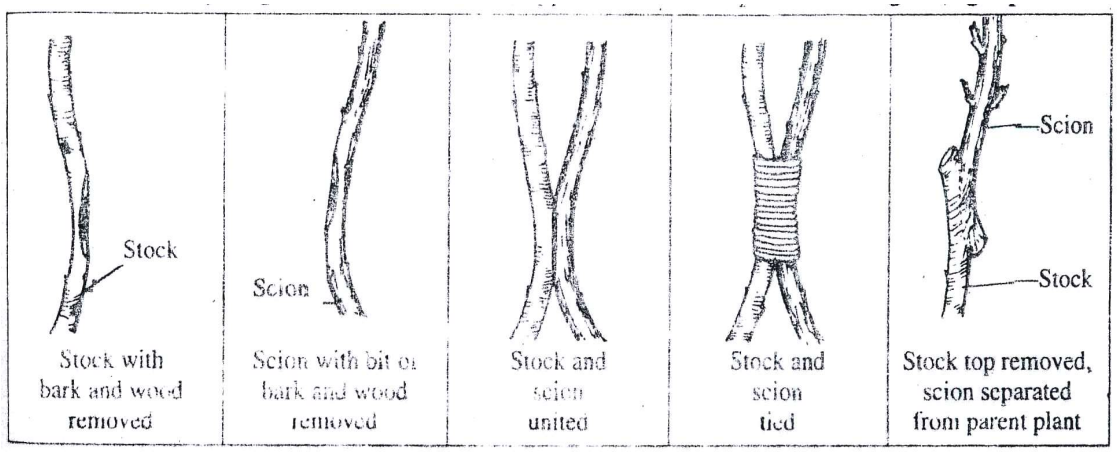


Fig. 19.4 Approach grafting

Cleft grafting

With a sharp knife, a stem without bends and side branches is cut across. The end of the stub is split like a 'V' to insert scion. Scions are prepared by cutting the end in a wedge shape with the help of a sharp knife. Two or three good buds of the scion above the stock are ensured. Similarly many slits are made towards outside of a large stock. In each slit only one scion is inserted. The point of union is held in position and a waxed tape is put around the junction.

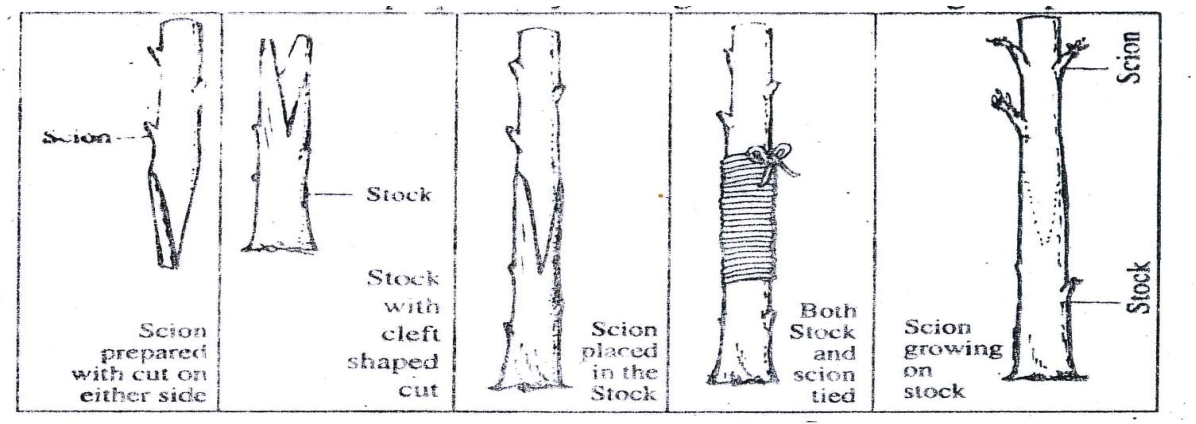


Fig. 19.5 Cleft grafting

Tongue grafting

This method is used on stocks that are relatively small. Top of the stock is cut diagonally and in an upward direction. Scion is cut diagonally in downward direction. Cut surfaces of both should be smooth. Uneven and wavy surfaces prevent proper contact and union. A second cut in given from above, downwards, which forms the tongue. This is done in a manner that the notch or tongue of the scion closely fits with that of stock. Later, it is wrapped and tied by twine and covered with a waxed tape.

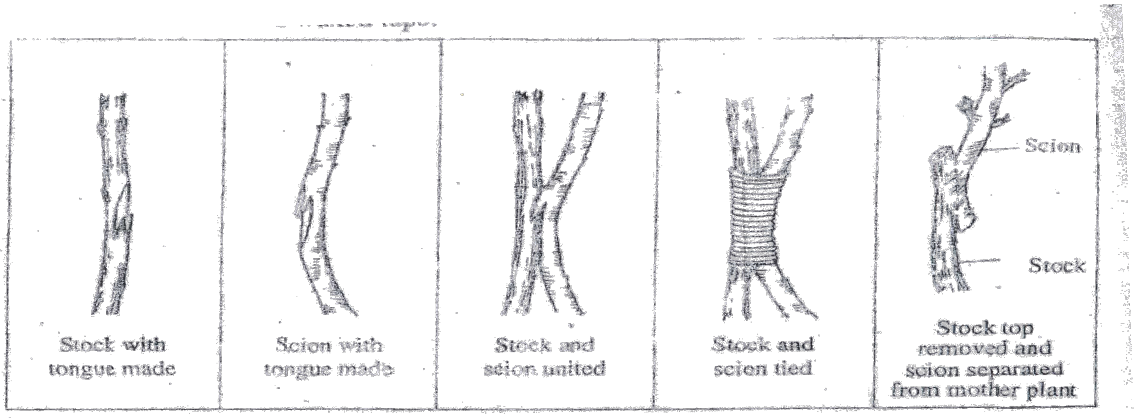


Fig. 19.6 Tongue grafting

Bud grafting

This method of grafting is used for propagating apple, orange, rose and other ornamental plants. A 'T'-shaped incision is made on the bark of the stock. The bark is made loose on either side of vertical cut. A single bud of the scion with a little wood is placed in the incision below the bark and held in position by applying tape. The bud draws water and mineral salts from the stock. As it grows, it synthesizes its own food. The buds of the stock will be removed so that they may not compete with the grafted bud. The bud, when cut, resembles a shield in shape. The cut resembles the letter 'T'. Active cells under the bud, and the exposed surface of the stock come together.

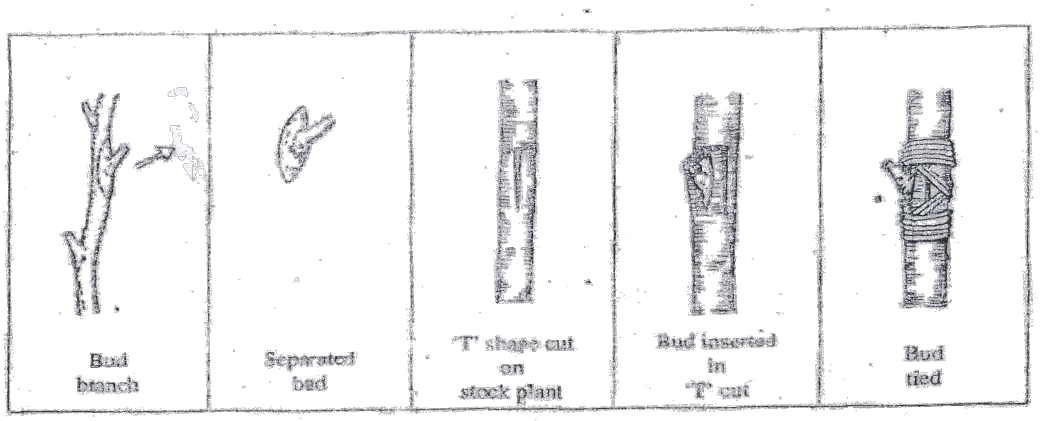


Fig. 19.7 Bud grafting

19.4 MICRO PROPAGATION

Propagation of plants by using miniature plant tissues called explants, is known as micropropagation. Tissue culture is used to grow plant parts like embryos, isolated cells and protoplasts. In micropropagation, very small pieces of tissue are grown in a sterile nutrient medium under aseptic conditions.

Many horticultural and timber crops are now propagated using tissue culture technique. These include orchids, citrus, strawberry, papaya, pine apple, potato, banana, teak and many ornamental plants and shade trees.

19.5 SUMMARY

Vegetative propagation helps to maintain fixed qualities and characteristic features of the parent plant. This method of multiplication is through bulbs, tubers, cuttings, layering and grafting. A large number of horticultural crops like guava, grape, pomegranate, orange, citrus etc. vegetable crops like potato, sweet potato and flowering plants like *Chrysanthemum* are produced through vegetative propagation. The cutting from roots and shoots develop into new plants eg. Rose, Sugar cane, Hibiscus etc. Layering is an alternative in plants where propagation through cuttings is difficult. In grafting, a part of one plant is inserted into another plant in a way that both of them will unite and continue their growth. The upper part of the union is called scion and the lower part as the stock. It is economically feasible to produce plants through grafts for those that are difficult to propagate by cuttings. At least four different method of grafting are practiced, which include approach grafting, cleft grafting, tongue grafting and bud grafting. Plant cell and tissue culture offers several advantages for plant propagation and also for crop improvement.

19.6 TECHNICAL TERMS

Grafting, Mutations, Green House, Stock, Scion, Foliar Buds

19.7 MODEL QUESTIONS

Long Answer questions

1) Economic importance of vegetative propagation
2) Different methods of vegetative propagation
3) Different types of layering and grafting

Short Answer questions

1) Cuttings
2) Layering
3) Grafting
4) Micro Propagation

19.8 REFERENCE BOOKS

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