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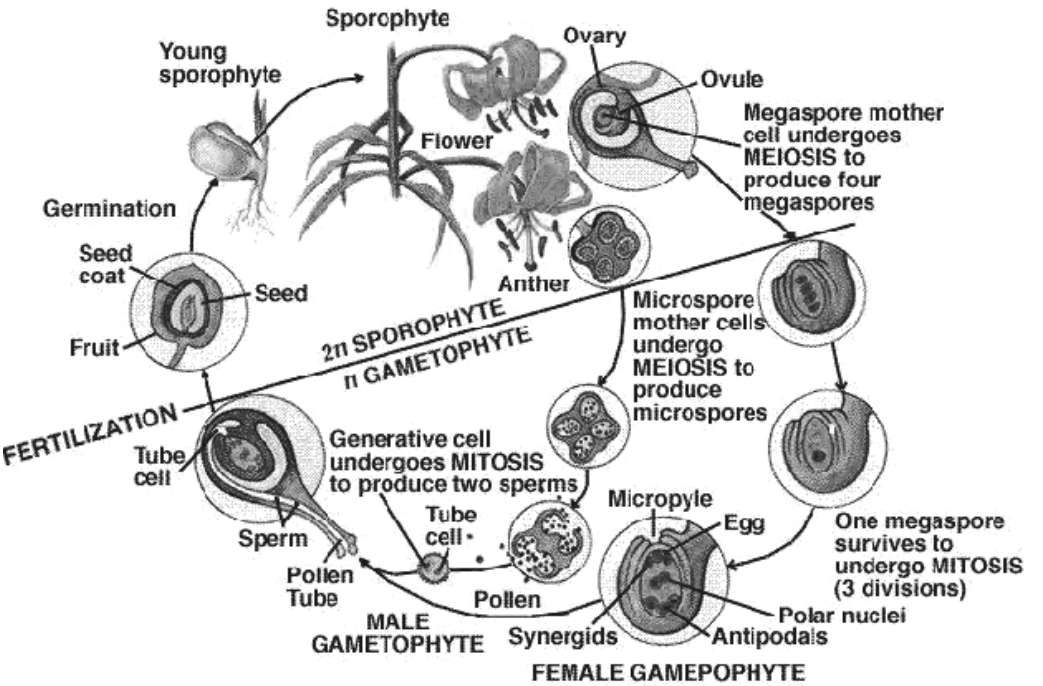

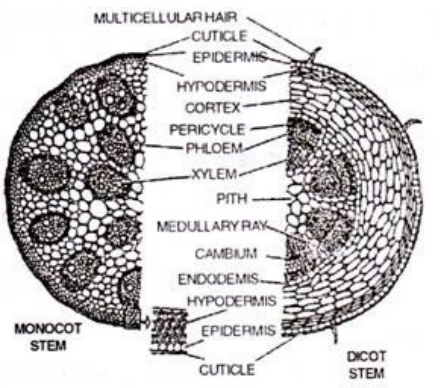

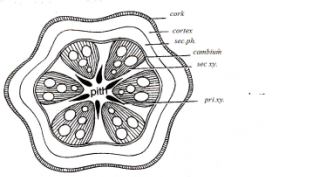

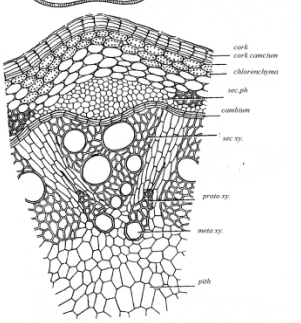

Biosystematics of Angiosperms, Plant Development and Reproduction

MBO-06

Vardhman Mahaveer Open University, Kota

Biosystematics of Angiosperms, Plant Development and Reproduction

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Editor

Dr. G.P. Singh
Department of Botany
University of Rajasthan, Jaipur

Writers:

- **Dr. Amit Kotia** 1,2,3,4
  Department of Botany
  University of Rajasthan,
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- **Dr. Praveen Mohil** 5,6,7,8
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Biosystematics of Angiosperms, Plant Development and Reproduction

Vardhman Mahaveer Open University, Kota

Preface

The present book entitled **"Biosystematics of Angiosperms, Plant Development and Reproduction"** has been designed so as to cover the unit-wise syllabus of MBO-06 course for M.Sc. Botany (Final) students of Vardhman Mahaveer Open University, Kota. The basic principles and theory have been explained in simple, concise and lucid manner. Adequate examples, diagrammes, photographs and self-learning exercises have also been included to enable the students to grasp the subject easily. The unit writers have consulted various standard books on the subject and they are thankful to the authors of these reference books.

Unit - 1

Origin of Intrapopulation Variation and Phylogeny of Angiosperms

Structure of the Unit:

1.0 Objectives

After going through this unit, you will be understood about:

- the relationship between the population and environment
- Ecads and Ecotypes
- Evolution and Differentiation of species-various models
- Ancestors of Angiosperms with their Time and Place of Origin
- Primitive Living Angiosperm
- Inter-relationship of Angiosperm with other existing groups

1.1 Introduction

The origin of species is based on the population study as we have to first learn about what kind of population exists in the habitat as they may be of same gene pool and may be some isolation has occured in their gene pool. After the study, the terminology *viz.* Ecads and Ecotype came, and the difference is that the morphological variation disappear in Ecads when they grow in same habitat which means that these variations are not coded genetically (*Euphorbia hirta*) where as in Ecotype, the variations do not disappear as the characters are coded genetically (*Potentilla glandulosa*).

This population isolation mechanism resulted in development of new species as the characters have been genetically modified and the speciation process is initiated. There are two methods of speciation *viz.* Allopatric and Sympatric patterns. Scientists described different models and concepts of species which help in underatanding the species at different levels like- The Biological Species Concept is the concept of species reflecting about the species that a population or series of populations those freely interbreeding here organisms reproductively isolated from other such population.

The origin of Angiospems is a massive subject to study and there are some lacunas to correlate the origin of Angiosperm because the fossil record has not provided conclusive information about the origin of angiosperms. But modern science has some how concluded the relation of the major groups and their phylogeny and where they have arisen. Some fossil records show their origin or relation with *Pteridospermales, Bennettitales, Cycadales* and *Pentoxylanean* group whereas some botanist show the affinities and origin of Angiosperms with lower group and primitive Angiospems like *Ephedrales, Gnetales, Amentiferae, Ranalesccc*, and *Isoetalean* members.

The time of origin of Angiosperm is not clear as they suddenly appeared in the *Cretaceous* period and since this age, angiosperms have became dominant vegetation on the earth. Even the place of origin is also described by different botanist like according to Seward (1931) the Arctic region, Smith (1970) located in South-East Asia, Retallack and Dilcher (1981) believed origin in the Rift valley system adjoining Africa and South America. The primitive living Angiospers are belong to some families this includes *Winteraceae, Magnoliaceae, Degeneriaceae, Himantandraceae, Eupomatiaceae, Annonaceae, Canellaceae,* and *Myristaceae* belongs to the orders *Magnoliales. Austrobaileyaceae, Amborellaceae, Monimiaceae* and *Calycanthaceae* belong to the order Laurales and in the order Trochodendrales with the famalis *Trochodendraceae* and *Tetracentraceae*. Some other like orders *Piperales, Nymphaeales, Illiciales* and *Ranunculales* also reported primitive members.

1.2 Origin of Intrapopulation Variation

1.2.1 Population and Environment

Generally, the population is defined in different ways for different purposes. Population is the group of individuals which are common to appear. The definition which is used largely by taxonomists is "The group of individuals considered to gene-pool together at any one time because of the features they share in common." It means that the group of plants, growing together, which look similar or a series of such plants which grow in a particular area. Usually in population genetics, the co-habitation of the particular group of individual allows them to exchange their genes. Therefore a breeding population can be defined as "a group of individuals less degree with of crossing and with consequent gene flow". Two points should be noted here

(i) Possibility of inter breeding
(ii) Genetic similarity

The population is not defined on the basis of morphological characteristics especially at gene level. Population is that in which the individual members share gene and become inbreeding there by sharing a common gene pool.

Sirels (1951) categorized population in two distinguished group as Plurispecific and Unispecific population. The Plurispecific population is constituted by the plants, animals and microorganisms which are living together in the same habitat. The area for that population may be a square yard, a square mile, a wood piece or an ant heap. Besides this, unispecific population is a group of individuals belonging to the same species and living in the same habitat. The populations always remain subjected to change as they can vary in size, composition and area. They may increase or decrease in size.

Several experiments by different workers have been performed on population-

A) Turesson's Experiment : Turesson was mainly concerned with the differentiation of population in different ecological condition. The genetical morphological and physiological nature of population samples from different parts are collected and grown in uniform condition in his experimental garden at "A karp" in Sweden. He noted the reaction of plant in cultivation over the year in term of habit, height and flowering time etc. and found that in some cases the difference noted in the field disappeared in other cases and the differences showed intergraduation, but in most instances they persisted. He showed that the genetically different races were often correlated with habitat differences and that similar selective forces in the habitat appeared to produce similar kinds of adaption in different species. The term '*ecotype*' was applied to these products of reaction between the genotype and the habitat.

B) Clausen, Keck and Hiersey's experiment: All these botanists are known for their transect experiment which consists of the gene ecological studies of populations of various species including *Potentilla glandulosa* (Rosaceae) and *Achittea milkfolium* (Asteraceae).

A transect was taken across Central California for climatic differences. Experimental gardens were established at varying altitude from about 30 meters to 500 meters, and 3300 meters and were maintained free from weed. These were extended for more than 30 years indicating the scope of the investigation. Populations of the species mentioned above were grown at various altitudes and their responses to climate conditions were noted. There was a superficial resemblance or parallelism in morphological as well as physiological features in the ecotype adapted to similar climatic conditions. Hybrids between the ecotypes were shown to be fully fertile and could produce new genotypes with new adaptive features.

Both these experiments clearly indicated that interaction between habitat and genotype could apparently produce ecotype.

C) Gregors's Experiment: Similar investigations were undertaken by Gregor and his co workers at the Scotissh Society for the Research in Plant Breeding. They called these investigators experiment taxonomy and its practical procedures were studied by Gregen, Dowey and Long (1936). They regarded orthodox experimental taxonomy as complementary. They had grown the races under uniform environmental conditions in the experimental garden so that the variation shown by races could be studied unaffected by the irregularities of their natural habitat. They worked on the races of *Plantago maritina* (Plantaginaceae) and concluded that the pattern of ecotype variation was more frequently continuous than discontinuous corresponding to gradients shown by the habitats.

D) Russion school: The Russion school is an important school of gene ecological studies. It is developed under supervision of Dr. Sinka Ja (1948) of the Institute of Plant Inducting at Leningrad.

He was responsible for much of the work on ecotype recognition. The Russian approach differs in same respect from those of other school and has paid great attention to geographical and zonal variability of population in wild and cultivated plant *(Onobrychis*).

Four vertical belts of population may be distinguished.

1. High mountain belt
2. Middle mountain belt
3. Sub mountain belt

4. Steppe mountain belt

The morphological features of the population show gradual changes with the environment.

1.2. 2 Ecads and Ecotypes

(i) Ecads

The term Ecads is used for the plant species differing in appearance, especially in morphological features like the size of vegetative part, number of stem erectness and reproductive vigour, but belonging to same homogenous genetic stock. Their variation is due to environmental influences. When different ecad or ecophenes are transplanted in to the same habitat, these variations disappear, for example, in *Haplopappus* species. The appearance in *Haplopappus* species, Asteraceae shrub was strongly affected by soil type. Many taxonomists considered *Haplopappus venetus* and *Haplopappus decumbens* as two different species but after plantation in the same habitat and same soil type, both the species were identified as ecads rather than different species.

Similarly in *Euphorbia hirta*, which in growing in prostrate in disturbed area but becomes erect in undisturbed area. These morphological variations are not fixed and upon transplantation the prostrate form may become erect thus a species can occupy a range of habitat.

(ii) Ecotypes

Ecotype is the basic unit of biosystematics adapted to a particular environment but capable of producing fertile hybrids with other ecotypes. Thus, ecotypes of the same species are not isolated by genetic barriers. It can be considered equvivalent to a variety or sub species of population.

The term ecotype was proposed by Turesson (1922). He defined it as "Ecotype is an ecological unit to cover the product arising as a result of the genotypic response of a species to a particular habitat".

Later onTuresson (1929) redefined the ecotype and emphasized the genetic cross ability between ecotype, there after Gregor (1939, 1945); Clausen, Keck and Hiersey's (1939, 1945) developed the concept and gave the definition as:

"A population distinguished by morphological and physiological characters, most frequently of quantitative nature, interfertile which other ecotype of the ecospecies but prevented from freely exchanging genes by ecological barriers".

Thus, important characters of ecotype are:

1. The variation are morphological, physiological or phenological (like differ in time of flowering and fruiting) or combination of two or all the three.
2. Variations are associated with certain distinctive habitat type.
3. Ecotypes are distinct and discrete entities.
4. They are genetically based or genetically fixed.
5. They are interfertile.

1.2. 3 Evolution and Differentiation of species-various models

Speciation is applied to the origin and evolution of population in terms of genetical processes, although the word itself suggests the evolution of morphological differences of the phenotype. Speciation is concerned with the formation of reproductively isolated gene pools. It would be *idle* to pretend today that such genetically defined units necessarily coincide with morphologically defined species (Walters, 1962). This being so, the term speciation seems most unfortunate for a process leading to the formation of units which taxonomists may not consider species. Taxonomic species, in so far as they represent populations, have of course evolved through various genetic processes, but they do not all represent reproductively isolated, intrabreeding units. On the contrary, what taxonomists designate as species represent many different evolutionary situations (Heywood, 1965). As Walters (1962) comments, it is surprisingly difficult to find an example of an "ideal" or uncomplicated species among the many cases that have been investigated genetically or biosystematically. Normally some complicating factor such as apomixis, inbreeding, polyploidy, hybridisation, or a combination of these, is operative. One of the most interesting and absorbing lines of research for a taxonomist today is a study of the relationship between taxonomic species and the micro-evolutionary situations or units with which they most nearly coincide. It is felt that this point needs considerable emphasis since any suggestions today with which illuminating the evolution and genetic nature of taxa provokes strong protest. But to advocate the usage of the term species in a practical morpho-geographical sense in no way denies the importance of micro-evolutionary, population and genetic studies on the nature of taxa. It simply regards them as separate although complementary approaches. The genetic basis of species is not important for their recognition and naming by taxonomists; to suggest otherwise is demonstrably false and to attempt a working classification with taxa based on gene pools is simply to misunderstand the essentials of practical classification. Then, what is the relevance of speciation to taxonomy? This is a far more difficult question to answer. An obvious answer is the one already suggested, namely that whenever possible the majority of taxonomists today wish to have an understanding of the evolutionary nature of the species and lower units that they describe in morphological and geographical terms, even though

such knowledge may in no way alter their actual classification procedures or the units recognised in practice. The following points have also to be considered:

(a) Since taxonomic species are not necessarily reproductively isolated populations, we cannot consider their origins as though they were.

(b) What we can do is to attempt to ascertain the evolutional situations which our species represent.

(c) In doing so we may find that taxonomic species may cover more than one gene pool, coincide approximately with one, or do not represent Mendelian populations at all, but much more complex and imprecise situations.

(d) As a result of such studies it may be found that a more natural and satisfactory means of delimiting the species may be possible, but this should only be employed if there are clearly defined morphological features supporting (i.e. correlated with) the new delimitation. In other words, species should make taxonomic sense and not be recast simply to meet gene pool definitions irrespective of the possibility of their morphological recognition. Speciation studies may show that a taxonomic species comprises diploid and tetraploid which cannot be morphologically separated; to recognise the different cytodemes as separate species simply because they represent different isolated gene pools which may satisfy the requirements of some geneticists but would make little taxonomic sense.

(e) It is a negetive assumption to believe that all plants are organised into clear cut, reproductively isolated gene pools. The various factors causing reproductive isolation occur at different intensities, evolve at different rates and operate in different parts of populations in different wave and with various degree of effectiveness.

(f) It is widely overlooked that speciation studies presuppose a classification into taxonomic species. While evolutionary or population taxonomists are often ready to criticize the outdatedness of orthodox taxonomic procedures and units, they are entirely dependent on the prior existence of a classification before they can themselves take their initial steps. If we could wipe the slate clean and start clasisification from scratch, it is debatable whether it would be possible for us to devise any more practical method than just attribute grouping which is morphologically expressible (Walters, 1962). However, we do not have such an opportunity and as a result there are repeated attempts to redefine species in genetic terms, attempts which are often doomed to failure. The fact that taxonomically and genetically defined species cannot necessarily be equated, clearly limits the role of speciation studies in classification, but it offers us a fascinating new field of relationships for study. It should also be remembered that

knowledge of the origins (phylogeny) of species or of biosystematic units, may give us little information about their present day relationships in respect of certain attributes, such as interfertility (cf. Jones, 1961).

Various concepts are available for the origin of the species but the basic knowledge as to, how the population has seprated from with their ancestors or the two groups have seprated from each other is cleare the actuale theory of speciation. The isolation in population may be in Allopatric or Sympatric pattern. The allopatric speciation is the differentiation and complete attainment of reproductive isolation of the population, which are completely geographicaly separated. Whereas sympatric speciation is the differentiation and complete attainment of reproductive isolation of the population, which are not completely geographicaly separated. There populations overlap in their distribution.

The Species concept

Biological Species:

The biological species is the species in which a population or series of populations are freely interbreeding organisms and which are reproductively isolated from other such populations.

“Group of actually or potentially interbreeding natural populations, genetically isolated from each other or often such groups by one or more reproductive isolating mechanisms”.

Grant (1971) defined a biological species as “the reproductively isolated system of breeding populations.”

Mayr (1969) defined species as “groups of interbreeding natural populations that are reproductively isolated from other such groups”.

Traditional concept of species

Many biologists believe that living organisms do not vary continuously over the whole range, but they fail into more or less well defined groups which is commonly called species.

Traditional taxonomists adhere to this philosophy of discrete species. They examine many morphological, anatomical and chemical characters from many specimens, but eventually select only a few morphological characters to serve in defining the various species.

Taxonomic species concept

A species consist of groups of morphological and ecological similar natural populations which may or may not be interbreeding, but which are reproductively isolated from other such groups. Three aspects are combined in this definition

1. External appearance

2. Breeding behavior
3. Habitat distinctiveness

Microspecies

Microspecies, as defined by Grant (1981) are populations in predominantly uniparental plant groups which are uniform among them selves and are slightly differently uniparental plant groups which are uniform among them and are slightly differentiated morphologically from one other. They are often restricted to a limited geographic area and are frequently of hybrid origin.

Microspecies (Grant 1981) fall into four main classes according to the mode of reproduction:

1. Clonal microspecies, reproducing by vegetative propagations in *Phragmities*.
2. Agamospermic microspecies, reproducing by agamospermy
3. Heterogamic microspecies, reproducing by *Oenothera biennis*
4. Autogamous microspecies, predominantly autogamous and chromosomally homozygous

Biosystematic concept of species

Biosystemics are interested in determining natural biotic units: populations of plant which maintain the distinctiveness because of biological barriers that genetically isolate them from other populations. These isolating brreiers may be breeding behavior or inability to form fertile hybrids with closely related groups.

Numerical Concept of Species

Another approach to taxonomy is to consider all forms of evidence with equal emphasis. This statistical approach is called numerical taxonomy. A basis tenet for this method is that a great deal of evidence is required to separate taxa. Some 50 to 300 characteristics are used for a given study, and they range from morphological to biochemical characteristics.

Typological Species Concept

According to Mayr (1957), this is the simplest and most widely held species concept. Typological thinking holds the dogma of the constancy of species. Variation, under this concept, is merely an imperfect manifestation of the idea implicit in each species. If the degree of variation is too great to be ascribed to imperfections of our sense organs, more than one species must be involved. Thus the species status is determined by degree of morphological difference.

The application of the typological species concept to practical taxonomy results in the morphologically defined species. The "degree of morphological difference" is the criterion of species status. Species are defined on the basis of their observable morphological differences.

Chronospecies (Palaeospecies)

The Successive species are replacing each other in a phyletic lineages which are given ancestor and descendant status according to the geological time sequence.

1.3 Phylogeny of Angiosperms

1.3. 1 Ancestors of Angiosperms

The Ancestors of Angiosperms/Development of Angiospermy: The fossil record has not provided conclusive information about the origin of angiosperms. Although absent in earlier Mesozoic strata, diverse and abundant angiosperms first appear in Cretaceous deposits in the form of leaves, flowers, pollens, seeds and fruits. In these specimens, angiospermy is clearly well established, so that no insight regarding its origin has been supplied by such fossil remains. Various groups of gymnosperms have been suggested as angiosperm precursors, but these hypothetical ancestors have not been universally accepted as authentic. But there are some groups which have some similarities with angiosperms. These groups are as follows:

1. **Amentiferae:** According to one hypothesis, the amentiferous flowering plants are the most primitive. This is because of the simplicity of their flowers, their lack of coloured petals, and their wind pollination. The stamens are also considered to be quite similar to the microsporphylls of gymnosperm. However, some botanists consider the simplicity of these flowers to be the result of reduction and specialization.
2. **Ranales:** Another hypothesis is based on the supposed strobiloid nature of certain angiosperm flowers such as *Ranunculus* and *Magnolia,* in which an indefinite number of microsporophyllus (stamen) and megasporophyllus (pistils) are spirally arranged on an axis with short internodes. Arber, Parkin, Bentham and Hooker, Bessey and Hutchinson supported Ranalian descent for angiosperms. This order also includes vessel-less families like Trochodendraceae and Tetracentraceae.
3. **Pteridosperm:** Oliver and Scott (1903) discovered the presence of seeds on the fronds of *Lyginodendron oldhamium*. They proposed the group name Pteridosperms to include those Cycadofilicales that bear seeds. According to Andrews (1961), some Pteridosperms may have given rise to angiosperms, like *Lagenostoma* possess cupulate seeds.

4. **Ephedrales theory:** Wettstein (1935) advocated for the origin of angiosperm from *Ephedra* like ancestors which resemble *Casuarina,* a member of the Amentiferae, in many respect. The male flowers of both *Ephedra* and *Casuarina* are simple and reduced. And there is certain similarity between *Ephedra* and *Casurina* as they both have single central microsporangiophore and megasporophyll; perianth is situated in the axil of a bract which may be in pairs of two or four. In *Casurina*, the bracteoles of the female flower become hard and form the protective covering.
5. **Gnetales:** Wettstein (1901, 1935) found that the Gnetales are closely related to Angiosperm and regarded it as transitional group between Conifers and Angiosperms. Later Markgraf (1930) and Fagerlind (1940) showed homology between the flowers of Gnetales and Angiosperms. The Gnetales resemble Angiosperm in many characters. The leaves are reticulately veined like dicotyledons and the wood is heteroxylous. The unisexual inflorescence of *Gnetum* is comparable to the catkin of many Amentiferous taxa as in *Ephedra* and *Casurina*. *Gnetum* possesses two unilocular anthers which often have a tandency towards their fusion. The similarities between *Gnetum* strobilus and catkin of Amentiferae are considered superficial. Molecular phylogenetic analysis also shows that Gnetales are not related to Angiosperms and thus contradicts this theory which is based mainly on morphological characters (Doyle 2001).
6. **Pteridospermales:** Andrews (1961) suggest about the origin of Angiosperm from some Pteridosperm which occurred in Palaeozoic period like *Lagenostoma* (having cupulate seeds), where as in *Gnetopsis* and *Calathospermum* the cupules are multiovulate. According to Long (1956), the first integument of an angiospermous seed is formed by the fusion of telomic units and second integument is an outgrowth of chalaza or first integument this theory support about the carpel origin from Pteridosperm cupule.
7. **Bennettitales:** The Bennettitales is Mesozoic Gymnosperm plant group which was growing in Jurassic to Cretaceous periods. The structure of strobilus or flowers of Bennettitales are bisexual and resemble to the flowers of present of *Magnolia.* The Bennettitales

1.3.2 Time and Place of Origin of Angiosperm

Age of angiosperms: The origin and early evolution of angiosperms are enigmas, for botanists, for over a century. Angiosperms or flowering plants form the largest group of plant kingdom, including about 411 families, 8000 genera and 3, 00,000 species. They are considered to be the highest evolved plants on the surface of the earth. Angiosperms are annuals or perennial herbs, shrubs, trees, climbers, twiners and lianas.

Fossil remains of the true angiosperm are found only in the later geological periods and as a group they are more modern than other vascular plants. The sudden appearance of the highly specialized angiosperms in the *Cretaceous* period is unexplainable. From this age angiosperms are now dominant vegetation of the earth with universal distribution.

Place of origin of angiosperm – It was earlier believed that angiosperms arose in the Arctic region (Seward, 1931), with subsequent southward migration. Axelrod (1970) suggested that flowering plants evolved in mild uplands (upland theory) at low latitudes. Smith (1970) located the general area of South East Asia, adjacent to Malaysia as the site where angiosperms evolved when Gondwana and Laurasia were undergoing initial fragmentation. Stebbins (1974) suggested that their origin occurred in exposed habitats in areas of seasonal drought. Takhtajan (1980), who believed in the neotenous origin of angiosperms, suggested that angiosperms arose under environmental stress, probably as a result of adaptation to moderate seasonal drought on rocky mountain slopes in areas with monsoon climate. Retallack and Dilcher (1981) believed that the earliest angiosperms were probably woody, small leaved plants occurring in the Rift valley system adjoining Africa and South America. Some of these angiosperms adapted to the coastal environments and became widespread following changings sea levels during the early cretaceous period. Although agreeing with the role of environmental stress, many authors in recent years (Hickery and Doyle, 1977; Upchurch and Wolfe, 1987; Hickey and Taylor, 1992) have suggested that early angiosperms lived along streams and lake margins (Lowland theory). Later they appeared in more stable back swamp and channel sites, and lastly, on river terraces. These sites would have been characterized by high nutrient levels and frequent loss of plant cover due to periodic disturbances.

1.3. 3 Primitive Living Angiosperm

Some flowering plants show primitive characters as in lower group in their evolution during initial periods. Scientist called them as 'living fossils'. These primitive plants survived due to favorable condition for them during present day, but their distribution is restricted now.

The primitive living angiosper belong to families Winteraceae, Magnoliaceae, Degeneriaceae, Himantandraceae, Eupomatiaceae, Annonaceae, Canellaceae, and Myristaceae which come under to the orders Magnoliales. Austrobaileyaceae, Amborellaceae, Monimiaceae and Calycanthaceae belong to the order Laurales and the order Trochodendrales with the families Trochodendraceae and Tetracentraceae. Some other orders like Piperales, Nymphaeales, Illiciales and Ranunculales are also reported as primitive members.

The most primitive families of flowering plants belong to the order Magnoliales with many primitive characters present in the eight families of the Magnoliales than any other order of the flowering plants. The primitive characters include being vessel-less, spirally arranged free floral parts, broad stamens undifferentiated into connective and filament, incompletely closed carpels, and monocolpate pollen like gymnosperms. These archaic features are always associated with some advanced features and some considered the Magnoliaceae to be the most primitive and others considered Winteraceae as that.

Some main characters of primitive angiosperms are listed below:

1. Woody habit
2. Leaves alternate, mostly simple, entire, pinnately nerved and stipulate.
3. Leaves folded in bud, showing conduplicate vernation.
4. The node with three or more gaps.
5. Primitively organized water conducting system vessel-less wood, tracheids with very oblique end walls; vascular rays heterogeneous.
6. Flower radially symmetrical and bisexual with large number of floral parts arranged spirally on an elongated floral axis.
7. Large solitary flowers, terminal on leafy branches.
8. All perianth parts morphologicaly similar.
9. Flowers insect pollinated.
10. Stamens broad laminar structures not differentiated into filament and connective.
11. Monocolpate pollen grains.
12. Carpels leaf- like, folded adaxially along the mid-rib with many ovules; a petiole like stipe is present, the carpels are not completely closed at the time of pollination; style is absent and the stigma is decurrent.
13. Laminar placentation with ovules situated some distance from the carpel margins.
14. Anatropous bitegmic ovules.
15. Fruit many- seeded follicles, developing from multicarpellary apocarpous gynoecium.
16. Seeds with abundant endosperm and a very small embryo.

Some genera are belonging to five most primitive families of angiosperms.

Following are the examples of primitive living Angiosperms:

Magnolia (Magnoliaceae)

The *Magnolia* genus is represented with 120 species dispersed in the tropical and subtropical area of the earth. Within India, around 12 species occur and the common species is *M. grandiflora* which is cultivated in gardens. The vessels occur of primitive type with scalariform perforations, showing scalariform intervascular pitting. The

parenchyma in wood is generally terminal apotracheal type and the rays are heterogeneous. The nodes are multilacunar due to presence of several leaf traces. They are evergreen/deciduous trees and shrubs. The leaves are alternate, entire and pinnately veined. The large deciduous stipules enclose the young buds. The flowers are large solitary or terminal, bisexual, actinomorphic and hypogynous. Receptacle is characteristic of the flower of *Magnolia,* where the floral parts are arranged spirally. The internodes are shortened with absence of vessels. The perianth (9-12 tepals) is arranged in trimerous whorls, the outermost whorl being sepaloid and calyx and corolla both appear but differentiation is not clear in some species. The several free and spirally arranged stamens are present which show primitive characters as three traced, laminar and produced above the microsporangia. The pollen of *Magnolia* is of monocolpate type, an ancestral gymnosperm type and also the sporoderm similar to that found in some gymnosperms. The genus *Magnolia* perhaps possesses the most primitive type of pollen in the Magnoliaceae. Small embryo and abundant endosperm is of primitive nature of *Magnolia*. The sarcotesta in the seed-coat is also of primitive nature.

Drimys (Winteraccae)

The *Drimys* genus is represented by 70 species distributed in Borneo, New Guinea, New Caledonia, Australia, Tasmania, NewZealand and South America. Fossil of *Drimys* are found from the west coast of North America, eastern Australia and Antarctica. The members had habit of trees and shrubs with alternate or subverticillate simple, entire, pinnately veined, dotted gland, aromatic exstipulate leaves. The nodes are trilacunar and the important character is the absence of vessels in the secondary xylem. The tracheids are thick walled with large circular bordered pits initially, then scalariform bordered pits, the rays are heterogeneous. The flowers are borne in fascicles and in their structure, they show extremely primitive features. They are bisexual, actinomorphic and hypogynous. The floral axis is short and the perianth is whorled, differentiated into calyx and corolla. The calyx is membranous sac enclosing the petals, split into two or four sepals. There are six or more petals, arranged in two whorls in imbricate manner. The stamens are arranged in spiral condition, the anthers are dithecous and introrse. The pollen grains are advanced as they are united in permanent tetrad, the distal aperture reduced to a circular pore and a reticulate exine. Few carpels are arranged in a single row and are free. The old world species of *Drimys* have very primitive type of carpels, reminiscent of young leaves, adaxially folded along the midrib. A petiole-like stock is also present. Stigma is decurrent. The fruits are many seeded follicles, and the seeds are with small embryo and copious endosperm.

Degenaria (Degeneriaceae)

Degenaria is a monotypic genus of the family Degeneriaceae with a very restricted distribution and endemic to Fiji Islands. The genus is only represented by *Degenaria vitiensis* species which is a small tree. The wood shows many primitive characters in its structure with the presence of thin walled vessel elements, with scalariform perforation with several perforation bars and also scalariform intervascular pitting, the rays are heterogeneous. The node is pentalacunar.The flowers are long pedicellate and supra axillary. There are 2-3 bracts on the pedicel and the presence of these bracts suggest that the solitary flower may develop from the inflorescence by the reduction of lateral flowers. The floral axis is short, perianth is cyclic, distinct into calyx and corolla. There is a single series of three persistent sepals and 12-13 fleshy and deciduous petals in 3-4 whorls. The stamens and carpels of *Degenaria* are very primitive. Stamens are many, laminar, undifferentiated into filament and connective. On the adaxial surface of the stamen a pair of long narrow microsporangia is present on each side between the median and lateral vein embedded in the sterile tissue of the sporophyll. Thus *Degenaria* has most primitive type of stamen in angiosperms. Pollen grains are also primitive type as they are monocolpate with sporoderm similar to the gymnosperm members. In gynoecium of *Degenaria* though the number of carpels and is reduced to one. The carpel is an extremely primitive with conduplicately folded structure. The margins of the carpel are completely free. The carpel is thus not completely closed in *Degenaria* at the time of pollination in primitive Angiosperms. Style is absent and the stigma is decurrent. The stigmatic margins are not fused at the time of pollination. Pollen grains are received by the outcurring glandular hairy carpellary margins. As fruits develop, the contiguous adaxial surfaces of the carpellary margins become concrescent. The fruit is large, leathery, asymmetrical and indehiscent.

1.3. 4 Inter-relationship among the major groups of Angiosperms

The Angiosperms are usually divided into two major groups the Dicotyledons and the Monocotyledons. We had already pointed out the inadequacy of criteria separating the two groups from each other. The overall evidence from morphology shows that the Monocots are derived from the Dicots. The single cotyledon of Monocot has arisen as a result of the loss of one of the two cotyledons of a Dicot ancestor, the surviving cotyledon quite often assume a pseudoterminal position. There are several Dicots in which one of the two cotyledons is partially or totally suppressed, such as in *Trapa natans, Carum bulbocastanum* and *Corydalis cava, Ranunculus ficaria* etc. On the other hand among the Monocotyledons the single cotyledon is not only lateral in position but the trace of the second cotyledon, the epiblast is present in quite a number of taxa, such as *Oryza*. How

one of the two cotyledons of Dicot might have become aborted is shown by the germination of certain species of the genus *Peperomia*. This is a typical Dicot plant. The embryo has got two equally developed cotyledons. The seed is endospermic and germination is of the epigeal type. The two cotyledons after absorbing endosperm, becoming raised above the ground by the elongation of the hypocotyl and forming the cotyledonary leaves. Hill (1908), however, observed that in a small group of geophilous individuals, one of the cotyledons remains within the seed and is entirely suctotial whereas the other cotyledon escapes from the seed, is carried high by the elongation of its petiole and assumes the appearance of the characteristic first leaf. The author suggests that the first leaf of a monocot seedling may therefore be of cotyledonary nature, thus accounting for both the cotyledons of their Dicot ancestors. According to Miss Sargant (1908), the single cotyledon of Monocot is the result of fusion between the two cotyledons of Dicots, brought about as a result of the geophilous habit of the Monocots. The fusion of the two cotyledons to form a cotyledonary tube is seen in many Dicotyledons, such as *Anemone coronaria, Erianthis hiernalis* and *Podophylum peltatum etc*.

1.4 Summary

Initially the population ecology study was carried out by botanists like Turesson (1922); Clausen, Keck and Hiersey; Gregen, Dowey and Long (1936) and Russion school (Sinka, 1948), they worked for the population structure with correlation to enviromnment. Generally, the Ecads are the population where the different of the morphological variations are disappear when they grow in same habitat that is these variation are not coded genetically (*Euphorbia hirta*) where as in Ecotype, the variation donot disapper as the characters are coded genetically (e.g. *Potentilla glandulosa*). This population isolation mechanism results in the development of the new species and there can be two methods of speciation *viz.* Allopatric and Sympatric pattern. Scientists have described different models or concepts of species which help to understand the species at different levels like the Biological, Ecological, Taxonomical, Numerical and Traditional *etc.* The origin of Angiosperms can be related to some fossil records which show their relation with Pteridospermales, Bennettitales, Cycadales and Pentoxylanean group whereas some botanist shows the affinities and origin of Angiosperms with lower group and primitive Angiosperms like Ephedrales, Gnetales, Amentiferae, Ranales and Isoetalean members. The time of origin of Angiosperm is believed to be in the *Cretaceous* period and the Origin place is described by different botanists to be in the Arctic region, South-East Asia and the Rift valley system adjoining Africa and South America. The primitive living Angiosperms belong to Magnoliales, Laurales, Trochodendrales, Piperales, Nymphaeales, Illiciales and Ranunculales.

1.5 Glossary

- **Population:** The group of individuals considered to gene-pool together at any one time because of the features they share in common
- **Ecads:** The term used for the plant species differing in appearance, specially in morphological features, their variation is due to environmental influences. When different ecad or ecophenes are transplanted in to the same habitat, these variations disappear.
- **Ecotypes:** Ecotype is an ecological unit to cover the product arising as a result of the genotypic response of a species to a particular habitat.
- **Allopatric and Sympatric Speciation:** The allopatric speciation is the differentiation and complete attainment of reproductive isolation of the population, which are completely geographicaly separated. Whereas sympatric speciation is the differentiation and complete attainment of reproductive isolation of the population, which are not completely geographicaly separated. There populations overlap in their distribution.

1.6 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. Which are possible places of Angiosperm origin?
2. Write the name of any two primitive Angiosperms.
3. The term ecotype was proposed by……
4. Who suggest about origin of Angiosperm from Pteridospermales?
5. Who define the biological species concept?

Section B : (Short Answer Type Questions)

1. Write the difference between the Ecads and Ecotypes.
2. Write the Bennettitales and Amentiferales theory of origin of the Angiosperm.
3. Write the note on the Turesson's Experimental work on population study.
4. Write a short note on interrelationship among the major groups of Angiosperms.

Section C : (Long Answer Type Questions)

1. Describe the palce and time of origin the Angiosperm with suitable ancestor theory.
2. Write a detail note on the primitive living Angiosperm

1.7 References

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Unit - 2

The Species Concept & Plant Nomenclature

Structure of the Unit:

2.0 Objectives

After going through this unit you will be understand about:

- hierarchy
- principles used in assessing relationship and determination of taxa and attribution of rank
- history and development of Plant Nomenclature (International Code for Botanical Nomenclature)

2.1 Introduction

The assembling of different categories (some common characters base) in a systematic enumeration can be defiened as taxonomic hirerarchy. The categories of higher level such as Division, Class, and Order etc. are referred to as the major categories while those at lower level such as secies, variety etc. are called the minor categories. According to the ICBN (1961), Article 2 and Article 3 indictes genus, family, order, class and division as principle rank of taxa in the ascending sequence. The plant kingdom is further divisible into the total 19 subordinate rank.

Plant nomenclature or the system of naming plants is an important part of taxonomy. It deals with the determination of correct name, to be applied to a known plant or taxon. It is well known that many plants have several common names in general use in various parts of the country; and often the same, common name is used for different plants and furthermore some plants have no common names, therefore it becomes essential to give to a particular plant a definite name which may be used all over the world. These are the basic problems which cause to assembls of all over word botanist together and developed a set of rule known as International Code for Botanical Nomenclature. The rules of ICBN is followed which divided and section and these sections further subdivided into articles and recommendation.

2.2 Taxonomic Hierarchy

The taxonomic hirerarchy means that assemblage of individuals or small distinct groups by some common characters and give them a position in a systematic enumeration. The taxonomic category is a level or rank assigned to that group. The taxonomic units can be defined as any group of indivisual sharing a highest number of characters among them and shown the greatest stability of characters within them.

Every category represents a group of plants population but no one of these is defined in a strict manner because their delimitation varies and each is subjective in character. The categories of higher level such as Division, Class, Order etc. are referred to as the major categories while those at lower level such as Species, Variety etc. are called the minor categories. A minor category may be considered to be one whose name is also a part of the name of a particular taxon but it is not necessary for mojor category bear any name which is not a part of the name of the plant included under it.

According to the ICBN (1961), Article 2 states that every individual plant is treated as belonging to a number of taxa of consecutively subordinate ranks, among which the rank of species is basic. And Article 3 indictes species, genus, family, order, class and division as principle rank of taxa in the ascending sequence. The number of rank of ranks can be increased by adding the prefix sub to the term denoting the ranks can be increased by adding the prefix sub to the term denoting the rank of by introducing supplementary term like subspecies, sub family, etc. or super class, infra class and so on. The plant kingdom is further divisible into the following subordinate rank as given below:

Plant Kingdom

Division

Sub-division

Class

Sub-class

Order

Sub-order

Family

Sub-family

Tribe

Sub-tribe

Genus

Sub-genus

Series

Species

Sub-species

Varieties

Subvarieties

Forma

Clona

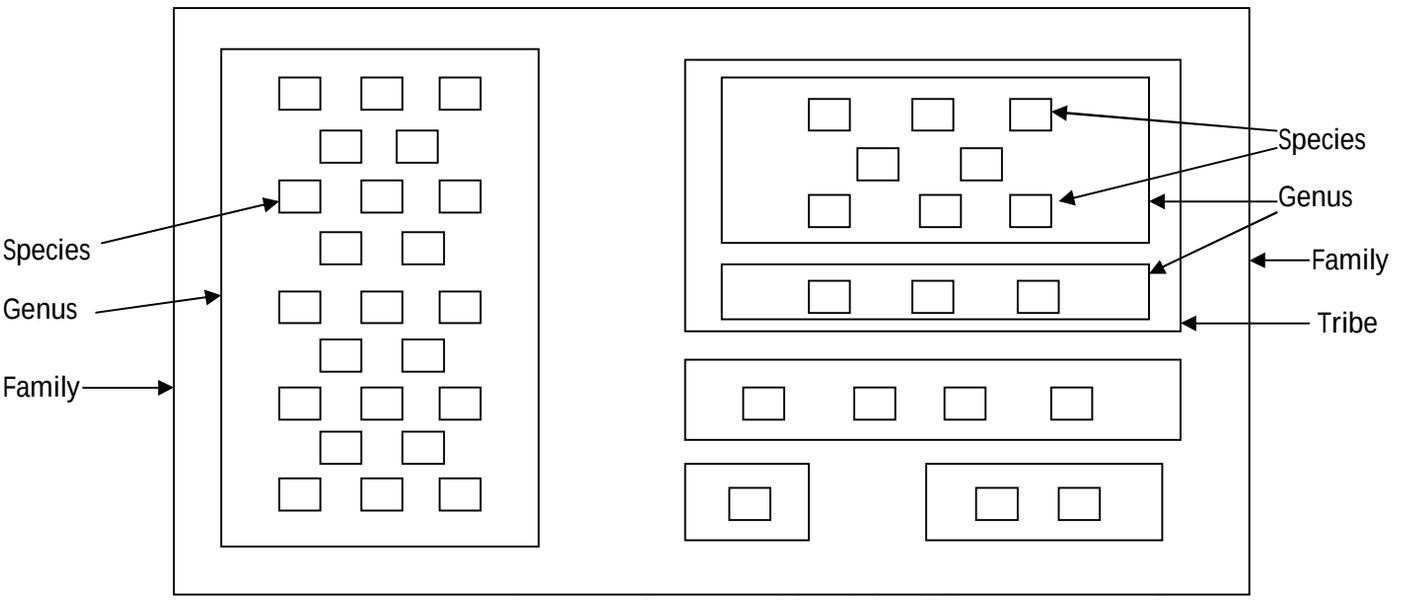

Fig. 2.1 : The Assembly of the different Categories

2.3 Principles used in Assessing Relationship and Determination of Taxa and Attribution of Rank

Quite apart from historical usage, a large number of factors are involved in preparing a natural system of classification. The principles can for convenience be discussed under three headings: Criteria, Guides, Practical Consideration. Whether or not they are followed is largely a matter of taste although some of them, at least, are widely accepted.

Criteria

It is of course impossible to discuss criteria without reference to characters which provide the criteria (i.e. principles taken as a standard of judging). Nevertheless, as characters are fully deal as some point discuss below:

1. ***Only genetically fixed variation should be used for classification:*** This was a principle accepted even by John Ray, and is one which causes us some difficulty only at and below the level of species where it is often not possible, in the absence of experimental evidence, to distinguish between environmental modifications and genetically fixed variations. In practice the taxonomist must be follow the character which are fixed if there is any doubt about the character are not consider for delimitation of taxa.
2. ***Natural classification should be firmly based on correlation of characters and discontinuity of variation,*** both for the assessment of relationship (in terms of overall similarity) and for the delimitation of taxa. As many characters as possible should be used in assessing relationship. For the any classification those characters which show the highest correlation, therefore these character are the most useful for classification. The vegetative characters are broadly use in the delimitation of groups above the species and sectional level than has been used previously. Character is not the "type" of a taxon but the range of variation and the number of intermediates which largely decide the delimitation of taxa. The assessment of relationship and the delimitation of taxa are more important than attribution of rank.
3. ***All available material should be used when classifying a group :*** Characters which appeared to provide good differentiate when the group was first described may no longer hold when more material is available this applies particularly to specific and generic concepts. Additional specimens frequently break down the differences used to delimit species in the pioneer phase of classification. New species described by various authors accumulate in genera originally based on far fewer species, so that a genus may become too broad and its content too heterogeneous for maintenance as a single genus; on the other hand, what were initially treated as two genera may have to be merged into one genera.

4. ***When any taxon is revised (be it species, genus or family), it should be studied throughout its entire geographical range :*** Above the species level this is often impossible when writing a Flora, but any taxonomic revision which entails the new delimitation of taxa and changes of rank should be done throughout the range of the group. The dangers attendant on neglecting to do so are illustrated in a paper by Tutin (1959).
5. ***Consistency of treatment :*** It is evident that no definitions of taxa or assignation of rank can be consistently employed throughout the plant kingdom. Nevertheless, there is room for more consistency than classification shows at present. Inconsistency of treatment is a very real problem and results from three main causes: lack of time for one taxonomist to study the whole group; psychological differences between taxonomists and between the different concepts they hold; revisions based on different material. Taxonomists should be as consistent as knowledge of the facts allows, within the taxon with which they are dealing. To divide the genera in one part of a family and leave the rest of them unconsidered may sometimes be necessary but is not good taxonomy. If we recognise *Pulsatilla* and *Ceratocephalus* as genera separate from *Anemone* and *Ranunculus* respectively, there is every reason (in fact rather better reasons) for recognising *Consolida* as a genus separate from *Delphinium.* Inconsistency of treatment is very noticeable in the preparation of Floras and makes comparison difficult a failing which the *Flora Europaea* should do much to alleviate for the European continent.

Guides

1. ***Ability to cross :*** The claim of many biosystematists that *species* should not be treated as such unless their members are potentially inferfertile or that species should be potentially intersterile with others and their components interfertile, is no longer acceptable, though in the majority of cases the factual basis of the claim may be true. It cannot be accepted as a general rule for several reasons:
   (a) Inability to cross does not imply lack of genetic relationship.
   (b) Morphological differences are not necessarily associated with inability to cross.
   (c) In polyploid complexes the polyploid derivatives may be able to cross unlike the diploids from which they are derived and this tendency increases with the level of ploidy. The derivatives are normally unable to cross with their diploid progenitor from which they may be morphologically inseparable. In such cases crossability is at variance with phyletic relationship.

   As species of different genera are usually intersterile (with exceptions in the *Orchidaceae,* etc.), the inability to cross is of little help in separating genera but if we shift the stress from the differences between genera to the resemblances between

the species within them, crossability (and resemblances in chromosome structure) can help in delimiting the genus as a natural group of species (Rollins, 1953); the same would apply to sections within it.

2. ***Chromosome morphology and number :*** The karyotype can prove a most useful guide both in classification and phylogeny. We have already discussed the difficulties of using chromosome number for the delimitation of species, the conclusion being that it should be associated with constant morphological differences before the population can be recognised as a separate species. The karyotype, however, can be particularly useful as a guide in the delimitation of genera and in assessing their genetic relationship. In many groups virtually no help can be derived from this source, but in others it either confirms the classification based on gross morphology or suggests how a more natural classification might be achieved.

   The work of Gregory (1941) on the karyotype of *Ranunculaceae* shows how the genera might be more naturally grouped on the basis of chromosome morphology and number, although this has not been translated into a revised classification.

   It must be stressed that chromosome morphology and number are *a guide* in classification, not an unequivocal criterion. By showing up likely phylogenetic relationships, they suggest more naturatral groupings. But it is up to the taxonomist to confirm these by finding correlated morphological characters so that a group can be satisfactorily defined and delimited.

3. ***Homologous variation :*** The law of homologous series workout by the Vavilov (1922). Homologous variation has its basis in a series of ancestral genes. Such variation is particularly obvious in crop plants. In the native flora natural selection often eliminates certain variations, so that a rare species usually shows only part of the variation found in a commonerally. Nevertheless, an appreciation of the law of homologous series can be of great help to the work of the taxonomist.

4. ***Geographical and ecological distribution :*** A great deal has been written, especially by German and Russian botanists, on the "geographical method" in taxonomy. All taxa (except *forme)* have a fairly well defined geographical distribution, the outcome of evolutionary history and climate. At the level of the species and below, geographical and ecological distribution play a rather different role in classification. Two plants growing in different areas (allopatric) may or may not be interfertile only experimental evidence can show us that. But if they grow in the *same* area (sympatric), and particularly when they also grow in the same habitat, the situation is different. If the two plants continue distinct, it is fair to assume that there is some reproductive

barrier between them. If we are in doubt whether to assign specific rank, information on their distribution and ecology may therefore be decisive.

5. ***Phylogenetic considerations :*** Monophyly is a desirable criterion of taxa, at least above the level of hybridogenous species. Evidence for monophyly (or polyphyty) must normally be indirect, and based upon the interpretation of other evidence. This involves the study of as many characters as possible, including their relationship to one another and their functional importance, the geographical distribution of taxa, their comparative karyology and ability to cross. By studying this evidence we may feel that we can distinguish between resemblance due to patristic affinity and resemblance due to convergence. In the latter case we should study the group again to see if it can be classified in a manner more consistent with its probable evolutionary history.

   Phylogenetic considerations, however, are interpretative and cannot be used to construct a primary classification. They can only be used to interpret an initial classification in evolutionary terms and by revealing undesirable features allow us to improve it as far as we can without abandoning the practical aspect of classification. We must, however, be very careful as to how we have inferred monophyly or polyphyly. If these inferences have been deduced from the classification, then it is logically incorrect to use them to alter the classification. Phylogenetic inferences drawn from cytology or geographical distribution, for instance, can only be used to support or improve a classification in so far as they have not been used in its construction.

Practical Considerations

If a general classification is going to be widely used, it needs to work. For the place taxa in higher taxa it needs to examine depratively. This means that practical considerations which mitigate against the 'objective' aspect of classification are very important in systematics and necessitate that science shall come to terms with art if biological classification is to be of maximum use to science.

1. ***Ease of observation :*** Although we have stressed the desirability of using as many characters as possible in classifications, this does not mean that all characters are equality suitable for ready differentiation or diagnosis. Botanists have, in general, felt that humanity is best served if flowering plants are distinguished by at least some difference in gross morphology, though they may be associated with internal differences. These "marker" characters are extracted from a much larger number considered in assessing the taxonomic position of the group and are often the one used in the construction of key.

2. ***Size of taxonomic groups :*** This is probably one of the maior and most neglected factors effecting classification. None of Linnaeus's genera was large in 1753 the biggest, *Carex,* contained only 29 species whereas today it contains well over 1000. The 100 families recognised by de Jussieu suggest that he considered this to be a convenient number for the division of the flowering plants, but since that time the number of plants known has enormously increased and many of the old genera and families have been divided up. J. Hutchinson recognises over 400 families, most of which are apparently based on genera which were already known to de Jussieu and were accommodated by him elsewhere. About one third of de Jussieu's genera are placed in the same families by Hutchinson, and the rest in different families. Though some scientists may find this state of affairs rather shocking, it is an inevitable effect of practical convenience which is a necessary part of classification; it is nothing for taxonomists to be ashamed of.

It is a quite commonly stated principle that *the morphological gap between two taxonomic groups should be inversely proportional to the size of the groups.* This is, in effect, a practical necessity, and was a principle adopted by de Caudate and by Bentham and Hooker and one which prevented them from recognising a very large number of monotypic genera and families. The *Scrophulariaceae* and *Gesneriaceae* are two large families which differ mainly in their placentation. It is the fact that they form such large and apparently natural groups that makes them worth recognising as different families, instead of tribes of the same family. The same may be said of *Labiatae* and *Verbenaceae, Caprifbliaceae* and *Rubiaceae.* On the other hand, one could point to small genera in the *Berberichiceae* or *Nymphaeaceae* which might be treated as separate families. From the point of view of delimitation and degree of divergence they are a good deal better as families than the pairs we have just mentioned, but because they are small there has been an understandable reluctance to raise their rank.

In less worked areas it is most probable that an increase in the amount of material available will eventually lead to a broadening of specific limits. To describe small species in the pioneer phase can hardly be avoided, and more confusion may be caused by too wide a species concept in this stage than by too narrow a one. But to split when abundant material does *not* justify it (i.e. by ignoring continuity of variation) is inexcusable; yet it is still done, even in sexual groups. Several arguments have been put forward in favour of splitting of which the following seem the most relevant:

i. No clear line can be drawn between specific and subspecific taxa. Therefore morphologically distinguishable populations which occupy a different place in the economy of nature should be accorded similar status. Specific rank is therefore accorded to all of them a return to the monotypic concept of species followed by Linnaeus.

ii. If nature shows a vast number of such populations, it is up to the taxonomist to express the fact in his classification. He should not hide or ignore that variation of nature. The first two arguments are tenets of the Komarovian School.

iii. Since many biosystematists have emphasised intersterility as the main criterion of species, numerous 'genetic' species can be described.

iv. At the level of the genus or family, small groups are more natural and easier to use than large ones.

v. The *Index Kewensis* includes no infra-specific taxa, so only species can be readily traced.

Taxonomy has two components: analysis and synthesis. The taxonomist must often ask himself the question: Is it more important to emphasise resemblances or differences? The answer will vary from group to group. The role of the family is one of synthesis the expression of relationship is therefore of overriding importance. It is at specific rank that we are most concerned with discrimination. Other things being equal, binomials are more convenient than trinomials because they are shorter. Provided some allowance is made for local hybridisation, we must be able, for communication, to refer most of our specimens to binomials. If variation is too great to allow us to do this, then our species concept is too narrow to provide a workable classiffication.

3. ***Economic importance :*** Like size of flower, this must have had a considerable bearing on classification. Groups which are economically important either for their products or for their place in agriculture or horticulture are likely to be extensively studied, while some humble group like the *Urticaceae* wait in vain for a botanist to open the cupboard and have a good look at them, let alone study them *in vivo.* It is not that one grudges the *Orchidaceae, Gramineae, Eucalyptus, Nicotiana* or *Gossypium* their economic importance; it is only that one wish *all* groups were economically as important, because the emphasis on economic groups is bound to unbalance classification. On the other hand, the extensive study of cryptic characters in economic groups has been extremely rewarding. The *Gramineae* provide a splendid example of what can be achieved in a difficult group where con-

vergence has often obscured phyletic relationship. Not only are the *Gramineae* classified in a more natural manner than ever before; we understand them infinitely better. The same cannot be said of the *Cyperaceae* which are economically relatively not important.

4. ***Stability :*** A great deal has been made of the desirability of stable classification. Gilmour has gone so far as to consider this of overriding importance. Although we agree that stability would be desirable, we certainly do not believe that changes and very considerable ones should not be made. The discoveries of the present century are providing a wealth of data that should be incorporated into classification. Not to use these data to improve a natural classification is too high a price to pay for stability. By all means let changes be made cautiously, and not before the evidence is strong.

There are three different ways in which changes in a system of classification can be brought about:

1. ***Relationship*** *:* If it is found that a plant is overall more nearly related to another genus than to the one in which it was originally classified, then it should certainly be removed either to another genus already accepted or to form one of its own, depending on the amount of divergence shown. Changes in this direction seem inevitable and science will not be served by ignoring them.

2. ***Rank*** *:* We have indicated that changes of rank are very largely matters of opinion. Rank is influenced by tradition, the size of the group and the degree of divergence. Yet if we are trying to classify consistently and to produce a well balanced classification, changes of rank cannot be avoided. In this connection it is obvious that the stability of species and genera are particularly desirable because they form the binomial which we must use for the designation of species. Yet changes in family status can be hardly less trying when we are looking up plants in a flora or herbarium. Changes in the rank of tribes, sections or subspecific taxa result in much less inconvenience. On the whole, however, we deprecate change of rank except for motives of consistency and in the achievement of a balanced natural classification. The effect of raising the rank of a taxon is to draw more attention to it.

3. ***Nomenclature :*** Changes that are not taxonomic, in the names of plants are strictly nomenclatural and result from the correct application of the rules of the Code, particularly those concerned with priority of publication. These legalities can prove very trying and take up a disproportionately large amount of a taxonomist's time which would be better spent in studying plants. However, a fairly stable set of rules

and it is to be expected that their consistent application will eventually result in a more stable nomenclature. Though we must follow rules as they stand, we cannot help feeling that name changes for anything except good taxonomic reasons are harm to biology.

2.4 History of Plant Nomenclature

History of Plant Nomenclature

Initially the rule of plants nomenclature was developed and formed modern sets of rules, these rules are known as International Code of Botanical Nomenclature. These rules of nomenclature started with Linnaeus "Critica Botanica (1737)" which was improved later (1751) in 'Philosophia Botanica' and established the actual beginnings of nomenclature for plants. Many botanist worked indivisually but the steps where the all botanist start gatherered for conclude the problus of nomenclature was initiated internationally at first time at Paris (1867) where the many European and Americal botanist came first time together.

Paris code (1867)

The first 'International Botanical Congress' meeting organised to formulation of rules for nomenclature at Paris (Aug, 1867). Around 150 European and American botanists gathered for this meet. Alphonse de Candolle prepared a manuscript 'Lois de la nomenclature botanique (the laws of botanical nomenclature)'. These laws were accepted as the best guide for nomenclature of plants and called as de Condolle rules. This meeting called as Paris code and accepted mostly all botanists only some English (Kew) and Americal botanist not admit they made their on rules (Kew rules, which is not written any where).

Rochester code (1882)

A set of rules of nomenclature prepared by some United States, botanists under the leadership of N.L. Britton, New York Botanical Garden (Rochester). This meeting is known as the Rochester Code which is based on Paris Code. The basic difference between Paris and Rochester codes were: (1) the concept of types as a scientific and fundamental unit for naming taxa (Type Specimen); (2) the Latin diagnosis also needed with the specimen.

Vienna code, (1905)

Third Inernatinal Botanical congress was held in the Vienna (1905) and this none as Vienna code. This based on Paris code but some modification and amplified considerably. The main rule is generic name should be conserved because their extensive use earlier

(Nomina generic conservanda). The rule of tautonymy (repetition of the generic name) was note accepted. New plant always describe with Latin dignoseis.

American Code, (1907)

This was the proposal against the Vienna code and refused all rule, this formed by the Rochester botanist. The main acceptance of the 'Princile of Nomina Conservanda' and the new plant always describe with Latin dignoseis. The revision of the Rochester code was published in 1907 under the name of American code.

International Rules of Botanical Nomenclature, (1930)

T.A. Sprague, M.L. Green and A.S. Hitchcock organized 5th International Congress at Cambridge (1930) England. The effort was made to harmonise the all difference between the Vienna and American codes. As the result the first time in botanical history, a code of nomenclature came into being that was international in function as well as in name.

Amsterdam Congress, (1935)

The Sixth International Botanical Congress was held at Amsterdan. The major change was to provided for the advance from January 1, 1932 to January 1, 1935 as the date after which all diagnoses of plants new to science (excepting bacteria) must be in Latin. The rules of nomenclature are divided into chapters and sections and these are divided into articles and recommendations. The third edition of this rules contained an enumeration of the nomina generica conservanda as of 1935.

Stockholm Congress

The Seventh International Botanical Congress met at Stockholm in 1950. Rules of nomenclature were discussion and considered like introduced a certain number of definitions on types.

Paris Congress, 1956

The eight Congress principles and recommendations of the rules were treated separately. The chief decisions of this Congress were like effective publications and the adoption of a list of *Nomina Specifica Rejicienda* which has the same stabilising effect as introducing a *Nomina Specifica Conservanda.*

Montreal Congress, (1961)

The ninegth Congress rules regarding nomenclature of fossils plant species were prepared and list of Nomina Familiarum Conservanda was accepted.

Edinburgh, (1964)

The 10th Congress had some changes in nomina familiera conservanda accepted in the

previous congress were done. List of recently discovered gymnospermous families was prepared and a list of the meanings of difficult words relating to nomenclature was prepared.

Seattle code or Seattle congress, (1972)

The eleventh congress (1969) was orgnized in Seattle and knon as Seattle code, where mainly discussion were made on species, genus and below levels. A committee was formed to deal with the problems relating to the position of taxa above the rank of family. This congress produced a Code published in 1972.

Leningrad, (1975)

The twelth congress was held at Leningard (1975) where some proposals regarding some changes in the type concept were made and work done prepration of list Nomina Generica Conservanda.

The Tokyo congress:

The 15th interational congress was orgniged at Tokyo (1993), which was published in 1994.

2.5 International Code for Botanical Nomenclature

For the naming of the plants species and systematic enumeration according to the taxomonic herarcy was followed, similarly the naming a plant species the rules of nomenclature are consider. The rules of International Code for Botanical Nomenclature is followed which divided into sections and these sections are further subdivided into articles and recommendation.

Principles of ICBN

1. Botanical nomenclature is independent of zoological nomenclature.
2. The application of names of taxonomic group is determined by means of nomenclatural types.
3. The nomenclature of a taxonomic group is based upon priority of publication.
4. Each taxonomic group with a particular circumscription, position and rank can bear only one correct name the earliest that is in accordance with the Rules, except in specified cases.
5. Scientific name of plants are treated as Latin regardless of their origin.
6. The rules of nomenclature are retroactive when expressly limited.

Chapter 1st: General Consideration and Guiding principles

Gives following articles provide in assence a preamble to rules

i. Establish the need for rule.
ii. Declare the independence of botanical from Zoological nomenclature.
iii. Provide for the basis of scientific names on the Latin language.
iv. Delimit nomenclature to dealing with terms of rank and name
v. Specify that the rule apply to all forms of plant life/

Chapter 2nd: Categories of Taxa and the term denoting the taxa

This chapter is composing of 4 article by which are specified the name of the categories from species through division and their order sequence. The ending of the name indicate the categories for example:

Kingdom
Division (-phyta) Spermato**phyta**/ Emryo**phyta**
Sub-division (-phytina) Ptero**phytina**
Class (-opsida) Magnoli**opsida**
Sub-class (-idae) Pter**opsidae**
Order (-ales) Ros**ales**
Sub-order (-ineae) Ros**ineae**
Family (-aceae) Ros**aceae**
Sub-family (-oideae) Ros**oideae**
Tribe (-eae) Ros**eae**
Sub-tribe (-inae) Ros**inae**
Genus {us (*Pyr**us***); All**ium** (um); Arob**is** (is); Ros**a** (a); polypog**on** (on)}
Sub-genus
Series
Species
Sub-species
Varieties (-var)
Subvarieties

Forma (-f)

Clona (-Cl).

The name of a family should be with suffix –aceae (like- Ros**aceae**) but there are 8 different families of angiosperm which not followed this rule so according to ICBN there name are changed:

Names under usage		**Names according to rules**
Cruciferae	-	Brassicaceae
Guttiferae	-	Clusiaceae
Leguminosae	-	Fabaceae
Compositae	-	Asteraceae
Gramineae	-	Poaceae
Labiatae	-	Lamiaceae
Palmae	-	Arecaceae
Umbelliferae	-	Apiaceae

Chapter 3rd : General Consideration and Guiding principls

This chapter cantains the major portion of the rules and divieded into 15 sections and among these following first three section are discuss below:

Section 1st : The principle of priority.

According to the principle of priority rule the first validly published name of a species becomes its valid name and if other names are published, subsequently for the same taxon then they become synonyms. This was decied that "Species Plantarum (1753)" for the flowering plants by Linnaeus was starting point.

Cleome gynandra Linn. (1753)

Cleome pentaphylla Linn. (1762)

Gynandropsis pentaphylla De Condolle (1824).

According to rule *Cleome gynandra* Linn. (1753) was correct.

Section 2nd: The type method.

The 5th article of section 2 give the detals for name of plant species based on nomenclatural type. Nomenclatural type is a herbarium specimen which is used by author for the naming of species. If the type specimen is not prepared by by author then only the drawing or photograph were used.

The kinds of type are depending on the way in which specimen selected these are followed:

1. **Holotype :** It is the particular specimen or material used by the author this always carried the name of the taxon which given by author.
2. **Lectotype: A** specimen or other material which is duplicate of the holotype.
3. **Syntype:** When two or more then specimens are cited and no holotype was describe by author out of them any one specimens were designated simultaneously as the syntype type.
4. **Neotype:** It is a specimen selected to serve as the nomenclatural type of a toxon in a situation when all material on which the taxon was describe is missing.
5. **Isotype:** It is a specimen which is duplicate of the holotype.
6. **Paratype:** It is a specimen given along with the the holotype.
7. **Topotype:** Specimen collected from the same locality from which the holotype was collected.
8. **Cotype:** Other material collected same plant from which holotype collected.

The following some other type also describe

1. **Clastotype:** The portion of the same clone.
2. **Paralectotype:** The specimens which were left after the selection of lectotype out of syntypes.
3. **Synonymotype:** It is that constituent element of a taxon quoted by the author as being identical with the newly described taxon.
4. **Schizotype:** Any fragment of a type is known as Scl1izotype.
5. **Merotype:** It is a fragment of original holotype which has been divided into two or more pieces after having been used as a basis for description.

Section 3rd: Limitation to the principle of priority

According to this section the name should be selected for the seed plants, Pteridophytes, Algae and Hepaticae up to 1/5/1753. After this date any species should be need ICBN rull criteria.

Rules of Botanical Nomenclature:

Some other rules are follows:

“213” All those plants which belong to one genus must be designated by the same generic name.

“214” All those plants which belong to different genera must be designated by different generic names.

"257" The specific name must distinguish a plant from all its relatives.

"264" The original place of plant does not give specific difference.

"285" The specific name should always follow the generic name.

Ranks of taxa

Taxon is a taxonomic group of plants or taxonomic unit of any rank. Taxa word is plural of taxon. For example the species *Brassica campestris* Linn. is a taxon. The family Brassicaceae is a taxon. These are coined as follows:

1. By adding the prefix 'sub' to the terms, For example - sub-family, subgenus, subspecies, etc.
2. By introducing supplementary terms, between two principal ranks of taxa; for example (i) between family and genus - tribe; (ii) between genus and species-section, series. (iii) Below species - variety and form.

Binomial System or Binomial nomenclature

The botanical name of any plant is in two parts, the first part is the name of genus name and second is the specific epithet *e.g.* for Pea is *Pisum sativum* write in italics when printed and underlined when it typed or hand written. The binomial names are more definite and precise than ordinary names. Being generally in Latin, they have a universal acceptance by people of all languages. They indicate the genetic relationship and descent of individual plant. The concept to write the generic name and species epithet with author citation rules is given below:

The Generic Name

It is always a noun, in the singular or nominative case and is always written with first capital initial letter. The generic name may be descriptive, aboriginal name of the plant or a name in honour of a person

These names have come from many sources *viz.*:

1. **Honour of Person:** Many genera have been named in honour of some person, usually a botanist as *Bauhinia,* honours the two Bauhin brothers; *Bignonia* the Abbe Bignon and *Nicotiana* for Jean Nicot, who introduced tobacco into Europe.
2. **Special Feature of Plant:** Many generic names are formed usually expressive of some feature of the plant. Examples are *Leucadendron,* ‘silver tree’ from the white leaves, *Xanthoxylum,* "Yellow-wood", *Oxydendrum,* "sour-tree", from the acidic

leaves.

3. **Land of its Discovery;** In many cases some names for plants existed in lands where they discovered, these names from the native languages and were converted into Latin generic names. Among these may be mentioned *Tsuga,* from the Japanese and *Pandanus,* from the Malayan *etc*.
4. **Fancy or Mythology;** Another type of genetic names includes those of fanciful, mythological, or poetic origin *e.g. Theobroma* (god's-food) was applied to the chocolate plant.

The Specific Epithet

The species may be regarded as morphological definable units, made up of groups of individuals, which are usually interbreeding. These names indicate the kind of plant and facilitate the members of a genus to distinguish from each other. They may be either an adjective indicating a distinguishing characteristic of the species. It is second part of the scientific name, called by Linnaeus the "trivial name". It may be derived from many sources.

1. **Descriptive Adjective :** It might be expected that this would continue to be the commonest form of the specific epithet, as indeed it is. These may be indicative of (a) colour, alba (white) and nigra (black); (b) may indicate the size, shape or habit of the plant, as *nana* (dwarf) and *gigantea* (giant); (c) may designate the plant's habitat, as *arvensis* (in fields) and *aquatica* (in water) (d) may indicate other characteristics, *fragrans* (fragrant), *spinosa* (spiny), *tomentosa (woolly), religiosa* (religious) and *toxicaria* (poisonous) (e) may indicate the region where the plant was found, as *chinensis* (Chinese), *Japenica* (Japanese), *nepalensis* (of Nepal) and *indica* (of India).
2. **Second Type :** Second type of specific epithet is a descriptive adjective formed by combining 2 or more Greek/ Latin words which making reference to some characteristic feature of the plant, as *angustifolia* (narrow-leaved), *cordifloia* (with heart-shaped leaves) and *quadriloculari*s (four-celled).
3. **Third Type** : A third type is formed from a noun, with a suffix indicating some resemblance or relationship, as *amaranthoides* (resembling *Amaranthus).*
4. **Fourth Type :** A fourth type is in honour of some famous botanist like roxburghii *(Yanda roxburghii)*.

In taxonomy all the names are treated as Latin names. The names are treated according to the principle of International Code of Botanical nomenclature (ICBN). The names are of following kind:

- **Homonym:** The names spelt identically but based on different types (Art. 64).
- **Naked name:** The name which is published without description or diagnosis or reference. These names are not validly published.
- **Synonym:** When one of two or more names considered applying to the same taxon.
- **Tautonym:** It is a name in which the specific epithet repeats exactly the generic name.

The Authority

The name of person or persons written after the botanical name is called the authority of the name. Authority is given when some person or persons have originally published the account of, and described each taxon whether family, genus or species and gave it a name. For example *Mimosa pudica* first named and described by Linnaeus becomes the authority for that name and it is written as *Mimosa pudica* L. Similarly *Eruca sativa* Lamk show that Lamarck first named the species. The authors names are write in abbreviated form like Linn. or L. for Carolus Linnaeus, Benth for G. Bentham and DC for A.P. de candolie etc.

2.6 Summary

The systematic enumeration of a taxon can be defiened as taxonomic hirerarchy. The categories of higher level such as Division, Class, and Order etc. are referred to as the major categories while those at lower level such as secies, variety etc. are called the minor categories. According to the ICBN (1961), Article 2 and Article 3 indictes genus, family, order, class and division as principle rank of taxa in the ascending sequence. The plant kingdom is further divisible into the total 19 subordinate rank.

Plant nomenclature is an important part of taxonomy, which deals with the determination of correct name, to be applied to a known plant. It is well known that many plants have several common names in general use in various parts of the country, a particular plant a definite name which may be used all over the world. These are the bsic problems which cause to assembls of all over word botanist together and developed a set of rule known as International Code for Botanical Nomenclature. The rules of ICBN is followed which divided and section and these sections further subdivided into articles and recommendation.

2.7 Glossary

- **Plant nomenclature:** Plant nomenclature is the system of naming plants according to therules of ICBN, nomenclature is an important part of taxonomy.
- **Holotype:** It is the particular specimen or material used by the author this always carried the name of the taxon which given by author.
- **Neotype:** It is a specimen selected to serve as the nomenclatural type of a toxon in a situation when all material on which the taxon was describe is missing.
- **Topotype:** Specimen collected from the same locality from which the holotype was collected.

2.8 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. The first 'International Botanical Congress' (Aug, 1867) meeting organized at…….
2. Write the new name of Guttiferae according to ICBN……..
3. A specimen or other material which is duplicate of the holotype is known as…….
4. Write the full from of ICBN.
5. Can a karyotype prove a most useful guide both in classification and phylogeny?

Section B : (Short Answer Type Questions)

1. Write the different between holotype and isotype.
2. Write the principle of ICBN.
3. Write the short note on taxonomic hierarchy.

Section C : (Long Answer Type Questions)

1. Describe the rules of ICBN.
2. Describe the principles used in assessing relationship & determination of Taxa
3. Describe the history of plant nomenclature.

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Unit - 3

Biosystematics

Structure of the Unit:

3.0 Objectives

After going through this unit you will be understand about:

- the aims and principles of Biosystematics
- the various components of Biosystematics
- the Alpha, Beta and Omega Taxonomy
- the Numerical Taxonomy: a modern concept in the classification of the plants
- prepration of Keys and their role in identification of taxa

3.1 Introduction

Camp and Gilly (1951) proposed some endeavor under "Biosystematy" to cover newer approaches in plant taxonomy. Later on in 1965 Lawrence introduced the term Biosystematics "*Biosystematics is a phase of botanical research that approves the study of living population and delimit the natural biotic units, and classify the taxa of different orders of degree*". The main aim of biosystematics is to distinguish the evolutionary units and prepare new taxa and check the limit of those that already exist.

The Alpha taxonomy is mainly the history of taxonomy which covers all initial the taxonomical work by different scientists. The main drawbacks of the alpha taxonomy are that, it is based just on form and shape of organisms and does not consider other features, so it is difficult to understand the evolution of species. Beta Taxonomy is the phase where the taxonomist takes in to account all morphological characters which appear during the lifecycle of the organisum and applied them to give a systematic position to the taxa. Omega Taxonomy which has widest correspondece of all disciplines of plant sciences, is based on all the information and data available for the plant species *viz.* Micro-Morphology, Anatomy, Palynology, Embryology, Cytology, Phytochemistry, Genome Analysis and Nucleic Acid Hybridization as Taxonomic Evidence; Serological, Computer (Internet/online data) and GIS *etc.*

Michel Adanson (1763) for the first time pointed out that equal weightage should be given to all characters while classifying any plant species. He initiated this concept first time and these principles are called as '*Adansonian Principles*'. The Adansonian principles have received great support since 1960 and developed new methods in taxonomy included under a general term Numerical Taxonomy, which is also known as modern classification method. The Numerical Taxonomy is analysis of various types of taxonomic data by mathematical or computerized method which is used to find out the similarities or affinities between taxonomic units, and then arrangement of these units in the order of higher ranking taxa on the basis of these similarities.

3.2 Biosystematics

Biosystematic studies include all the relationships with the emphasis on phylogenetic and phenetic relation. These relations are:

1. Relation of descent
2. Relation of similarity
3. Spatial relationship
4. Trophic relationship

Biosystematics is defined some scientist as an application of genetics (cytogenetics), statistics chemistry to the solution of systematic problems in order to provide explanations about the diversity of the organism within the frame of the theory of evolution. Investigation of variation through experiments in comparative morphology, anatomy, seed coat microsculptural features, reproductive biology, phytochemistry, embryology, karyotype, meiotic behaviour studies and to correlate these characters with adaptation of organisms existing in that locality; and to describe the diversity of organisms to delimit it to assess the evolutionary adoption processes which occur within the population. Further to provide a hypothesis for the systematized diversity for the sake of devising ways to prove that hypothesis is called as biosystematics.

3.2.1 Aims of Biosystematics

The main aim of biosystematics is to delimit evolutionary units and devise new taxa and check the limit of those that already exist. The object is to work out the operative processes of speciation rather than the speciation itself. At a particular time interval, phenotype is the final result of actions and reactions between and the factors of the environmental complex on one hand and genotype on the other. Biosystematics emphasizes on this relationship and evaluates the phenotypic expression, the result of the variation in the factors of the environmental complex and genotypic compound.

3.2.2 Principles of Biosystematics

Biosystematics include the investigation of plant variations and to differentiate between the genotypic and phenotypic variation patterns and the way that variation will be converted for adaptation as to how polyploidy especially allopolyploidy, amphiploidy aneuploidy and polymorphism will promote the evolutionary mechanisms. Due to different geographical and ecological places certain physiological heritable adaptive characters are produced in an organism which will be selected by either biotic or abiotic selection.

The species can be recognized morphologically but the characters merely serve as markers for the boundaries determined by the genetic methods. Morphological evidences are used to recognize species which are defined in genetic terms. Instead of creating morphological boundaries around species, geographical, ecological adaptational characters, changed chromosomal data of cytological and genetical evidences are also used to create clear boundaries around the species.

Biosystematics analyzes breeding system, studying patterns of variation, determines the evolutionary potential and does the pertinent work in the chemical, numerical, cytological, anatomical, embryological and palynological aspects of systematics for selected groups of

organisms. According to Mayer (1959) subspecies category is not suitable for describing the population structure of the species which is the aim of many biosystematists. Further more, he did not suggest that it should be abandoned as a satisfactory unit as, although subspecies is not a unit of evolution but conceals inter and intra population variability of species. The experimental taxonomist strives to arrive at an overall picture of biological relationships by attempting to correlate the relationship evidenced by ecology, cytogenetics, gross micro-morphology, palynology, phytogeography and physiology. There are advantages and limitations in each of these approach that but it is significant that the findings of taxonomy (classical as well as modern) and that of biosystematics, each contributes to the other to build a stronger single disciplines. Lawrence (1964) further identified biosystematics with experimental taxonomy. Some scientists equated the study of population genetics with experimental systematics. The experimental systematist usually begins with classical interpretation of species and works backwards so as to understand the genetic mechanisms of its material. The population geneticist begins with the raw population, discarding in his own mind any classical concept of species and works from there into a series of group concepts which may or may not be comparable to the taxonomist concept of species. Experimental Taxonomy does not mean the use of experimental procedures but it is the taxonomic position of organisms in respect of its population rather than individuals, and the evolutionary process which occur within populations.

Based on the data of cytology, genetics, ecology and morphology the biosystematist developed a classification for experimentally investigated natural taxa and such categories are not intended as substitutes for the units used in classical or practical taxonomy. They are not equivalents, but they may be counterparts of them which do not have nomenclatural status; each category provides a single term for a biosystematic situation. The category terms should be applied either to a plant or population only after they have been experimentally proved to exist. None of the category terms should be applied to a plant or population unless its right to the category has been established and recorded.

3.2.3 Components of Biosystematics

Methods of Biosystematics Study

1. The first step in biosystematics is an investigation or a sampling of the taxon and its populations and the cytological study of the chromosomes of many populations within geographic races, species, genera and so on. Difference in morphology, chromosome number, and behaviour in meiosis usually indicates genetic differences or similarities of taxonomic significance.

2. The determination of the ability of the different populations to hybridize, and to study the different populations which hybridize, and to study the vigour and fertility of the hybrids which may disclose the presence or absence of natural breeding barriers between groups.
3. In meiotic behaviour of hybrids the homologies of chromosomes will be determined and pairing behaviour indicates the degree of genetic relationship in the material. The cytological data is obtained from different geographical populations and races, to establish fertility relations by making some crosses among geographical races and to study meiotic behaviour of homologous chromosomes obtained from hybrids. This will be compared with the data obtained from comparative morphology and geographical distribution. When it is applied, it has an increased objectivity over one obtained through a consideration of morphology and distribution alone. According to Clausen (1945), the data obtained from genetics, cytology, comparative morphology and ecology when applied together to the study of organic evolution make up biosystematics. Biosystematics is concerned with the variation and evolution of species. It is concerned with the process of evolution than it is with classification itself.

Biosystematic Categories

The major objective of biosystematic studies is to arrive at a better understanding of the natural relationships of plants, particularly those of the rank of genus and below. These categories are not intended as substitutes for the units used in classical or practical taxonomy, and they are not necessarily the equivalent of these, although they may be counterparts of them. Each provides a single word term for biosystematic situation, and in no case should the term be applied to a plant or a population unless the situation for which the term stands has been proved experimentally to exist for the particular taxon. The four most widely accepted categories of the biosystematics are, in order of ascending phyletic value: ecotype, ecospecies, coenospecies and comparium.

1. **Ecotype :** The term ecotype was proposed by Turesson (1922) and was well defined by Gregor et al. (1936) as "a population distinguished by morphological and physiological characters, most frequently of a quantitative nature; interfertile with other ecotypes of the escospecies, but prevented from freely exchanging genes by ecological barriers". Ecotype is the basic unit in biosystematics.
2. **Ecospecies :** The ecospecies was defined first by Turesson as “a group of plants comprised of one or more ecotype, within the coenospecies, whose members are able to interchange their genes without detriment to the offspring". Related ecospecies are usually separated by incomplete genetic barriers, which, in addition to ecological barriers, are adequate to preclude free interchange of genes with any other ecospecies.

When ecospecies of one coenospecies are crossed, the resultant hybrids are either partially sterile, or if fertile, they produce many weaklings in the F2 generation (as slow growing dwarfs, individuals highly susceptible to disease against which parents enjoyed immunity, and teratological misfits). Such weaklings are unable to compete, and fail to reproduce. A few such hybrid segregates may possess sufficient vigor to survive. These may be reabsorbed by interbreeding into one or the other parental ecospecies. Related ecospecies generally inhabit different but often contiguous ecological or geographical areas, thus retaining a relative genetic purity. In general, the ecospecies approximates the conventional and conservative taxonomic species.

3. ***Coenospecies :*** The coenospecies is a group of plants representing one or more ecospecies of common evolutionary origin, so far as morphological, cytological and experimental facts indicate. Coenospecies of the same comparium are separated by genetic barriers so nearly absolute that all the hybrids between them are sterile unless amphiploidy (amphidiploidy) occurs. For this reason, distinct coenospecies may exist in a single environment without genetic intermixing. Rather often the coenospecies parallel the taxonomic sections or subsections of the genus.
4. ***Comparium :*** The comparium is the biosystematic unit that often is comparable to the genus. It is composed of one or more coenospecies that are able to intercross. Distinct comparia are unable to intercross and complete genetic incompatibility prevails between them. There are however numerous taxa, accepted by orthodox systematist as genera, that may contain two or more comparia (e.g., in the Fabaceae). In some families the accepted and conventional genera are not equivalents of comparia or even of coenospecies (e.g., some Crassulaceae, Orchidaceae, and Brassicaceae).

Two separate schemes have been devised by Turesson (1922) and Danser (1929). In Turesson's system there are three ranks (Coenospecies, ecospecies, and ecotype). In Danser's system, the definitions of the terms are such that none of the six is synonymous with any other (Comparium, Commiscuum, and Convivium). Danser's lowest term convivium covers the definition of both ecospecies and ecotype while his higher terms Comparium and Commiscuum define the higher and lower aspects of coenospecies. In addition, Turesson employed a lower term still, the ecophene, which denotes the ecological variant, purely the product of environmental modification of the phenotype. F.E. Clement used the term ecad for such variation.

Deme Terminology

Deme Terminology was used with a prefix, denoting any group of individual of a specified taxon. The nature of the association of group of individuals or the kind of population unit which they may represent must always be specified by the addition of an appropriate prefix, second order and compound prefixes may also be used to specify relationships more precisely.

The word, deme used alone simply means a group of individuals of specific taxon and shows no other relationships unless this is specified in the context (Davis and Heywood, 1963, Heslops Harrison, 1967). Various basic deme terms are indicated below: The deme terminology equivalents are also indicated where appropriate in connection with other concepts described in this section. A new system of deme terminology was proposed by Gilmour and Gregor (1939) and developed by Gilmour and Heslop- Harrison (1954).

Deme is denoting any group of specified taxon, orthodox categories can be used as frame work into which other classification for a particular purpose can be fitted to provide an infinite flexible series of categories which could be used to define any group of individuals on the basis of any set of criteria. This deme terminology system in its original concept, non-hierarchial and it falls outside the scope of formal taxonomic categories (genus, species etc). This concept avoids the use of root words such as species and type which are associated with the latter.

The main purpose is the use of natural root, deme which implies nothing in itself except a group of related individuals of a particular taxon. The precise meanings of the terms are provided by various prefixes of which only three were originally proposed.

Topodeme: A deme occurring with in a specified geographical area.

Ecodeme: A deme occurring within a specified kind of habitat.

Gamodeme: Deme composed of individuals which interbreed in nature.

Later Gilmour and Heslop-Harrison (1954) expanded these suggestions and provided following basic demes.

1. Phenodeme: A deme differing from others phenotypically.
2. Plastodeme: A deme differing from others phenotypically but not genotypically denoting reproductive behaviour.
3. Genodeme: A deme differing from others genotypically.
4. Autodeme: A deme composed of predominantly self fertilizing (autogamous) individuals.

5. Gamodeme: A deme composed or individuals which are situated spatially and temporally that wthin the limits of breeding system all can interbteed.
6. Endodeme: A deme composed of predominantly closely inbreeding (endogamous) but dioecious individuals.
7. Agamodeme: A deme composed of predominantly apomictic (non-sexually reproducing) individuals.
8. Clinodeme: A deme which together with other such demes forms gradual variational trend over a given area.
9. Cytodeme: A deme which differs from others cytologically.

3.3 Alpha, Beta and Omega Taxonomy

3.3. 1 Alpha Taxonomy or Classical Taxonomy

Alpha taxonomy is initial phase of classification where the scientists started the categorization of species with the basic ideas or we can say that it is an 'Artificial Classification' where the taxonomist classified an object on the basis of one or two easily recognizable characters (Turrill, 1938). The scientist like Theophrastus (370–285 BC) classified the plant species into tree, shrubs, undershrub and herbs. Similarly other scientist *viz.* Aristotle, Otto Brunfels, J. Bock (Hieronymus Tragus), Charles de Ecluse, John Gerard, Andrea Caesalpino, Jean (Johann) Bauhin, Gaspard (Casper) Bauhin, John Ray and J.P. de Tournefort *etc*. has classified the plants or animals.

A similar Artificial System was based on reproductive characters of the plants, known as the sexual system, developed by father of taxonomy Carolus Linnaeus (1707-1778). Linnaeus is the taxonomist who developed the binominal nomenclature system which is started by the *Caspar Bauhin*. Alpha taxonomy is mainly the history of taxonomy which covered all the initial taxonomical work by different scientist. The main disadvantages of alpha taxonomy are that it relies just on form and shape of organisms and does not consider the other features, which makes it difficult to understand the evolution of species.

3.3.2 Beta Taxonomy or Explorative Taxonomy

Beta taxonomy is the phase of taxonomy developed after the disagreement of the Alpha taxonomy. The phase where the taxonomist, succeed more explorations and categorizations for all the species. Botanist uses most of all the morphological characters which appear during whole lifecycle and applied them to give a systematic position to the taxa. Here we can say that this is a system of classification where scientist classified the taxa based on their natural similarities of vegetative and floral characters. So Beta taxonomy can be considered the phase when the 'Natural system of classification' was

going on. *de Jussieu* (1748-1836) and Augustin Pyramus de Condolle (1778-1841) and the Casimir de Condolle were the botanists who initiated this type of work. George Bentham (1800-1884) and Joseph Dalton Hooker (1817-1911) are known for proposing the Natural System of Classification. The virtue of the Beta taxonomy is that the identification of plant species is easier because the plants morphological characters are used as much as possible and even some embryological, cytological and anatomical characters are also applied. This is the phase of tradition where the exploration and identification (the categorization of species into relevant groups and their arrangement in a hierarchy of higher categories was carried out by botanists which resulted in the development of most of the herbarium and botanical gardens.

3.3.3 Omega Taxonomy or Encyclopaedic Taxonomy

The 'Alpha and Beta Taxonomy' are mainly descriptive and based on morphological features which correspond to the identification of taxa by Turrill (1938). With the advancement of sciences plant systematics is also developed as taxonomist have started to use different discipline of modern plant sciences such as Micro-Morphology, Anatomy, Palynology, Embryology, Cytology, Phytochemistry, Genome Analysis and Nucleic Acid Hybridization as Taxonomic Evidence; Serological, Computer (Internet/online data) and GIS *etc.* So the 'Omega Taxonomy' has widest correspondece of all disciplines of plant sciences which is based on all the informations or data available for the plant species.

The omega taxonomy is the phase where the botanist classified the plant species on the basis of evolutionary and genetic relationship of the plant in addition to morphological characters. The omega taxonomy is the phase where the phylogenetic system of classification was considered, that is, mean most of the botanists identified the species with their origin and the phylogenetical relationship with other species. A.W. Eichler, Adolph Engler and Karl A. Prantle, Charles Bessey, John Hutchinson, Armen Takhtajan, Arthur Cronquist, Rolf F. Dahlgern and Robert F. Thorne are those who developed the phylogenetics classification and apply to relationship among the species for their origin.

Present scenerio is totally changed with the advancement of molecular biology, applied computer sciences and internet data matrix, as now the botanists starts to trace the origin of the species and their phylogenetic relationship with the other species. Kare Bremer initiated the APG system which concludes the taxonomic problems day by day.

3.4 Numerical Taxonomy

Initially the plants were classified on the basis of their morphological characters and taxonomists classified plants in reliance of a particular character *e.g.* Monocotyledones (one cotyledon present) and Dicotyledon (two cotyledons present).

The first time Michel Adanson (1763) pointed out that equal weightage should be given for all characters while classifying any plant species. He initiated this concept first time and these principles are called as '*Adansonian Principle*s'. The Adansonian principles have received great support since 1960 and developed new methods in taxonomy included under a general term Numerical Taxonomy, which is also known as modern classification method.

The mathematical or computerized method supports to analysis of various types of taxonomic data and its helps to find out the similarities or affinities between taxonomic units, and then arrangement of these units into taxa on the basis of their affinities.

According to the Heywood (1967) the Numerical Taxonomy may be defined as "The numerical evalution of the similarity between groups of organism and the ordering of these groups into higher ranking taxa on the basis of these similarities.

Numerical Taxonomy is a developing branch of the taxonomy, which received importance with the development and advancement of the computer. This field of study is also known as mathematical taxonomy (Jardin & Sibson, 1971), Taxometrics (Mayer, 1966), Taximetrics (Rogers, 1963), Multivariant morphometrics (Blackith & Reyment, 1971) and Phenetics and Numerical Taxonomy (Sneath & Sokal, 1973).

Principles of Numerical Taxonomy

The philosophical methods of the modern Numerical Taxonomy are based on ideas that were first proposed by a French naturalist Michel Adanson (1763). Adanson rejected this idea to giving more importance for certain characters and believed that natural taxa are based on the concept of similarity which is measured by taking all the characters into consideration.

The principles of modern Numerical Taxonomy were developed by Sneath & Sokal (1973), these are based on the modern interpretation of the Adansonian principles and as such are termed Neo-Adansonian principles. These Principles are as follow:

1. The greater the content of information in the taxa of a classification and the more characters it is based upon, the better a given classification will be.
2. Every character has equal weightage for creating of new taxa.
3. Over all similarity between any two entities in a function of their individual similarities in each of the many characters in which they are being compared.
4. Distinct taxa can be recognized because correlation of characters can be different in the group of organisms.

5. Phylogenetic suggestion can be made from the taxonomic structure of a group and also from character correlation, this given certain hypothesis about evolutionary pathway and mechanisms.
6. Taxonomy is viewed and practiced as an experimental science
7. Classification is based on phenetic similarity.

There are two aspects of Numerical Taxonomy; the construction of taxonomic group and their discrimination.

Operational Taxonomic Units (OTUs)

Any individual, species, genus, family, order, or class are can be basic unit in Numerical Taxonomy and this known as Operational Taxonomic Units (OTUs). Generally when the OTU is supra individual (above the level of an individual), there should be adequate representation of various polymorphic form i.e. when genera are compared they should be represented by different species, when families are compared, they should be represented by different genera and so on. This is always remember that comparison of OTUs of equal rank is made in Numerical Taxonomy like species with species and genus with genus.

Taxonomic characters

A conventional definition of a taxonomic character is a "characteristic that distinguishes one taxon from another." Thus white flower may distinguish one species from another with red flowers. Hence, the white flower is one character and red flower is another character. A more practical definition given by numerical taxonomists like Micher and Sokal (1957), "as a feature, which varies from one organism to another," and by this second definition, flower color (not white or not red) is a character, and the white flower and red flower are its two character states.

Some authors used the term attribute for characterstate but the two are not always synonymous. When select a character for numerical analysis it is important to selecting a character for numerical analysis as it is important to select a unit character. Unit character is defined by Sokal and Sneath (1963) as a taxonomic character of two or more states, which within the study at hand cannot be subdivided logically, except for subdivision brought about by changes in the method of coding.

Like the trichome type may be glandular or eglandular. A glandular trichome may be sessile or stalked. An eglandular trichome may branch or unbranched. In such a case, a grandular trichome may be recognized as unit character.

The first step in the handling of character is to make a list of unit characters. The list should include all such characters concerning which, information in available.

All the characters should be weighed equally. Some authors advocate that some characters should subsequently be assigned more weightage than others, such consideration generally get nullified when a large number of characters are used. It is a general opinion that numerical studies should involve not less than 60 characters, but more than 80 are desirable. For practical consideration there may be some characters concerning which information is not available (like many plant not bear fruit) or the information is irrelevant (trichome type where as many plants are without trichome), or the characters which show a much greater variation within the some OTUs (flower colour in Lantana). Such characters are omitted from list. This constitutes residual weighing of characters.

Coding of Characters

Many as characters are used for analysis of OTUs. So a huge amount is generated and this makes the use of computer essential. Before the data is fed into the computer, it should be suitably coded. The characters most suitable for computer handling are two state characters (binary or presence/ absence), like habit is woody or herbaceous. However, all characters may not be two states. They may be qualitative multistate (flower white, red, blue) or Quantitative multistate character (leaves two, three, four or five at each node). Such multistate characters can be convered in to two states (flower white or coloured, leaves four or less than four). Some time the character may be splite (flowers white/not white, red/not red, leaves two/not two, three/not three and so on).

The two state or binary characters are best coded as 0 and 1 for the two alternate states. The coded data may be entered in the form of a matrix with t number of rows (OTUs) and n numbers of columns (characters) with dimension of the matrix being t x n.

Residual weighting involves excluding character from the list when enough information of OTUs is not available or is irrelevant. If these characters are used than NC code (Not Comparable) is entered in the matrix. For data handling by computers, the Numerical Taxonomy code (Not comparable) is entered in the matrix. Code is assigned a particular (not 0 or 1) numeric value. Such residual weighting should how ever be avoided but is appreciable for a particular character.

Although other form of coding could also be handled, these are often more difficult. One solution is to give a separate symbol to each character state as explained below:

Flower colour

White A

Red B

Yellow C

Purple D

There may be some symbol occuring in two OTUs otherwise mismatch is recorded.

Character / OTUs	Habit Herbaceous-1 Woody-0	Leaves Simple-1 Compound-0	Habitat Terrestrial-1 Aquatic-0	Ovule Unitegmic-1 Bitegmic-0	Corolla Free-1 United-0
1	1	0	0	1	0
2	0	0	1	0	0
3	1	NC	0	1	0
4	1	1	0	0	1
5	1	0	0	1	1

Measuring Resemblance

Once the data have been coded and entered in the form of a matrix, the next step is to calculate the degree of resemblance between every pair of OTUs. A number of formulae have been proposed by various authors to calculate similarity or dissimilarity (taxonomic distance) between the OTUs. If we calculate the similarity (or dissimilarity) based on binary data coded as 1 or 0, the following combinations are possible.

		OTUs K	
		1	0
OTUs J	1	A (1x1)	Ob (1x0)
	0	C (1x0)	Od (0x0)

Number of matches = M= a+d

Number of mismatches = U= b+c

Sample size n = a+b+c+d= M+U

The data is in resemblance with the using of formula given by different scientist *viz.* Sokal and Michener (1958) and Jaccard (1908).

3.5 Taxonomic Keys

For plant exploration, a large amount of specimens are assembled and this collection is further needed to place a specific location in Herbarium. For this a proper identification of these plant specimens is needed. First of all identification has to be done at family level but thereafter it needs to go to the genera, species, and varietie etc. Initially botanists compared the plant specimens with previously determined material which is harmful for the herbarium sheet, to present this damage some botanists prepared keys and provided with floras and manuals. Some botanists who prepared keys, especially for the identification of plants, are Davis and Cullen (1965), Hutchinson (1967), Saldanha and Rao (1974) and Naik (1977) *etc*.

According to Davis and Heywood (1963), the main object of the key is identification of plant specimen and save the time to separate it from the different group by using one or two easily observable characters.

The keys are categorized in two types Punched card keys and Dichotomous keys. The Punched card keys are generally used for schools and college students which is an interesting learning exercise for taxonomic problems and identification but for the further study purpose, the dichotomous keys are used by most of the botanists.

Punched card keys: These keys are made up by cards of suitable size with the name of all taxa (all families, genera and species) printed on card for the identification of the taxa which is present in keys. Every card is given a number and one character printed near one of the corners. All the taxa showing this character are indicated by a perforation in front of their name and those do not have that character are without any perforation. So there are as many as cards as the characters chosen for the purpose. The plant specimen identified shows that only card which possessed character by the specimen for its recognition. Characters combination present in specimen will allow only one perforation through, in which the set of card selected. The specimen is then identified in belonging family by which the cards indicate the perforation.

Dichotomous key: These types of keys consist of pair of contrasting characters and each statement of which leads to recognition of taxa. The leads are numbered and both begin with the same word as far as possible. The character which used for key preparation should be easily observable and constant. Qualitative characters are preferred over quantitative characters. In closely related taxa with overlapping characters, it is useful to user more then one contrasting character so that the combination of character are

concidered to decide the similarity or difference between the taxa. The following two examples were presented for the dichotomous keys:

1. The bracket or parallel key

In the bracket or parallel key, the two leads of the characters are always next to each other in successive lines on the page. At the end of each line there is a name or a number referring to character later in the key. An example of the bracket or parallel key to eight genera is showen below:

Bracket or parallel Key

1. Aquatic herbs; leaves alternate, corolla lobes Induplicate valvate in bud……. 1. *Limnamthemum*
1. Terrestrial herbs; leaves opposite, corolla lobes Induplicate valvate in bud……. 2.
2. Ovary with 2 celled ……. 2. *Exacum*
2. Ovary with 1 celled……. 3.

3. Corolla lobes eglandular at the base……. 4.

3. Corolla lobes with one or two glandular Nectaries each at the base……..……. 8. *Swertia*

4. The corolla regular …….. 5.

4. the corolla irregular ……. 7.

5. Style linear …….. 6.

5. Style very short or absent ……... 5. *Gentiana*

6. Flowers in sessile axillary clusters …….. 3. *Enicostema*

6. Flowers in dichotomous cymes…..…….. 4. *Erythraea*

7. Flowers yellow; stigma subentire……. 6.*Hopea*

7. Flowers pink or white; stigma deeply 2- lobed……. 7. *Canscoria*

2. The indented or yoked key

The indented or yoked key is widely used for taxonomic literature. In this type of key, the collateral leads of a character are arranged in yokes and each lead is identified by a number of letters. Each of the consecutive characters is indented at a fixed distance from the margin of the page. An example of the indented or yoked key for eight genera of the Gentianaceae from a geographical region is given below:

Indented or Yoked Key

1. Aquatic herbs; leaves alternate; corolla lobes induplicate valvate in buds…….. *1. Limnathemum*
1. Terrestrial herbs; leave opposite; corolla lobes contorted in bud
2. Ovary with 2 celled…….. *2.Exacum*

   Ovary with 1 celled…….
3. Corolla lobes eglandular at the base
4. The corolla regular
5. Style linear
6. Flower in sessile axillary clusters 3. *Enicostema*
6. flowers in dichotomous cymes 4. *Erythraea*
5. Style very short or absent 5. *Gentian*

4. the corolla irregular

7. Flower yellow; stigma subentire….. 6. *Hopea*

7. flowers pink or white; stigma deeply

   2 –lobed……. 7. *Canscora*

   3 – corolla lobes with one or two glandular

   Nectaries each at the base 8. *Swertia*

Advantages and Disadvantages of indented and Bracket Key

There are advantages and disadvantages of each of these two types of dichotomous keys. The advantage of the indented key is that the similar elements are grouped so that they can be seen visually and this makes the identification easier. On the other hand, in the indented key the lines become indented for each character, the lines shortening from the right. This can be uneconomical since more page space is required. In bracket key the group of similar element is not immediately apparent as there is no opportunity to group similar elements having one or more characters in common. However, the bracket key make good use of page space as all leads are approximately of the same length as only alternate couplets indent. Some authors even do not indent alternate couplets and all couplets have a common margin at the left of the page.

Rules for Construction of Key

The following are some rules which are to be considered when preparing a key.

1. The key should be dichotomous.

2. The first word of each lead of the character should be identical (*e.g.* if the first lead of a character begins with the word flowers, the second lead should also begin with the same word).
3. Two consecutive characters should not begin with the same word. If it becomes necessary to repeat the same word it should be preceded by the article "the.
4. Two parts of the character should be made up of contradictory statements, one part of which will apply to the situation and the other will not.
5. Avoid the use of overlapping ranges (e.g. leaves 4-6 cm verses 5-8 cm long).
6. The leads of the character should be read as opposite statements, e.g., leaves opposite verses leaves not opposite.
7. Use readily observable morphological features and avoid distinction on the basis of geographic distribution.
8. It may require two sets of key in some cases. For e.g. in dioecious plants, one key using characters of male plant and the other of the pistillate plant.

3.6 Summary

Biosystematics is a phase of botanical research that approves to the study of living population and delimits the natural biotic units as it classifies the taxa of different orders of degree. The main aim of biosystematics is to demarcate the evolutionary units and formulate new taxa and check the limit of those that already exist.

The Alpha, Beta and Omega taxonomy are the different phases of taxonomy like artificial, natural and phylogenetic classification. The Alpha taxonomy is mainly where taxa are described on the basis of only few morphological characters. Beta Taxonomy is the phase where the taxonomist takes into account all morphological character that appears during whole lifecycle and applied them to give a systematic position to the taxa. Omega Taxonomy has widest correspondece to all branches of modern plant science which is based on all the informations or data available for the plant species.

The Numerical Taxonomy is analysis of various types of taxonomic data by mathematical or computerized method which is to find out the similarities or affinities between taxonomic units, and then arrangement of these units in the ordering of higher ranking taxa on the basis of these similarities.

3.7 Glossary

- **Biosystematics:** Biosystematics is a phase of botanical research that approves to study of living population and delimit the natural biotic units as it classify the taxa of different orders of degree.

- **Alpha Taxonomy:** The Alpha taxonomy is mainly the history of taxonomy which covered all the initial taxonomical work by different scientist. The alpha taxonomy relies just on form and shape of organisms and does not consider the other features, so it is difficult to understand the evolution of species.
- **Beta Taxonomy:** The phase where the taxonomist succeeds for all morphological character which appears during whole lifecycle and applied them to give a systematic position to the taxa.
- **Omega Taxonomy:** Omega Taxonomy has widest correspondece of all discipline of plant science which is based on all the information or data available for the plant species *viz.* Micro-Morphology, Anatomy, Palynology, Embryology, Cytology, Phytochemistry, Genome Analysis and Nucleic Acid Hybridization as Taxonomic Evidence; Serological, Computer (Internet/online data) and GIS *etc.*
- **Numerical Taxonomy:** The Numerical Taxonomy is analysis of various types of taxonomic data by mathematical or computerized method which is to find out the similarities or affinities between taxonomic units, and then arrangement of these units in the ordering of higher ranking taxa on the basis of these similarities.

3.8 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. A deme occurring within a specified geographical area is known as……..
2. Adansonian Principles' for Numerical Taxonomy are given by……..
3. Write the full from of OTUs……..
4. Which Taxonomic Key is important to use for schools and college students?
5. Alpha, Beta or Omega Taxonomy, among these which is follow the modern system?

Section B : (Short Answer Type Questions)

1. Write the difference between punched card keys and dichotomous keys.
2. Write the aims and principles of Biosystematics.
3. Write the short note on the Omega taxonomy.
4. How the multistate charcter can be changed into binary characters.

Section C : (Long Answer Type Questions)

1. Describe the Numerical taxonomy and its importances.
2. Write essay on Biosystematics.

3. Describe the taxonomic keys.

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Unit – 4 (Part-A)

Taxonomic Evidence and Taxonomic Tools - I

Structure of the Unit:

4.0 Objectives

After going through this unit you will be understand about:

- to solve the taxonomic problems by using the modern taxonomic tools : Cytology, Anatomy, Palynology, Embryology, Phytochemistry, Molecular Techniques, Serology etc.

4.1 Introduction

The classification was started with categorization of the species on the basis of their morphological characters. The floral morphological characters are majorly used for classifications, besides floral, other morphological characters also play important role in identification of specific groups of plants. But in recent era, with the development of sciences different branches of botany have developed. These branches are used as modern

tools to give a systematic position and examine phylogenetic relationships. The anatomical work keeping taxonomy in view was mainly carried out by Bailey et al. (1951). Carlquist (1996) has described trends of xylem evolution especially for primitive angiosperms. The study of nodal anatomy has considerable significance in angiosperm systematics. The vascular system of floral parts is arising in the recepticale and can be seen in its cortex. *Paeonia* is removed from family Ranunculaceae and keep into a distinct family Paeoniaceae, this separation has been supported by floral anatomy.

The structure of pollen grains is higly significant in plant taxonomy because every genra has specific characterstics in their pollen wall which help to establish the evolutionary history of angiosperms. The NPC formula of pollen grain study is of great significance in the modern taxonomy. Cytology deals with the study of Cell Morphology and Karyology. Study of the Nucleus is an important tool biosystematics problem and also concludes about evolution.

The chromosomal characters and their behavior during cell division provide relevant information for species identification and their systematic position. The chemical composition of plants play very important role in plant identification. There are various chemical compounds which are used to establish taxonomy such as alkaloids, amino acids, betalins, fatty acids, carotenoides, flavonoids, polysaccharides, tannins, terpenoids and aromatic compounds. The chemical constituent is termed as chemical character. Chemical characters of plants are used mainly in classification or in solving taxonomic problems and are therefore, called Chemotaxonomy, Plant Chemo-systematics and Chemical Plant Taxonomy.

The DNA Hybridisation, DNA Polymorphism and Electrophoresis are important molecular techniques to study at molecular level to sort out the systematic problems and phylogenetic relationship among the different taxa. In *Triticum, Hordeum* and *Secale,* the results of DNA hybridization show that the *Triticum* (75%) is more closely related to *Secale* (100%) than *Hordeum.* The DNA sequence data is applied for analysis of phylogenetic relation and the identification of unique sequences show certain variations in different organisms. The study of antigen and antibody reactions is called serology. The application or utility of serology in solving taxonomic problems is called serotaxonomy. Serology helps to recognize the similarities and dissimilarities amongst different taxa which help to solve and determine the degree of similarity between species, genera, families, etc. For e.g. while studying the serological assessment of relationships within Solanaceae, Hawkes and Tucker (1968) observed a strong serological relationship between *Solanum, Nicotiana*, *Lyopersicum* and *Datura*.

4.2 Morphology

When the classification studies were in initial phases, gross morphological characters used to be the base for categorization of species such as the earlier classifications like Artificial and Natural systems Floral morphological characters are majorly used for classifications and besides there other morphological characters also play important role in identification of specific groups of plants. Some of the important morphological characters which are used to distinguish the taxa are discussed as follows:

Habit

Plant habit is an important character while categorizing taxa. For e.g. Hutchinson (1973) used the woody or herbaceous character to distinguish Lignosae and Herbaceae series in his classification. The unbranched stem of Arecaceae and the liana/climber form in Convolvulaceae are the characteristics of these groups. Some herbaceous plant with specific features *viz.* Amarylidaceae, Iridaceae and Liliaceae can be categorized on the basis of the presence of bulb, the whereas the unbranched stem of Arecaceae and the underground rhizome of Araceae, Musaceae and Zingiberaceae is helpful in distinguishing them from other groups. Presence of rhizome is an important characteristic for categorization of genus *Iris*. Similarly the structure of the bulb in genus *Allium* is an important taxonomic character for identification.

Leaves

Leaf morphology is also an important character to distinguish one taxa from the other, for *e.g.* on the basis of this feature, the Palms, *Salix* and *Populus* species are identified. The genus *Azadirachta* has been split from *Melia* on the basis of the presence of unipinnate leaves whereas in *Melia* bipinnate leaves are present. Similarly, genus *Sorbus* from *Pyrus* and genus *Sorbaria* from *Spiraea* has been separated on the basis of leaf morphology. Interpetiolar stipules are a distinguished and identification character for the family Rubiaceae. The stipules play an important role in identification of *Viola* and *Salix*. Similarly, leaf venation is also a valuable character in identification of *Ulmus* and *Tilia* species.

Flowers

Floral characters are extensively used for segregation of different taxa. On the basis of certain floral characters following taxa can be identified, *e.g.* calyx in Lamiaceae and corolla of Fabaceae and *Corydalis* species, the stamens of different plants families *viz.* Lamiaceae, Fabaceae (Mimosoideae), Malvaceae, Asclepiadaceae and Orchidaceae can be studied, whereas the carpels is an important character for distinguishing Caryophyllaceae. Some of the characteristic features are gynobasic style of Lamiaceae, gynostegium of

Asclepiadaceae, cyathium inflorescence of *Euphorbia* and the capitulum inflorescence in Asteraceae.

Fruits

The characteristics related to fruits in plants have been widely used for identification of taxa *e.g*. Coode (1967) classified genus *Valerianella* on the basis of the fruit. Similarly, Singh *et al.* (1972) classified Indian genera of Asteraceae on the basis of fruit morphology, that is cypsela (achene), presence or absence of pappus, presence or absence of beak, scales or bristles, number of ribs on cypsela. These characters are valuable identifying features. *Melandrium*, *Silene*, *Cerastium* are the members of family Caryophyllaceae which are categorized on the basis of numbers of valves present in the capsule. Genus *Veronica* is identified on the basis of seed characters.

4.3 Anatomy

Anatomical work keeping taxonomy in view was mainly carried out by Bailey (1951) and his Co workers, Metcalfe (1954) etc. Carlquist (1996) has described the trends of xylem evolution especially for primitive angiosperms. With the advancement of sciences, the anatomical characteristics have played an important role in solving the taxonomic problems and understanding phylogenetic relationships between different taxonomic groups.

Wood anatomy

The wood is made up of secondary xylem formed by cambium in a stele. Wood anatomy is significantly used to establish the systematic position of primitive vesselless Angiosperm families like Winteraceae, Trochodendraceae, Tetracentraceae, Amborellaceeae and Chloranthaceae. Vessels are absent in Gymnosperms (except Gnetales) but they occur in some Angiosperms. There are two hypothesis one of which suggests that origin of Angiosperm is from vesselless Angiosperm and other one suggests that the origin is from the advanced Gymnosperms (Gnetales).

Studies on wood anatomy have suggested that Amentiferae represent a relatively advanced group and that Gnetales are not ancestral to angiosperms. Bailey (1944) concluded that vessels in angiosperms evolved from tracheids with scalariform pitting, whereas in Gnetales they developed from tracheids with circular pitting, therefore Bailey (1944) suggested that vessels have independent origin in these two groups. The wood anatomy is the basis for solving of certain taxonomic problems such as separation of *Paeonia* into a distinct family Paeoniaceae and *Austrobaileya* into a separate family Austrobaileyaceae is done according to this feature.

Nodal anatomy

According to Bailey (1965) and Takhatajan (1964), the study of nodal anatomy has considerable significance in angiosperm systematics. The number of vascular traces entering leaf base and leaf gap in the vascular cylinder of node are distinctive for several groups. The unilocular 2 traced node is the most primitive type and all the other type is derived from it. This type of node is quite common in Magnoliales but not seen in Amentiferae.

Floral anatomy

The flower is modified shoot and the bract, bracteoles, sepals, petals, stamens and carpels are modified leaves. The vascular system for supply of all floral parts can be seen in the recepticale cortex. The study of the vascular supply to a flower is defiend as floral anatomy. It is one of the important tools which have led to many significant contributions in understanding the phylogeny of angiosperms.

The vascular supply in the carpels of different genera of the family Ranunculaceae have confirmed the origin of achene (*Ranunculus*, *Thalictrum, etc.*) from follicle (*Delphinium*, *Aquilegia,* etc.) by the successive reduction in the number of ovules ultimately to one. Floral anatomy also supports the separation of *Menyanthes* from Gentianaceae into a distinct family Menyanthaceae. The genus *Centella* is separated from *Hydrocotyle* on the basis of ovules receiving vascular supply from alternate bundles, whereas in *Hydrocotyle* the ovules receive vascular supply from the fusion of two adjacent bundles. *Paeonia* is removed from family Ranunculaceae and kept into a distinct family Paeoniaceae. The separation has been supported by floral anatomy as both sepals and petals have many traces, carpels have five traces and the stamens are centrifugal.

Trichomes

Trichomes are appendages of epidermis which may be non-glandular or glandular. Non glandular trichomes (simple unicellular or multicellular hairs) generally occur in Brassicaceae, Lauraceae and Moraceae. For e.g. the peltate hairs (*Olea*) or flattened scales, the branched dendroid hairs, stellate (*Styrax*) or candelabrum like (*Verbascum*). Glandular trichomes may be sessile or stalked and present a variety of forms. Glandular hairs of *Atriplex* are bladder like and unicellular with few celled stalk and basal cell and they secrete salt, some may secrete nectar (calyx of *Abutilon*). The *Urtica* genus has stinging hairs which are highly specialized with silica tip which readily breaks when hair is touched. The broken tip is sharp like a syringe which injects to skin which irritating cell contents in the skin.

4.4 Embryology

Embryology data plays a significant role in plant taxonomy. Earlier Hafmeister and Strausburger later Maheshwari (1950) and Johari (1963) suggested the possibility of using embryological characters to solve taxonomic problems. Certain embryological characters are very important in solving the taxonomic issue, *viz.* micro and mega sporogenesis, Pollen grain development structure of ovule, Embryo sac development, Endosperm and Embryo development and Seed coat.

Families distinguished on the basis of specific embryological features

Some of the flowering plants families are characterized by unique embryological features. Some of them are described below:

Cyperaceae

Cyperaceae family is distinguished from other flowering plants on the basis of development of single pollen grain rather than four. During the formation of microspores from microspore mother cell via meiosis, there of the microspore get degenerated and a single pollen grain is formed from the remaining microspore.

Loranthaceae

Maheshwari (1964) has split Loranthaceae into two subfamilies Loranthoideae and Viscoideae on the basis of their embryological characters. Loranthoideae has triradiate pollen grains, *Polygonum* type of embryo sac, early embryogeny is biseriate, embryo suspensor is present and viscid layer is outside the vascular supply in fruit whereas, Viscoideae has spherical pollen grains, *Allium* type of embryo sac, early embryogeny many tiered, embryo suspensor absent, and viscid layer inside the vascular supply of fruit. Later on these two subfamilies were raised as distinct families Loranthaceae and Viscaceae.

Onagraceae

Onagraceae family is distinguished from other flowering plants families due to the presence of the *Oenothera* type of embryo sac. This type of embryo sac is absent in other Angiosperms. This is a four nucleate embryo sac which is derived from the micropylar megaspore of the tetrad formed by megaspore mother cell meiosis.

Podostemaceae

Podostemaceae family members are perennial aquatic herbs. These members have a unique embryological character of formation of a pseudoembryo sac due to the disintegration of the nucellar tissue. The family is also characterized by the occurrence of

pollen grains in pairs, bitegmic tenuinucellate ovules, bisporic embryo sac, prominent suspensor haustoria and absence of triple fusion.

Specific examples suggesting the role of embryological data in taxonomy

There are a few examples which show that embryological data has been very useful in the interpretation of taxonomic affinities:

Trapa

The genus *Trapa* was earlier kept in the family Onagraceae but according to modern taxonomic tools, it was subsequently removed from Onagraceae and placed in the family Trapaceae. The following embryological features support this separation:

Table - 4A.1 : Embryological Features of Trapaceae & Onagraceae Family

S. No.	Trapaceae	Onagraceae
1.	Ovary semi-inferior, bilocular with single ovule in each locule	Superior ovary, trilocular, with many ovules in one locule
2.	*Polygonum* type of embryo sac	*Oenothera* type
3.	Endosperm absent	Nuclear or absent
4.	One cotyledon extremely reduced	Both not equal

Paeonia

The genus *Paeonia* was earlier kept in the family Ranunculaceae but on the basis of embryological characters, Worsdell (1908) suggested that it should be placed in a distinct family, Paeoniaceae. The embryological characters which support this view are Centrifugal stamens (centripetal in Ranunculaceae), pollen with reticulately pitted exine with a large generative cell (smooth exine with small generative cell in Ranunculaceae), free nuclear endosperm in which later only the peripheral part becomes cellular and seeds are arillate.

Exocarpos

Exocarpos genus was initially placed in the family Santalaceae. On the basis of its embryological characters (articulate pedicel, 'naked ovule' and presence of a pollen chamber) Gagnepain and Boureau (1947) suggested its removal to a distinct family Exocarpaceae near Taxaceae under Gymnosperms. Ram (1959) studied the embryological

characters of this genus and concluded that the flowers show the usual angiospermic character as the anther has a distinct endothecium and glandular tapetum, pollen grains get shed at 2-celled stage, embryo sac is of the *Polygonum* type, endosperm is cellular, and the division of zygote is transverse. These characters confirm that *Exocarpos* is indisputably an angiosperm, and a member of the family Santalaceae.

4.5 Palynology

The study of the sturacture of pollen grain (mainly the pollen wall) is very significant in plant taxonomy because every genera has a specific characterstic pollen wall. This has helped to establish the evolutionary history of angiosperms. The taxa shows single morphological pollen type is considered stenopalynous, whereas taxa which have different type of pollen grain are considered as eurypalynous. The stenopalynous groups are of considerable significance in plant taxonomy.

The following characteristics of pollen grains are considered by taxonomists to segregate different taxa:

The number of nuclei present at the time of dispersal: The primitive angiosperms are shed at 2-nucleate stage, whereas in advanced groups, pollens are shed at 3 nucleate stages. In most angiosperms pollen grains are radial symmetry. Bilateral symmetry only occurs in some primitive groups like gymnosperms. In most angiosperm, the pollen grains are globose in shape but in some other members they are boatshaped, ellipsoidal and fusiform in shape.

Initially pollen grains form tetrads and the outer end of grain is termed distal pole, whereas the inner end where grains joint as proximal pole. The line running around the pollen at right angles to the polar axis is termed as equator. The pollen grains of angiosperms are separate prior to release, these single pollen grains are called as monads, in rare cases pollen grains are dispersal in fused pairs or four they are known as dyads and tetrad respectivly.

Following five different types of tetrads are differentiated:

Tetrahedral tetrad: Four pollen grains form a tetrahedron are arranged in a sphere e.g. family Ericaceae.

Linear tetrad: Four pollen grains arranged in a straight line e.g. *Typha*.

Rhomboidal tetrad: 4 pollen grains are in a single plane.

Tetragonal tetrad: Four grains are in one plane and equally spaced as in *Philydrum*.

Decussate tetrad: Four grains in two pairs, arranged at right angles to one another, as in genus *Lachnanthes*.

In *Calliandra* of Mimosoideae, the pollen grains are attached in a group of more than four. These pollen grains represent a polyad which is made up of 8-10 pollen grains. Whereas in some members of family Orchidaceae, e.g. genus *Piperia,* pollen grain are connected in a irregular group known as massulae. In subfamily Asclepiadaceae and several members of orchidaceae, all pollen grains of a theca are fused into a single mass which is called as pollinium.

Pollen wall

The pollen grain wall is made up of two layers, outer exine and inner intine. The exine is hard due to sporopollenin which is resistant to decay. Pollen wall sculpturing is structure present on the outer surface of exine wall. The common types of sculpturing include: baculate, echinate, spinulose, foveolate, reticulate, fossulate, verrucate, gemmate, psilate, and striate.

Pollen aperture is a region where the pollen tube comes out, the exine may be inaperturate (without an aperture) or aperturate. Aperturate may be single pore (monoporate), a single slit running at right angles to the equator (monocolpate), three slits (tricolpate), three pores (triporate), three slits each with a geminate pore in middle (tricolporate) and with many pores (multiporate). Pollen with one or more slits located at the polar end is accordingly termed, **monosulcate**, **disulcate** and **trisulcate**, depending on the number of slits. Pollen grain with slits joined at poles is termed **syncolpate**.

The role of pollen grains in systematics in *Nelumbo* is a good example which is separated from Nymphaeaceae and kept into a distinct family Nelumbonaceae and this is largely supported by the tricolpate pollen of *Nelumbo* as against the monosulcate condition in Nymphaeaceae.

4.6 Cytology

Cytology deals with study of Cell Morphology and Karyology. The Nucleus study is an important tool to solve biosystematics problems and also conclude about evolution. Chromosomal characters and their behavior during Cell division provide relevant information for species identification and their systematics. The chromosomes vary from species to species and this is due to the result of different evolutionary history, These variation are very small in related species but they may be quite larger in various related groups. The cytological differences provide important evidence for species evolution.

The characteristics of chromosomes have greatest significance in biosystematics and in evolutionary studies. Solbrig (1968) pointed out that most of the cytotaxonomic studies have largely made use of following aspects:

1. Chromosome number
2. Morphology of Chromosomes
3. Behaviuor of Chromosomes during the Cell division

Chromosomal number

Extensive work on chromosome numbers in plant species were carried out by Darlington and Janaki-Amal (1945), Darlington and Wylie (1955), Federov (1969) and Löve *et al.* (1977). *Regnum vegetabile,* a series for the **Index to Plant Chromosome Numbers** published by The International Association of Plant Taxonomy (IAPT, 1967 and 1977), is published in 9 volumes which is mostly forming annual lists of chromosome numbers. The Missouri Botanical Garden maintains the update of records on chromosome numbers and this can be inquired for online information about recorded plant species.

The chromosome numbers are recorded as diploid number (2*n*) in somatic cell and in a germ cell (gamete) numbers are recorded as haploid (*n*). The gametophytic chromosome number in diploid species is designated as basenumber (*x*). In a diploid species represented $n = x$, whereas in a polyploid species n is always multiples of x. In a hexaploid plant species with $2n = 42$ will thus have $n = 21$, $n = 3x$ and $2n = 6x$. The angiosperm shows a great variation in their chromosome number. The highest chromosome numbers are recorded in Poaceae member *Poa littoroa* ($n = 132$) and the lowest number in Asteraceae member *Haplopappus gracilis* ($n = 2$). The highest chromosome number in a plant species is occurs in Pteridophytes member *Ophioglossum reticulatum* ($n = 630$).The duplication of chromosome numbers (polyploidy) also have taxonomic significance *e.g.* the grass genus *Vulpia* species contains hexaploid ($2n = 42$) and tetraploid ($2n = 28$) where as diploid ($2n = 14$).

On the basis of chromosome numbers many scientists worked to solve some taxonomic problems for many taxa *e.g.* Raven (1975) worked on chromosome numbers at the family level in angiosperms and concluded that the original base-number for angiosperms is 7*x*.

The 5*x* in *Paeonia* with large chromosomes has been separated to others into the family Paeoniaceae, this placement is supported by other morphological, anatomical and embryological characters. Similarly in Poaceae subfamily Bambusoideae has 12*x*, whereas Pooideae has 7*x*. In *Mentha* genus the chromosome numbers provide strong support to subdivision *viz. Audibertia* (9*x*), *Pulegium* (10*x*), *Preslia* (18*x*) and *Mentha* (12*x*).

Morphology of Chromosomes

Generally the chromosomes morphology is considerd on the basis of chromosome size, position of centromere and presence of secondary constriction position. The centromere position causes the different size of both arms some time they may be equal or unequal, as

metacentric (centromere in middle), **submetacentric** (away from middle), **acrocentric** (near the end) or **telocentric** (at the end). The whole chromosomal structure in a species is called karyotype and their diagrammatically represented is term as idiogram*l* karyogram. The idiogram*l* karyogram are specific for the each species *e.g. Lyris* (n=12: 6- metacentric and 6- submetacentric); *Pseudosago* (n=22: 1-telocentric, 6- submetacentric, 5- metacentric) and *Pseudolyris* (n=12: 2- metacentric and 22- telocentric). Family Agavaceae is an interesting example of utilization of chromosomal morphology as the *Agave* members keep in Amaryllidaceae for their inferior ovary and *Yucca* keep in Liliaceae due to superior ovary). The genera were shifted into Agavaceae on the basis of their similarity. The bimodal karyotype (mean 5 large and 25 small chromosomes) is also support to keep them in Agavaceae.

Behavior of Chromosomes during the Cell division

The chromosomes behavior during pairing (synapsis) and subsequent separation is useful sometimes in studying the interrelation of taxa. Meiosis also sometime provides taxonomic information *e.g.* Juncaceae and Cyperaceae families have small Chromosomes with unlocalized centromere.

The genus *Paeonia* was traditionally put in the family Ranunculaceae. On the basis of cytological data of the *Paeonia* is different from the Ranunculaceae members, as the basic chromosome numbers 5*x* which is unlike to other Ranunculaceae members like they possess 6*x*, 7*x* and 8*x*.

Similarly the *Yucca* and *Agave* shows same type karyotypes which are asymmetrical, comprising 10 large and 30 small chromosomes. On the basis of morphological characters similar habit and secondary growth both genus keep in a same family Agavaceae as both are separately put in distinct families.

4.7 Phytochemistry

The chemical component of plants play very important role to plant identification. There are various chemical compounds used in the taxonomy. Alkaloids, amino acids, betalins fatty acids, carotenoides, flavonoids, polysaccharide, tannins, terpenoids and aromatic compounds are the important examples. The chemical constituent is termed as chemical character. Chemical characters of plants are used mainly in classification or in solving taxonomic problems are called Chemotaxonomy, Plant Chemo-systematics and Chemical Plant Taxonomy.

In this phase classification of plants are based on their chemical ingredient i.e., on their molecular characteristics. The method of chemical taxonomy is thus exact and simple in

principle consisting of investigations of the distribution of chemical compounds or groups of biosynthetically related compounds in series of related or supposed related plants.

Greshoff (1909) suggested the use of comparative chemistry in taxonomy. According to McNair (1935) plants can be classified chemically in accordance with the substances made by them. Such chemical classification may be compared with or used as supplement to morphological classification and may be of some importance in the development of true natural system of angiosperm phylogeny.

Mentzer (1966) categorized three types of chemical constituents for taxonomy:

(1) Primary constituents: like proteins, nucleic acid derivative, chlorophylls and polysaccharides.

(2) Secondary constituents: who lack nitrogen and are not involved in the basic metabolism of cells.

(3) Miscellaneous substances.

Turner (1969) categorized these compounds according to their molecular size into following type:

(1) Micro molecules: Compounds having low molecular weight eg alkaloids, amino acids, cyanogenic glucosides, glucosinolates (mustard oil glucosides), pigments (anthocyanins, betalains, and so on), phenolics (flavonoids), and terpenoids.

(2) Macromolecules: Compounds having high molecular weight (over 1,000) eg Proteins, DNA, RNA, cytochrome-c, ferredoxin complex polysaccharides etc.

Naik (1984) classified these compounds as follow and chemical characters can be used in taxonomy as

(1) Directly visible like starch grains, raphides etc.

(2) Secondary plant products like alkaloids, flavonoids and terpenoids

(3) Proteins.

Following some example are given here on the basis of various Chemical character there data have practical solved same taxonomic problems at the order lavel to generic level. These data have been treated as characters used in classification.

Tannin is present in Sapindaceae, alkaloids in Solanaceae (e.g. *Nicotiana, Datura*), aromatic compounds in Lamiaceae (Cronquist, 1981). Protopine is present in Papaveraceae. This alkaloid protopine is not found in any plant of other families (Manske 1954).

According to Hutchinson Fumarioideae is quite distinctly separated as a group from Papaveraceae proper and closely allied to certain genera of the family Berberidaceae e.g., *Epimedium, Aceranthus* and *Bongardia*.

Tannins: Tannins bearing or tanniniferous families are Anacardiaceae, Polygonaceae, Punicaceae, Rhizophoraceae, Tiliaceae, Ulmaceae, Urticaceae, Vitaceae, Winteraceae, Casuarinaceae, Lauraceae, Lythraceae, Sapindaceae, Sapotaceae, Sterculiaceae, Magnoliaceae, Meliaceae, Moraceae, Myrtaceae, Oxalidaceae, etc.

Some example of families which are without tannins : Acanthaceae, Amaranthaceae, Convolvulaceae, Portulacaceae, Solarraceae, Cucurbitaceae,Basellaceae, Campanulaceae, Capparidaceae, Caryophyllaceae, Chenopodiaceae, Lamiacae, Papaveraceae, Brassicaceae, Verbenaceae, Violaceae, Zygophyllaceae etc.

Steroids: Steroids are high molecular weight solid alcohols. Cardenolides are cardiotonic glycosides and these are present in the following taxa *Euonymus* (Celastraceae); *Mallotus* (Euphorbiaceae); *Digitalis* (Scrophulariaceae); *Isoplexis* (Ranunculaceae); Apocynaceae and Asclepiadaceae.

Terpenoids: Terpenoid compounds are found in Citrus plants, Mints and Umbellifers etc. Many tribes of the Asteraceae (Compositae) are characterized by the sesquiterpene lactones they produce. Sesquiterpene lactones have been used to show that in the tribe Vernonieae. *Ambrosia, Iva, Franseria* and *Xanthium* are removed from the tribe Heliantheae and placed in a separate tribe.

Flavonoids: Various types of flavonoids include flavones, flavoiumes, isoflavones and isoflavonoids, flavonols, anthocynidins, chalcones, aurones and biflavonyls. There are some examples showing the significance:

1. Thorne considers that Fabaceae is allied to Rutales by the presence of phenylated flavinoids in both Fabaceae and Rutaceae.
2. Presence of flavinoids indicates that the Arecaceae is closely related to the Poaceae, due to the presence of tricin and lutealin (leaf flavonoids) in both the families.
3. Sterculiaceae is closely related to Malvaceae by the presence of Cyanidin and gossypetin in both the families.

Sulphur Compounds: The Sulphur compounds having significance in taxonomy. Disulphides are largely responsible for the odour of plants e.g. Onions and Garlics etc. Glucosinolates (mustard oil glucosides) occur in Brassicaceae, Capparidiaceae Tovariaceae, and Moringaceae etc. cleare odour of sulphur can observ in these plants. The Cappariaceae are placed in order capparales on the bases of chemical characters which

were previously placed in one order Rhoeadales alongwith Fumariaceae and Papaveraceae.

Alkaloids: Alkaloid protopine is present in family Papaveraceae. Protopine is also present in family Fumariaceae, showing their close relationship with Papaveraceae. The lupin alkaloid is found in Fabaceae and tropane derivatives occur in the Solanaceae. Morphine is restricted to the *Papaver somniferum*, Coniine to a few members of Apiaceae similarly Strichnine to the some members of Strychnos.

Non-Protein Amino acids: Presence of Cyclo Propyl Amino Acids in Sapindaceae and Aceraceae show their close relationship. Canavanine shows a close analogue of arginine is found only in the Fabaceae. The seven intrageneric groups in *Lythyrus* and fourteen intrageneric groups in *Vicia* are recognised on the basis of distribution of amino acids.

4.8 Genome Analysis and Nucleic Acid Hybridization (Biochemical and Molecular Technique)

1. Electrophoresis

The Electrophoresis is important molecular technique to study mainly the proteins and to separate and identify proteins. The amphoteric properties are based to seprate the proteins due to the positively or negatively charged in various extents according to the pH of the medium protein travel in the gel at various speeds across a voltage gradient this reaction is carried out in a polyacrylamide gel (polyacrylamide gel electrophoresis—**PAGE**). The protein bands which covered same distances are mostly similar. Johnson (1972) studied on hexaploid wheat (*Triticum aestivum*) and supported on the bases of electrophoresis that the origin of *Triticum aestivum* from *Aegilops tauschii* and *Triticum dicoccum*. Crawford and Julian (1976), studied to assess species relationships in *Chenopodium* by combining data from flavonoids with proteins. *Chenopodium atrovirens* and *C. leptophyllum* had identical flavonoid patterns but could be distinguished by their different seed protein spectra. *C. desiccatum* and *C. atrovirens,* on the other hand, were closely similar in seed proteins but differed in flavonoids.

2. DNA and RNA Hybridisation

DNA is extracted from the organism to convert to a single strand polynucleotide chain and the amount of ressocation (annealing) with similarly treated DNA from another taxon which occur on mixing the two is taken as a measure of similarity of the nucleotide sequences. This utility and importance of DNA values in taxonomy may perhaps best be demonstrated in *Scikka bifolia* alliance a group of plants which a costant basis number x = 9 and little evidence for major structure rearrangement. The *Triticum, Hordeum* and

Secale are the results of DNA hybridization in three genus shows the *Triticum* (75%) is more closely related to *Secale* (100%) then in *Hordeum.*

3. DNA Polymorphism

Utilization of DNA sequence data for phylogenetic relation analysis involves the identification of unique sequences which show certain variation in different organisms, these sequences which can be use as genetic markers and identification of the target taxa and ultimate construction of phylogenetic trees. This procedure is also known as DNA Fingerprinting or DNA polymorphism. This method is now widely used in forensic investigations. A variety of techniques have been developed to detect this polymorphism and each method has its own advantages and limitations, and suitable for a particular situation. New methods are being continuously developed. Some of the commonly used procedures are Single Nucleotide Polymorphisms (SNPs) and Restriction Fragment Length Polymorphisms (RFLPs) *etc.*

4.9 Serology

The application or utility of serology in solving taxonomic problems called serotaxonomy. Any substance which is capable of stimulating the formation of an antibody is called antigen and the study of antigen and antibody reactions is called serology. Nuttal (1901) was the scientist who compares the immunochemical specificity of serum proteins for systematic point whereas Kowarski (1901), Bertarelli (1902) and Magnus (1908) are other notable serologists. Dunbar (1910) point out that proteins from pollen, seeds and leaves of rice were serologically distinct. A school of serology was founded in 1914 by Gohlke at Koenigsberg in Germany this became the centre of serological studies. Serology According to Boyden (1964) it is the branch of biology which `deals with "the nature and interactions of antigenic material and antibodies". According to Smith (1976) serology is the "study of origin and properties of antisera." Agglutinogens antigens are also called agglutinogens.

Serology help to recognize the similarities and dissimilarities amongst different taxa which is help to solve in the taxonomy problem and also determines the degree of similarity between species, genera, families, etc. It helps in comparing non-morphological characteristics this information is useful in taxonomy. Any single proteins from different plant species are also compared by serology techniques. Antigen & antibody reaction that results in visible clumps of organisms or other material is known as Agglutination

General steps for serological reaction:

1. The reaction is specific, an antigen combining only with its homologous antibody and vice versa.

2. Entire molecules react and not the fragments.
3. Antigens or antibodies do not pass through denaturation during these reactions.
4. Combination occurs at the surface, and it is firm but reversible.
5. Both antigens and antibodies take part in the formation of precipitates or agglutinates.
6. Antigens and antibodies can combine in varying proportions.

Serological results basis Mez and Ziegenspeck (1926) prepared a family tree for the entire plant kingdom. Here following some examples which conclude on the basis of serology and these examples are implication for classification of angiosperms:

1. According to Fairbrothers (1983) serological data have been used in the classification of orders and the assignment of families in Apiales, Caryophyllales, Capparales, Fagales, Cornales, Magnoliales, Juglandales, Papaverales, Rubiales, Ranunculales, Scrophulariales, Typhales, Primulales, etc.
2. Fairbrothers and Johnson (1959) separated six species of Bromus on the basis of serological studies.
3. According to Jensen (1967), serological characteristics within Ranunculaceae show a close similarity between Aconitum-Delphinium, Actaea-Cimicifuga, Anemone-Clematis and Ranunculus-Myosuru, and suggest a common ancestry for Aquilegia, Leptopyrum and Thalicttutn.
4. While studying the serological assessment of relationships within Solanaceae, Hawkes and Tucker (1968) observed a strong serological relationship between Solanum, Nicotkina, Lyoscyamus, Datum and Salpiglossis.
5. Simon (1971) showed a close relationship between Nymphaeaceae and Nelumbonaceae on the serological ground.
6. Serotaxonomic findings of Pickering and Fairbrothers (1970) in Umbelliferae support the classification of the family into Apioideae, Sanicul-oideae and Hydrocotyloideae, and also suggest that Apioideae is more close to Saniculoideae than to Hydrocotyloideae.
7. Gartner (1978) suggested some serological evidences for assigning Phaseolus aureus and P. mungo to the genus Vigna.
8. Hydrastis of Berberidaceae has more serological similarities with Ranunculaceae than Berberidaceae.

4.10 Summary

The classification on the basis of morphological characters was a traditional system, now a days botanist follows the different branches of the botany to identified the correct systematic position of the any taxon and also conclude the phylogenetic origin of the taxon. No doubt about the morphology is the basic need to aproch directly to identified any taxa but in recent era the following different modern tools also have some valuable information for identified the phylogenetic relationship. Like the Anatomical work for taxonomic view is the trends of xylem evolution especially for primitive angiosperms, also the nodal anatomy and floral anatomy. The polynology is also now higly significant in the plant taxonomy because each of genera has some specific characterstic in their pollen wall, and also helped to establish the evolutionary history of angiosperms. Similarly the Cytology is the study of Cell Morphology and Karyology. The Chromosomal characters and their behavior during Cell division provide relevant information for species identification and their systematic. On the other hand chemical characters of plants are used mainly in classification or in solving taxonomic problems are called Chemotaxonomy. The chemical component of plants play very important role to plant identification. The advances techniques like DNA Hybridisation, DNA Polymorphism and Electrophoresis are important molecular techniques to study at molecular level to sort out the systematic problems and phylogenetic relationship among the different taxa. Similarly the serological study help to recognize the similarities and dissimilarities amongst different taxa which help to solve in the taxonomy problem and also determines the degree of similarity between species, genera, families, etc.

4.11 Glossary

- **Karytypes:** The whole chromosomal structure in a species is called karyotype and their diagrammatically represented is term as idiogram*/* karyogram.
- **Chemotaxonomy:** Chemical characters of plants are used mainly in classification or in solving taxonomic problems called as Chemotaxonomy.
- **Pollinium:** In Asclepiadiaceae and some members of orchidaceae, the pollen grains of a theca are fused into a single mass which is called as pollinium.
- **Serological:** The application or utility of serology in solving taxonomic problems called serotaxonomy.
- **Serology:** Any substance which is capable of stimulating the formation of an antibody is called antigen and the study of antigen and antibody reactions is called serology.

4.12 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. Match the following:

Gynobasic style	Euphorbiaceae
Gynostegium	Asteraceae
Cyathium inflorescence	Asclepiadaceae
Capitulum inflorescence	Lamiaceae

2. According to modern Taxomomic tools Trapa is keeping in which family?
3. Pollinium is occur in which family?
4. Glucosinolates (Glucosides oil) occur in which family?

Section B : (Short Answer Type Questions)

1. Write the systematic position of the Peonea on the basis of embryological characters.
2. Write the short note on systematic position of Centrospermales on the basis of Chemotaxonomy.
3. Write the role of Karyotype in taxonomy.
4. Write a short note on DNA hybridization and its role in plant taxonomy.

Section C : (Long Answer Type Questions)

1. Describe the role of Chemotaxonomy in plant systematic.
2. Write a detail note to describe the embryology as a taxonomic tool to solve the plant taxonomic problems.
3. Write a detail note on molecular techniques as a modern tool in plant taxonomy.
4. Describe the Serology and its methods to study for solving taxonomic problems.

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Unit – 4 (Part-B)

Taxonomic Evidence and Taxonomic Tools - II

Structure of the Unit:

4.0 Objectives

After going through this unit you will be able to understand about-

- the concept and importance of Herbarium,
- list out important herbaria in the World and India,
- the methodology of plant collection and preservation of specimens and
- the arrangement and Maintenance of Herbarium.
- the Flora

4.1 Introduction

Man has been dependent on the plants for all his basic needs like food, clothing, shelter and medicine since human civilization. The knowledge and life long experience about the utilization of plants need to be transferred generation to generation, lead to develop methods of preservation of plants and plant materials. There must have been requirement of long term preservation of plant collections. The word 'Herbarium' was originally coined to a book about dried medicinal plants. Herba in Latin referred to slender weak-stemmed plants. A collection of herbs came to known as herbarium. Tournefort (c.1700) used the term 'Herbarium' for collection of dried plants, the method which earlier known as Hortus siccus. Luca Ghini (1490-1556), Professor of Botany at the University Bologna, Italy is thought to have been the first person to dry plants under pressure and mount them on paper and recommended this method of preservation. The oldest preserved herbarium specimen is of Gherardo Cibo, a student of Luca Ghini. By the time of Linnaeus, the technique was widely known in Europe. Luca Ghini paved the way for the establishment of the first herbarium of the world in Italy who also responsible for the first modern botanic garden (Orto botanico di Pisa) in the world at Pisa. It is interesting to note that many of the early herbaria made their beginning with botanical gardens. In the beginning, the plant specimens were mounted on sheets of paper which bound into book-like volumes. Linnaeus started arranging herbarium sheets separately according to groups. Earlier, the size of sheets was according to the size of the plants. The present concept of herbarium collections is a net result of the efforts of the botanists all over the world. There is huge change in collection, pressing, drying, and preserving the plant specimens in the recent years. Not only plant specimens but seeds, wood sections, pollen, microscope slides, fluid preserved flowers or fruits also can be preserved in herbaria. Herbaria cover

all plant categories. There are herbaria especially established for algae, fungi, bryophytes and pteridophytes. According to Jain and Rao (1977), herbarium is a storehouse of plant specimens collected from far and wide, mounted on appropriate sheets, arranged according to some known system of classification and kept in pigeon holes of steel or wooden cup boards, usually specially prepared for the purpose. Lawrence (1951) defined herbarium as a collection of plant specimens that usually have been dried and pressed, arranged in sequence of an accepted system of classification and are available for scientific study. A modern herbarium may be defined as a great filing system for information about plants primarily in the form of actual specimens and secondarily in the form of recorded notes, published information and pictures.

4.2 Functions of a Herbarium

Herbarium is a conservatory of material and data. It is a store house of vast collections of plants of different localities. They are quite helpful in determining plant locations and ranges, abundance, habitat, flowering and fruiting periods. The simple form of indexing of plant specimens in the herbaria helps them to be retrieved quickly.

The Herbariums are remarkable and irreplaceable sources of information about plants. They provide the comparative material that is essential for studies in taxonomy, systematics, conservation biology, anatomy, morphology, ecology, biodiversity, ethnobotany, and paleobiology, as well as being used for teaching, education and training. Functions of herbaria can be listed as:

1. Identification of plants: Herbarium serves as a fundamental source of identification of plants. Those engaged in taxonomic studies, can personally identify their engaged collection by comparison with already identified herbarium specimens.
2. Education: Herbarium helps in teaching for students, they can learn about different plants without going different regions. Its save both time and money.
3. Research: The specimens in the herbarium are identified correctly and give visual information about species. It is the basic tool for taxonomic studies. It serves as a source material for research in anatomy, morphology, agronomy, forestry, anthropology, entomology, ecology and for other environmental studies. Herbarium is a wonderful resource for identification of crude drugs in pharmaceutical research.
4. Biodiversity Database: Herbarium serves as a source for biodiversity information which serves in biodiversity estimation in different parts of the world.
5. Population study: Herbarium helps to study the population biology of the species. Herbarium collections in large numbers of a particular species representing different

areas help in assessing the population variations exhibited by a species in its distributional range and hence help in population studies.

6. Other functions:

- Most herbaria have specimens collected from different parts of the world and, thus their scrutiny can provide information on the geographical distribution of taxa. These broad geographical ranges of collections in herbarium are helpful in identifying the centers of endemism, centre of origin and migration of species.
- Voucher specimens preserved in various herbaria, provide an index of specimens on which studies on chromosomes, phytochemistry, ultrastructure micromorphology, etc. have been undertaken
- Herbarium serves as a repository of historic collections. In some national and international herbaria like that of KEW, London has vast collections way back to 17th century.
- Herbarium serves as a source for search of new genetic material. Herbarium material is sometimes used for DNA extraction after proper treatment.
- Herbarium facilitates and promotes the exchange of new material among institutions (taxonomy).

Herbarium helps in developing computer databases on plants. Recently electronic herbaria in form of digitized information are getting ready for which herbarium material is must.

4.3 Types of Herbarium

Herbaria can be classified broadly into two types: general and special

4.3.1 General Herbaria

On their status of collections and mode of operation General Herbaria may be further classified at different scales: international, national and local based.

a. **International Herbaria:** These are very large herbaria normally with more than 4 million specimens and have global representation of taxa, i.e., specimens collected from different countries. With the rich in type specimens. They help in writing floral monographs, in identifying rare specimens especially new taxa and provide facilities for visiting botanists. They provide loan specimens to other reputed herbaria on request. e.g. Royal Botanic Garden Herbarium, Kew.

b. **National Herbaria:** Generally these herbaria consist mainly of the plants of the country concerned and may also cover the plant material of neighboring countries. As

far as possible the representatives of all taxa in the country kept in these type Herbaria. Most of national herbaria are old and contain good number of collections. National herbaria have a significant role in, writing national and regional floras. They also provide facilities to visiting botanists. e.g. Central National Herbarium, Howrah.

c. **Local Herbaria:** These Herbaria cover a region in a country such as State, District or even a small area like a nature reserve. They relatively have short history and contain few type specimens. Local herbaria are useful in production of local floras. They contribute to national flora and of much help in catering the needs of local educational and research institutions.

4.3.2 Special Herbaria

These are generally small herbaria established for a specific purpose and have limited scope. They are of several types.

a. **Historic herbaria:** They are maintained separately within general herbaria and are very rich in type specimens e.g., Wallich herbarium at Kew.

b. **Herbaria of limited scope:** These herbaria house taxa of a particular group of plants. They may be large and usually associate with large herbaria or institutions like universities. e.g. Algal herbarium at National Institute of Oceanography , Goa; Forest Herbarium at FRI, Dehra Dun.

c. **Herbaria for teaching purpose:** These herbaria are seen with educational institutes like Universities or colleges. Sometimes these herbaria are large and should be considered as National or local Herbaria. They cater the needs of teaching and also research. These Herbaria usually contain the plants representing the surrounding areas and economically important species such as crop plants and ornamentals. e.g. Rajasthan University herbarium .

With the advent of information technology, new techniques have been adopted for the fast retrieval of Herbarium specimen information. This includes the microherbaria in the form of photographs and the electronic herbaria in the form of digitized databases.

4.4 Important Herbarium of the World and India

Herbaria are usually associated with universities, museums and botanical gardens. As stated earlier the first herbarium in the world in 1570 in Bologna, Italy by Luca Ghini. Presently around 4000 major herbaria are recognized in over 165 countries. Index Herbariorum is a world catalogue of public herbaria is published periodically by International Association for Plant Taxonomy (IAPT) and New York Botanical Garden. The recent edition is edited by Patricia Holmgren in 1990. Index Herbariorum lists the

world's herbaria. Each herbarium in Index is assigned an official acronym (code) that is used as a standard reference in the world. The Index Herbariorum (IH) entry for a Herbarium includes its physical location, URL, contents (e.g., number and type of specimens), founding date, as well as names, contact information and areas of expertise of associated staff. Only those collections that are permanent scientific repositories are included in IH. New registrants must demonstrate that their collection is accessible to scientists, and is actively managed. Each institution is assigned a permanent unique identifier in the form of a one to eight letter code, a practice that dates from the founding of IH in 1935. With an estimated 300 million specimens are housed in herbaria of different countries in the world out of which over 50% of the specimens are preserved in Herbaria of Europe.

4.4.1 Important Herbaria of the World

The Museum of Natural History is world's largest herbarium in Paris having of about 9 million specimens. Followed by the Royal Botanic Garden Herbarium at Kew, London which harbours over 7 million specimens including 3, 50,000 type specimens.

Table- 4.B.1 : The Major Herbaria of the World

S.N.	Name of the Herbarium	Year of Establishment	Acronym	No. of specimens
1	Museum National d' Histoirie Naturelle, Paris (Natural History Museum)	1635	P	8,900,000
2	Royal Botanic Gardens, Kew	1853	K	7,000,000
3	New York Botanical Garden, New York	1891	NY	6,500,000
4	Komorov Botanical Institute, Leningrad	1823	LE	5,700,000
5	Missouri Botanical Garden Herbarium,	1859	MO	5,200,000

Royal Botanic Gardens (Kew, England)

Royal Botanic Gardens (Kew): The Herbarium has a central role for research on plant biodiversity. The collection at Kew is still growing with a yearly addition of around 30,000 new specimens through a programme of joint work with overseas colleagues, expeditions, gifts and exchanges with other institutes at home and abroad. The Herbarium is also the repository of many voucher specimens. Such specimens are the only tangible

record by which species used in experimental research can be compared. The Herbarium contains over 350,000 type specimens the original specimens on which new species descriptions have been based. These specimens, some dating back to the eighteenth century, typify and fix a species name for all time, and are invaluable to researchers. Type specimens are vouchers for plant names and, as such, are the essential reference point for a name that botanists consult in seeking to apply names correctly.

Komorov Botanical Institute, Leningard

Komorov Botanical Institute, Leningard Herbarium has 6 million specimens from all over the world Latin America. This Herbarium has approximately 200 000-250 000 specimens from Latin America in its holdings. The most important and famous collections from Latin America are those of G. Langsdorff (1814-1817 & 1821-1829) and L. Riedel (1821-1828 & 1831-1836) from Brazil, G. Mertens (1826) and W. Karwinsky (1841-1843) from Mexico, and plants collected in expeditions of the Soviet Rubber Trust in 1926-1928. These collections are rich in type and other original specimens. The Herbarium has separate collections from, West Europe, Asia, Africa, North and South America, Australia (General Sector), Eastern Europe (Russian Sector), Middle Asia and Kazakhstan (Middle Asian Sector), Siberia and the Russian Far East (Siberian Sector), Caucasus (Caucasian Sector), Mongolia, China, Korean Peninsula, and Japan (Central Asian Sector), with separate type collections for each.

4.4.2 Important Herbarium of the India

Most of the major herbaria in India are managed by the Botanical Survey of India (BSI). The Central National Herbarium is the largest one with about 2 million specimens including 1500 type specimens. The most important herbaria of India with approximate number of specimens are listed here under.

Table - 4.B.2 : The Major Herbaria of India

S.N.	Name of the Herbarium	Year of Establishment	Acronym	No. of specimens
1	Central National Herbarium, Howrah	1795	CAL	2,000,000
2	Forest Research Institute, Dehra Dun	1816	DD	330,000
3	BSI, Southern Circle,Coimbatore	1874.	MH	259,000
4	Blatter Herbarium, St. Xavier College, Bombay	1906-07	BLAT	200,000
5	BSI, Western Circle, Pune	1956	BSI	170,000
6	National Botanic Research Institute, Lucknow	1948	NBG	120,000
7	BSI, Northern Circle, Dehra Dun	1956	BSD	102,169
8	BSI, Industrial Section, Calcutta	1887	BSIS	70,000

Central National Herbarium, Howrah

The Central National Herbarium (CNH) is a national repository with about 2 million plant species. William Roxburgh established this herbarium in 1795. This building is located on the bank of the river Hooghly. The CNH has collections from India as well as from other parts of the world. It maintains exchange programs with various international Herbaria. More than 15,000 type specimens, the Wallichian collections, with more than 12,000 specimens, and one of the few lithographed copies of Wallich's catalogue are housed in this herbarium. Microfiches of the Linnean herbarium and 26 other important herbaria are also preserved here. There is a large collection of coloured illustrations, with the 2583 coloured illustrations forming the Roxburgh Icones. Graphic material is continually added in the form of photo negatives and Cibachrome prints of types and authentic materials obtained from Kew Herbarium, England.

Several universities, colleges and research institutions are also maintaining the Herbaria. The Rajasthan University Botany Herbarium (RUBL) was founded in 1963 by eminent taxonomist Prof. Shiv Sharma. The herbarium has been given the official international recognition (acronym-'RUBL'). At present it has 20033 specimens belonging to 157 families, 700 genera and 1370 species. It is proposed to update the herbarium to include all the angiosperm plants available in all Districts of Rajasthan.

4.5 Making Herbarium

4.5.1 Preparing for Field Work

Plants shall be in depends on the purpose of study. Before moving to field work careful planning and arrangements have to be made for stay and cooperation from local authorities is must. Identity cards and route map of the area also given. Earlier published floras should be studied. Field trips should be taken up in the morning and to be completed by afternoon.

Field work can be classified into three types.

1. Collection trips: these are of short duration usually a day or two to near by places.
2. Exploration: for any floristic study, the area is intensively studied and collections are made regularly in different seasons. Usually explorations are of 2-4 weeks duration.
3. Expedition: expeditions are usually undertaken to remote or difficult area and are usually of several months duration.

4.5.2 Materials for Field Work

The equipment for field work depends on purpose of study. Items essential for collection include plant press, field notebook, collection containers like vasculum, big polythene

bags, cutter, pruning shears, scissors, knife and a digging tool such as trowel or pickaxe, hand lens, camera, measuring tape, pencil, scale, ball point pen, pocket lens, old news papers, blotting papers, ropes, identity cards, torch with batteries, binoculars, compass and altimeter (to note the altitude), cans containing preservatives, bottles containing formalin and first aid box,. The important ones are field notes, vasculum and plant pressers. Field Note book: Detailed notes should be entered in the field note book at the time of collection in the field itself. It has 100 leaves i.e. the pages are serially numbered (called as field numbers). Each and every page of the field notes have the detachable slips (usually 4 -8) at the right end of the sheet which are separated by perforated lines. All the slips of a page carry the same field number of the respective pages. The detached slips or number tags will be tied to the specimens with the thread provided. The field note book cantain the following information:-

Date of Collection

Genus, species and family names with vernacular names, uses locality. The name of the place or locality with reference to were known place which can be easily located in a map and traced.

Habitat and other relevant notes: This denotes the condition under which the-plant is growing, such as moist places, dry open areas, grasslands, forests, rocky crevices, steep slopes etc.

Habit and other description information: A brief description of characters which cannot be observed in pressed and processed specimens should be recorded for example habit of the plant (tree/climber, liana etc.), nature of the inflorescence, colour of flowers, fruits, leaves (as the parts usually get decolourize after alcohol treatment), pubescence, presence of aroma, latex and flowering or fruiting period also recorded.

Abundance and species association: The relative abundance and frequency of the plants should be noted. This infor-mation is valuable for commercial collections. Species association with other plant species is also recoreded which help to study the Ecology.

Specific local uses: Information can be gathered from the local people *viz*. medicinal values, authentic information should be obtained such as the ailments cured, mode of administration of the medicine.

Collector's name and date: Collector's names should be written in full with date of collection of speciemes.

Equipments

Vasculum

It is a metallic box made up of with tin or aluminum with a tightly fitted lid and with a shoulder sling, generally painted white to deflect heat. It is used to store specimens temporarily prior to pressing, and store bulky parts and fruits. For bulky part the vasculum is now commonly substituted by polythene bags which easy in storage, and readily made airtight using a rubber band to retain freshness for many hours.

Plant Press

A plant press consists of two wooden plywood sheets or wire mesh planks, each 45 cm x 30 cm. Many times a press is made with a lattice of wooden strips or small iron rods. Between the two pressers corrugated sheets, blotters or newspaper sheets are placed. Two canvas straps, belts or cotton ropes are used to tighten the press. Corrugated sheets or ventilators are made of cardboard, which help ventilation and the consequent drying of specimens. The plant press used for subsequent pressing and drying of specimens is kept at the base camp or the organization, is called the drying press. It is much heavier and has more number of corrugated sheets.

4.5.3 Collection of Specimens

The specimens collected should be in good condition. The size of the herbarium-sheet is approximately 28x42 cm. In case of herbs, the whole plant with underground parts should be collected. In case of woody plants like trees, shrubs or liana twigs with leaves and reproductive parts should be collected.

A pruning shears is used to cut the twigs of tall trees. The collecting twigs is essential for digging up rhizomes, deep seated bulbs or corms and the roots of most herbaceous plants. In case of grasses and sedges the whole plant including underground part should be collected. If the plants are uni-sexual care should be taken to collect both sexes, collections should contain at least flowers or fruits or preferably both. In case of grasses, sedges, and other herbaceous plants, the whole plant including the underground part should be collected. Generally better to collect quadruplicates for a single field number and the same taxon from other areas. This helps in studying the population of a species across the range of variation in characters is better than collecting a single plant.

Field Number

The field numbers given to collections are a very important record. Some workers have their own serial numbers. Generally institutes maintain one continuous serial number for collections. Continuous serial number either of the institute or of an individual for exploration in a particular area till that work is completed should be maintained. Each

specimen should be numbered before it is put in the collection bag. Only one series of numbers should be used and sequence be maintained. For each species the number should be entered in the field-book and a tag bearing that number should be attached to the specimens. Duplicates of the same species should be collected in 4-5 samples and the same number given to all duplicates. After tying the number and recording notes in the field notebook, the specimens are kept either in the vasculurn or polythene bags or in other containers or in the field press.

4.5.4 Collection Techniques for Special Kinds of Plants

It is used for Succulent plants like members of Cactaceae, Crassulaceae, and Euphorbiaceae etc. due to difficulties in making herbarium specimens by normal process. It shall be dried by artificial heating or frequent changing of dryers, they are prone to damage and catch fungal infection. There tissues should be killed by dipping in boiling water for few seconds or excess of tissues removed by hollowing out the thick organs or can be used alcohol or strong formalin.

1. **Collection of Ferns:** Ferns should be collected with their basal portion as the shape of rhizome, and hairs and scales on rhizome are important taxonomic characters if the frond is too big the exact size noted in the field note book along with other details.
2. **Collection of Aquatic Plants:** Free floating minute aquatic plants like *Lemna*, *WoIffia*, etc. are collected by inserting a wire press or sieve plate with white paper or muslin cloth below the specimen in water and taken out. Then the paper or cloth is lifted slowly with both hands and placed between the dryers. The plants should be changed along with the paper or muslin cloth. For the first one or two days, the changing should be more frequent.
3. **Collection of Plants having Mucilage, Gums and Resins:** The plants species containing mucilage or resin they stick to the dryers and cause difficulty while changing. Such specimens should be placed in a folder of muslin or any other thin cloth and pressed. Only the dryers should be changed and not the muslin cloths eg. *Hibiscus*.
4. **Collection of Aroids and Orchids:** Aroids (Araceae members) frequently have a virtually impenetrable epidermis and without killing properly they continueing to grow even in the press similarly some bulbous orchids. The killing is done either by heating with proper ventilation or by use of alcohol and formalin.
5. **Collection of Large Plants:** Due to their large size, bamboos, palms and bananas and tree ferns like Cyathea require special methods for collection. The habit of the

plant and the approximate size of the culms (or pseudo stem like banana), leaves and inflorescences must be recorded. Either the whole plant or its main parts should be photographed with a scale or their sketches must be made on the spot. Effort should be made to collect leaves or portion of leaves with noting the actual magerment; in case of bamboos few ligules must be collected.

4.5.5 Processing of Specimens

Poisoning the specimens for avoid deterioration and damage to collected specimens, the specimens are duly poisoned for long preservation. Two methods are employed for poisoning.

1. **Dry Method:** The specimens are poisoned at the time of collection; the whole plant is dipping the saturated solution of mercuric chloride in ethyl alcohol (usually 20 gm in a litre of alcohol). The plant is again put in the presser till it gets completely dried. Lauryl Pentachlorophenate (LPCP) is also used for poisoning the specimens. 3.75% of LPCP is mixed in white spirit.
2. **Wet Method:** This method is highly suited for tropical countries. When the day collections are so numerous it is not always possible to dry the collection by changing of blotters. In such cases, the collections are spread out in ordinary old newspapers and bundled up. Each bundle is then placed in a large polythene hag. Ten per cent formalin is poured over the bindles, so that the bundles just get soaked thoroughly. Care should be taken to avoid excess of formalin in the bags. The bags are then tied airtight. No further change of folders is necessary till reaching the headquarters. By this method, it is possible to bring the collections made even over 3-4 months. On reaching the headquarters, the bundles are opened out and the specimens are open for further action.

   Pressing of Specimens: Pressing is the process of placing specimens between the absorbents under moderate pressure. The main object of pressing is to flatten and dry the specimens. This is done by keeping the straps tight and by changing the blotters every day for 6-10 days depending on weather. The plants gradually lose their moisture and finally get dried.
3. **Drying of Specimens:** After poisoning and pressing of the specimens are needed to dry completely and this can be achieved by two methods.

   Natural drying: It is a slow process which may take not less than 15 days to specimens for complete desiccation. The specimens kept in the presser are transferred to new blotter or news paper on alternative days and again tied tight and allowed dry at room temperature.

Artificial drying: In this method, artificial heat is used for drying. Specimens in the field presser are transferred to drying presser containing more number of corrugated sheets after a day of collection. The presser is then kept in a drier. Use of a hot air blower in the drier cabinet speeds up the hot circulation of heat and specimens get completely dried within 2 days. Recently solar-powered driers are also in use which attain a temperature of 60°C and are capable of drying 100 specimens on a sunny day.

4.5.6 Mounting and Labeling

Collected, poisoned, pressed and dried specimens are mounting on herbarium sheets. Fixing the processed plant specimen on herbarium sheet is called as mounting. A standard herbarium sheet is 28cm x 42 cm and usually made up of heavy long lasting white hand made paper.

The specimens should be neatly and uniformly spread and fixed on the sheet. Parts of the plant should be exposed for study.

For small herbs, the whole plant with roots or underground parts should be accommodated on one mounting sheet.

For larger herbs, though it is possible to collect the whole plant, yet it has to be cut into two or three parts.

For woody plants like trees, shrubs or liana with reproductive parts should be pasted on the herbarium sheet. Longer than the size of mounting sheet can be folded in the shape of V, N, M or W.

Glueing and Stitching: The common technique is pasting specimens to sheet with glue (usually Gum-Arabic). The glue paste is made by adding flakes of glue to boiling water, gradually and in small quantities, till it makes a thin paste. As the paste become thick and hard after cooling, the vessel containing glue should be kept on low heat during mounting work. Small quantity of mercuric chloride or thymol crystals or copper sulphate may be added as insect repellent, nowadays fevicol is frequently used. There are two methods to fix the specimen on the herbarium sheet. The glue paste is applied to the backside of the specimen which is later pressed onto the mounting sheet and allowed to dry in presed condition for few hours. The method for delicate specimens or grasses is to smear the glue on a glass plate or thick flat plastic sheet and specimen be placed on the sheet and the glued specimen is transferred to a mounting sheet. After fixing the specimen on the sheet with glue, the specimen may be further sewed with thread here and there for more secure fixing.

Herbarium Label: Mounting of the specimens is followed by pasting of herbarium label on the right hand side at bottom. The size and design of herbarium labels slightly vary according to need. General size is about 8x12 cm.

(1) Field number (the number given from the field notes). (2) Name of the family (3) Name of the genus and species. (4) Locality of collection (actual place of collection of the plant). (5) Altitude (important for hilly regions). (6) Date of collection. (7) Description/Notes (habit, habitat, features not seen with the herbarium specimen). (8) Collector's name and (9) Identification (person who identifies the specimen) is required for label.

4.6 Arrangement and Maintenance of Herbarium

4.6.1Sorting and Filing of Specimens

Before the mounted specimens are incorporated in the herbarium, they are stamped with the distinctive mark (seal) of the herbarium or institution and an accession number is given. Field number and accession numbers of a specimen are different. Stamping is usually done on the top right hand corner. This stamp carries the name of the institution and space for accession number and date of accession. The sheets are also listed in an accession register along with their, accession number and then incorporated in the main herbarium. The well mounted, identified and accessioned herbarium sheets are incorporated in specially prepared almirahs in pigeon holes folder vise. All the sheets of the same species are placed in lighter covers called the 'species cover' or folders; and all the species (with species covers or folders) belonging to one genus are placed in one or more folders of heavy paper, called the genus cover.

The specimens are usually arranged in the herbarium according to some accepted system of classification. Generally in Indian herbariums are followes Bentham and Hooker classification. For other groups of plants, the specimens have to be arranged according to some standard system of classification. The pigeon-hole, where bundles of a new family start, is marked by a fixed label or by hinged flap-board separator cardboard. The name of the family is printed or written on this in bold letters.

4.6.2 Indexing the Specimens

An index card system should be maintained in the herbarium for easy reference for identify the species of interest. It provides an abstract of the specimens preserved in the herbarium.

The index cards should contain information of the specimens' field number, dates of collection, localities of collection, name of collectors etc. Nowadays computerized database are being maintained in the herbaria.

4.6.3 Special Arrangments for Certain Specimens

Segregation of Cultivated Plants: Specimens of common cultivated plants of a region should be separated from others and kept in the general herbarium at the end of each family. It is better to there in genus covers of a different colour.

Segregation of Sheets for Students and Beginners: If a herbarium is frequently visited by inexperienced students and other beginners, it is advisable to maintain a set of collection of species commonly encountered in surrounding areas separately. Undetermined Specimens: if some plants are not identified, these should not be mixed with others, but should be kept in separate bundles for further study by specialists of those groups whenever such experts visit the herbarium.

Bulky Herbarium: Many large specimens such as bamboos or large rhizomes and fruits cannot be mounted or displayed on mounting sheets and require special drawers or cupboard; this section of the herbarium is called 'Bulky herbarium'.

Handling or Sorting of Unmounted Duplicates: After mounting 1 or 2 specimens of each collection, the remaining duplicates should be preserved as stock carefully, as these may be needed for various purposes. The bundles of these duplicates should also be fumigated at regular intervals and stored in closed or open racks. The specimens should be packed in old news-papers and bundles arranged according to serial numbers. The bundle should bear the labels or tags showing the contents which will help in tracing the duplicates plants.

4.6.4 Decontamination Methods

Any dried material enters a herbarium must be subjected to a process of decontamination. This can be done by heating, fumigation, deep freezing or by using insect repellents. The specimens are generally in dried conditions and hence not prone to any damage to fungi, bacteria, but may be easily attacked by pests like silverfish and beetles. The pest control can be done by heating the sheets at 60C for 4-8 hrs in heating cabinet or kept in deep-freezers where the temperature maintained in the range of 20ºC to 60ºC. Microwave ovens are also used for pest control. Insect repellent chemicals such as Para dichlorobenzene (PDB) and Naphthalene are used for pest control. Usually these are powdered and sprinkled on sheets are the balls are put in small muslin cloth bags and kept in pigeon holes. Since both the chemicals are toxic at high concentration, care should be taken with the time of exposure. Further PDB should not be used along with naphthalene. Fumigation is done for killing pests in mounted as well as unmounted duplicate speci-mens. This process involves use of any one of the volatile poisonous liquids like methyl bromide, carbon disulphide or carbon tetrachloride. A mixture of ethylene dichloride (3 parts) and

carbon tetra chloride (I part) is commonly used. These are placed in small saucers or petri dishes in each herbarium case and the cases kept closed for about a week

4.6.5 General Instructions for Herbarium Users

1. Do not shuffle the sheets kept in folders.

2 Store the specimens properly and perfectly. Do not crowd too many specimens into a box or in a pigeon hole.

3. Do not write anything on herbarium sheets.

4. Do not lay books or other heavy objects on specimens.

5. No flower and fruit is to taken for dissection from herbarium sheets.

6. Those specimens which need repair should be brought to the notice of the curator.

7. While handling, full bundles should be taken out form the racks; pulling one or more sheets will surely damage the specimens.

8. Putting bundles again in the racks after use should be done with extreme care.

9. Specimens should not be kept on tables for longer periods to prevent.

10. Alimarahs should be closed after taking and while keeping back the bundles.

11. Open flames, smoking is strictly prohibiting in the herbarium.

12. If any incorrect identification is there it should get notice to the curator of the herbarium. It will help in regular updatation.

4.7 Flora

A flora is systematic enumeration of plant species which grow in a given geographical region. It provides key, discretion, photograph and linear diagram of the plant species. The illustration is well in determinate of the plant species which describe in the flora. The first chapter is introductionary and describe fitures of the region like, geographical, topography, soil type maps, climate etc. relatively than it discuss with taxonomic characters of the plant species which offen deccribe in it. One of the main functions of flora is plant identification. Flora may cover any suitable area from a small forest, city, district, state, country or a continental. A flora is describes the general physiognomy.

A flora usually been with the flowering plants, include native species as well as establish species and cultivated plants are mantened after end of the family. Name of the texa was given according to ICBN (International Code of Botanical Nomenclature), scientific name, author citation, reference to source of original publication. The synonyms of the

species also provided and description of the taxa are the given in technical terms, the flowering and fruiting periods and local name also mention.

Depending on the scope and the area covered the floras categorized as:

(1) Continental flora:

(a) Flora of Tropical Africa by D. Oliver *et al*. (1868-1937).

(b) Flora of Australiensis by G. Bentham (1863-78).

(2) Regional flora:

The Flora of British India (1-7 Volume) by Sir J.D. Hooker (1872-97).

(3) Local flora:

(a) Flora of Delhi by J.K. Maheshwari (1963).

(b) Flora of the upper Gengetic plain by J.F. Deathie (1903-22).

(c) Flora of the Indian Desert by M. M. Bhandari (1978).

(d) Flora of Jaipur District S. Sharma (1976).

(4) Comprehensive Treatment (Have much broder scope):

(a) Genera planterum of G. Bantham and J.D. Hooker (1862-83)

4.8 Summary

Herbarium is storeroom of plant specimens collected from various areas, dried, mounted on appropriate sheets and arranged in sequence according to some known system of classification. Herbarium serves as original source in identification of plants, helps in teaching and research and also a repository of historic collections. Herbaria can be type general or special. General herbaria are at different level like international, national and local based as their collections status and mode of process. Special herbaria are small and established for a specific purpose and have limited scope. Index Herbarirum listed as the world's important herbaria but the largest herbarium of the world is at Paris (collection of 7 million specimens). Central National herbarium at Howrah is the largest (collection of 2 million specimens) Indian Herbarium. For the prepration of Herbarium the plant collection should be done in a well planned manner, plant containers, field notes, plant pressers are must for field work and all the details of the species should be entered in the field notes book. Special care is needed for collecting special groups of plants like succulents, bulky specimens etc. after the collection specimens properly poisoned, dried, pressed and preserved. Specific methods for pasting of specimens on herbarium sheets should be adopted and label pasted on the sheet must be cover abstract information of the species. The specimens incorporated with a distinctive herbarium number and seal and accessioned herbarium sheets are sorted out family, genus and species vise and kept them in appropriate folders.

A flora is systematic enumeration of plant species which growing in a geographical region. It provides key, discrition photograph, linear diagram of the plant species. One of the main functions of flora is to provide a mean of plant identification.

4.9 Glossary

- **Herbarium:** A collection of plant specimens that usually have been dried and pressed, arranged in sequence of an accepted system of classification and are available for scientific study.
- **Flora:** A flora is book of systematic enumeration of flowering plant species which occurs in a given geographical region.

4.10 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. Write the name of biggest Herbarium of world.
2. Write the name of biggest Herbarium and its number of specimeans in India.
3. Write is the standard size of Herbarium Sheet?
4. Write the name of any local Flora of your area.

Section B : (Short Answer Type Questions)

1. Write the importance of herbarium.
2. Write an account on the methodology of collecting plant specimens.
3. Write the different technique of plant processing.

Section C : (Long Answer Type Questions)

1. Write the technique for succulents and bulky plant processing.
2. What should be the contents of a field notes?
3. What are the things to keep in mind during mounting the specimens on sheets?

4.11 References

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Unit - 5

Classification Systems of Angiosperms

Structure of the Unit:

5.0 Objectives

Following are the objectives of this unit:

- to name all plants of the world and fix their specificity to their habit, habitat, distribution, characters etc.
- to arrange them according to their characters in their particular place in the classification.
- to name the taxon according to ICBN.

5.1 Introduction

The taxonomic literature reveals three types of classification. As a whole system are artificial system, natural system and phylogenetic system. History of classification of angiosperms can be classified into two major systems which can be further subdivided as follows.

A. Pre Darwinian system:

Period-I: Classification based on habit, which starts with Theophrastus (370-285 BC).

Period-II: Classification based mainly on number of floral parts, especially stamens. This period was characterized by system of classification that was built to be artificial with the sole purpose of plant identification. Example Carolus Linnaeus (1707-1778).

Period-III: Classification based on morphological characteristics or form relationship. Example Bentham and Hooker

B. Post Darwinian system:

Period-IV: Classification based on phylogeny of plants; starts with Engler and Prantl.

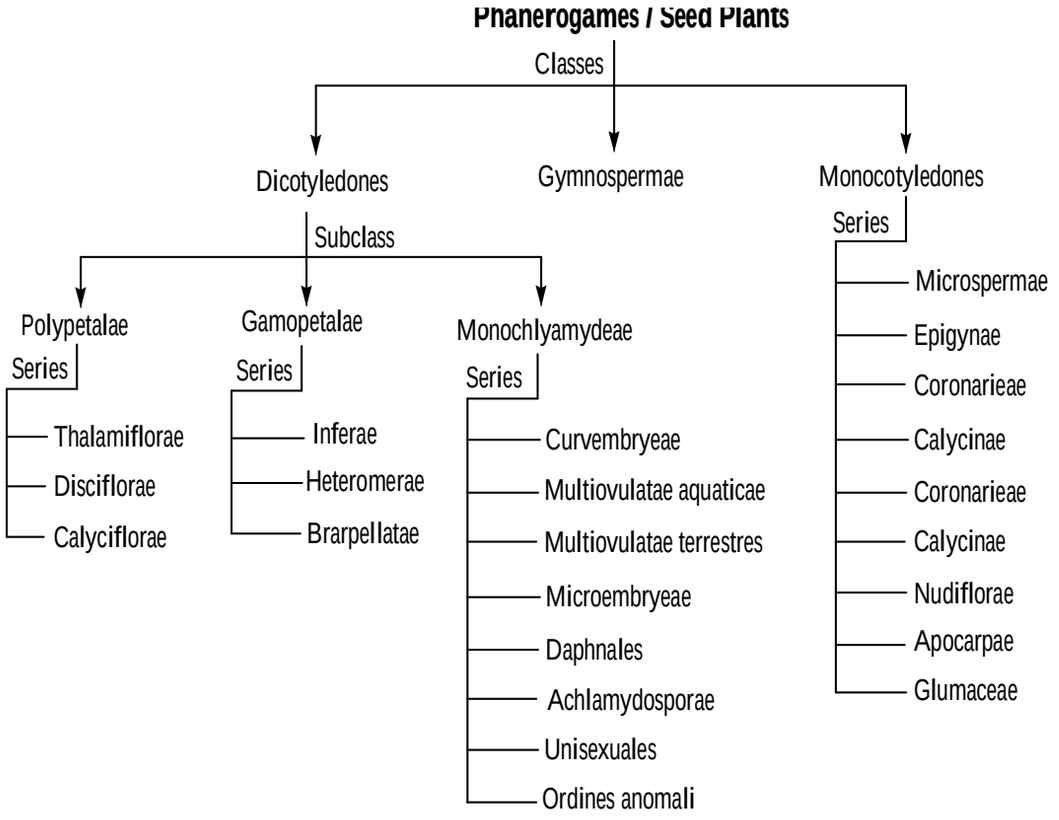
5.2 Bentham and Hooker's Classification

The system of classification of seed plants presented by Bentham and Hooker is well developed natural system. The classification was published in *Genera plantarum* (1862-83).

The *Genera plantarum* provides the classification of seed plants which divided into 202 families, 7569 genera and 97,205 species. Many important herbaria of the world (including India and England) have specimens arranged according to this system.

Seed plants (Phanerogams) divided into three classes viz., Dicotyledones having cotyledones 2, reticulate venation and tap root system. Gymnospermae and Monocotyledones having cotyledon single, parallel venation and adventitious root system. The Class Dicotyledones were further subdivided into three subclasses; Polypetalae, Gamopetalae and Monochlamydeae (Mainly based on the presence or absence of petals and their fusion). These subclasses were further subdivided into series, orders and families. Class - Monocotyledones and Subclass - Monochlamydeae (incompletae) described by series and families directly. A broad outline of the classification is presented in Table 5.1

Table 5.1: Classification of Bentham and Hooker



Key characters of some important taxa:-

Class: Dicotyledones- It is characterized by two cotyledons, reticulate venation in leaves and tap root system.

Sub class: Polypetalae- Petals are free.

Sub class: Gamopetalae- Petals are fused.

Sub class: Monochlamydae- Flowers have perianth *i.e.* calyx and corolla are not distinguished

Class: Gymnospermae- It is characterized by naked seed.

Class: Monocotyledones- It is characterized by single cotyledons, parallel venation in leaves and adventitious root system.

Merits

1. The system has great practical value for identification of plants. It is very easy to follow for routine plant identification.
2. The system is widely followed for the arrangement of specimens in the herbaria of many countries including Britain and India.
3. Dicotyledones are placed before the Monocotyledones.

4. Placements of gamopetalae after polypetalae is supported by evolutionary condition.
5 Heteromerae is rightly placed before bicarpellatae.
6. Ranales were considered as most primitive dicots by most of the leading authors.
7. Keys for the identification are very useful. Each family had a synopsis at the beginning which is very useful in identification.
8. The description of families and genera are precised.
9. Large genera have been divided into small (subgenera) in order to facilitate identification. .
10. The arrangement of taxa is based on overall natural affinities decided on the basis of morphological features, which can be easily studied with the naked eye or with a hand lens.

Demerits

1. The system does not give any idea about phylogeny.
2. Gymnosperms are kept between Dicotyledones and Monocotyledones which are extremely anomalous.
3. The taxon Monochlamydeae is entirly artificial, due to based on one whorl of perianth.
4. Unisexuales is a loose assemblage of diverse families, which share only one major character i.e. unisexual flowers. Cronquist (1988) separates these families under two distinct subclasses Hamamelidae and Rosidae and Takhtajan (1987) under Hamamelididae and Dellenidiidae.
5. Bentham arid Hooker did not know the affinities of the families placed under Ordines anomaly.
6. Compositae (Asteraceae) is highly advanced family and placed in inferae at the beginning of Gamopetalae.
7. Orchidaceae is an advanced family with inferior ovary and zygomorphic flowers, but the family is placed towards the beginning of Monocotyledones.
8. In gamopetalae, inferae with an inferior ovary is placed before the other two series having a superior ovary. The inferior ovary is now considered to advanced derived from a superior ovary.
9. The position of series apocarpae is unsatisfactory due to its free and superior carpels.

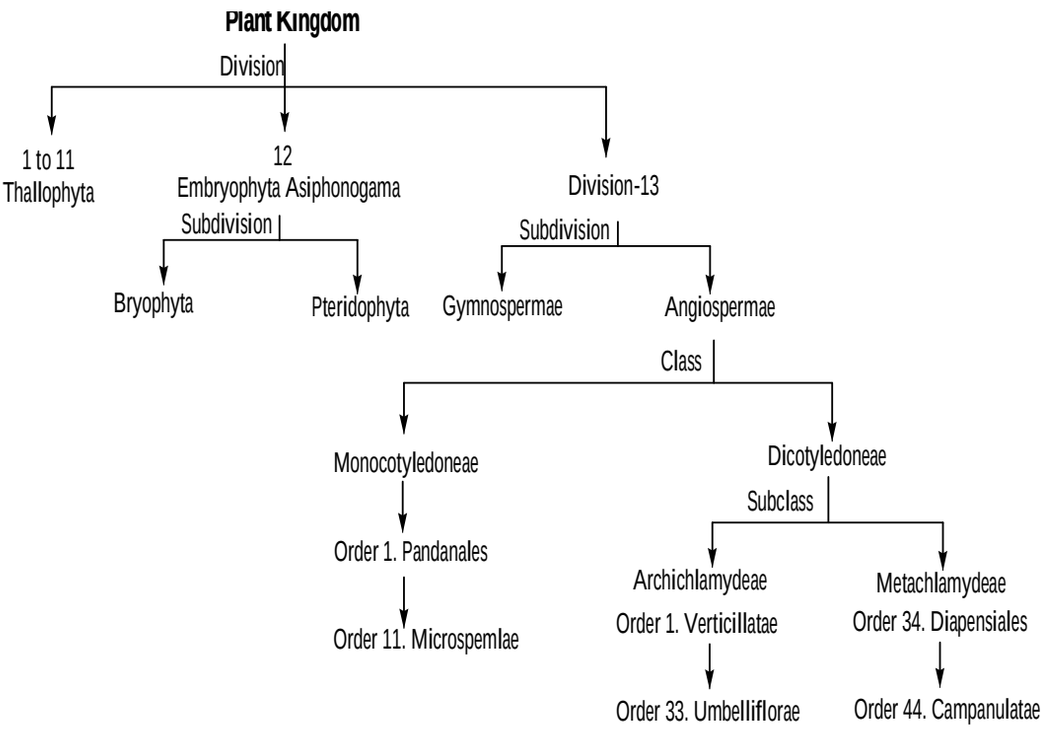
5.3 Engler and Prantl's Classification

This system of classification covers entire plant kingdom. It is proposed by Adolph Engler (1844-1930) and Karl A. E. Prantl (1849-1893). The classification was published in a monumental work *Die Naturlichen pflanzenfamilien* in 23 volumes (1887-1915).

It was considered as first phylogenetic system of classification. Families are arranged according to increasing complexity of characters. In this system of classification, the plants are divided into 13 groups and ranked the divisions. The first 11 divisions covers Thallophytes, the 12th divisions of Embryophyta Asiphonogama covers Bryophytes and Pteridophytes. For the seed plants there was last 13th division termed Embryophyta Siphonogama.

Embryophyta Siphonogama was further divided into subdivision Gymnospermae and Angiospermae. Angiosperms are further divided into classes Monocotyledonae and Dicotyledonae. Monocotyledonae is further divided into 11 order and 45 families starts with Pandanceae while ends at Orchidaceae. Dicotyledone's family starts with Casuarinaceae to Compositae having total 258 families of 44 orders. Order Pandanales and Verticillatae are starting order of Monocotyledons and Dicotyledons respectively (Table 5.2).

Table 5.2 : Engler and Prantl's Classification



Key characters of some important taxa:-

Division (1-11) : Thallophyta - Plants with thallus. In thallus plant body is not differentiated into root, stem and leaf.

Division (12) : Embryophyta Asiphonogama – This group includes Bryophytes and Pteridophyta

Division (13) : Embryophyta Siphonogama – This group includes seed plants which further divided into Gymnospermae and Angiospermae. Angiosperms are classified into Monocotyledones and Dicotyledones.

Merits

1. This was the first phylogenetic system of classification.
2. Gymnosperms are separated and placed before angiosperms.
3. Many large families have been splited into small and natural families, like Urticaceae into Urticaceae,Ulmaceae and Moraceae.
4. The artificial group Monochlamydeae (Bentham and Hooker) was merged into Archichlamydeae.
5. Families with epigynous flowers were treated as advanced in Archichlamydeae and metachlamydeae.
6. The evolution is considered as hypogyny to epigyny.
7. Compositae (Dicots) and Orchidaceae (Monocots) are rightly placed towards the end of Dicotyledones and Monocotyledones, respectively.

Demerits

1. Monocotyledones are placed before Dicotyledones.
2. Amentiferae (including Betulaceae, Juglandaceae and Fagaceae) with reduced unisexual flowers having few floral members and borne in catkins, were considered primitive; but recent studies reveals that it is advanced taxon.
3. Origin of Dichlamydeous forms (distinct calyx and corolla) from the monochlamydeous forms (single whorl of perianth) is not supported by majority.
5. Polyphyletic origin of Angiosperms is not accepted due to recent evidence which indicates monophyletic origin.
6. Helobiae (including family Alismaceae, Butomaceae and Potamogetonaceae) is a primitive group but not placed properly.

5.4 John Hutchinson's Classification

John Hutchinson (1884-1972) was associated with the Royal Botanic Gardens, Kew, England. Classification of Angiosperms was published in the book "*The Families of Flowering Plants".* It is published in two volumes separately; Volume I was on Dicotyledones and Volume II was on Monocotyledones in the year 1926 and 1934 respectively. The classification was published and updated regularly, second edition in 1959 and the 3rd in 1973, one year after his demise.

Hutchinson classified only the flowering plants and ranked Phylum Angiospermae as distinct from Phylum Gymnospermae. Dicotyledons are divided into division Lignosae and Herbaceae having 54 and 28 orders respectively. Division Lignosae starts with Magnoliaceae and ends with Verbenaceae. In case of Herbaceae it start with Paeoniaceae and ends with Lamiaceae. Monocotyledons were divided into division Calyciferae, Coroliferae and Glumiflorae. Family Butomaceae placed at starting while Poaceae at the end of Monocotyledons. The classification was based on 24 principles (Table 5.3). Some of them were mentioned below:-

i. Evolution takes place in both direction i.e, upwards (sympetaly) and downwards (apetaly).

ii. Evolution does not necessarily involve all the organs in all the directions of a plant at the same time.

iii Dicotyledones possess collateral vascular bundles arranged in a cylinder are more primitive in origin than Monocotyledones having scattered bundles.

iv. Simple leaves precede compound leaves. Spiral arrangement of foliage and floral leaves precedes that of opposite and whorled types.

v. Solitary flower is more primitive than the inflorescence and bisexual precede unisexual flowers.

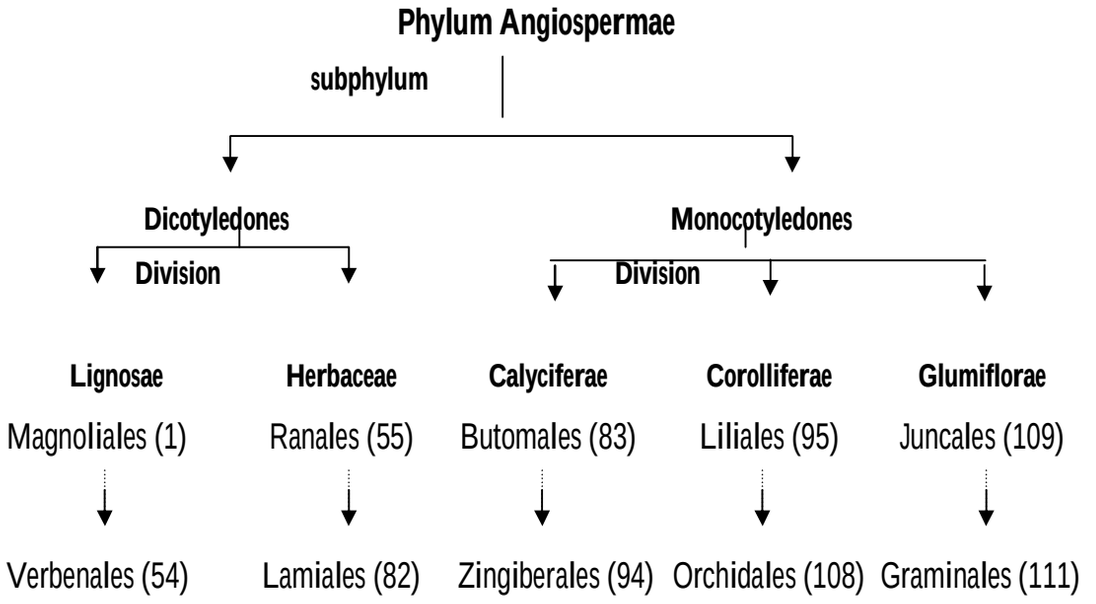
vi. Polypetalous conditions are more primitive than gamopetalous.

vii. Hypogyny is primitive than the perigyny followed by epigyny.

viii. Endospermic seed is primitive than non-endospermic.

ix. Many stamens is primitive than few stamens.

Table-5.3 : Classification of Hutchinson.



Key characters of some important taxa:-

Division: Lignosae- Starts with Magnoliales, including woody dicots.

Division: Herbaceae- Starts with Ranales, including herbaceous dicots.

Division: Calyciferae- It is a monocotyledons group starts with Butomales **to** Zingiberales.

Division: Corolliferae- It is a monocotyledons group starts with Liliales to advance Orchidales.

Division: Glumiflorae- It is a monocotyledons group starts with Juncales to most advance and last taxa Graminales.

Merits

1. This system of classification is considered to be more phylogenetic. It is based on generally accepted phylogenetic principles.
2. Magnoliales kept at the starting point with family Magnoliaceae.
3. The abolition of previous commonly used terms e.g., Polypetalae. Gamopetalae, Monochlamydeae, Archichlamydeae and Metachlamydeae etc.
4. Monocotyledones are considered to be more advanced than Dicotyledones.
5. The arrangement of families of Monocotyledones is widely accepted. Keys to the identification of genera have been provided.
6. The placement of Alismatales towards the beginning of Monocotyledones finds general acceptance.

7. Advanced family Poaceae has been kept at the last.
8. Taxon classified up to the generic level, together with identification keys.

Demerits

1. The system is not useful for practical purposes.
2. Angiosperms originated from hypothetical group "Proangiosperm" But information not provided about this group.
3. The division of Dicotyledones into Lignosae and Herbaceae is mainly based on habit; due to this closely related families were separated eg. Araliaceae and Apiaceae.
4. The related families on the basis of floral structure were separated, *e.g.,* closely related families of Ranales as Ranunculaceae and Magnoliaceae were kept far away.
5. The members of Liliaceae and Amaryllidaceae were clubbed together; all plants having umbel inflorescence are placed in Amaryllidaceae.
6. The plants having phylloclades like *Ruscus* are placed in Ruscaceae.

5.5 Armen Takhtajan's Classification

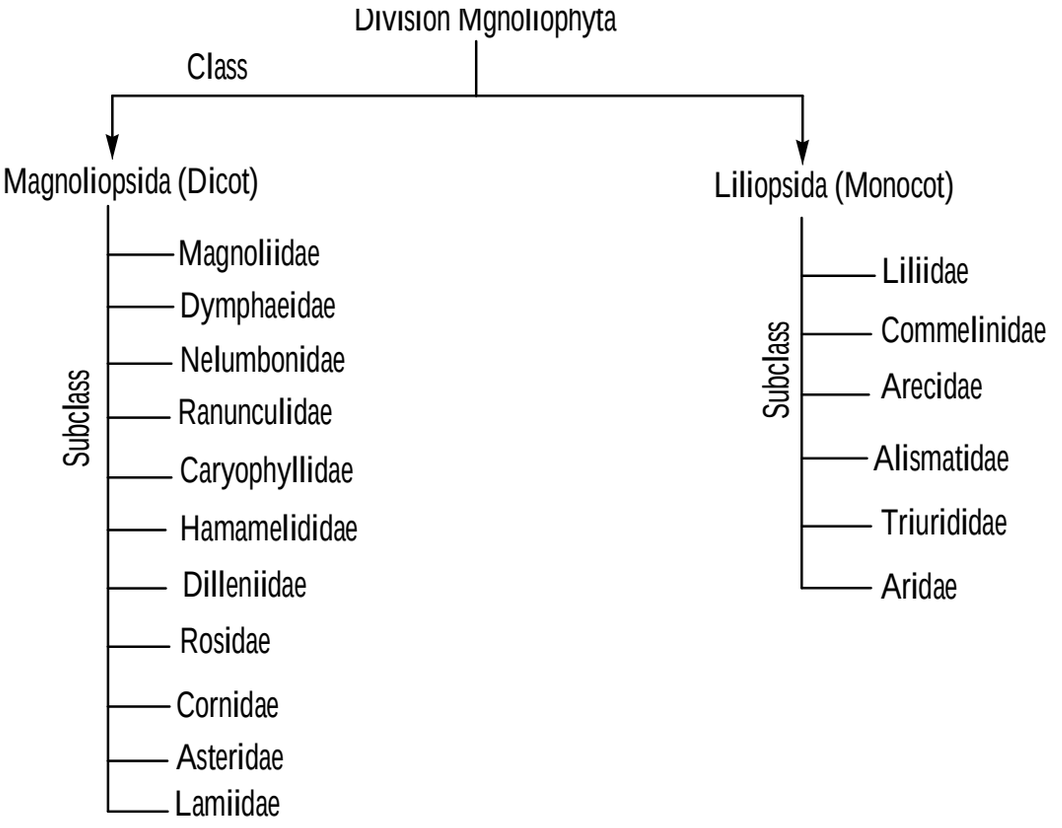
Armen Takhtajan (1910-2009) was associated with V.L. Komarov Botanic Institute, Leningrad (now St. Petersburg). Takhtajan published classification of flowering plants in *Die Evolution der Angiospermen* (1959) which was explained in detail in his system and Phylogeny of Flowering Plants (1966). Recent classification presented in Outline of the *Classification of Flowering Plants* (Magnoliophyta) in Botanical Review in 1980.

Takhtajan classified Angiosperms as Division Magnoliophyta and subdivides that into the class Magnoliopsida (Dicotyledones) and Class Liliopsida (Monocotyledones).

Class Magnoliopsida consists of 7 subclasses, 20 superorders, 71 orders and 333 families; Class Liliopsida consists of 4 subclasses, 8 superorders, 21 orders and 77 families. He used the rank of superorder as a supplementary rank between subclass and order, derived superordinate names from generic names and with an ending - *anae*; order suffixed by - *ales* and suborders - *eae.*

Takhtajan classified angiosperms up to the family level. He considered the monophyletic origin of angiosperms, the group having evolved from ancient group of gymnosperm, called seed ferns (Lyginopteridophyta). Takhtajan proposed revised system in 1997 (Table 5.4).

Table-5.4 : Armen Takhtajan's classification



Key characters of some important taxa:-

Class : Magnoliopsida- Include all dicotyledones. Dicots are grouped into 11 subclasses on the basis of phylogenetic concepts.

Class : Liliopsida- Include all monocotyledones. Monocots are grouped into 06 subclasses on the basis of phylogenetic concepts.

Merits

1. The system is more phylogenetic than other earlier system.
2. It is based on widely accepted phylogenetic principles.
3. Abolition of artificial group like Polypetalae, Gamopetalae, Lignosae etc. has resulted in more natural grouping of taxa.
4. The families are small homogenous units comprising closely related genera. For example Lamiaceae and Verbenaceae are under the order Lamiales. Similarly Caryophyllaceae, Chenopodiaceae and Portulacaceae are under the order Caryophyllales.
4. The system starts with the Magnoliidae which are the most primitive

Dicotyledones.

5. Liliopsida starts with Alismatidae which are the most primitive living Monocotyledones.
6. Dicotyledones precede monocotyledons.
7. Dicotyledones are believed to be more primitive than the monocotyledones.
8. The subclasses are based on all available information.
9. It is a phylogenetic system with satisfactory relations of various groups.

Demerits

1. The system is not suitable for plant identification.
2 The system is not useful for adoption in herbaria.
3. Keys to the identification of taxa are not provided.
4. Extremely narrowly defined taxa have resulted in the unwarranted splitting of related groups.
5. Derivation of the Monocotyledones from the stock ancestral to Nymphaeales.

5.6 Arthur Cronquist's Classification

Arthur Cronquist (1919-1992) was associated to the New York Botanical Garden. He proposed classification of angiosperms in *The Evolution and Classification* of *Flowering Plants* (1963). The system was further elaborated in 1981 in his book An *Integrated System* of *Classification* of *Flowering* Plants.

Angiosperms were ranked as division (named Magnoliophyta) while Dicotyledones and monocotyledones as classes. Classes were further divided into subclasses. Class Magnoliopsida further divided into six subclasses namely Magnoliidae, Hamamelidae, Caryophyllidae, Dilleniidae, Rosidae and Asteridae. Whereas Liliopsida (Monocots) into Alismatidae, Arecidae, Commelinidae, Zingiberidae and Liliidae.

Magnoliopsida (Dicots) one classified into 64 order and 320 families. In Monocotyledones (Liliopsida) are distributed under 19 order and 66 families. Dicotyledones starts with Magnoliales with ending at Astrales. Alismatates is the starting order of Liliopsida while ends by Orchidales. Family Asteraceae and Orchidaceae is Dicotyledones and Monocotyledones are rightly placed towards the end of each group.

Cronquist considered that closely related groups of organisms evolved in similar patterns because they have similar evolutionary potentialities and are likely to produce similar mutations.

Pteridosperms (seed ferns) were considered as the probable ancestors of Angiosperms. The Magnoliidae were considered as the basal complex and the remaining five subclasses were derived from it separately. Liliopsida were considered to have arisen from aquatic ancestors and a group probably close to the modern Nymphaeales. All the five subclasses of Liliopsida have some ecological coherence.

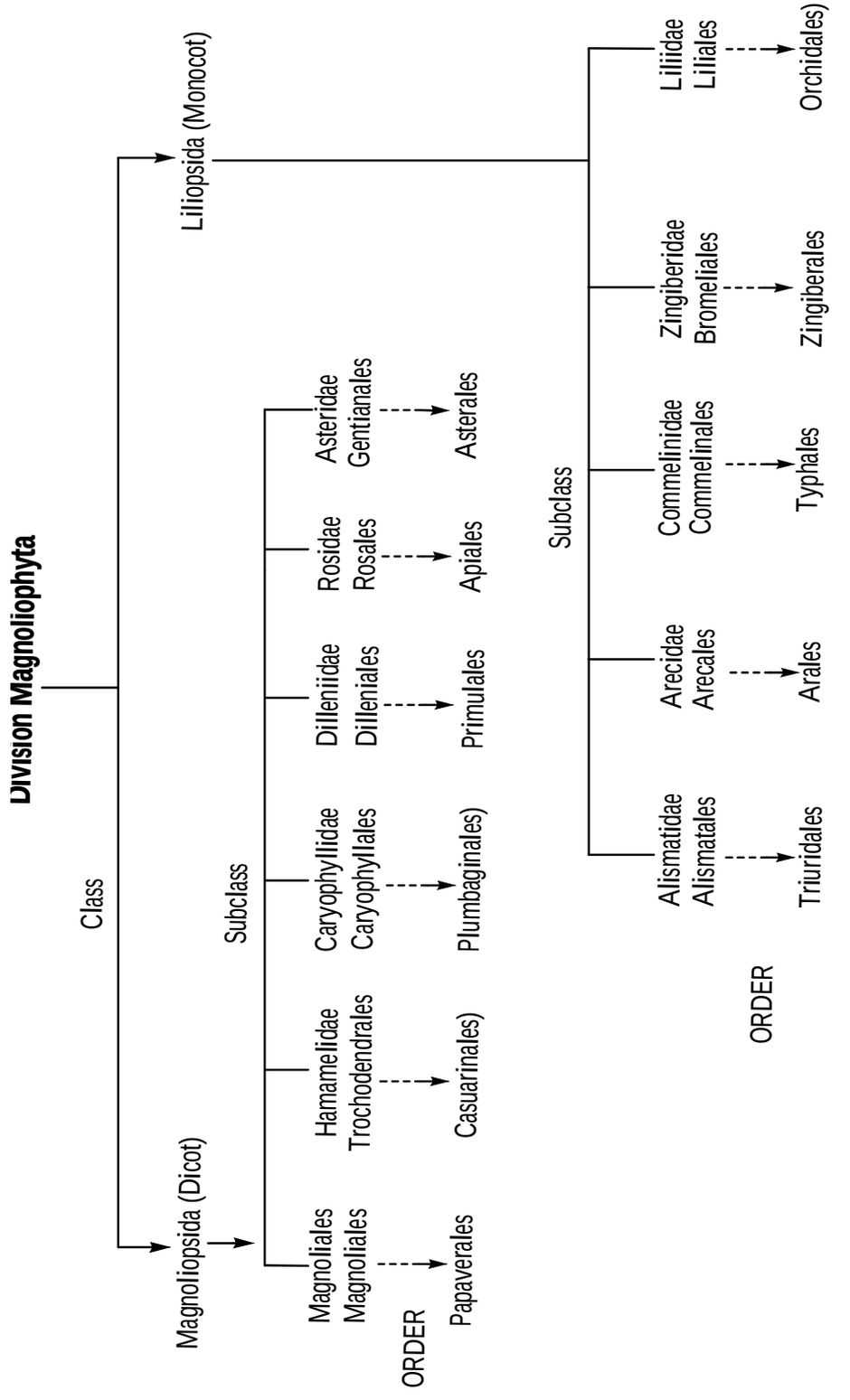


Table-5.5. Arthur Cronquist's Classification

Key characters of some important taxa:-

Class : Magnoliopsida- This term used for dicotyledones. Dicots are grouped into 06 subclasses. Subclasses are further categorized into orders. Orders start with Magnoliales to Asterales.

Class : Liliopsida- All monocots are kept in this group. Monocots are grouped into 05 subclasses. Subclasses are further categorized into orders. Orders start from Alismatales to orchidales.

Merits

1. The classification is mainly based on principles of phylogeny and widely used in the USA.
2. Classification emphasized on morphology, phytochemistry, anatomy, ultrastructure, chromosomes etc.
3. Keys for each group up to families were provided.
4. Abolition of artificial group named such as polypetalae, Gamopetalae, Lignosae, Herbaceae etc. has resulted in more natural grouping of taxa.
5. Verbenaceae and Lamiaceae are kept under the order Lamiales. Caryophyllaceae, Chenopodiaceae and Portulacaceae are placed under Caryophyllales is justified.
6. Placement of Magnoliidae as the most primitive group of angiosperms, dicotyledones before monocotyledons.
7. Magnoliales at the beginning of Magnoliidae and Butomaceae at the beginning of Liliopsida, finds wide acceptance.
8. Family Compositae in Dicotyledones and family Orchidaceae in Monocotyledones are rightly placed towards the end of respective group.

Demerits

1. The system of classification is not useful for plant identification
2. Identification keys are not given.
3. Distribution and description for genera are not provided.
4. Asteridae has loose assemblage of several diverse sympetalous families.
5. Aquatic ancestry of monocotyledones is not supported by scientific communities.

5.7 Rolf Dahlgren's Classification

Rolf Dahlgren (1932-87), was associated with the University of Copenhagen, Denmark. He proposed a new phylogenetic system of classification and a new method of illustrating imaginary phylogenetic shrub.

It was first appeared in A *Text Book of Angiosperm Taxonomy* in 1974. This system was however later revised and a revised system with a diagram was published in 1975. After this complete revision published in 1980 wherein author stated that the new classification is still provisional.

According to Dahlgren angiosperms are monophyletic in origin and have evolved from one particular line of gymnosperms. System covers entire characters emphasizing anatomy, embryology and chemistry. Magnoliflorae and Alismatiflorae were considered most primitive in dicotyledones and monocotyledones respectively.

Angiosperms are named Magnoliopsida and ranked as class. It is further divided into subclasses namely magnoliidae and liliidae for dicotyledones and monocotyledones respectively. Subclass magnoliidae has been further divided into 24 super order, 80 orders and 346 families. Liliidae is classified into 7 super order, 26 order and 92 families (Table-5.6).

After Dahlgren's death in 1987, his wife Gertrud Dahlgren continued his work and finally published the work on dicotyledones, followed by a classification of monocotyledones, both in 1989, incorporating the latest ideas of Dahlgren, and bringing up an updated classification of angiosperms.

Angiosperms are classified into Magnoliidae (Dicotyledones) and Liliidae (Monocotyledones). Magnoliidae include 25 superorders, 7 orders and 343 families. Liliidae includes 10 superorders, 24 orders and 104 families (Table-5.6).

Table-5.6 : Rolf Dahlgren's Classification

An Outline of Dahlgren's system of classification (1980)

Class : **Magnoliopsida** (Angiosperm)

Subclass : **Magnoliidae** (Dicotyledones)

Superorder-1 Magnoliiflorae → Annonales

↓

Magnoliales

Superorder-2 Nymphaeaflorae → Nymphaeales

&

Piperales

Superorder-3 Ranunculiflorae → Papaverales & Ranunculales

Superorder-4 Caryophylliflorae → Caryophyllales

Superorder-5 Polygoniflorae → Polygonales

Superorder-6 Malviflorae → Paeoniales ↓ Rhamnales

Superorder-7 Violiflorae → Violales ↓ Salvadorales

Superorder-8 Theiflorae → Theales & Droserales

Superorder-9 Primuliflorae → Ebenales & Primulales

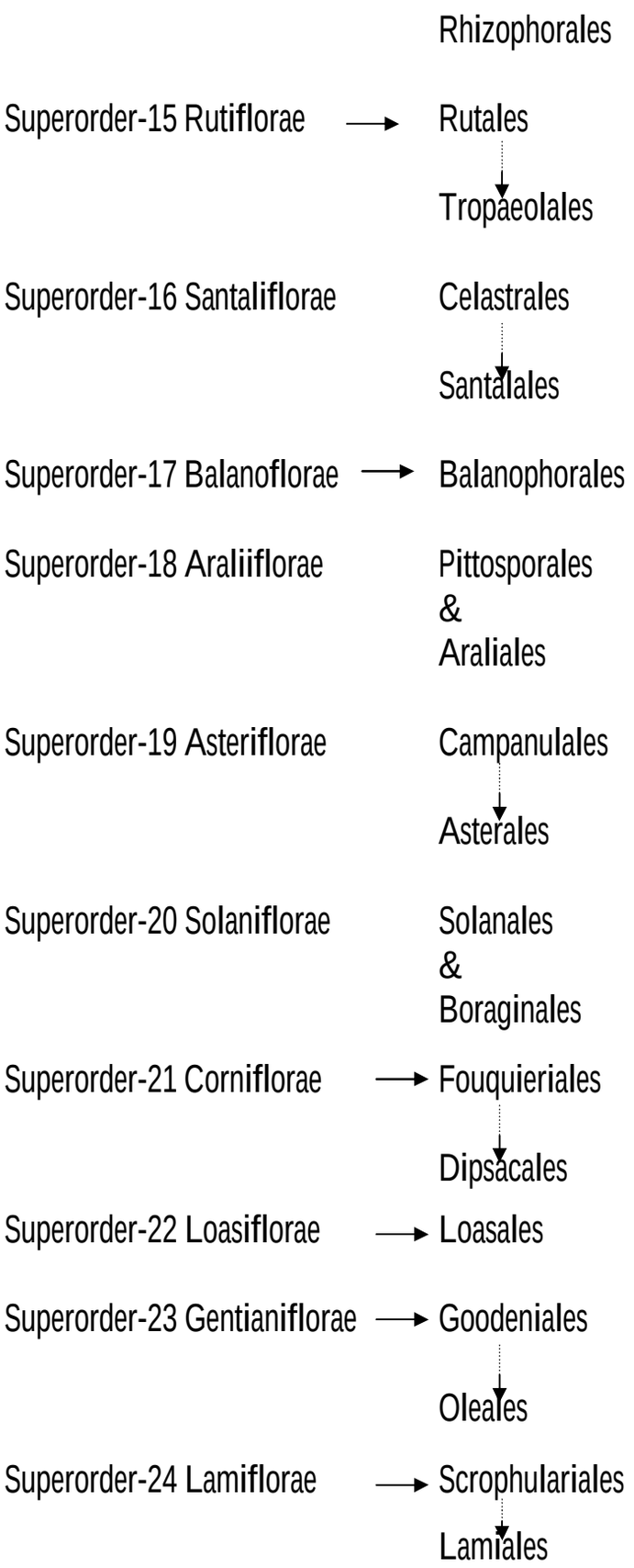
Superorder-10 Rosiflorae → Trochodendrales ↓ Malales

Superorder-11 Podostemiflorae → Podostemales

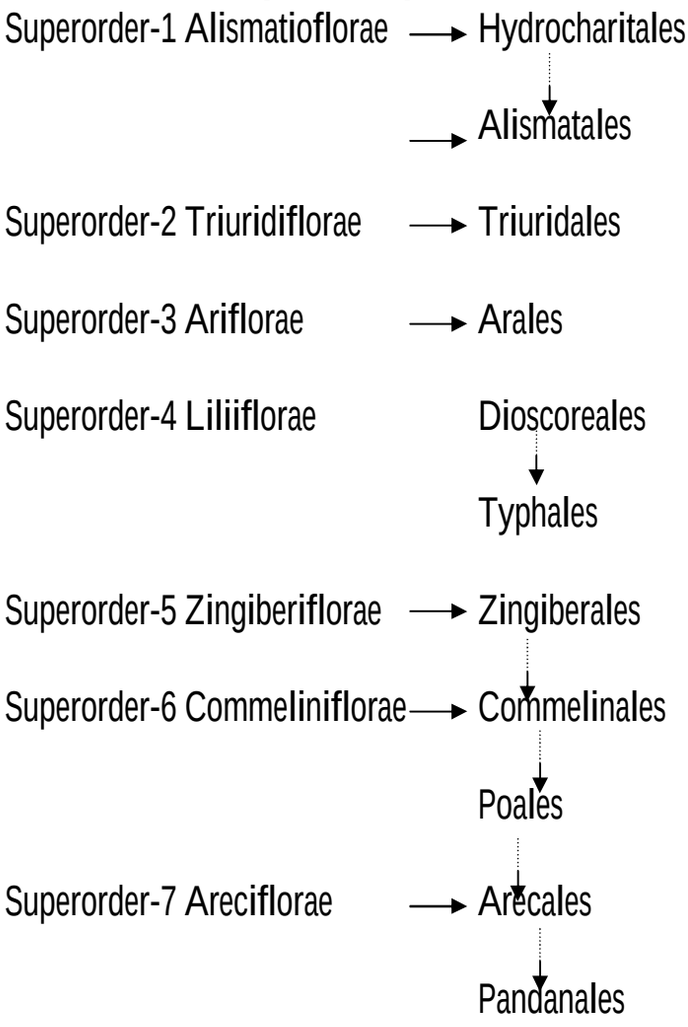
Superorder-12 Fabiflorae → Fabales

Superorder-13 Proteiflorae → Proteales

Superorder-14 Myrtiflorae → Myrtales ↓


Rhizophorales
Superorder-15 Rutiflorae
Rutales
Tropaeolales
Superorder-16 Santaliflorae
Celastrales
Santalales
Superorder-17 Balanoflorae
Balanophorales
Superorder-18 Araliiflorae
Pittosporales
&
Araliales
Superorder-19 Asteriflorae
Campanulales
Asterales
Superorder-20 Solaniflorae
Solanales
&
Boraginales
Superorder-21 Corniflorae
Fouquieriales
Dipsacales
Superorder-22 Loasiflorae
Loasales
Superorder-23 Gentianiflorae
Goodeniales
Oleales
Superorder-24 Lamiflorae
Scrophulariales
Lamiales

Subclass: Liliidae (Monocots)



Merits

1. The system is highly phylogenetic. Morphology, phytochemistry and embryology were employed for classification.
2. The angiosperms (named magnoliopsida) ranked as a class.
3. Dicotyledones placed before monocotyledones.
4. All families were placed properly.
5. The separation of Brassicaceae and Capparaceae has been justified.
6. Phylogenetic shrub presented.

Demerits

1. The system is not useful in plant identification
2. Plants are classified up to family level.
3. The classification is not useful for herbarium arrangement.
4. System includes unnatural order and super orders.

5.8 Robert F.Thorne's Classification

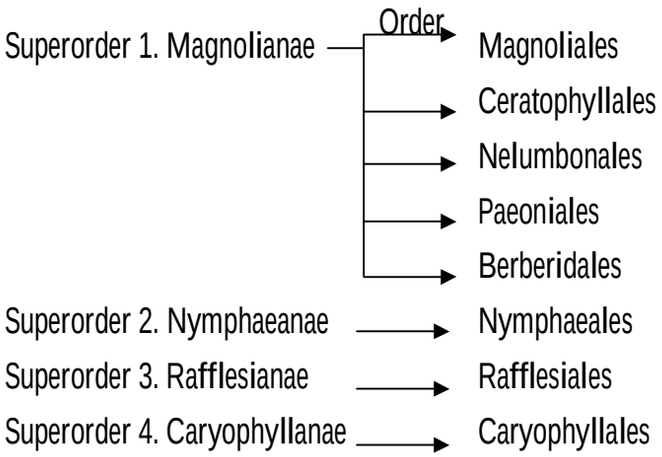
Robert F. Thorne associated with the Rancho Santa Ana Botanic Garden, California (United States) published a phylogenetic system. He proposed certain guiding principles of phylogeny and believed in monophyletic origin of Angiosperm. Thorne (1983) proposed a pure phylogenetic system on the basis of characters of comparative morphology, cytology, embryology, paleobotany, phytogeography, pollen and seed morphology ultrastructure with host-parasite relationship.

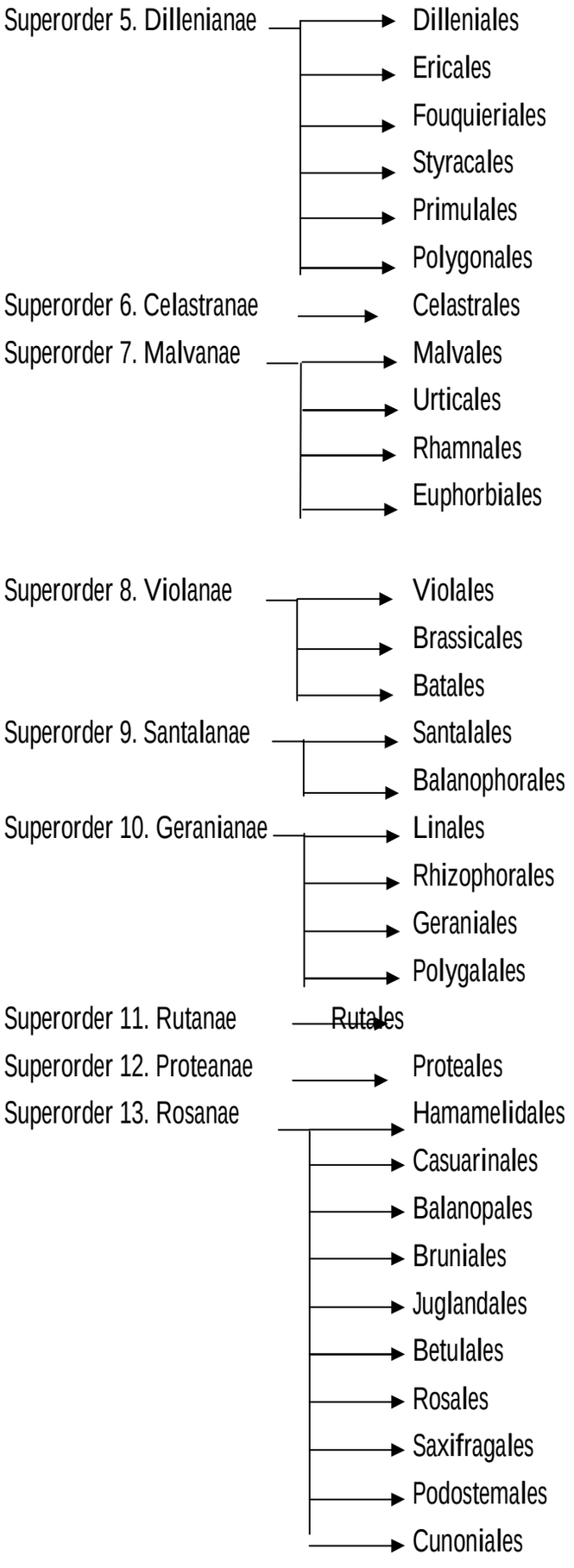
Angiospermed are treated as a class rank with an initial bifurcation into two subclasses namely dicotyledons (magnoliidae) and monocotyledons (liliidae). Related orders are placed within each subclass as super order. In super order related families are kept into suborders.

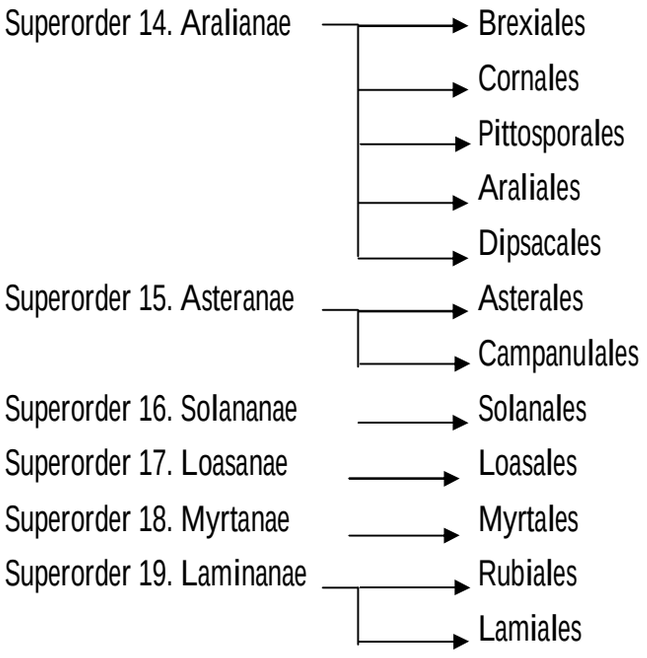
In proposed system class Annonopsida (Angiospermae) classified into 2 subclasses magnoliidae (dicotyledonae) and liliidae (monocotyledonae). Both these group were further divided into 28 super order, 54 order, 73 suborder, 350 families. System covers 2,25,490 species of 12255 genera (Table-5.7).

Table-5.7 : Robert F.Thorne's Classification

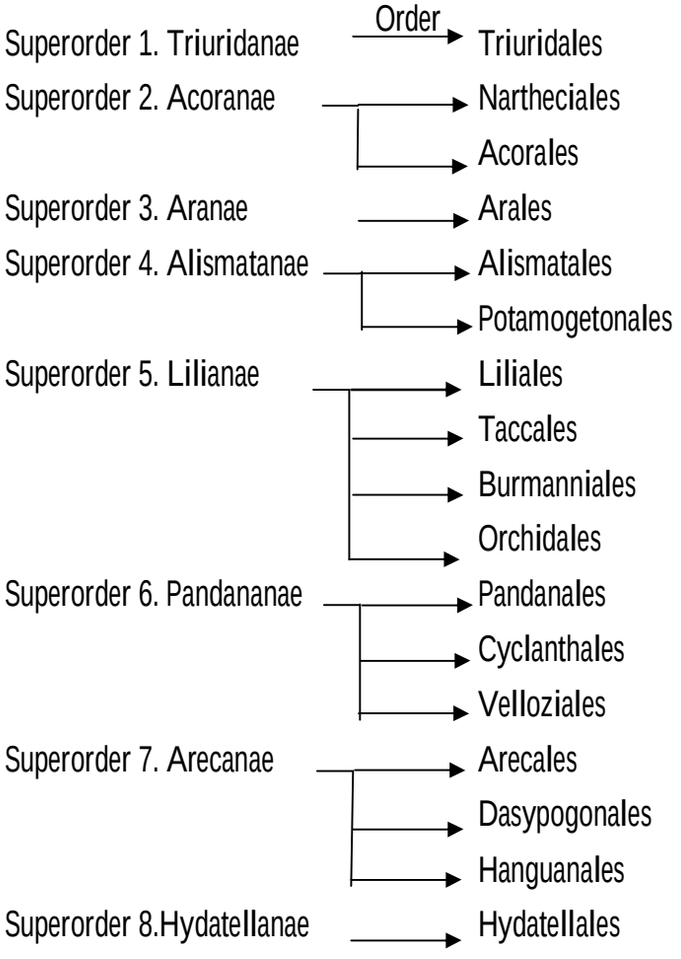
Subclass Magnoliidae (Dicots)

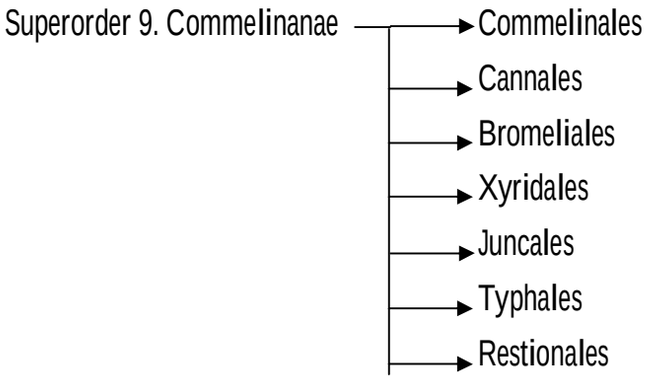



Superorder 5. Dillenianae
Dilleniales
Ericales
Fouquieriales
Styracales
Primulales
Polygonales
Superorder 6. Celastranae
Celastrales
Superorder 7. Malvanae
Malvales
Urticales
Rhamnales
Euphorbiales
Superorder 8. Violanae
Violales
Brassicales
Batales
Superorder 9. Santalanae
Santalales
Balanophorales
Superorder 10. Geranianae
Linales
Rhizophorales
Geraniales
Polygalales
Superorder 11. Rutanae
Rutales
Superorder 12. Proteanae
Proteales
Superorder 13. Rosanae
Hamamelidales
Casuarinales
Balanopales
Bruniales
Juglandales
Betulales
Rosales
Saxifragales
Podostemales
Cunoniales



Subclass Liliidae (Monocots)





Merits

1. This system is highly phylogenetic; emphases given on recent tools like molecular systematics and chemotaxonomy with other characters
2. The angiosperms are ranked as a class.
3. The system is well explained and elaborated. Larger families were divided into subfamilies as well as order in suborders.
4. Abolition of traditional groups Dicotyledones s and Monocotyledones.
5. Angiosperms were directly divided into various subclasses.
7. The subclass Magnoliidae was kept at the beginning of angiosperms including the paleoherbs.
8. Group Amentiferae has been abolished. The families of Amentiferae (a group of unrelated families) have been distributed into different orders.
9. The separation of Brassicaceae and Capparaceae has been supported by recent studies as well as morphological data.

Demerits

I. This system is highly phylogenetic but it is not very useful for plant identification.
2. For the use in herbaria; Identification keys, distribution and description of genera was not provided.
3. It is believed that angiosperms originated from some Pteridospermous members in early Cretaceous times are not accepted by several taxonomists (Mondal, 2011).
4. Affinities of five genera namely *Emblingia, Guametela, Haptanthus, Heteranthia*, and *Pteleocarpa* of angiosperms are not given.

5.9 Summary

Classification is an important biosystematics. Botanical classification deals with the grouping of plants on the basis of characters. Including characters may be morphological, anatomical, cytological etc. In natural type of classification there is only one or two morphological characters are included. Classification of Linnaeus is the best example for this. In case of natural type of classification i.e., Bentham and Hooker's system, all the morphological characters considered. Such type of classification has great practical utility for plant identification purposes. Modern systems like phylogenetic classification cover all type of characters. In other words we can say that phylogenetic classification includes phylogeny. Every system of classification has their own merits and demerits. Classification of Bentham and Hooker are followed by India and England in practical as well as in herbaria arangement.

5.10 Glossary

- **Monochlamydeous:** Having single whorl of perianth.
- **Phylogeny:** The evolutionary development of a population or of parts or organs of members of a given population.
- **Phylogeny:** The evolutionary development of a population or of parts or organs of members of a given population.
- **Taxon (pl. *taxa*):** A general term applied to any taxonomic element, population, or group irrespective of its classification level.

5.12 Self -Learning Exercise

Section A : (Very Short Answer Type Questions)

1 Who is the author of book families of flowering plants?

2 Indian herbaria follow ……..of classification.

3 Write an example of natural classification.

4 Classification of Thorne is the example of…….classification.

5 Define Phanerogames.

Section B : (Short Answer Type Questions)

1 Write characters of Ranales.

2 What are the difference between Dicotyledones and Monocotyledones?

3 Write demarits of Bentham and Hooker classification.

Section C : (Long Answer Type Questions)

1 What is Classification? Explain the difference between artificial and natural classification with suitable examples.

2 Define phylogeny. Write in detail on phylogenetic classification studied by you.

5.12 References

- Davis, P.H. and Heywood, V.H.1963. Principles of Angiosperm Taxonomy. Oliver and Boyd, London.
- Sharma, A. K and Sharma, R.2010. Taxonomy of Angiosperms. Pragati Prakashan, Meerut.
- Singh, G. 2004.Plant Systematics: Theory and Practice. Oxford and IBH Publishing Co. Pvt.Ltd. New Delhi.

Unit - 6

Phytogeography

Structure of the Unit:

6.0 Objectives

Followings are the objectives of this unit:

- to study role of geographical factors in plant taxonomy.
- introduction to Endemism including hotspots and hottest Hotspots.
- to study the concept of Phytogeography.
- to study the phytogeographical regions of India as well as vegetational diversity of Rajasthan.
- to know about Plant Exploration, Invasion and Plant Introduction.

6.1 Introduction

Taxonomy ensures nomenclature, classification and identification of plants. Classifications are mainly based on natural affinities between taxa. For the purpose of more acceptable and natural classification, it should be based on much evidence as is available for construction of groups. The excellent significance of interrelationship of plant taxa can be appreciated only after it is understood that the related plants have common ancestory as well as interrelated geographical distribution. Plant geography is a branch of botany that deals with the distribution of plants in present and the past (Good, 1974). It concerned with patterns and process in plant distribution.

Geography is the study and description of the differentiation and distribution of earthly phenomena, its physical features, climates and products living or inert. Study of the physical structure and inhabitants play an important role in plants as well as animal life, particularly its distribution. Phytogeography is the branch of botany which deals with the study of origin and distribution of plants both in time and space. Plant geography deals with the ranges (Geographical distribution) of plants on or near the surface of the earth and its waters. In other words, phytogeography is a subject that has developed with and because of progress in basic biological sciences.

Phytogeography is itself a relatively modern, although distributional studies of plants are as old as botany itself. The first real structural approach to Phytogeography was made by Humboldt (1817). "Father of Phytogeography" credit goes to Humboldt, for study of relationship between plants and environment, both latitudinally and altitudinally.

Phytogeographers are concerned with patterns and process in plant distribution. There are biotic and abiotic interactions to influence the distributions, origin, dispersal and extinction of taxa. Traditionally phytogeography concerns itself largely with floristics and its classification.

Phytogeography can be classified into two categories.

A. Descriptive or Static Phytogeography: This deals with the actual description of floristic or vegetational groups found in different parts of the world. Early plant geographer described floras and attempt to divide earth into floristic and botanical zones. It is related with distribution of plant communities. There are affecting factors i.e. inherent and geographic. The inherent factors deal with evolution of individual while geographic factors are climatic and barrier factors. Descriptive phytogeography is of major significance to taxonomy and ecological studies. Knowledge of the distributions of families, genera and species of plants adds to one's understanding of these groups.

B. Interpretive or Dynamic Phytogeography: It deals with the dynamics of migration and evolution of plants. It explains the causes of plant distribution. Good (1931) suggested the theory of tolerance in plant distribution. Mason (1936), Cain (1944) and some others have pointed out the factors involved in the distribution of plants. Dynamic phytogeography is another means of denoting interpretive phytogeography. It represents the synthesis and integration of accumulated and ascertainable facts of the related sciences of cytology, genetics, paleobotany, ecology, evolution, taxonomy, comparative morphology and phylogeny. In the words of Cain (1944) it search for causes of distributional phenomena, both modern and historical, it finds its explanations in the material which is more particularly the province of special sciences.

Interpretive phytogeography is a borderline science which depends for its materials and some of its concepts upon the more specialized sciences and it derives its distinction as a field of study from synthesis and integration. Interpretive phytogeography is second phase that follows naturally after descriptive phytogeography (Lawrence, 1967).

6.2 Phytogeographical regions of India

Plant geographer has divided the world into several phytogeographic divisions depending on the nature of soil, climate and types of plant growing in the areas. Good (1964) divided the world into six kingdoms and 37 smaller areas under them. Phytogeographic region is defined as an area of uniform climatic condition and having a distinctly recognizable type of vegetation.

Hooker marked nine distinct areas on the basis of nature of vegetation in British India. It included the subcontinent of India, Burma, Malaya, Sri Lanka and the Maldiva and Laccadiva Islands. The nine divisions are as follows:- Eastern Himalayas, North West Himalayas, Indus plain, Malabar, the Deccan, Ceylon, Maldiva islands, Burma and Malaya.

In 1937 Calder described the vegetation of India. He distinguished six phytogeographic divisions and later on the divisions are named as regions which are follows- Eastern Himalayas, Western Himalayas, Indus plain, Gangetic plain, Malabar and Deccan. Chatterjee (1962) while making a study of the endemic flora of India divided the subcontinent into eight divisions excluding Burma. These subdivisions are as follows-

1. Deccan- Mysore, Chennai and major part of Hyderabad, i.e. the states of Karnataka, Tamilnadu and Andhra Pradesh.
2. Malabar- Comparable to Malabar division of Hooker and Calder consisting of the narrow strip of land south of Sind running to the Southern most part of the country and including the Western Ghat hills.

3. Indus plain- Area on both sides of the Indus, divided into a drier tract of Sind, Baluchistan with Rajasthan and a comparatively humid tract of Punjab.
4. Gangetic plain- The area extends from East Delhi to Sundarbans of Bengal passing through Bihar, Orissa and U.P. Divided into three smaller areas as (i) comparatively drier tract of Punjab including Delhi up to Allahabad of Uttar Pradesh (ii) a comparatively humid tract including the rest of U.P., Bihar, Bengal (i.e. West Bengal and Bangladesh) and (iii) the Sundarbans.
5. Assam – The area includes valleys of Brahmputra, Jaintia, Khasi and Garo Hills, Mishmi hills, North Himalayas, Santosh river, Naga Cachar and Mizo hills, Mizoram, Meghalaya, Nagaland, Manipur, Tripura and Assam region. A good rainfall and dense vegetation is the characteristic feature of this area.
6. Eastern Himalayas- It includes Darjeeling, Sikkim, Bhutan and eastewards to Mishmi. This is divided into three zones according to altitude,viz (i) Tropical zone- stretching up to 1800 mtr, (ii) Temperate zone- stretching from 1800-3500 Mtr and (iii) Alpine zone- above 3500 Mtr.
7. Central Himalayas- This region is restricted to the Nepal. They are joined to Kumao Himalaya in west and Sikkim Himalaya in east.
8. Western Himalayas- Kumaon to Kashmir and N.W. frontier province.

6.3 Endemism

The term endemism is commonly used for regular occurrence in a specified area. Endemism means the confinement of a particular species, genus, or groups of plants and animals to a particular area. Endemism normally applied only where there is a considerable restriction in the area of distribution. Cosmopolitan in distribution is extreme opposite to the endemism.

Tropical forest shows a high degree of species richness and endemism (Orians and Groom, 2005). Endemic taxa are restricted to specific areas such as oceanic islands, peninsular regions, mountain peaks and unique geographical areas. Globally, botanically interesting areas are rich in endemics, especially islands (Richardson, 1978). Regions with high concentration of endemic species are classified as Biodiversity Hotspots by the Conservational International (CI).

Taxa occurring only in restricted geographical area are known as endemics. The term endemic in botany indicates to a species or any other taxon confined to a particular region like state, district, city etc. For the exact meaningful the area under consideration should be smaller one.

There are two basic types of endemism, namely, Neo-endemism and Paleo-endemism. Types are:-

(a) **Neo-endemism:** A taxon is evolutionarily young and not yet spread over the new area, because taxon has originated recently. Common examples are *Senecia cambrensis, Crepis fuliginosa* etc.

(b) **Paleo-endemism:** An ancient taxon which was widely distributed in the past, but now has restricted distribution due to climatic changes or human influence. *Gikgo biloba* is common example, once it was widespread in the Northern Temperate Zone but now restricted to China.

Theories of Endemism

There are two main theories to explain the endemism. The first theory believes that the last survivors of once flourishing flora which is now declining are the relics or epibiotics which are endemics. However, second theory believes that these are recent and youthful forms in course of gradual extinction. The theory is also known as Age and Area hypothesis. In support of age and area hypothesis, examples of *Primula, Impatiens, Rhododendron* etc. have been given. The first theory is supported by examples of *Sequoia semiperirens* of the central valley of California & Oregon and *S. gigantea* of Sierra Nevada which are endemic to their respective native homes were extensively distributed in Cretaceous and Tertiary periods.

According to the age and area hypothesis, area is directly proportional to its age in the scale of evolution. So, a small area of distribution shows relatively young in age e.g., *Coleus* is distributed on the summit of the dry Ritigala mountains in Sri Lanka, with two species *C. elongatus* and *C. barbatus*. *C. elongatus* is endemic and *C. barbatus* is widely distributed in tropical Asia and Africa.

Factors Responsible for Endemism

1. When species remain poor in adaptation to different types of environmental conditions.
2. These remain restricted to a particular place due to geographical barriers like sea, mountains etc.
3. Their reproductive organs produce less efficient reproductive cells like propagules, seeds etc. due to this they are unable to make distribution in far off places.

There are some others factors influence the degree of endemism of a concerned area. These include the distance from other similar areas and the period of area isolation. According the Wulff, 85% of Flora of St. Halene, 80% of Hawaii Islands and 72% of New

Zealand is endemic. Mountains have more endemic species as they are isolated e.g., 70% sp. of Himalayas are endemic.

According to Chatterjee (1962) the percentage of endemic species of Dicotyledones plants in India is more than 50. Maximum endemic plants are found in the Himalayas and South India. Indo-Gangetic plains have a very small number of endemic species. Regarding total number of endemic genera in india, there are only 49 genera are endemic to India that belongs to 22 families (Irwin and Narasimhan, 2011)

6.4 Hot Spots and Hottest Hotspots

Geographical locations, physiography, edaphic and climatic factors affects the diversity of organisms. These factors influence the flora and fauna. The Areas with high levels of species richness, threat and endemism are called as Hot spots. The bio-diversity of the particular geographical area when comes under threat it is called Hot spot or sensitive spot. Indian subcontinent has unique geographical positions, distinct physiographic, edaphic, climatic zones and gradients. This subcontinent is abodes a very rich and diverse flora and fauna with-high percentage of endemism. It is placed 10^{th} among the plant rich nations of the world and 4^{th} among the Asian countries. Among 25 most important global Hot spots of biodiversity two are present in India, namely, Eastern Himalayas and Western Ghats. Based on the uniqueness of the phytogeographical zones and pattern of endemism 25 Micro Hot spots also have been identified in India.

Hot Spots include Mediterranean basin, tropical Andes in South America, Madagascar and Indian Ocean islands, South Africa's Cape floristic province, south west Australia, large areas around Indonesia, Malaysia and the Philippines and Eastern Himalaya. There are no designated Hot spots in the area of the former Soviet Union. Though large extent of forest is found but in terms of Bio-diversity priority it is much lower than the tropical rainforests.

Myers (1988) for the first time identified 10 tropical forest hot spots with high level of plant endemism and a serious level of habitat loss. Myers (1960) added 8 more hot spots including 4-Mediterranean-type ecosystem. Again in 2000 Myers *et al.* identified more hot spots and identified 25 biodiversity hot spots. These 25 hot spots contain 44% of the world's plants as endemics and 35% of the terrestrial land vertebrates in an area of 1.4% of earth's land surface. Recently 34 Biodiversity Hot Spots have been identified.

Hot spots cover less than two percent of the planet's land surface but contain more than 50 percent of its terrestrial biological diversity. In these Hot spots the ecosystems are at greatest risk. Less than 10 percent of other original natural habitat is remaining. In the Mediterranean basin, only two percent is present. One aspect of the hot spots is that

almost 40 percent of all terrestrial plants and at least 25 percent of vertebrate species are endemic to these areas.

Global warming and extinction of endemic species from Bio-diversity hot spots was studied in detail and published in Conservation Biology. The potential impact of climate change and global warming on 23 of the world's tropical hot spots of biodiversity places are identified. It includes Southwest Australia, Caribbean, New Zealand, Madagascar, Tropical Andes and Mountains of China

For being a Hot spot a region must contain at least 1500 species of vascular plants *i.e.,* 70.5% of the world's total flora as endemics and it has to have lost at least 70% of its original habitat. The 34 Hotspots distributed in various regions of the world are listed below:

A. North and Central America

(1) California Floristic Province, (2) Caribbean Islands, (3) Madrean Pine-Oak Woodlands, (4) Mesoamerica

B. South America

(5) Atlantic forest, (6) Corrado, (7) Chilean Winter Rainfall-Valdivian forest

(8) Tumbes-Choco-Magdalena, (9) Tropica Andes

C. Europe and Central Asisa,

(10) Caucasus, (11) Irano-Anatolian,(12) Mediterranean Basin

(13) Mountains of Central Asia

D. Africa

(14) Cape floristic region, (15) Coastal forest of eastern Africa, (16) Eastern Afromontane, (17) Guinean forests of West Africa, (18) Horn of Africa, (19) Madagascar and Indian Ocean islands, (20) Maputaland-Pondoland-Albany, (21) Succulent Kareo.

E. Asia Pacific

(22) East Malasian Islands, (23) The Himalaya, (24) Indo-Burma, (25) Japan , (26) Mountains of South West China, (27) New Caledonia, (28) New Zealand, (29) Philippines, (30) Polynesia-Micronesia, (31) South West Australia, (32) Sundarland, (33) Wallacea, (34) Western Ghats and Sri Lanka.

Hottest Hotspots

For the consideration of hottest hot spot, there are five key factors viz, number of endemics and endemic species per area ratios for both plants and vertebrates and habitat loss. These factors do not carry equal weight, so they cannot be combined into a single quantitative ranking. Medagascar, Philippines, Sundaland, Brazil's Atlantic Forest, Caribbean, Indo- Burma, Western Ghats or Shri Lanka and Eastern Arc and Costal forest of Tanzania or Kenya are the eight 'hottest hotspots', which appear at least three times in the top ten listings for each factor. The leaders are Madagascar, the Philippines and Sundaland, appearing for all five factors, followed by Brazil's Atlantic Forest and the Caribbean, appearing for four. Three of these hotspots, Madagascar, the Philippines and the Caribbean, have small areas, which further highlights their importance.

Two additional hotspots, the Tropical Andes and the Mediterranean Basin, should be considered as hyper-hot candidates for conservation support in light of their exceptional totals of endemic plants: 20,000 and 13,000, respectively. The Tropical Andes is at the top for endemic vertebrates too, and the Mediterranean third after Sundaland for endemic plants, with 34% more than the fourth hotspot. But they do not rank in more than two of the five factor listings. Similarly, Mesoamerica is second for endemic vertebrates (49% more than the third highest), but it scores only tenth for endemic plants.

6.5 Plant Exploration, Invasion and Plant Introduction

Studies of plant sciences are started in remote past during Rig-Veda times. Literature explains the plant exploration in India by Portuguese plants or its parts like seed, propagules etc. are transported from one place to another or new place for particular purpose. Purpose may be for agriculture, research, food etc. Naturalization starts when favourable conditions are available. Migrated or introduced species may affect negatively on local flora and fauna. Plant exploration and introduction is one of the fundamental activities of mankind. Introduction of genes from one place of origin to a new area is important. This may be is part of state, country, continent etc.

6.5.1 Plant Exploration in India

Literature indicates that the Science of Botany in India had its beginning in the remote past during Rig-Veda times. Parasara's Vrikshayurveda reveals the science of medicine, probably before the beginning of the Christian era. Extensive work on plant exploration must have preceded the writing of this work. However, due to the complete isolation of Indian culture from development of taxonomy in Europe, after Renaissance the earlier development of taxonomy in India remained ignored (Tiagi and Kshetrapal, 1995).

In India the Portuguese were the first to Explore botany. A book on Indian flora named "*Coloquois dos simplese Drogas da India*" describing a large number of drug plants were published in1563 by Garcia d Orta. He was the first Portugese explorer. In the year 1578, work published on Indian plants "*Tractades de las Drogas*" by Acosta.

A Dutch botanist Hendrick Ven Rheede (1660-1699) was the governor of Malabar published Hortus Indicus Malabaricus in 13 volumes with 794 plates. Poradiscus Botanicus was published in 1687 by Paul Hermann. Nicholaus Burman in 1768 published Flora Indica.

John Gererd Koenig - A Danish botanist, an earlier pupil of Linnaeus arrived in India in year 1768. He built a society, called, "The United Brothers'' in order to promote the study of Indian Botany. Koenig made explorations in the Madras region."The plants of the Coast Coromandel" was published in 3 volumes (1795, 1798, and 1819) with 300 coloured plates.

'Flora Indica' an important classical on Indian plants were published by Roxburgh (1820-29) with 2533 coloured drawings. Buchanan succeeded Roxburgh (1814) as the third Superintendent of the Royal Botanical Garden, Sibpur, and Calcutta. Robert Kyd was the founder (1787) and the first Superintendent of Royal Botanical Garden, Sibpur, Calcutta.

William Roxburgh was known as The Linnaeus of India. He toured India extensively for plant exploration. His collections were published in 1825 by D. Don in "Prodramus Florae Napalensis". Griffith (1810-1845) worked in Assam Valley, Buma, Bhutan, Sikkim and Central India and Malacca with collecting 9000 species. Nathaniel Wallichi made extensive plant exploration in Nepal, Western India etc. He was the 4th Superintendent of the Royal Botanical Gardens, Calcutta from 1815-1835.

Joseph Dalton Hooker wrote "Flora of British India" on the basis of Griffih and Thompson's collections which were sent to him at Kew. Hooker along with Thomson made extensive exploration in Himalayas. Discovering many new species of *Rhododendrons* and published a monograph also in 1949.

Santapau (1958) reviewed the work of exploration in India by various workers and published "Systematic Botany of Angiosperms" by Indian Botanical Society.

6.5.2 Plant Invasion

Transportation of plants or its parts like seed, propagules etc by humans across a major geographical barrier is termed as introduction. Naturalization starts when abiotic and biotic barriers to survival are surmounted and when various barriers to regular reproduction are overcome. Invasion further requires that introduced plants produce reproductive offspring in areas distant from sites of introduction. Taxa that can cope with

the abiotic environment and biota in the general area may invade disturbed, semi natural communities. Invasion of successionally mature, undisturbed communities usually requires that the alien taxon overcomes a different category of barriers (Richardson, 2000).

Plant domestication is a co-evolutionary process by which human selection on the phenotypes of promoted, managed or cultivated plant population's results in changes in the population's genotypes that make them more useful to humans and better adapted to human intervention in the landscape.

According to Darwin (1882) human selection may be either unconscious or directed (Heiser, 1988). For plant domestication to take place there must be selection and management to cause differential reproduction and survival.

The degree of change in the targeted population can vary:

1. Wild A naturally evolved population whose genotypes and phenotypes have not been modified by human intervention.
2. Incidentally Co-Evolved (incidental domestication): A population that volunteers and adapts in a human disturbed environment, possibly undergoing genetic change, but without human selection.

   Many weeds are examples of incidentally co-evolved species, which can also enter the domestication process if humans start to select for their useful traits and start to manage or cultivate them (Harlan 1992:90).
3. Incipiently Domesticated (specialized domestication): A population that has been modified by human selection and intervention (at the very least being promoted), but whose average phenotype is still within the range of variation found in the wild population for the trait(s) subject to selection. The variance of this average is probably smaller than that of the original wild population, however, as selection has started to reduce genetic variability.
4. Semi-Domesticated: A population that is significantly modified by human selection and intervention (at the very least being managed) so that the average phenotype may diverge from the range of variation found in the wild population for the trait(s) subject to selection. The variance of this phenotypic average may be larger than that of the wild population, because the phenotypic variation now includes both types that are common in the wild population and types that are novel. Underlying genetic variability [e.g., isozyme variation (Doebley, 1989)], however, continues to decrease because fewer individuals meet the selection criteria and are therefore included in the next generation. The plants retain

sufficient ecological adaptability to survive in the wild if human intervention ceases, but the phenotypic variation selected for by humans will gradually disappear in the natural environment.

A plant population similar to Semi-Domesticated, but whose ecological adaptability has been reduced to the point that it can only survive in human-created environments, specifically in cultivated landscapes (Harlan 1992). Genetic variability is generally less than in Semi-Domesticated because of increased selection pressure and loss of ecological adaptation. If human intervention ceases, the population dies out in short order, depending upon its life history, stature and the type of vegetation that invades the abandoned area.

In clonally propagated crops, a single genotype may be the domesticate, but also is lost soon after it is abandoned (Clement, 1999)

6.5.3 Plant Introduction

Plant exploration and introduction is one of the oldest activities of mankind. Seeds and seedlings were routinely included as part of the household. Genetic diversity is the key to maintaining and improving agriculture. The invaders in early periods were the first to bring plant material like seeds; from one place to other places. It was first introduction of plants including cotton, maize, jowar, apple, papaya, cashew nut, potato, groundnut, mustard etc. British people introduced Tea, Litchi, etc. from China and Cabbage, Cauliflower, etc. from Mediterranean to India.

Plant Introduction is introduction of genotype from its place of origin to a new area. This may be in part of state of within the country (Indigenous) or from different country (Exotic). In India wheat's variety Sonara 64 and Lerma Roza were introduced from Mexico in 1960.

The introduction becomes secondary if an already introduced material is hybridization with indigenous material to get new another and improved germplasm *e.g.,* Kalyan sona (Wheat) and IR8 (Rice).

Several botanical gardens in India are conserving the rare plants introduced from outside in living condition or in the form of herbarium. National Botanical Research Institute (NBRI), Lucknow; Central Botanical Garden), Howarah; Royal Botanical Garden, Kew (England) are some of the important examples.

Various other agricultural and horticultural Institutes are playing also an important role in plant introduction. It started in 1946 after the establishment of Plant Introduction Division in IARI (New Delhi). After this, division was changed to National Bureau of Plant Genetic Resources (NBPGR) in 1976. NBPGR is presently involved in exploration,

collection, conservation, evaluation, documentation maintenance, exchange and distribution of germplasm by various research stations.

They are situated at Akola (Maharashtra), Jodhpur (Rajasthan), Kanya Kumari (Tamil nadu), Shimla (Himachal Pradesh) and Shillong (Meghalaya) covering whole country.

There is certain procedure. First of all the desired material is procured from donor country or state on exchange or purchase basis through a particular agency. In India it is carried out by NBPGR. The concerned material should be strictly certified for being free of pathogen *i.e.* disease, pest, weeds etc. Certification must be as per plant protection and quarantine laws.

After this, material is given particular number indicating exotic, indigenous, wild and catalogued. The details of material are included such as scientific name, species, variety, place of origin, place of collection, date of collection and record of the all concerned data.

(a) Getting new varieties, *e.g.,* Lerma Roja, FR 8, Sonara 64 etc.

(b) Crop Improvement.

(c) Introduction of new ornamental plants.

(d) To study inter-relationship among crop plants and their wild relatives.

Demerits of Plant Introduction

The non-seriousness at the time of plant introduction *i.e.,* mainly on the part quarantine gives rise to serious problems of disease, pests and weeds.

(a) Disease: Late blight of potato was introduced form Europe is 1883, Coffee rust was introduced from Sri Lanka in 1876 and Bunchy top of banana was introduced from Sri Lanka in 1940.

(b) Pests: Potato tuber moth was introduced from Italy in 1900. Wooly aphids of apple were introduced in North India with some exotic accessories. Fluted scale of *Citrus* was introduced from Australia through Ceylon in 1928.

(c) Weeds: *Parthenium* or congress grass is a deadly weed and was introduced with programme PL 480.

6.6 Plant Diversity and Vegetation of Rajasthan

6.6.1 Plant Diversity

Seed plants are Angiosperms and Gymnosperms. In Rajasthan, majority of plants are belonging to angiosperms. *Ephedra foliate* is only natural occurring gymnosperm. There are 1911 indigenous and naturalized plant species belonging to 780 genera under 154

families. Other than wild plant species there are approximately 274 cultivated species are found in Rajasthan.

Poaceae is the largest family followed by Fabaceae and Asteraceae. Except Poaceae (296 species) and Cyperaceae (100 species) monocotyledons are very poorly represented. The remaining 118 species belong to 29 different families. Some families represented by single genera and single species like Burmanniaceae, Cannaceae etc.

Among dicotyledones, Polypetalae is dominant having 624species belonging to 257 genera and 68 families. Gamopetalae finds second position with 276 genera and 594 species. In monochlamydae, 177 species are belonging to 69 genera and 20 families (Shetty and singh, 1993).

6.6.2 Vegetation of Rajasthan

The arid and semi arid region of India covers ca. 3,17,090 km^2 area and is mainly spread over seven states viz., Rajasthan, Gujarat, Haryana, Maharashtra, Karnataka, Andhra Pradesh and portions of Jammu & Kashmir. Climatically, the semi-arid region (annual rainfall 400 – 1000 mm) is further divisible into two zones (Rodgers and Panwar, 1988). The first zone lies in Rajasthan, Gujarat, Western Madhya Pradesh, Punjab and Haryana, immediately west of which lies the Indian Desert. The second zone extends to the rain-shadow area of Western Ghats in Maharastra, Andhra Pradesh, Karnataka and Tamil Nadu. The former zone has been recognized as Zone 4^{th} (semi arid) while the latter forms a part of Zone-6 (the Deccan Plateau). Zone 4 is further divisible into the Punjab Plains and Gujarat-Rajputana. The semiarid region in the country is generally demarcated based on 400mm isohyets. The semiarid zone in India represents 'Savannah' vegetation and extensive xerophilous grasslands rich in legumes and shrubs.

Rajasthan is the largest state of India. It is located in the north-western part of the India. The Aravalli Range forms a line across the state running roughly from Guru Peak (1722 meters), near the town of Abu (Mount Abu) in the south-west to the town of Khetri (Singhana) in the north east. About three-fifths of the state lies northwest of this line, leaving two-fifths in the southeast.

There are the two natural divisions of Rajasthan due to the Aravalli hills. The north-western part is generally arid and unproductive, although its character shifts gradually from desert in the far west and north-west to comparatively fertile and habitable land toward the east. The area includes the Thar or The Great Indian Desert.

The Aravali Range splits the state into two geographical zones (1) desert at one side and (2) forest belt on the other. Only 9.56% of the total geographical region lies under forest vegetation. The arid region of India occupies nearly 9% geographical area and covers

208,751 km² in Rajasthan. Biogeographically, it is divisible into two provinces viz., Thar Desert of Rajasthan and Katchh Desert of Gujarat. The Thar Desert comprises southern Dune systems, northern Dune systems, eastern transition or 'Bagar' located in Jalore, Jodhpur, Nagaur and Churu districts. The sand covers an irregular rocky floor, but occasionally local prominences and ridges rise above the level of the sand. Covering an area of around 62% in 12 districts of Rajasthan, this desert extends in the eastern part of Pakistan.

The average annual rainfall in the region is 275 mm and it is less than 100 mm in the extreme west. The vegetation is extremely sparse in most of the land forms and generally described as tropical thorn scrub and desertic formations. The flowering plants in Indian desert including both indigenous and naturalized plants comprised 682 species belonging to 356 genera of 87 families. Poaceae, Fabaceae, Asteraceae and Cyperaceae are the dominating families having about 111, 65, 41 and 36 species respectively (Bhandari, 1990). The flora of the Thar Desert is adapted to exploit the special microhabitats influenced by peculiar topography, geology, edaphic and climatic conditions.

A. Arid region

The arid region of India lies between 24° to 29° N latitudes and 70° to 76° E longitudes. It occupies nearly 9% of India's geographical area and covers 208,751 km² in Rajasthan alone and about 62180 km² in Gujarat (Rahmani 1997). Biogeographically, it is divisible into two provinces, namely, Thar Desert of Rajasthan and Katchh Desert of Gujarat (Rodgers & Panwar 1988). The Thar Desert comprises southern dune systems, northern dune systems, eastern transition or Bagar located in Jalore, Jodhpur, Nagaur and Churu districts. The Thar Desert is the eastern extension of the vast Persio-Arabian desert, which joins the great Sahara desert. It is about 640 km long and 160 km wide. It is covered by a depth of several meters of sands which are constantly shifted by winds blowing from the southwest (Krishnan 1982). The sand covers an irregular rocky floor, but occasionally local prominences and ridges rise above the level of the sand. Covering an area of around 62% in 12 districts of Rajasthan, this desert extends in the eastern part of Pakistan. Sand dunes are present in nearly 58% of the area (Shankarnarayan 1988). A major portion of western Rajasthan falls under Sand dunes and sandy plains. Occurrence of saline-sodic soil with pH upto 9.0 is a common feature. The soils of the desert plains are sandy loam with a significant proportion of lime within 150 cm of soil profile (Dhir and Jain 1982). Luni River forms the main drainage system during rainy season. The climate of the desert region also shows extremes with temperatures ranging from sub-zero in winter to as high as 52°C in summer. Dust storms in the desert tracts may reach a velocity of 130 km/h and may deposit 50-75 mm of dust on the ground per storm. The relative humidity varies from

25% (April) to 85% (July). Frosts are generally severe during winter, particularly in the sandy areas. Tree saplings upto 2 m in height, sometimes, succumb to such frosts. The vegetation is extremely sparse in most of the land forms and generally described as Tropical Thorn Scrub and Desertic formations.

Vegetation of Sandy Plain and Sand Dunes: It is covered by sandy plains and sand dunes with varying degree of stability. The area is characteristic for its various vegetation associations in different regions. Generally, the stabilized sandy plains and sand dunes are covered with vegetation of trees and shrubs viz., *Leptadenia pyrotechnica, Crotalaria burhia, Sericostoma pauciflorum, Calligonum polygonoides, Capparis decidua, Lycium barbatum, Zizyphus nummularia, Aerva tomentosa, Calotropis procera, Acacia jacquemontii, Acacia senegal, Prosopis cineraria, Salvadora oleoides, S. persica, Tecomella undulata and Maytenus emarginatus. Common herbs include Argemone mexicana, Tephrosia purpurea, T. falciformis, Farsetia hamiltonii, Convolvulus microphyllous, Boerhavia diffusa, Heliotropium* sp., *Indigofera linifolia, I. cordifolia, Tribulus terrestris, T. rajasthanensis* and *Echinops echinatus.* There are some typical climbers, namely, *Citrullus colosynthis, C. lanatus, Cucumis* sp., *Cocculus pendulus, Mukia maderaspatana, Momordica dioica, Coccinia grandis* and *Pergularia daemia* are present.

Regarding monocotyledones particularly member of Poaceae known as grasses *viz., Aristida funiculata, A. adscensionis, Cenchrus ciliaris, C. biflorus, C. prieurii, C. setigerus, Eragrostis* sp., *Cyperus* sp., *Dactyloctenium sindicum, D. aegyptium and Lasiurus scindicus* are common. Inter dune areas also support similar vegetation but abundance of species may vary depending upon the availability of moisture and stability of sand dunes. Sandy and unstable areas lack any vegetation but some ephemerals can be seen during the rainy seasons e.g., *Cleome viscosa, Polycarpaea corymbosa, Corchorus tridens, Triumfetta pentandra, Tribulus terrestris, Gisekia pharnaceoides, Mollugo cerviana, Mollugo nudicaulis, Pedalium murex, Sesamum indicum, Martynia annua, Amaranthus spinosus and Euphorbia* sp. *Cistanche tubulosa* a root parasite usually seen in this habitat and commonly associated with *Calligonum polygonoides, Capparis decidua and Calotropis procera.*

Gravelly pediments or rocky plains: these areas locally known as Magra. It is an important part of desert. It is located at Pokaran - Jaisalmer, Barmer, Jodhpur and Bikaner. Common species of this habitat are *Cleome vahliana, C. viscosa, C. gracilis, Fagonia indica, Rostellularia procumbens* and *Sericostoma pauciflora.* With some common prostrate plants growing in the gravelly ground are *Tribulus terrestris, Indigofera linnaei, I. linifolia, I. cordifolia, Heliotropium rariflorum, Euphorbia prostrata, E. granulata, E.*

clarkeana, Mollugo cerviana and *Mollugo nudicaulis.* Common shrubs of gravelly habitat include *Euphorbia caudicifolia, Leptadenia pyrotechnica, Capparis decidua, Calotropis procera, Zizyphus nummularia, Acacia senegal, Prosopis cineraria, Salvadora oleoides* and *Maytenus emarginatus.* Following are the example of some common grasses viz. *Dactyloctenium sindicum, Blepharis sindica, Melanocenchris jaquemontii, M. abyssinica, Oropetium thomaeum* and *Tragus roxburghii*. The rocky areas represent different geological formations ranging from shallow depressions to elevated areas and foot hills. The rocky slopes have typical species such as *Anogeissus pendula*, *Asparagus racemosus, Balanites aegyptiaca* and *Euphorbia caudicifolia*.

B. Semi Arid Region:- Some of the wildlife protected areas in this region attain very high ungulate biomass e.g., Ranthambore, Sariska, Gir, Velavadar, Nalsarovar, Jessore, National Chambal Sanctuary, Karera Sanctuary, Kuno - Palpur Sanctuary, Mount-Abu Sanctuary, Sitamata and Kumbalgarh Sanctuary. *Dicliptera abuensis, Strobilanthes hallbergii, Berberis asiatica, Ceropegia odorata, C. hirsuta, Ceropegia vincaefolia* are some of the interesting plants of this zone.

Vegetation in Semi arid area in Rajasthan:- In Rajasthan, Aravalli Hill range separates semiarid tract from the arid zone. The average annual rainfall to the east of Aravalli ranges between 525-675 mm and reaches 1000 mm at some locations. Eastern Rajasthan has rich alluvial soil that supports good forests and agricultural crops. The vegetation comprises Tropical Dry Deciduous Forests, Savannah Woodland and Tropical Thorn Forest. *Anogeissus pendula* is the dominant species with association of *Buchanania lanzan, Diospyros melanoxylon, Mitragyna parvifolia, Cassia fistula, Schrebera swietenioides, Pterocarpus marsupium, Holoptelea integrifolia, Butea monosperma and Mallotus philippensis. Boswellia serrata* occupies hill crests of *Aravallis, Terminalia arjuna* forms riparian forests. *Sterculia urens* is distributed throughout Aravalli range and *Anogeissus latifolia* is mainly found in open forest of Aravallis range in southern Rajasthan. Other locally available common species in the region are *Kydia calycina, Mangifera indica, Woodfordia fruticosa, Launea coromandelica, Cochlospermum religiosum, Flacourtia indica, Tectona grandis and species of Bauhinia, Albizia, Acacia, Ziziphus, Capparis, and Ficus* (Sharma & Tiagi 1979; Shetty & Singh 1987).

6.7 Summary

The distribution of plants in past geographical region with its evolution and explanation of its distribution in term of environment and other ecological aspect form phytogeography. Calder (1937) described the vegetation of India. He distinguished six phytogeographic divisions and later on the divisions are named as regions as follows- Eastern Himalayas,

Western Himalayas, Indus plain, Gangetic plain, Malabar and Deccan. Chatterjee (1962) divided the subcontinent into eight divisions excluding Burma.

Taxon occurring only in restricted geographical area is known as endemics. Tropical forest shows a high degree of species richness and endemism (Orians and Groom, 2005). Endemic taxa are restricted to specific areas such as oceanic islands, peninsular regions, mountain peaks and unique geographical areas. Globally botanically interesting areas are rich in endemics, especially islands (Richardson, 1978).

Hot spots cover less than two percent of the planet's land surface but contain more than 50 percent of its terrestrial biological diversity. In the Mediterranean Basin, only two percent is present. One aspect of the hot spots is that almost 40 percent of all terrestrial plants and at least 25 percent of vertebrate species are endemic to these areas. Hot spots include Mediterranean basin, tropical Andes in South America, Madagascar and Indian Ocean islands, South Africa's cape floristic province, south west Australia, large areas around Indonesia, Malaysia and the Philippines and Eastern Himalaya. Regions covering less than one percent of the world's total land area but holds the world's highest biological diversity both in terrestrial and marine ecosystems, in addition to an amazing diversity of human cultures and languages is known as hottest hot spot.

In India the Portuguese were the first to Explore botany. A book on Indian flora named "Coloquois dos simplese Drogas da India" describing a large number of drug plants were published in1563 by Garcia d Orta. William Roxburgh was known as The Linnaeus of India. Griffith (1810-1845) worked in Assam Valley, Buma, Bhutan, Sikkim and Central India and Malacca with collecting 9000 species. Joseph Dalton Hooker wrote "Flora of British India" on the basis of Griffih and Thompson's collections which were sent to him at Kew. Hooker along with Thomson made extensive exploration in Himalayas. Discovering many new species of *Rhododendrons* and published a monograph also in 1949. Santapau (1958) reviewed the work of exploration in India by various workers and published "Systematic Botany of Angiosperms" by Indian Botanical Society.

Transportation of plants or its parts like seed, propagules etc by humans across a major geographical barrier is termed as introduction. Naturalization starts when abiotic and biotic barriers to survival are surmounted and when various barriers to regular reproduction are overcome. Invasion further requires that introduced plants produce reproductive offspring in areas distant from sites of introduction. Plant exploration and introduction is one of the oldest activities of mankind. Seeds and seedlings were routinely included as part of the household. Genetic diversity is the key to maintaining and improving agriculture.

The flora of the Indian desert is an admixture of the western (65.8%) as well as the eastern (34.2%) migrated taxa while only 9.4% species are endemic. The flowering plants in Indian desert including both indigenous and naturalized plants comprises of 682 species belonging to 356 genera of 87 families. Poaceae, Fabaceae, Asteraceae and Cyperaceae are the dominating families having about 111, 65, 41 and 36 species respectively (Bhandari, 1990). The flora of the Thar Desert is adapted to exploit the special microhabitats influenced by peculiar topography, geology, edaphic and climatic conditions.

6.8 Glossary

- **Phytogeography:** The branch of botany dealing with the study of origin and distribution of plants both time and space.
- **Endemic: Confined** to a given region.
- **Flora**: Species content of plants in an area.
- **Hotspots**: Biodiversity rich places.
- **Invasive species**: An invasive species is defined as an organism (plant, animal, fungus or bacterium) that is not native and has negative effects on our economy, our environment or our health not all introduced species are invasive.
- **Scrub**: Vegetation dominated by shrubs with few all trees.
- **Vegetation**: The collective and continuous growth of plant in space.

6.9 Self learning Exercise

Section A : (Very Short Answer Type Questions)

1. Define endmism.
2. What is plant exploration?
3. What is invasion?
4. Define hot spot.
5. What is the full form of NBRI?

Section B : (Short Answer Type Questions)

1. Write the name of any two hot spot of India.
2. Define hottest hot spot and also write the name of two hottest hot spot.

3. Write any two merits and demerits of plant introduction.
4. Write the name of four plants which are found in Rajasthan desert.

Section C : (Long Answer Type Questions)

1. Write an essay on vegetation of Rajasthan.
2. Write a detail note on invasion and plant introduction.

6.10 References

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Unit-7

General Account of Families: I (Ranales, Centrospermae and Amentiferae)

Structure of the Unit:

7.0 Objectives

Following are the objectives of this unit:

- To understand systematic position of taxon (Ranales, Centrospermae and Amentiferae) under various system of classifications like natural, phylogenetic as well as APG.
- Phylogenetic relationship of taxon included families in concerned groups with their characteristic features.
- To study of various characters like morphology, embryology etc. in Ranales, Centropermae and Amentiferae.

7.1 Introduction

System of classification of angiosperms deals with plants grouping on the basis of their similarities. There are some natural groups of taxon, which treated under various categories in system of classification. Similar characters of plant group termed as natural taxon having similar origin. On the basis of characters, plant or plant group has been ranked and placed variously like Ranales at starting point. Natural and phylogenetic

classification have same purpose but places and ranked has changed accordingly.

7.2 Ranales

Ranales is a natural taxon. It is characterized by the parts typically spirally arranged (cyclic in advance member), numerous, distinct, the perianth often not differentiated into calyx and corolla. Gynoecium is apocarpous and multicarpellate (i.e., composed of many unilocular unicarpellte pistils).

Ranales have been considered as the most primitive due to the absence of vessels in large number of families like Alismataceae. Magnoliaceae, Winteraceae etc. Peculiar floral morphology, anatomy as well as binucleate pollen at the time of release from the microsporangium in several families like Magnoliaceae, Ranunculaceae, Winteraceae etc.

Benthum and Hooker (1862-83) kept this order at the starting of classification. Ranales of Benthum and Hooker comprises eight families: Ranunculaceae, Dilleniacea, Calycanthaceae, Magnoliaceae, Annonaceae, Menispermaceae, Berberidaceae and Nymphaeaceae. It is characterized by numerous floral parts, especially stamens and carpels; floral parts arranged in spiral, cyclic or hemicyclic manner with hypogynous flower.

Key to the Families of Ranales

1. Trees, ethereal oil cell present

2. Sepals 3:

3. Stipules present, endosperm not ruminate…….Magnoliaceae

3. Stipules absent, endosperm ruminate……Annonaceae

2. Sepals 5-numerous:

4. Sepals 5 ……..Dilleniaceae

4. Sepals numerous……..Calycanthaceae

1Herbs, ethereal oil cell absent:

5. Flowers unisexual, dioecious…….Menisperemaceae

5. Flowers bisexual:

6. Plants aquatic, perianth of numerous tepals…. Nymphaeceae

6. Plants terrestrial, perianth biseriate:

7. Stamens numerous, spirally arranged, carpels numerous, leaves usually compound…. Ranunculaceae

7.Stamens 4-8, in two whorl opposite the petals, carpels 2- 3, leaves simple or

compound…… Berberidaceae

Engler and Prantl (1938), have included 19 families namely Nymphaeaceae, Ceratophyllaceae, Cercidiphyllaceae, Trochodendraceae, Ranunculaceae, Lardizabalaceae, Berberidaceae, Menispermaceae, Magnoliaceae, Calycanthaceae, Lactoridaceae, Annonaceae, Eupomatiaceae, Myristicaceae, Monimiaceae, Gomortegaceae, Lauraceae, Hanandiaceae and Himantandraceae in 21st order of subclass Archichlamydeae.

Hutchinson (1959) has included seven families namely, Paeoniaceae, Helleboraceae, Ranunculaceae, Nymphaeaceae, Podophyllaceae, Ceratophyllaceae and Cabombaceae in 55th order of II phylum Herbaceae.

Ranales (Magnoliales) are considered, as the most primitive order according to Arber and Parkin (1907), Bessey (1915), Hutchinson (1959, 1969), Takhtajan (1980) and Cronquist (1988). But Engler and his associates considered that the Ranales are advanced and kept them in the middle of the Dicotyledons.

Hutchinson (1959, 1969) kept the woody members in Lignosae and herbaceous members in Herbaceae in 7 orders. Magnoliales, Annonales, Laurales, Dilleniales and Rosales under Lignosae while Ranales and Berberidales under Herbaceae.

Engler and Prantl (1897-1915) kept all these families in his order Ranales except Dilleniaceae, which was kept in the order Parietales. Cronquist (1988) kept these families in 7 orders in three subclasses. Magnoliales, Laurales, Illiciales, Nymphaeales and Ranunculales (Magnolidae), Dilleniales (Dillenidae), and Trochodendrales (Hamamelidae).

Takhtajan (1980) kept them in orders in four subclasses

Magnolidae : Magnoliales, Illiciales, Laurales, Nymphaeales, Nelumbonales

Ranunculidae : Ranunculales

Hamamelidae : Trochodendrales

Dillenidae : Dilleniales, Paeoniales

7.3 Centrospermae

Centrospermae is a taxon usually characterized by haplochlamydous and sometimes diplochlamydous flowers, basal or free central (with reference to the name of order) axile placenta (in which partition walls are dissolved), unilocular ovary, campylotropous ovule and seed with coiled or curved embryo and perisperm.

It is also called Curvembryae due to presence of curved embryo. The Centrospermae

(Caryophyllales) group represents one of the most controversial orders in the Angiosperms. For a very natural group the term Centrospermae is given by Eichler (1876).The curved campylotropous ovule frequently becomes a kidney shaped seed round the wall of which lies the embryo.

There are some other characterstic features also found like superior ovary, single whorl of perianth, P-III sub type of sieve element plastid and Betalins (Except Molluginaceae and Caryophyllaceae).

Bessey designated the order as Caryophyllales and supported its Ranalian ancestry indirectly through Rosales; in the Caryophyllales he included the families Podostemaceae and Salicaceae.

Eichler (1876) included following orders as follows-

Order-1. Oleraceae: Includes family Polygonaceae, Nyctaginaceae, Chenopodiaceae and Amaranthaceae.

Order-2. Caryophyllales including single family Caryophyllaceae.

Order-3. Oputinae which includes families Phytolaccaceae, Portulacaceae, Aizoaceae and Cactaceae.

Centrospermae order of Engler and Prantl (1897-1915) included the families Aizoaceae, Amaranthaceae, Chenopodiaceae, Nyctaginaceae, Phytolaccaceae, Cactaceae, Basellaceae, Caryophyllaceae, Cynocrambaceae and Portulacaceae. After some time Engler (1924-1936) separated the family Cactaceae and converted into order Opuntiales of single family Cactaceae.

Mabry (1976) classified the order Centrospermae or Caryophyllales on the basis of all available data from different disciplines like morphology, embryology, serology, ultra structural etc. into two sub order and 11 families.

A. Sub-order: Chenopodinae includes families like (i) Aizoaceae, (ii) Amaranthaceae, (iii) Basellaceae, (iv) Cactaceae, (v) Chenopodiaceae, (vi) Didiereaceae, (vii) Nyctaginaceae, (viii) Phytolaccaceae and (ix) Portulacaceae.

B. Sub-order: Caryophyllinae includes only Caryophyllaceae and Molluginaceae.

The families of sub order Chenopodinae contains a unique N-containing pigments the Betalins and the other two families of sub order Caryophyllinae shows Anthocyanins. There is some evidence of ultra-structural research on sieve-tube plastids (Behnke and Turner, 1971; Behnke, 1976) and pollen morphology (Nowicke; 1976; Skvarla and Nowicke, 1976) has revealed the close relation between the betalin families and the Caryophyllaceae and the Molluginaceae.

The pollen studies including exomorphic (Nowicke, 1976) and endomorphic (Skvarla and Nowicke, 1976) have related that exines with spinulose and tubuliferous or punctate surface pattern were structurally very similar between the betalin families and Caryophyllaceae and Molluginaceae and indicate a close phylogenetic relationship.

Bentham and Hooker (1862-1883) kept the above families at three different places. The families Caryophyllaceae and Portulacaceae have been kept in order Caryophyllinae under the series Thalamiflorae of sub-class Polypetalae. Cactaceae and Aizoaceae are included in order Ficoidales under the series Calyciflorae of subclass Polypetalae. Family Amaranthaceae, Nyctaginaceae, Chenopodiaceae and Phytolaccaceae have been kept in the series Curvembryae of sub-class Monochlamydae. The family Molluginaceae has been merged in the Aizoaceae as a sub-family Molluginoideae and Basellaceae in Chenopodiaceae as a subfamily.

Hutchinson (1969, 1973) includes families Phytolacaceae, Basellaceae, Chenopodiaceae and Amaranthaceae at different places within the order Chenopodiales of division Herbaceae. Molluginaceae, Caryophyllaceae, Ficoideae and Portulacaceac have been included in the order Caryophyllales, of Herbaceae. The family Nyctaginaceae and Didiercaceae are included in order Thymeleales and Sapindales respectively in division Lignosae. New order Cactales has been created for Cactaceae only under the division Lignosae.

It seems that there are 11 basic families, in spite of them some taxa of diverse origins assign to this order. Status of certain taxa with example discussed below.

The endemic Australian family Gyrostemonaceae has sometimes been included in the order Caryophyllales; sometimes it has even been included with the Phytolaccaceae (Cronquist, 1968). Palynological studies; by Eckardt (1964), Phytochemical tests, chromosome numbers and the presence of S-type sieve-element plastids in Gyrdstemonaceae (Behnke, 1916) have proved that this family should be separated from Centrospertnae. The families Polygonaceae and Plumbaginaceae have been included in the Centrospermae by various taxonomists. They differ from Centrospermae in the absence of Betalins and in having S-type sieve element plastids (Behnke, 1976).

7.4 Amentifereae

The group Amentiferae (Catkin possess scaly bract, usually flexuous spike or spike like inflorescence) are characterized by trees or shrubs, flower small, unisexual, at least the staminate in elongated catkins, Anemophilly and exalbuminous seeds. Distribution occurs mainly in the North temperate Zone. Catkin (Ament) is a scaly bracteated, usually flexuous spike or spike like inflorescence of cymules.

It is a distinctive order with the characterstics of its single family, Salicaceae. There is much evidence that the family has been derived from advanced ancestral stocks. The order Salicales through Fagales were designated by Eichler (1883) as a single order, Amentiferae. They were accepted as a phyletically homogenous group of primitive Dicots by Engler and Randle, both of whom rejected Eichlers's name and considered them to represent several orders.

Hutchinson transferred all these amentiferous orders (Plus the Urticales) to a phyletic position that treats them as these descendants of hamamelidaceous ancestors. The plants were restudied by Hjelmquist (1948) and treated by him as a natural assemblage of several orders comprising a single taxon, Amentiferae.

Plants are deciduous trees or shrubs. Leaves are simple, alternate, stipulate and deciduous. Flowers are small in size and unisexual, those of each sex in dense erect to pendulous catkins, each flower subtended by a scale (or bract). Perianth is absent or vestigial, represented by a copular disk or small nectary. Male flowers are represented by 2-30 stamens, free or connate Female flowers having 2-4 unilocular carpels, with numerous ovules. Placentation is parietal or basal with inferior ovary. Anemophilous type of pollination is found. Fruit is 2-4 valved capsule. Seeds exalbuminous covered with silky hairs arising from the funicle.

Eichler (1883) classified the order Amentiferae, incuding the following families like Salicaceae, Garryaceae, Myricaceae, Balanopsidaceae, Leitneriaceae, Juglandaceae, Batidaceae, Betulaceae and Fagaceae. They were accepted as a phyletically homogenous group of primitive dicotyledons by Engler and by Rendle both of whom rejected Eichler's name and considered them to represent several orders.

Hutchinson transferred all these Amentiferous orders (plus Urticales) to a phyletic position that treats them as descendents of Hamamelidaceous ancestors. Cronquist and Takhtajan kept these families in their subclass Hamamelidae except Salicaceae and Garryaceae which were kept in Dilleniidae and Rosidae respectively. A wide range of evidence has, presented in support of the view that these dicotyledons are neither natural nor primitive taxa. Engler considered it as the primitive most group of Dicotyledons.

In the year 1957, Benson divided group Amentiferae into 10 orders and12 families. These are (1) Salicales: Salicaceae, (2)Leitneriales:Leitneriaceae, (3)Batidales:Batidaceae, (4) Balanopsidales: Balanopsidaceae, (5) Myricales: Myricaceae, (6) Fagales: Betulaceae, Fagaceae, (7) Garryales: Garryaceae, (8) Julianales:Julianaceae, (9) Juglandales : Rhoipteleaceae, Juglandaceae, (10) Casuarinales: Casuarinaceae.

According to one hypothesis, the amentiferous plants are the most primitive. This is

because of the simplicity of their flowers, their lack of coloured petals, and their wind pollination. The stamens are also considered to be quite similar to the microsporophylls of gymnosperms. However, some botanists consider the simplicity of these flowers to be the result of reduction and specialization (Subrahmanyam, 1997).

7.5 Summary

There are some natural taxon angiosperms like Ranales, Centrospermae and Amentiferae. These groups are generally considered as controversial. There are so many attempts has been made by systematists to solve the controversy. The order Ranales is a natural taxon. Floral parts are many and spirally arrangement is a unique feature. In general the calyx and corolla are not distinguished. Both the reproductive parts of flower are numerous and free. Ranales is also named as Magnoliales by many systematists in various system of classification.

The centrospermae is characterized by the presence of basal or free central placentation in superior ovary with curved or coiled embryo. It's also known as by Curvembryae. Perianth typically biseriate, ovary superior and unilocular are the important characters. There are eleven families have been kept at different places by different systematists.

The group Amentiferae is characterized by trees or shrubs. Flowers are small, unisexual and at least the staminate in elongated catkins. Pollination is anemophillous and seeds are exalbuminous. Plants are mainly distributed mostly in the North Temperate Zone. There are approximately twelve families in this group. All families are treated variously by botanist. Among all taxon, Amentiferae is the most controversial.

7.6 Glossary

- **Albumen:** Starchy or other nutritive material accompanying the embryo; commonly used in the sense of endosperm, for the material surrounding the embryo.
- **Bract:** A leaf subtending a flower.
- **Bracteole:** A secondary bract, usually on the pedicel of a flower.
- **Carpel:** Female reproductive part of a flower consisting of ovary, style and stigma.
- **Catkin:** A scaly-bracted, usually flexuous spike or spikelike inflorescence of cymules.
- **Corolla:** The inner whorl of a biseriate diplochlamydeous perianth.
- **Corolline:** Like a corolla.

- **Deciduous:** Falling at the end of one season of growth or life, as the leaves of non-evergreen trees.
- **Habit:** General appearance of a plant eg. Herbs.
- **Habitat:** Kind of locality in which a plant grows.
- **Monochlamydous:** Having a perianth of a single series.
- **Ovule:** The body which, after fertilization, becomes the seed; the egg-containing unit of the ovary.
- **Placentation:** The arrangement of ovules within the ovary.
- **Pollination:** The transfer of pollen from the dehiscing anther to the receptive stigma.
- **Whorl:** Three or more leaves or floral leaves at one node in a circle.

7.7 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. Write the place of Ranales in Natural classification (Benthum and Hooker)?
2. What is the meaning of *Ament* in Amentiferae?
3. What is placentation?
4. Define curvembryae.
5. Define polyandrous condition.

Section B : (Short Answer Type Questions)

1. Write key characters of Ranales.
2. What are the basic characters of Centrospermae?
3. Write any two names of families belonging to the Amentiferae.

Section C : (Long Answer Type Questions)

1. What are the primitive characters? Discuss the primitive characters of Ranales.
2. Write a comparative statement of Centrospermae and Amentiferae.

7.8 References

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Unit - 8

General Account of Families: II (Tubiflorae, Helobiales and Poales)

Structure of the Unit:

8.0 Objectives

Following are the objectives of this unit:

- To calculate systematic position of taxon (tubiflorae, helobiales and poales) in different systems of classifications in course of development.
- To study interrelationship among families of Helobiales and Poales on the basis of characteristic features as well as floral characteristic in tubiflorae.
- To describe position and total number of families in system of classification starts from Bentham and Hooker to APG.

8.1 Introduction

In Angiosperm classification system, plants are grouped into taxon on the basis of characters. A group of plants having similar characters constitutes the taxon. Most of the taxa are natural, like tubiflorae. Taxon can categorised as a primitive and advance. Poales is considered an example of advanced taxon among monocotyledones. In order tubiflorae, helobiales and poales included families are placed variously by different taxonomists. This taxon is treated variously in natural as well as in phylogenetic system of classification. In

phylogenetic and APG system families are arranged according to their phylogeny.

8.2 Tubiflorae

Tubiflorae is a natural taxon. Plants are characterized by herbs with regular or irregular tetracyclic flowers, gamopetalous corolla, hypogynous and epipetalous stamens and unitegmic ovules.

Tubiflorae is kept under the 7th series of Metachlamydeae by Engler & Prantl (1884-1930). Its plants are grouped into 22 families. Further the Engler and Diels set up the order to contain eight sub-orders and 22 families, viz. Convolvulineae (Convolvulaceae, Polemoniaceae, Fouqueriaceae), Lennonineae (Lennoaceae), Boragineae (Hydrophyllaceae, Boraginaceae), Verbenineae (Verbenaceae, Labiatae), Solanineae (Nolanaceae, Solanaceae, Scrophulariaceae, Bignoniaceae, Pedaliaceae, Martyniaceae, Orobanchaceae, Gesneriaceae, Columelliaceae, Lentibulariaceae, Globulariacea), Acanthineae (Acanthaceae), Myoporineae (Myoporaceae) and Phrymineae (Phrymaceae).

Tubiflorae of Wettstein has same circumscription as given by Engler (except that the Fouquieriaceae were placed in the Parietales and the Plantaginaceae were included here). Rendle supported Engler's circumscription, but the Convolvulaceae was segregated as the Convolvulales, the Fouqueriaceae and Lennoaceae omitted and the Selaginaceae created. Lawrence treated all the families in the order, but excluded the Columelliaceae.

Hutchinson (1969) kept some of these families in Lignosae and Herbaceae within the different orders namely Bignoniales and Verbenales of Lignosae. Solanales, Personales, Boraginalesmand Lamiales are under herbaceae. Cronquist (1981, 1988) kept these families in the subclass Asteridae. Cronquist kept them in 3 orders viz., Solanales, Lamiales and Scrophulariales.

Bentham and Hooker included these families in the Series Bicarpellatae under three orders as follows-

1. **Polemoniales:** Polemoniaceae, Hydrophyllaceae, Boranginaceae, Convolvulaceae and Solanaceae.
2. **Personales:** Scrophulariaceae, Orobanchaceae, Lentibulariaceae, Columelliaceae, Gesneriaceae, Bignoniaceae, Pedaliaceae and Acanthaceae.
3. **Lamiales:** Myoporineae, Selagineae, Verbenaceae and Labiatae.

The taxon Tubiflorae possesses several illustrating lines of floral development. In the Convolvulaceae, the flowers are actinomorphic with ovary bilocular having two ovules in each loculus or tetralocular having one ovule in each loculus. It is related to the Polemoniaceae, but differs from it by the terminal style and ovary structure. The flowers

of the Polemoniaceae tend to be slightly zygomorphic, but the carpels may be two, three or five and ovules few to many in each loculus. The floral plan of the Hydrophyllaceae is similar to that of the Polemoniaceae, except that the ovary is bicarpellary and bilocular or unilocular due to the failure of the large marginal placentae to meet in the centre.

Critical studies of the morphology of the order Personales reveals that Verbenaceae and Labiatae are very closely related, similarly Acanthaceae, Bignoniaceae, Pedaliaceae and Martyniaceae are related; Scrophulariaceae, Orobanchaceae, Lentibulariaceae, Gesneriaceae bear close relationship.

Floral range of Tubiflorean families

Flowers actinomorphic hypogynous, corolla funnel shaped to semi-tubular, rotate, tetracyclic, calyx persistent, ovary bicarpellate, often with false septa. Stamens-5. predominantly twining herbs or shrubs, with milky latex often. …… Convolvulaceae

Flowers as above, calyx the same as above but consisting of almost free sepals. Ovary with false septa sometimes, carpels are oblique. Stamens-5. Predominantly herbs, rarely shrubs. No milky latex…… Solanaceae

Flowers actinomorphic bisexual, pentamerous, corolla similar to Convolvulaceae in aestivation. Stamens-5. Ovary pentacarpellary with longitudinal septa. Fruit like nutlet…….

NolanaceaeFlowers bisexual, actinomorphic or slightly zygomorphic; corolla mostly tubular, aestivation of the petals contorted. Ovary with 3- carpels, rarely with 5 or 2 carpels, mostly trilocular, rarely bi- or pentalocular, placentation axile. Predominantly herbs…….Polemoniaceae

Flowers bisexual, actinomorphic. Corolla rotated, gamo with various shaped(bell or funnel), Petals imbricate. Ovary bicarpellary, bilocular, sometimes apparently unilocular, placentation axile usually, tending to be parietal. Herbs ……. Hydrophyllaceae

Flowers bisexual, actinomorphic, rarely zygomorphic. Corolla bell shaped or tubular or funnel shaped, petals imbricate, rarely contorted. Ovary of 2 carpels, originally two celled but finally separating into 4 one ovuled portions with gynobasic style, style rarely terminal. Predominently herbs……..Boraginaceae

Flowers usually medianly zygomorphic, stamens usually 4, didynamous rarely only 2. Carpels, median; ovules numerous on thick axile placenta. Calyx persistent. Endosperm present.

…….Scrophulariaceae

Flowers zygomorphic, corolla bilabiate or personate. Stamens 4, didynamous. Parietal placentation. Endosperm present. ……..……..Orobanchaceae

Flowers zygomorphic, corolla personate. Stamens-2.Anthers monothecal. Ovules many on free central placenta. Endosperm absent. ……..Lentibulariaceae

Flowers zygomorphic, corolla bilabiate or irregular campanulate, Stamens 4, didynamous. Ovary bilocular, rarely unilocular. Axile placentation. Endosperm present or scanty…… Bignoniaceae

Corolla bilabiate. Stamens 4 or 2; anthers spurred or hairy. Pollen various shaped. Placentation axile, ovule on retinacula. Endosperm absent…….Acanthaceae

Corolla bilabiate. Calyx persistent and 5-lobed. Stamens 4, didynamous or sometims 5, rarely 2. Ovule 1 in each locule ……Verbenaceae

Corolla bilabiate. Calyx persistent, bilabiate usually, many toothed. Stamens 4 or 2, connective much developed. Style gynobasic. Ovules 4 in apparently 4-celled ovary …….. Labiatae

8.3 Helobiales

Helobieae or helobiales is a primitive taxon. It is also known as by Najadales or Fluviales. This is characterized by the aquatic or marshy habitats, often completely submerged, flowers hemicyclic to cyclic, actinomorphic, bisexual to unisexual, stamens 1 to many polyandrous, carples free and helobial endosperm.

Helobieae represents the Engler's second order of Monocotyledoneae. It includes seven families namely Potamogetonaceae, Najadaceae, Aponogetonaceae, Scheuchzeriaceae, Alismataceae, Butomaceae and Hydrocharitaceae.

Helobieae is considered as the most primitive taxon in the Monocotyledons. There are some reasons of primitiveness –

(i) Floral parts are free and disposed in many whorls.

(ii) Abortion of perianth and reduction in number of stamens of certain members of the family Najadaceae.

(iii) Number of stamens and carpels are many having transition from the spiral to whorled arrangement.

(iv) Occurrence of wide lamina like filaments with basifixed anthers in the Juncaginaceae and Potamogetonaceae.

(v) Presence of laminal placentation in some members of family Alismataceae and

Hydrocharitaceae.

(vi) Incomplete closure of carpels in certain members of the family Hydrocharitaceae.

Cronquist (1968) created three orders and twelve families for Helobial plants:

1. Alismatales:-Butomaceae, Limnocharitaceae, Alismataceae.
2. Hydrocharitales:- Hydrocharitaceae.
3. Najadales:- Aponogetonaceae, Scheuchzeriaceae, Juncaginaceae, Najadaceae, Potamogetonaceae, Ruppiaceae, Zannichelliaceae, Zosteraceae.

Stebbins (1974) supported Cronquist (1968) system but Takhtajan (1969) included two extra families namely Posidoniaceae and Cymodaceae under the Najadales.

Dahlgren (1975) removed the Butomaceae from Alismatales and placed in the Hydrocharitales along with the Hydrocharitaceae and Aponogetonaceae; here the Najadales was meant to accommodate the Najadaceae and seven families viz., Scheuchzeriaceae, Juncaginaceae, Potamogetonaceae, Zosteraceae, Posidoniaceae, Zannichelliaceae and Cymodoceaceae.

Thorne (1976) accepted three orders and ten families:

1. Alismatales:-Butomaceae, Alismataceae and Hydrocharitaceae.
2. Zosterales:-Aponogetonaceae, Scheuchzeriaceae (including Juncaginaceae), Potamogetonaceae, Posidoniaceae, Zannichelliaceae, Zosteraceae.
3. Najadales:- Najadaceae.

Hutchinson (1969) placed them in six orders - Butomales, Alismatales, Juncaginales, Aponogetonales, Potamogetonales and Najadales. Takhtajan (1980) included Alismataceae, Butomaceae and Hydrocharitaceae in the order Alismatales while the other families have been segregated into a separate order Najadales, while Cronquist kept them in three orders-Alismatales, Hydrocharitales and Najadales, all in subclass Alismatidae.

Dahlgren and Clifford (1982) proposed the Hydrocharitales for the Butomaceae, Aponogetonaceae, Hydrocharitaceae (including Thalassiaceae and Halophilaceae), the Alismatales for the Alismataceae (including Limnocharitaceae) and the Zosterales for the Scheuchzeriaceae, Juncaginaceae, Najadaceae, Potamogetonaceae (including Ruppiaceae), Zosteraceae, Posidoniaceae, Cymodoceaceae and Zannichelliaceae

In the Helobiae both Potamogetonaceae and Najadaceae are very advanced in terms of evolution. According to Engler and Rendle family Najadaceae is the earliest family in terms of evolution. In the other families of group, flowers are confirm to the structure that were derived from the regular trimerous types having the formula of P 3+3, A 3+3, G 3+3.

There is trend of multiplication of the members of the androecium and gynoecium either by doubling or by increasing the number of whorls.

The order is important from the consideration of some taxonomists who believe that other orders of Monocotyledons were derived from it. There are some supporting features like aquatic habit representing submerged to semi-submerged condition, large embryo with large hypocotyl for storing food, absence of endosperm in seeds and free carpels (apocarpy).

Taxon Helobieae is considered most primitive ones by most of the systematist. Wettstein has also given first place to the order Helobieae. Hutchinson splitted Helobieae into several independent orders such as Butomales, Alismatales, Juncaginales, Aponogetonales, Potamogetonales and Najadales.

In views of Wettstein and Hutchinson the primitive monocotyledonous order Helobieae was derived from early Ranales (Dicotyledons). So Helobieae forms the possible connecting link between Dicotyledons and Monocotyledons.

There is possible line of evolution, indicating that Butomoideae, Alismaceae and other monocotyledonous families had their origin from Ranales (Mitra 1974).

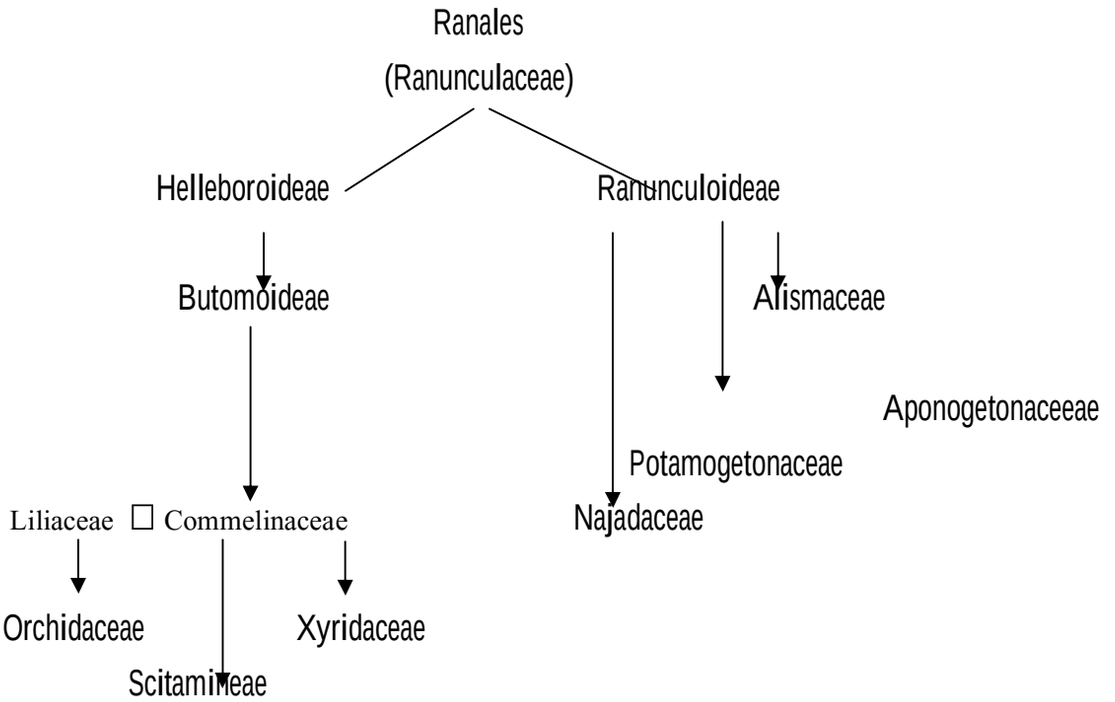

Fig. 8.1 : Possible origin from Ranales

Ranalian origin of Monocotyledons is based on the following:

1. The single solitary bracteole characteristic of Monocotyledons is posterior, it springs from flower stalk, this bracteole may be median posterior to the first

perianth leaf if the arrangement is spiral, or this bracteole is median lateral to odd perianth (first perianth leaf); this type of bracteole is rare in Dicotyledons but occurs in some Ranunculaceae, Nymphaeaceae; so some authors give importance to Helobieae as direct descendant of early Dicotyledon as well as direct ancestor of other Monocotyledons.

2. Ranunculaceae and Alismaceae have apocarpous pistil (carpels free) in common. Cabomboideae (tribe of Nymphaeaceae, Dicot) and Butomoideae (of Alismaceae) have similar placentation and trimery of flowers with apocarpy. Both Ceratophyllaceae (Dicot, Ranales) and Najadaceae (Helobiean member) have similar habits and common floral structures to some extent.

3. The families Alismaceae and Hydrocharitaceae of the order Helobieae, specially the subfamily Butomoideae of Alismaceae were all derived from Ranunculaceae. The subfamily Butomoideae of Alismaceae represents the most primitive type in the Monocotyledons. The morphology of carpels, placentation resemble that of Cabomboideae of Nympheaceae; regarding the structure of seeds, embryo, etc. it bears similarity with Ranunculaceae. Butomoideae and Cabomboideae are very similar in trimerous flower and aquatic habit also.

The origin of Potamogetonaceae from Alismaceae is indicated in the similar floral construction of *Wiesneria* (Alismaceae) and *Potamogeton.*

8.4 Poales

The grasses and sedges; flowers very small and usually termed florets, arranged in spikelets, without a conventional perianth (sometimes represented by bristles or reduced to minute scales), in axil of dry chaffy bracts, stamens typically 3 or sometimes 6 or rarely more, the ovary superior and 2-3 carpellate; fruit a caryopsis or achene, and with endosperm present and abundant.

The plants placed in this order have been considered by most botanists to compose a natural assemblage, and one with which some phylogenists in the past included other families (i.e. Juncaceae, Centrolepdaceae, Thurniaceae and Restionaceae). The taxonomic validity of placing both the grasses and the sedges into 1 order was rejected by Hutchinson(1934,1948), who segregated them into separate order (Graminales and Cyperales) and placed into a third and related order (Juncales) the other 4 families cited above. The results of morphological studies by Belk(1939) and by Blaser (1940,1944) supported the segregation of the grasses and sedge into 2 separate orders, but failed to provide substantianting evidence to justify the transfer into this general association of the plant compromising the Juncales of Hutchinson.

Monocotyledons have faced major changes over recent decades, especially as the result of phylogenetic advances (Chase *et al.* 2000). Relations among orders have changed while new insights and data on Monocot evolution are continuously updated. It represents a major part of Angiosperm and Monocot diversity. The families encompass approximately 20,000 species which comprises about 7% of the Angiosperms and 33% of the Monocots.

Gramineae or Poaceae is a large family comprising about 850 genera and more than 11,000 species. Members of this family commonly called as grasses and ranked as largest family among monocotyledons. They are cosmopolitan in distribution occurring from the Arctic to the Antarctic, from sea level to high elevations, forming the dominant vegetation of Savannas, Prairies and Steppes, usually preferring open type of habitats but frequently forming a part of forest undergrowth. They occur in dry situations on rocks, in deserts and in wet situation (Tiagi and Kshetrapal, 1995).

Regarding systematic position of Poales; initially they did not got name Poales. Actually history of classification starts with natural type of classification. In this connection Benthum and Hooker (1862-1883) has been kept the family Gramineae under the series Glumaceae of Monocotyledons with family Cyperaceae. Engler and Prantl's (1897-1915) system, both the families viz. Gramineae and Cyperaceae has been included in order Glumiflorae.

Various names have been employed for this order, that of Glumiflorae being the oldest. Hutchinson (Evolution and Phylogeny of flowering plants, 1969) gave ranke division to Glumiflorae; consisting of three orders named Juncales, Cyperales and Graminales. Takhtajan created the individual order Poales and Cyperales. Cronquist (1988) in the evolution and classification of flowering plants kept under the sub class Commelinidae and ranked order Cyperales including Poaceae and Cyperaceae.

The Poales is considered as an advance taxon of monocoptyledons. It is a large order including families commonly known as grasses, bromeliads, and sedges. Sixteen plant families are currently recognized by botanists under Poales. The order probably originated in late Cretaceous period about 66 million years ago (Bremer, 2002).

The earlier APG system (1998) adopted the same placement, although it used the spelling "commelinoids", and used the following circumscription (i.e., it did not include the plants in families Bromeliaceae and Mayacaceae in the order); including the families Anarthriaceae, Centrolepidaceae, Cyperaceae, Ecdeiocoleaceae, Eriocaulaceae, Flagellariaceae, Hydatellaceae (now transferred out of the monocots; recently discovered to be an 'early-diverging' lineage of flowering plants.), Joinvilleaceae, Juncaceae, Poaceae, Prioniaceae, Restionaceae, Sparganiaceae (now included in family Typhaceae), Thurniaceae, Typhaceae, and Xyridaceae.

The order represents about one third of the monocots, with ca. 20,000 species distributed in 16 families (APG III 2009). The APG III system (2009) accepts the order and places it in a clade called commelinids, in the monocots. It uses this circumscription. order Poales include the following families viz., Anarthriaceae, Bromeliaceae, Centrolepidaceae, Cyperaceae, Ecdeiocoleaceae, Eriocaulaceae, Flagellariaceae, Joinvilleaceae, Juncaceae, Mayacaceae, Poaceae, Rapateaceae, Restionaceae, Thurniaceae, Typhaceae, Xyridaceae.

8.5 Summary

There are some special groups of angiosperms like Tubiflorae, Helobiales and Poales. Tubiflorae is a large group of herbaceous plants with gamopetalous corolla. Regular or irregular tetracyclic flowers, hypogynous and epipetalous stamens and unitegmic ovules are the main characters. Generally it is composed of approximately 23 families. In general tubiflorean families reveals a range in floral characteristics. Various attempts have been made by systematists to assign its phylogenetic position.

Helobieae is considered as the most primitive taxon in the Monocotyledons. Helobieae or helobiales is natural group characterized by the aquatic or marshy habitats,often completely submerged, flowers hemicyclic to cyclic, actinomorphic, bisexual to unisexual, stamens 1 to many, polyandrous, carples free and helobial endosperm. Helobieae represents the Engler's second order of Monocotyledoneae. It includes seven families viz. Potamogetonaceae, Najadaceae, Aponogetonaceae, Scheuchzeriaceae, Alismataceae, Butomaceae and Hydrocharitaceae.

There are some features which reveal its primitiveness are (i). Floral parts are free and disposed in many whorls. (ii).Abortion of perianth and reduction in number of stamens of certain members of the family Najadaceae. (iii). Number of stamens and carpels are many having transition from the spiral to whorled arrangement. (iv). Occurrence of wide lamina like filaments with basifixed anthers in the Juncaginaceae and Potamogetonaceae. (v). Presence of laminal placentation in some members of family Alismataceae and Hydrocharitaceae. (vi). Incomplete closure of carpels in certain members of the family Hydrocharitaceae.

Members of this family Poaceae or Gramineae are commonly called as grasses and ranked as largest family among monocotyledons. They are cosmopolitan in distribution occurring from the Arctic to the Antarctic, from sea level to high elevations, forming the dominant vegetation of Savannas, Prairies and Steppes, usually preferring open type of habitats but frequently forming a part of forest undergrowth. They occur in dry situations on rocks, in deserts and in wet situation (Tiagi and Kshetrapal, 1995).

This order was named by Takhtajan and Thorne as Poales. In this connection Benthum and Hooker (1862-1883) has been kept the family Gramineae under the series Glumaceae of Monocotyledons with family Cyperaceae. Engler and Prantl's (1897-1915) system, both the families viz. Gramineae and Cyperaceae has been included in order Glumiflorae. Hutchinson (Evolution and Phylogeny of flowering plants, 1969) gave ranke division to Glumiflorae; consisting of three orders named Juncales, Cyperales and Graminales. Takhtajan created the individual order Poales and Cyperales. Cronquist (1988) in the evolution and classification of flowering plants kept under the sub class Commelinidae and ranked order Cyperales including Poaceae and Cyperaceae. Poales is a large order including families commonly known as grasses, bromeliads and sedges. Sixteen plant families are currently recognized (Bremer, 2002).

8.6 Glossary

- **Caryopsis:** A fruit where seed and pericarp are fused.
- **Didynamous:** Androecium having four stamens in two pairs of two different lengths.
- **Gamopetalous:** with a corolla of one piece, the petals united at least at the base where the corolla removable as a single structures.
- **Locule (Loculus):** Compartment or cell of an ovary, anther or fruit.
- **Ovule:** The body which, after fertilization, becomes the seed; the egg-containing unit of the ovary.
- **Phyllogeny:** The evolutionarydevelopment of a population or of part or organs of members of given population.
- **Spike:** A usually unbranched, elongated, simple, indeterminate inflorescence whose flowers are sessile.
- **Spikelet:** A secondary spike; 1 part of a compound inflorescence which of itself is spicate; the floral unit, or ultimate cluster, of a grass inflorescence composed of flowers and their subtending bracts.

8.7 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1 Write one characteristic features of Helobiae.

2 What is the meaning of Tubiflorae?

3 What is polypetalous corolla?

4 Define Spikelet.

5 Define Caryopsis.

Section B : (Short Answer Type Questions)

1 Write key characters of Tubiflorae.

2 What are the basic characters of Helobiales?

3 Write any two names of families belonging to the Glumiflorae.

Section C : (Long Answer Type Questions)

1 What are the advance characters? Discuss the advance characters of Poales.

2 Write a comparative note on Tubiflorae and Helobiales.

8.8 References

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Unit-9

Introduction to Plant Development and Seed Germination & Growth

Structure of the Unit:

9.0 Objectives

After studying this unit you will be understand the unique features of plant development and the process of seed germination in terms of-

- Stages of seed germination
- Metabolism of Nucleic Acid, Proteins and Mobilization of Food Reserves
- Tropisms and Hormonal Control of Seed Germination

9.1 Introduction

By studying this unit students will learn about the unique features of plant development which differentiate it from animal development. Besides, they will also know about the germination process and effects of environment factors and its control by various hormones.

Seed germination is the first process with which a plant begins its life. It occurs only when the environment conditions become favorable to support the life and growth of plant. Various developments take place in a sequential manner when a seed germinates. Reserved food materials are mobilized and consumed until self-dependent seedling stage is established. Various plant growth hormones cross talk with each other and control seed germination. The detailed account of these facts has been included in this unit.

9.2 Unique features of Plant Development

Unique features of plant development which also differentiate it from animal development are as follows:

- Occurrence of genetically active haploid (gametophyte) and diploid (sporophyte) phases of the life
- Lack of a germline
- Occurrence of double fertilization in the course of sexual reproduction to produce both an embryo and a nutritive tissue-Endosperm
- Plant cells are non-motile. The rate of cell division and direction of elongation determine plant shape and forms. Although plants have 3 basic tissue-systems (dermis, ground tissue and vascular tissue), they don't depend on on gastrulation to form these layer structures.
- Plants grow continuously. Fresh organs develop during the course of their entire life cycle by groups of actively dividing cells known as meristem.

- Plants have marvelous developmental plasticity. Lost plant parts and even entire plants can be regenerated from only one cell. Furthermore, environmental factors significantly affect overall plant form.

Common features between plant and animal development:

- Fertilization of a 1N egg by a 1N sperm.
- Cell division and growth make the shape of the embryo.
- Molecular mechanisms responsible for generation of diverse cell types.

In plants, natural selection can take place on the haploid genome because of the presence of active gametophytic phase, deleterious alleles are removed from the population which are otherwise get masked in diploid organisms.

A key difference between plants and animals is regarding the germline. In animals, germ cells are established early during embryo development and they continue as a distinct stem-cell population all through the animal life; only the derivatives of the germ-cell undergo meiosis to produce gametes. While in plants, the stem cell population (shoot apical meristem) which produce shoot (leaves and stem) during vegetative growth switches to produce flowers during reproduction. Developmental and environmental signals trigger the switch for flowering. Conversely, the nonexistence of a germline and the late switch of plant shoot from somatic lineages into floral development allow genetic mutations to accumulate and be represented in the gametes. Consequently, plants produce greater genetic diversity, however, through haploid selection, impose strict selection than animals.

In plants, reproduction is inherently more elastic. There are many cases of asexual plant reproduction, ranging from the regeneration of tiny plantlets on leaf margins to apomixis in which seeds can develop in flowers without fertilization. Additional developmental flexibility is observed in vitro, as diploid somatic cells and haploid pollens can form embryos in tissue culture. Given the present-day domination of flowering plants on earth, the double fertilization event appears to have been an evolutionary innovation of great importance.

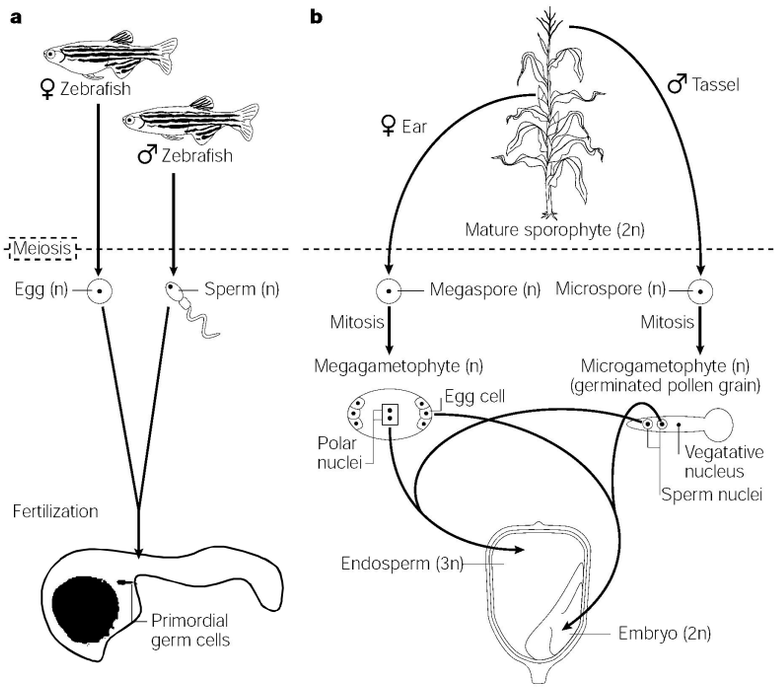

Fig. 9.1 : Comparison of Plant and Animal Life Cycles (Source: Walbot and Evans 2003)

a) In Animal life cycle, gametes are produced directly through meiosis and gene expression is restricted to diploid cells. In the embryo, the germline is set aside early (primordial germ cells) and these are the only cells that are competent to undergo meiosis in the adult.

b) Typical flowering plant life cycle (maize), in which meiosis, followed by mitotic divisions, produces two types of haploid organism that are genetically active — the female mega-gametophyte and the male microgametophyte (pollen grain). Mitotic division in the microgametophyte results in a pair of sister cells that differentiate into sperm. In the megagametophyte, a haploid egg differentiates and is fertilized by one sperm cell to produce an embryo, and two haploid nuclei fuse to form a diploid central cell that is fertilized by another sperm cell to become triploid endosperm. The endosperm is a terminal nutritive tissue that contributes to normal embryo development in flowering plants.

9.3 Seed Germination

Imbibition of water by seeds triggers a series of metabolic procedures that finally results in the emergence of the radicle, thus bringing to an end of germination. Afterwards, the stored reserve foods are mobilized to support early seedling growth. Seed germination involves following steps:

9.3.1 Water Uptake

Germination includes events that start with the uptake of water (imbibition) by the quiescent dry seed and ends with the emergence of the embryonic axis, typically the radicle. Mature dry seed uptakes water in 3 phases (fig. 9.2). The very low water potential of the dry matrices of the seed (cell walls and storage components) drives the first influx (Phase I, imbibition). After rapid hydration of seed matrices, a plateau (Phase II) is reached. Water is again taken up only after completion of germination, as the embryo develops into a seedling (Phase III). This kinetics of water uptake is affected by the seed structure, for example in cereals water does not enter all parts equally, but is directed preferentially towards the embryo or its radicle. Temporary structural perturbations occur to membranes during the phase I of water upkake. It results in a rapid leakage of ions and low-molecular weight metabolites from the seed. Lectins, proteinase inhibitors and seed-surface proteins working as protective agents against bacterial or insect invasion, are resealed by some seeds.

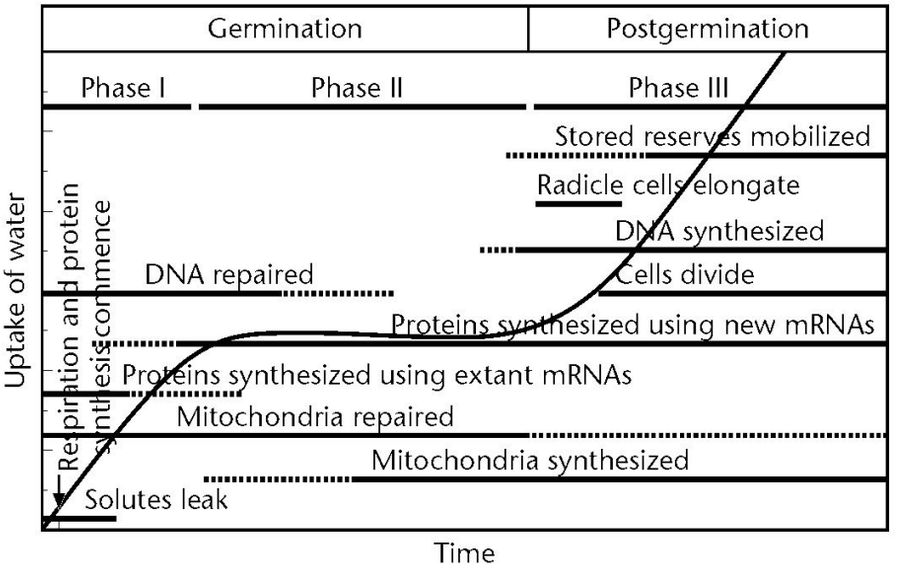

Fig. 9.2: The time course of events associated with seed germination and subsequent post germinative seedling growth (Source: Bewley, 2001).

The time required for the events to be completed varies from several hours to many weeks, depending upon inherent genetic factors and the prevailing germination conditions, particularly temperature and water availability.

9.3.2 Onset of Metabolism

Metabolic activities are rapidly resumed Imbibed seeds. The dry seed contains all the cellular structures and enzymatic machinery (that survived the desiccation phase of seed maturation) essential for the commencing the metabolism. Respiration is one of the first activities to resume. It can be observed within minutes after imbibition (Fig. 9.2). Respiratory pathways- the glycolytic and oxidative pentose phosphate pathways and Krebs' cycle restart during Phase I. Even though tissues in dry seed contain poorly differentiated mitochondria (survived after maturation), they contain enough Krebs' cycle enzymes and terminal oxidases to yield sufficient amounts of ATP to sustenance metabolism for several hours after imbibition. Afterwards two distinct patterns of mitochondrial development are observed: 1) in starch-storing seeds, pre existing mitochondria are repaired and activated. 2) Whereas in oil storing seeds characteristic mitochondrial biogenesis takes place, involving genomes of both the mitochondria and nucleus.

Like respiratory machinery, all the components of protein synthesis are present within the cells of mature dry seeds. Within minutes of rehydration protein-synthesizing factors and ribosomes recruited onto mRNAs stored in the dry seed. New ribosomes are synthesized and used for translation. Fresh mRNA molecules (some identical to old ones and other new ones) progressively replace previous pool of transcripts. The majority of mRNAs encode proteins that are required for normal cellular metabolism. As germination precedes, new proteins such as protein kinases, ATPases are also synthesized.

9.3.3 Completion of Germination

Radicle protrusion from the seed terminates germination and marks the commencement of seedling growth. This happens as a result of cell extension, which may be accompanied by cell division. DNA gets damaged during desiccation process of seed maturation; therefore its repairing takes place soon after the start of imbibition (Fig. 9.2). Mitochondrial DNA is also synthesized. New DNA synthesis occurs in actively dividing cells after radicle protrusion. Tubulin protein, a constituent of microtubule required for spindle formation in cell division, is also synthesized in them. Emergence of the radicle by cell extension is a turgor dependent procedure. Cell walls of radicle become more stretchable by the action of expansins which break the hydrogen bonds connecting cell wall polymers.

9.4 Mobilization of Food Reserves

Food reserves are mobilized from the storage organs after the completion of germination to support the seedling growth until it becomes autotrophic by starting photosynthesis. Cotyledons and endosperm are the seed storage organs in dicot and monocot plants

respectively. High-molecular-weight food reserves stored within them are changed into easily movable low-molecular-weight metabolites. Thereafter they are directed towards the growing regions (Fig. 9.3). The hydrolysis and utilization of these reserves generally occurs parallel. Carbohydrates, oils and proteins are main food reserves. Seeds contain considerable amounts of two or more of these major reserves.

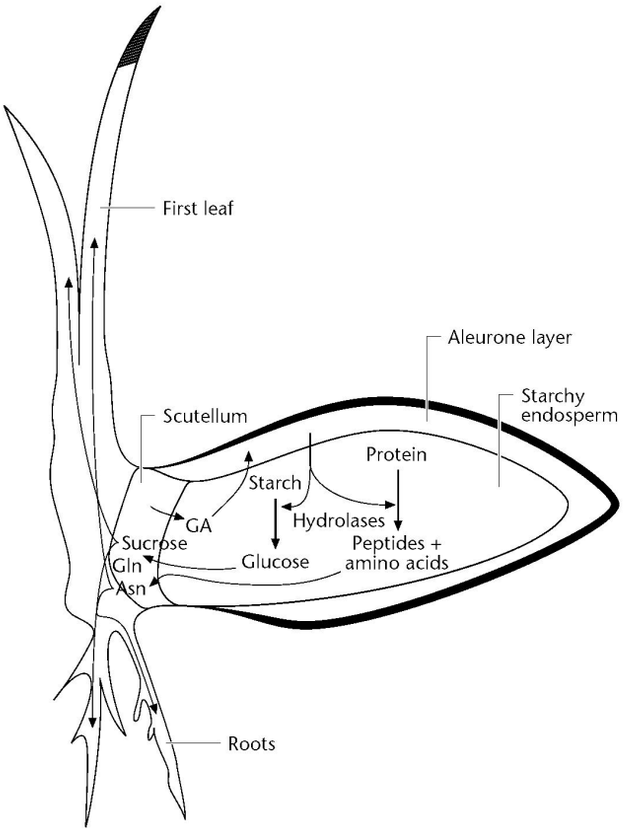

Fig. 9.3: The major events taking place during mobilization in a young cereal (barley) seedling following germination (Source: Bewley, 2001)

The plant hormone gibberellic acid (GA) is released from the scutellum and diffuses to the living cells of the aleurone layer where it promotes the synthesis of several hydrolytic enzymes. These are secreted into the nonliving cells of the starchy endosperm where the starch and protein reserves are stored. α-Amylase and maltase are key enzymes in the degradation of starch to glucose, and the proteinases hydrolyse proteins to short peptides and amino acids. The hydrolytic products are absorbed by the scutellum, which is part of the growing embryo. There the glucose is converted to sucrose, and the products of protein mobilization to the amino acids glutamine (Gln) and asparagine (Asn). These are transported throughout the seedling via the vascular system as a supply of nutrients to support growth.

9.4.1 Carbohydrates

Starch is found as major reserve carbohydrate in the endosperm of cereals, e.g. rice, wheat and maize. Starch contains amylose and amylopectin which are first hydrolysed by α-amylase. This enzyme is arbitrarily disruptions the (1-4) glycosidic bonds between the glucose (Glc) residues. β-amylase, another enzyme involved in starch degradation, splits off consecutive disaccharide maltose residues (Glc-Glc) from the nonreducing terminal of the large oligomers released by previous α-amylase action. These enzymes produce glucose and maltose residues from amylaose. Apart from these two residues, highly branched short chains of glucose, known as limit dextrins are also derived from amylopectin. A debranching enzyme- limit dextrinase hydrolyses the (1-6) branch points and releases the short chains, which are further cleaved by the amylases. Maltose is hydrolyzed into glucose units by maltase (Fig. 9.3). In monocots, all the above mentioned enzymes except β-amylase are synthesized in the embryo (scutellum) and surrounding aleurone layer. Thereafter, they are released into the storage cells of the endosperm to breakdown starch following germination. β-amylase already exists in the storage cells and is activated when required. Finally glucose is directed into the growing embryo via the scutellum and is converted to sucrose for transport to, and consumed by, the growing regions of the seedling. In certain dicot seeds, starch phosphorylase is found in place of α-amylase. It adds a phosphate group across the (1 - 4) linkage between the second last and last glucose residues at the nonreducing end of the polysaccharide chain. The other enzymes are still essential to accomplish full hydrolysis of the starch.

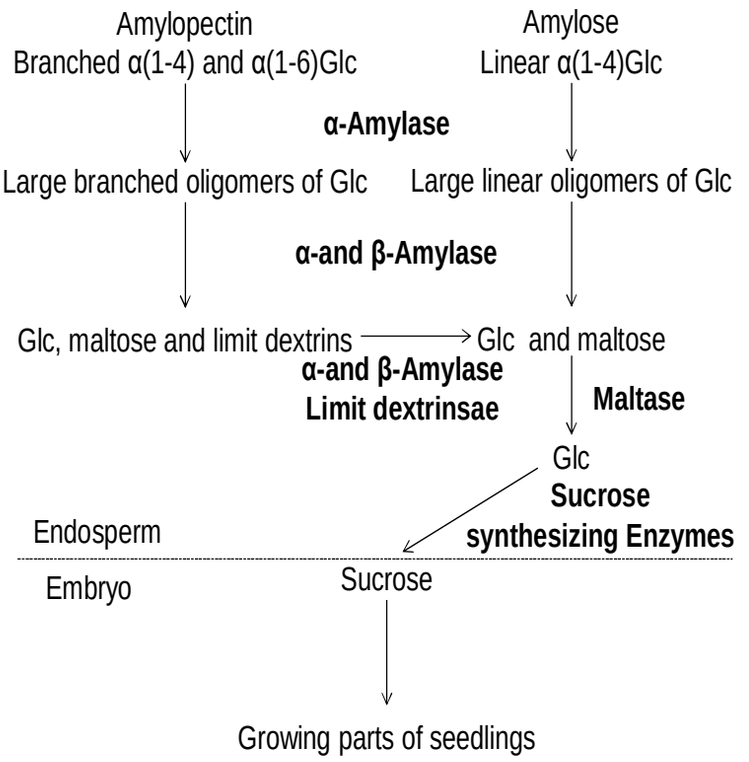

Fig. 9.4: Hydrolysis of starch grains in cereals by amylolysis (Source: Bewley, 2001).

In legumes amylose and amylopectin may be converted initially to glucose 1- phosphate (Glc-1-P), maltose and limit dextrins by starch phosphorylase and β-amylase, and then further to glucose (Glc) by limit dextrinase, bamylase and maltase.

9.4.2 Protein

Storage proteins are hydrolyzed by proteinases. These enzymes have been classified according to their hydrolytic activity. Endopeptidases hydrolyze inner peptide bonds to produce short oligopeptides, which are cleaved further by peptidases into amino acids. Carboxypeptidases and aminopeptidases release the terminal amino acid from the free carboxyl or amino end, respectively, of a polypeptide (Fig. 9.5). In cereals, the main sites of storage proteins is the endosperm and above mentioned enzymes are produced by the aleurone layer after germination. Pre-synthesized proteinases are also found within the endosperm that gets activated soon after water uptake by the seed. Protein hydrolytic products- small peptides and amino acids are transported to the growing axes.

In most dicot seeds, cotyledons contain storage proteins found within separate protein bodies. In the beginning after germination endopeptidases belonging to 'proteinase A' group, slice short-chain peptides from the storage proteins, making them soluble, and vulnerable to additional proteolysis. These susceptible chains are hydrolyzed to small oligopeptides and amino acids by the action of 'proteinase B' class endopeptidases and

carboxypeptidases. All of these reactions take place within the protein bodies. Thereafter, oligopeptides are released into the cytoplasm where aminopeptidases and other peptidases capable of acting on di and tripeptides, degraded them to individual amino acids (Fig. 9.6). Amino acids are converted to glutamine and asparagine prior to their mobilization to the seedling axes.

Above mentioned proteolytic enzymes are synthesized on the rough endoplasmic reticulum (RER). Thereafter, they are packaged into vesicles which are transported to the protein bodies, with which they fuse, discharging the hydrolases to initiate their action on the storage proteins. As protein digestion continues, the protein bodies fuse to form big vacuoles into which a range of hydrolases are secreted. Finally, they develop into autophagic vesicles accountable for eventual degeneration and senescence of the expended cotyledons.

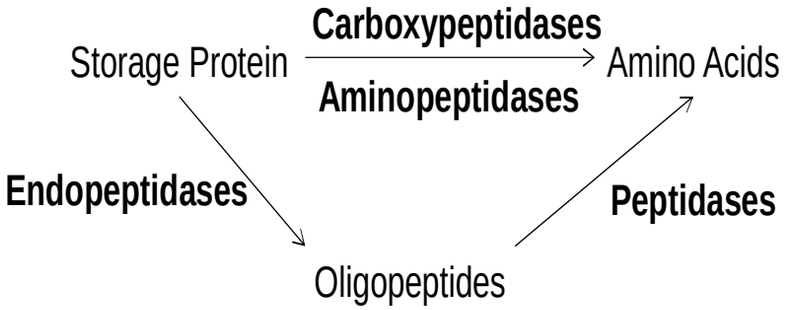

Fig. 9.5: Hydrolysis of storage proteins to their constituent amino acids by proteinases

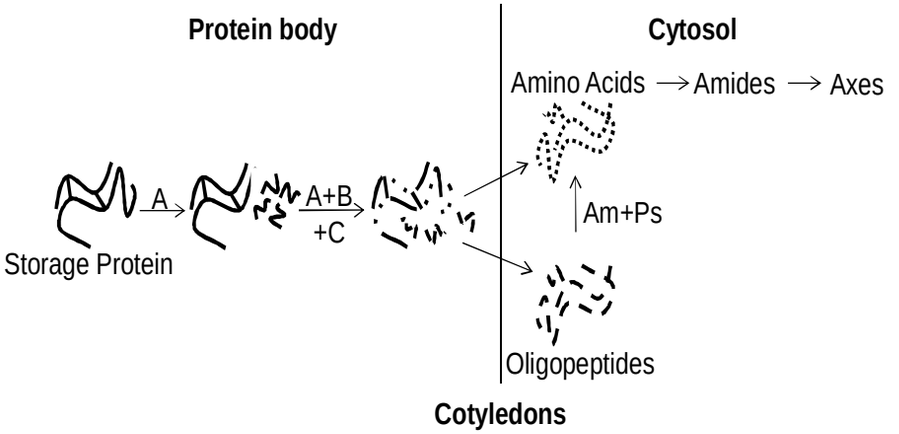

Fig. 9.6: A generalized pathway for the mobilization of storage proteins in the protein bodies of dicot seeds (Source: Bewley, 2001)

The native storage protein, here a disulfide-linked insoluble legumin, is initially trimmed by proteinase A (endopeptidase) activity (A) to render it more soluble as small oligopeptides are released. Further proteinase A activity, and that of proteinase B endopeptidases (A and B) result in hydrolysis of the protein to amino acids and small peptides which are transported into the cytosol. Hydrolysis of the polypeptides in the protein bodies is aided by the carboxypeptidases (C). The released oligopeptides are degraded further by aminopeptidases (Am) and peptidases (di- and tripeptidases, Ps) within the cytosol to yield amino acids. These are converted to the amides, glutamine and asparagine, and transported to the growing axes to support seedling growth (Based on Wilson (1986).

9.4.3 Oils

Triacylglycerols (TAGs) are the key storage oils present within the oil-storing cells in seeds. Their mobilization includes three distinct organelles: 1. the oil body, where TAGs are hydrolyzed to free fatty acids (FFA) and glycerol; 2. the glyoxysome, in which the FFA oxidation takes place and succinate is formed via the glyoxylate cycle; 3. the mitochondrion, where succinate is changed to oxaloacetate or malate. The latter two are processed further in the cytosol to produce sucrose. First TAG is hydrolyzed (lipolysis) by lipases, which cleave the fatty acids from the glycerol backbone (Fig. 9.7). Glycerol in due course moves in the glycolytic pathway in the cytosol after its phosphorylation, and converted to hexoses by the reverse glycolysis. From the oil body, free fatty acids (FFA) enter the glyoxysome, an organelle which is found in a nascent form in most oil-storing seeds, and expands as its membranes become augmented with phospholipids and its medium with enzymes. In the glyoxysome, FFAs are subjected to β-oxidation in which acetyl–CoA residues are sequentially removed. If the FFAs are unsaturated or consist of an odd number of C atoms additional enzymes also participate in catabolism. Directly attached to the β-oxidation pathway is the glyoxylate cycle, which receives the acetyl–CoA and in succession enzymatic reactions links this via enzymes in the mitochondria to the cytosolic glycolytic pathway, which then operates to yield hexose. The important enzymes in the glyoxysome are isocitrate lyase and malate synthase, which produce succinate and malate, respectively, for further catabolism in the mitochondria and cytosol. The final steps take place in the cytosol, where the hexoses are converted to sucrose, which is transported to the growing points of seedling. In nonpersistent storage tissues the glyoxysomes degenerate but in cotyledons that turn green during seedling growth their enzyme complement changes as they become transformed to peroxisomes; these play an important role in photorespiration.

Triacylglycerol ⟶ Diacylglycerol +FFA ⟶ Monoacylglycerol + 2FFA

↓

Glycerol + 3FFA

Fig. 9.7: Triacylglycerol hydrolysis by lipases

9.5 Seedlings Responses to Environmental cues -Tropisms

Seedlings show responses to environmental cues such as light and gravity in order to maximize their growth. Shoots of growing seedlings exhibit positive phototropism by bending in the direction of light which permits shoots to capture more light. Auxin is responsible for it.

Plant growth response to gravity is called gravitropism. Roots and shoots show positive and negative gravitropism respectively. Bending away from gravity i.e. negative gravitropism of shoot is arbitrated by auxin which causes lower part of stem to elongate. Gravity sensing cells are found in root cap which mediated positive gravitropism by roots.

Shoot apical meristems and immature parts of plant produce auxin- the hormone responsible for phototropism. Charles and Francis Darwin speculated what triggered plants to curve toward light. They established that growing tips of plants perceive light. Went proved that a chemical formed in the shoot tip is accountable for the shoot bending - he named it "auxin". An agar block can absorb compounds underneath a growing shoot tip. When the block is applied to an immature shoot, the shoot lengthens more on the side where the agar block is placed (Fig. 9.8).

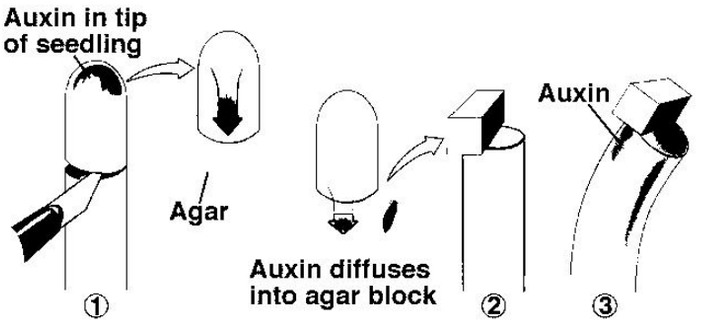

Fig. 9.8: Charles and Francis Darwin' shoot bending experiment

Auxin is formed evenly by developing shoot tips but is transported to the unlighted side of the shoot. That's why cells on the shaded side elongate more in comparison to cells on the lighted side (Fig. 9.9). It ensures this by making cell walls softer and more stretchable by increase of the cell's cytoplasm.

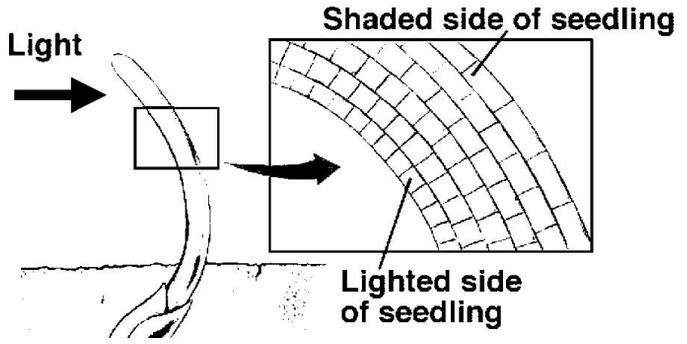

Fig. 9.9: Cells in shaded side show more elongation in comparison to those facing light directly

9.6 Hormonal Control of Seed Germination

Gibberellins and ABA are the most significant plant hormones for seed germination which have stimulatory and inhibitory effects on it respectively. Ethylene and brassinosteroids also exert enhancing effects on seed germination. Though auxin by itself may not be essential for seed germination, its interactions and crosstalk with ethylene and gibberellins may affect the procedures of seed germination. Roles of different plant hormones in context to seed germination are as follows:

9.6.1 ABA

ABA is popularly known as stress hormone. Apart stress response, it also control stomatal activity, seed dormancy. It has negative affect on seed germination process. It delays radicle expansion and weakening of endosperm. It shows inhibitory effect on expression of transcription factors which are required for the seed germination.

9.6.2 Ethylene

Though ethylene controls flowering, fruit ripening, aging and other plant activities, but there are different concepts regarding its role in seed germination. According to some scientists ethylene is required for seed germination while others suggest that ethylene is produced as a result of seed germination. An increase in the amount of ethylene during the germination has been reported in many plant seeds including rice, corn and wheat.

Ethylene is capable of germinating dormant seeds. It can induce rupturing of testa and endosperm. It antagonistically interacts with the inhibitory effects of ABA on seed germination. It has been proposed that ethylene controls the expression of genes which encode enzymes responsible for degrading seed-storage proteins during the initial phase of germination. These enzymes include cysteine-proteinase, proteasome.

9.6.3 Gibberellin

Gibberellin is essential for seed germination. This hormone helps in seed germination by weakening of endosperm, breaking coat dormancy and expansion of embryo cell. The loosening of endosperm is accomplished by modifications of cell wall proteins. Still, ABA can inhibit the weakening of endosperm. Gibberellin induces the synthesis of hydrolases particularly amylase. At molecular level, gibberellin stimulates variety of genes necessary for the production of amylase, glucanases and proteases. Gibberellin signaling takes place in aleurone layer which surrounds endosperm. Cross talk between hormones significantly affects seed germination.

9.6.4 Auxin

Auxin by itself is not essential for seed germination. Nevertheless, it is found in the seedling growing tip. It is required for the growth of young seedlings through regulation of the phototropism as described above.

9.6.5 Cytokinin

Cytokinins remain active in all phases of germination. They control (1) embryo growth by regulating the cellular division, (2) seed size, (3) seed production and germination, (4) hypocotyls and shoot growth. Cytokinins also improve seed germination by the alleviation of stresses.

9.6.6 Brassinosteroids

Brassinosteroids exhibit positive effect on seed germination by regulating the inhibitory effects of ABA. Gibberellic acid, ethylene and brassinosteroids enhance rupturing of endosperm and increase the capability of embryos to grow out of the seed. They antagonistically interact with ABA. These hormones have their own signaling pathways. Gibberellins along with light relieve seed photodormancy while brassinosteroids increase seed germination by increasing the growth of embryo.

9.7 Summary

Non-motile nature, genetically active haploid gametophyte, double fertilization, totipotency and enormous developmental plasticity are some unique characters of plant life.

Seed germination starts with water uptake and terminates with emergence of radical. Seeds contain food reserves in endosperm or cotyledons which are mobilized to support the initial growth of seedling until it becomes auxotrophic.

Cross-talk among different plant hormones mainly gibberellic acid, ABA, ethylene and brassinosteroids regulate different events during seed germination. Environmental factors light, gravity not only affect seedling germination as well as it further growth.

9.8 Glossary

- **Seed Germination:** the emergence and development from the seed embryo of those essential structures which, for the kind of seed in question, are indicative of the ability to produce a normal plant under favorable conditions.
- **Carbohydrate:** Neutral compound of carbon, hydrogen and oxygen. Sugar, starch and cellulose are carbohydrates.
- **Seed dormancy:** Seed dormancy can be considered a protective mechanism which helps prevent the seed of some species from germinating at the wrong time of year.
- **Hormone:** Chemical substance that modifies the growth and development of a plant. Root-inducing hormones help cuttings root faster than they would naturally.

9.9 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. Mention the unique feature of plant development regarding fertilization which also distinguishes it from animal development.
2. The most important character of the plant life is the presence of genetically active .....phase.
3. The first event during germination of a seed is……
4. In cereal grains, food reserves are stored in…..
5. Cotyledons contain main reserve food in seeds of…..
6. The plant hormone responsible for phototropism is…..
7. Name the two plant hormones which act agonistically in seed germination.

Section B : (Short Answer Type Questions)

1. Write short note on unique features of plant development.
2. Write short note on seed germination.
3. What are protein bodies?
4. What is the mechanism of phototropism shown by seedlings?
5. Write the role of ABA in seed germination.

Section C : (Long Answer Type Questions)

1. Mention about the unique features of plant development.
2. Write notes on cross-talk among different plant hormones regarding seed germination.
3. Describe the process of seed germination.
4. Describe the mechanism of mobilization of carbohydrates, oils and proteins during the process of seed germination.

Answer key of Section – A

1. Occurrence of double fertilization
2. Haploid (Gametophyte) Phase
3. Water Uptake
4. Endosperm
5. Dicots
6. Auxin
7. Gibberellin and ABA

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Unit – 10

Shoot Development

Structure of the Unit:

10.0 Objectives

After going through this unit you will be able understand to:

- correlation between various part of plant
- Structural variation in stem of various groups of plant
- structural advantage to plant in response to habitat
- secondary growth in plants and its utility

10.1 Introduction

Shoot is an important part of plant system .It bears leaf, branches, inflorescence, flower and fruit and seed in later stage. When leaf originates from the shoot apex some changes occurs in related tissues. During the process of stem elongation various tissues are produce which help to the plant development. After attaining certain level of growth plant require strengthful tissue to help its increase weight so role of secondary growth is very important.

10.2 Shoot Apical Meristem

Shoot with leaf primordia is the shoot apex; at this place primary organization of shoot is start up.It is usually small in angiosperms while wider in gymnosperms.

There are many theories have been given by various workers from time to time to explain the type of growth found in shoot apical meristem.

1. **Apical cell theory**. This was proposed by Hofmeister (1857) and supported by Nageli (1878). According to the theory, a single apical cell is the structural and functional unit of apical meristem which governs the entire process of growth. However, such organisation has been found only in algae, bryophytes, pteridophytes and not in phanerogams.

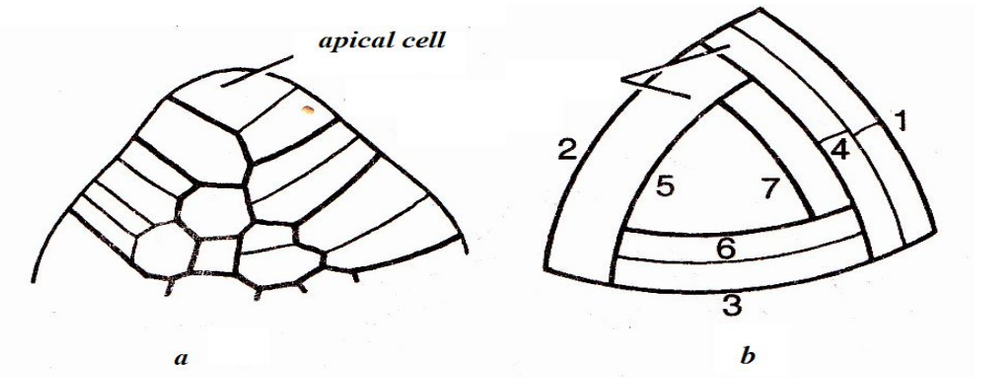


Fig. 10.1 : Shoot apex showing single Apical cell

a—LS of Shoot apex of Equisetum ; b- apical cell and most recent derivative

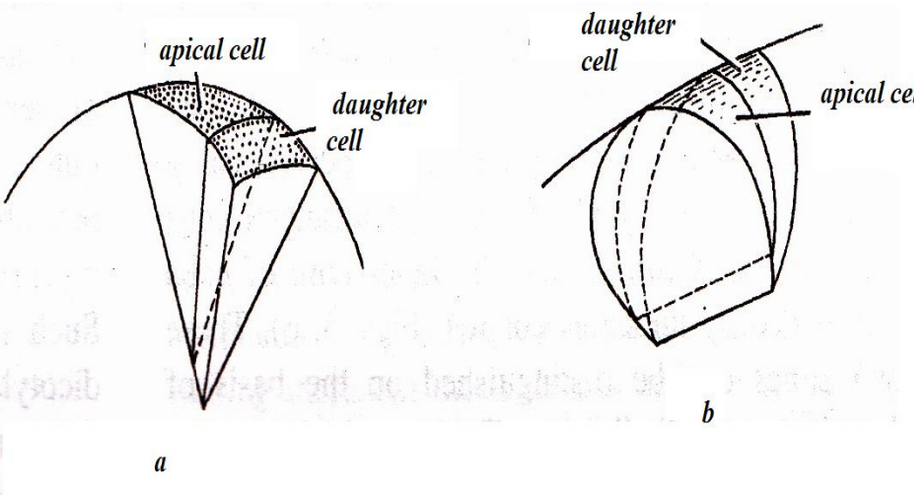


Fig. 10.2 : Forms of Apical cell

a---pyramidal b---Lenticular

2. **Histogen theory**. This was proposed by Hanstein (1868) according to him that three distinct meristematic zones (layers) are found in apical origion. Each zone consists of a variable number of layers called *histogen or tissue builder*. The histogens arise from separate group of initial cells and have different mode of development.

   These three histogens named are **(a) Dermatogen,** the outermost layer, **(b) Periblem**, the middle region composed of cells and **(c) Plerome,** the innermost. Each histogen has definite function. The **dermatogen** cells divide and form epidermis, the **periblem** forms the cortex and the **plerome** forms the stele (vascular tissues, pericycle, pith, rays etc.). Later on studies revealed that these layers have no morphological significance because these are not specific in their function. Also, it is not possible to distinguish these three histogen layers in gymnosperms and angiosperms groups. Hence this theory was not applied for shoot apical meristem.

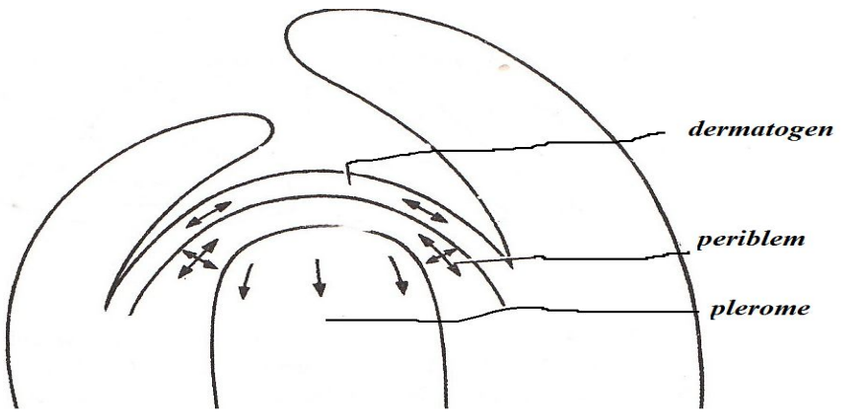


Fig.10.3 : Growth of Shoot Apex(Hanstein Histogen Concept)

3. **Tunica-corpus theory**. This was proposed by S**chmidt** (1924) to explain shoot apex organisation. According to this theory, there are two zones in apical meristem **(a) Tunica**, the outer zone consisting of one or more peripheral layers of cells, forming the outer region by anticlinal divisions, and **(b) Corpus** the central undiferentiated multilayred mass of cells surrounded by tunica which forms the central part of shoot by irregular divisions.

   Tunica and corpus zones have separate initials which are contiguous with one another at the tip of apex. The tunica cells are smaller and corpus cells are larger. The number of tunica layers may vary even in same species due to influence of seasonal growth changes.

Tunica term is somewhat a restricted term as regards to cell division which is always anticlinal. Some workers considered all mantle like layers as part of tunica and interpret the other stratified layer as corpus. Popham and Chan (1950) introduced **mantle-core hypothesis** without emphasising the importance of planes of division. According to them **mantle** included all the outer layers of apex and the **core** to the mass of cells surrounded by mantle.

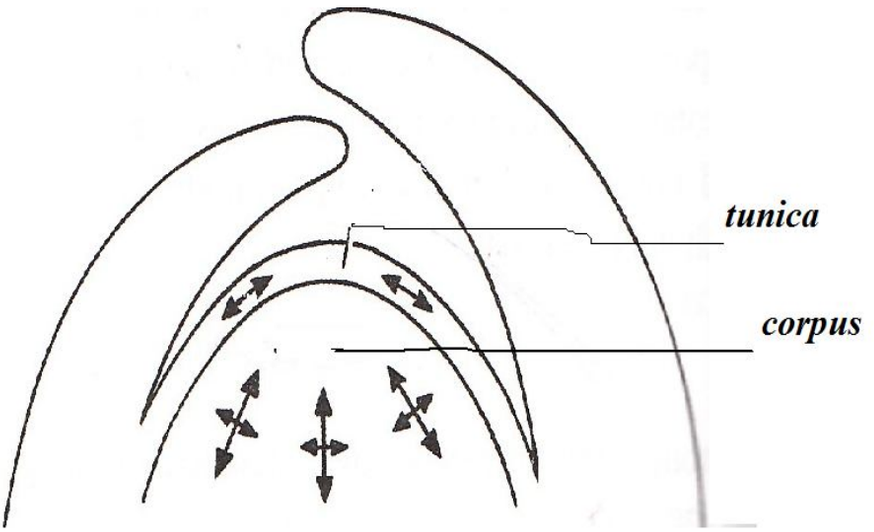


Fig. 10.4 : Growth of Shoot Apex(Tunica-corpus Concept)

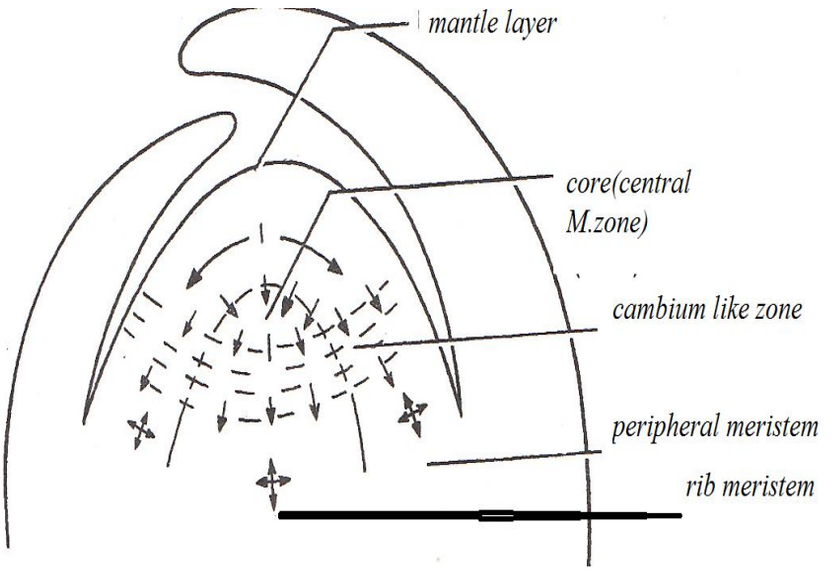


Fig 10.5 : Growth of Shoot Apex in dicot (Mantle-core concept)

4. **Histogenic layer theory**. This was proposed by Dermen (1947) rejecting tunica corpus theory proposed the concept of primary histogenic layer. He recognised three such basic layers in angiosperms and referred to them as L-I, L-II and L-III layers. Of these epidermis of leaf and stem is derived from L-I hypodermis, Cortex and sometimes part of vascular tissues from L-II and vascular tissues and pith arise from L-III. Presence of such three layers in fact suggests the theory seems as a modified version of Hanstein's histogen theory.

5. **Anneau Initial and meristeme d'attente theory**. This was proposed by Buvat (1955) according to him that peripheral and subterminal regions are real initiating zones whereas distal zones are inert in nature recognised three distinct regions in apical meristem, (a) *the anneau initial* (the peripheral active zone) (b) *the meristeme d`attente* (the waiting meristem which becomes active only during formation of terminal flower or inflorescence) and (c) *the meristeme medullaire* (the central pith region)

   Meristeme d'attente consists of two parts, the *promeristeme sporogene* and *promeristeme receptaculair* which thus contain part of tunica (except *anneau initial*) and part of corpus (except *meristeme medullaire*) i.e.,

   Tunica { *anneau initial*; *promeristeme* sporogene } *meristeme d`attente*

   Corpus { *promeristeme receptaculaire*; *promeristeme medullaire* } *meristeme d`attente*

   This theory later on becomes controversial mainly because it considers meresteme d'attente as waiting meristem which was previously regarded as apical initials. In several cases, this region has been found dividing very actively.

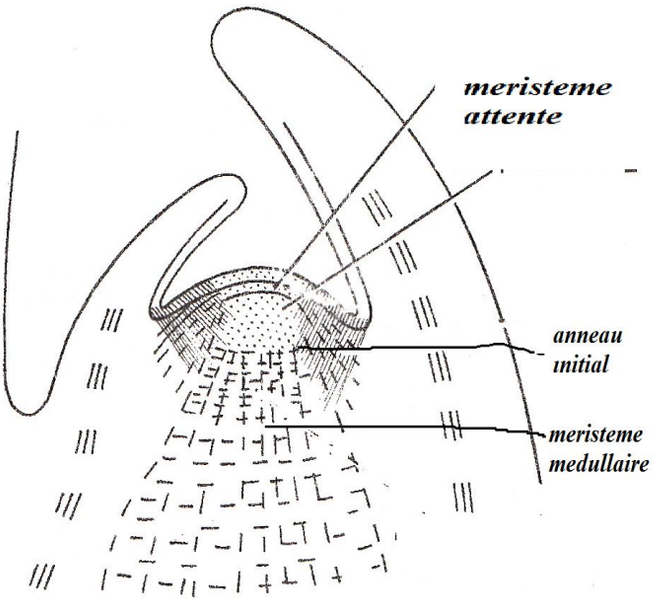


Fig- 10.6 : Growth of Shoot Apex in dicot
(anneau initial meriteeme d attente concept)

6 **Cytohistological zonations concept--**This was proposed by Foster (1939.According to him that in gymnosperms, tunica like layer is not found and so Foster divide shoot apex organisation of *Ginkgo biloba* on basis of cytohistological zonations, which react differently to staining. He recognised four inter-related zones, **(a) Apical initial group, (b) Central mother cell zone, (c) Rib meristem and (d) Peripheral meristeme** or flank meristem .These zones have also been found to show differences in biochemical activities. Such zonations have been reported in several angiosperms.

Later on Popham and Chan (1950) added one more zone, the **cambium like zone** found just below the youngest leaf primordium. However, it has not been found in all cases, such as *Chrysanthemum morifolium* (Popham and Chan, in 1950) and *Ricinus communis* (Singh and Singh, 1976).

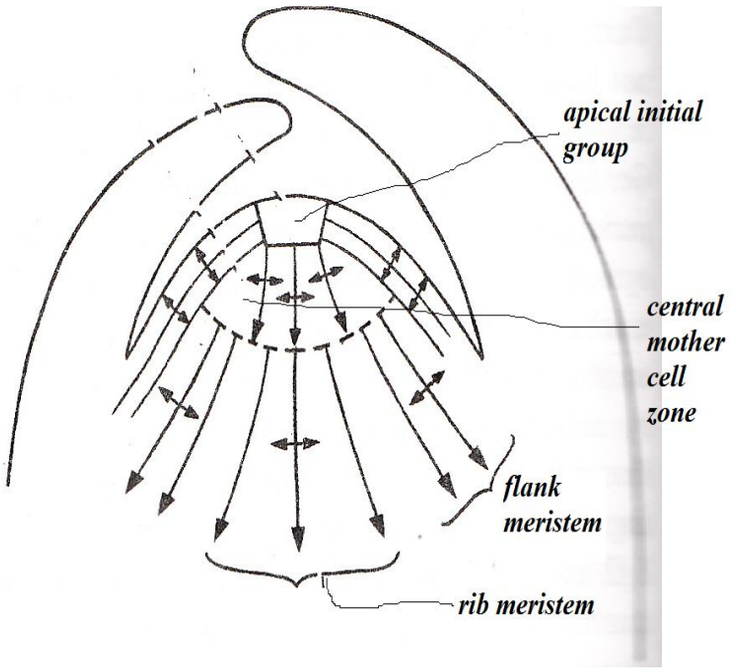


Fig- 10.7 : Cytological zonation seen in Shoot Apex of *Ginkgo biloba*

7. Regions of shoot apex concept----This was proposed by Wardlaw (1957) he recognised five regions in the shoot apex

(a) **Distal region -** a group of initial cells arranged in one or more tiers.

(b) **Sub-distal region** - superficial layers of meristematic cells where growth centres are located.

(c) **Organogenic region** - region of leaf initiation and tissue differentiation.

(d) **Sub-apical region** - region characterised by continued cell division, cell elongation and further internal cellular differentiation.

(e) **Region of maturation** - region of maturation with no division or differentiation of tissue.

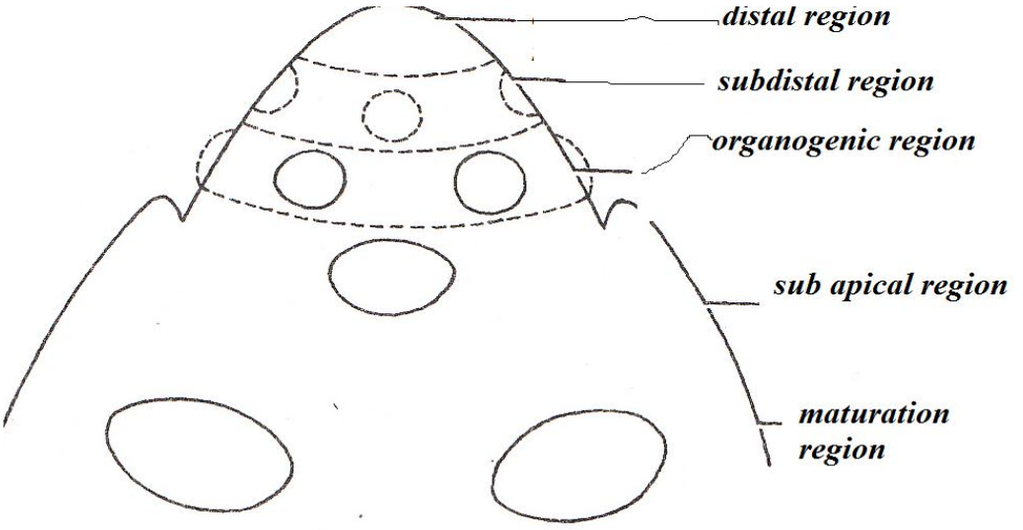


Fig. 10.8 : Shoot Apex with whorled phylotaxis

7. Types of shoot apices(Newman Concept)----Newman (1956) used the term **continuing meristematic residue** to refer to a group of permanent initial cells in the shoot apex and on the basis of form of this meristem, he classified shoot apices into three main types.

1. **Monoplex type**. In this case, meristem is the superficial layer only which is pointed inwardly and a single division is required for growth. This type is found in ferns and their related genera.

2. **Simplex type**. It is is characterised by a parallel sided meristematic residue limited to the superficial layer. It is commonly found in gymnosperms.

3. **Duplex type**. In this the continuing meristematic residue is parallel-sided but at least in two layers, the *superficial layer (s)* which divides anticlinally and *innermost layer(s)* which divides both anticlinally and periclinally . It is commonly found amongst angiosperms.

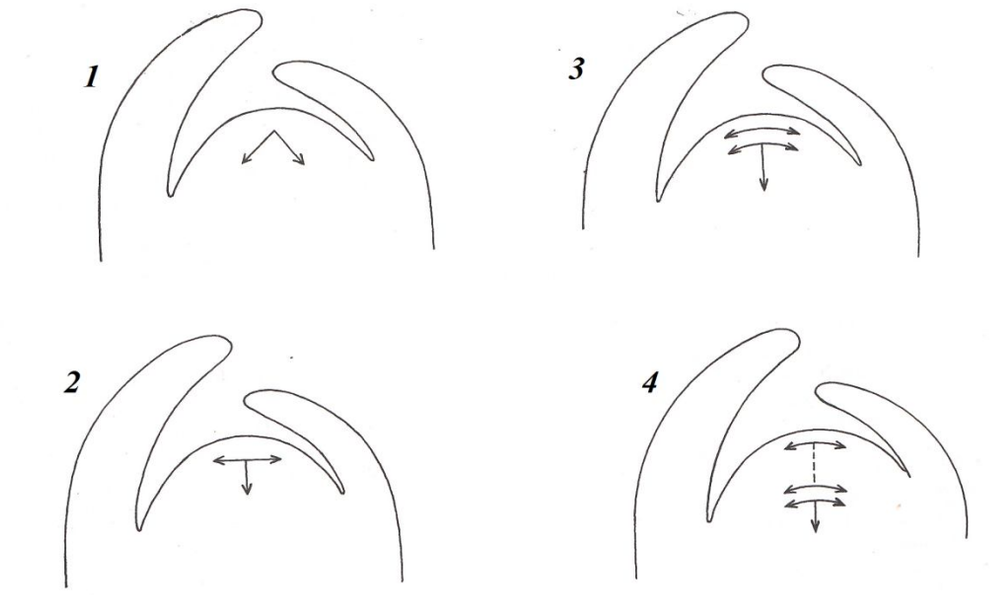


Fig 10.9 : Type of Shoot Apex: 1-Monoplex, 2-Simplex,3-4 Duplex

10.3 Primary Structure of Stem- Dicot & Monocot

The embryo is the starting stage in the life cycle of plant which later develops into plant which having three important organs—the stem, leaves and root. Apical meristem occurring at the tips of the axis produces new cells. Later on these cells become parmanant tissues, the body made up of permanent tissues is known as primary body.

In the lower vascular plants like Psilotales, there is no sharp demarcation between the stem and leaf.In the seed plant also the boundary between stem and leaves is not certain both part, originated from same meristem and are interdependent in the process of growth and differentiation.

Here primary structure of stem of vascular plants is described.

Primary Structure of the stem may be distinguished into three tissue systems: ---

(a) **Dermal system**, (b) **Ground tissue system**, and (c) **Vascular system**

The dermal system originates from the surface layer of the shoot apical meristem. The ground tissue from the flank and rib meristems and the vascular tissue system originates from the procambium.

[I] Dermal system

Stem is bounded by an **epidermis**, made of closely arranged rectangular cells with relatively thin primary walls. It is derived from the outermost layer of the shoot apex, the **dermatogen**. The outer tangential and radial walls of epidermal cells are impregnated

with **cutin** which is fatty/waxy substances.. The substance constitutes a special layer called, the **cuticle**. The thickness of the cuticle varies with the environment in which the plant grows. It is usually thicker in the xerophytes and relatively thin or even absent in the plants of deeply shaded and aquatic habitats. The epidermal cells are living cells with highly vacuolated protoplast containing numerous cell organelles. Chloroplasts are usually absent in the epidermal cells but they may occur in the epidermis of the plants growing in deeply shaded and aquatic habitats. The epidemial cells are capable of divisions in radial plane and also tangential enlargement. This characteristic enables epidermis to withstand the stress during increase in thickness of the stem at the time of secondary growth as describe later on.

Continuity of epidermal cells is broken-up by the presence of **stomata**. They play a important role in gaseous exchange .Various types of **trichomes** are also present on the epidermis.

Epidermis protects the inner tissues from desiccation, from excessive heat or cold, and from the attacks of organisms. As cuticle is impervious to water, it checks excessive loss of water by transpiration.

In herbaceous plants, epidermis usually persists throughout the life of the stem, but in arboreal species it is ultimately replaced by a secondary tissue, the **periderm** during secondary growth.

[II] Ground tissue system

In dicotyledonous stems the ground tissue is differentiated into **cortex** and **pith**. But in monocotyledons there is, however, no clear demarcation between cortex and pith. In dicotyledonous stem the cortex is usually distinguished into three distinct regions - an outer **hypodermis**, a middle parenchymatous **cortex** and an inner **endodermis**.

In herbaceous stems, the **hypodermis** usually consists of a few layers of collenchymatous cells immediately below the epidermis. In cylindrical stems, it forms a continuous layer .But in angular stems it occurs in the form of patches below the ridges (e.g., *Cucurbira, Peristrophe).* The primary function of hypodermis provide mechanical strength to the young system against compression and pulling due to wind Often the collenchymatous cells of the hypodermis possess chloroplasts and then they also perform the function of photosynthesis :

The region of the **cortex**, next to hypodermis is made of oval, round or polygonal parenchymatous cells with intercellular spaces. A few outer layers of this region may contain chloroplasts and then it is called **chlorenchyma** or **assimilatory parenchyma**. In some xerophytic plants where leaves are ephemeral or very much reduced, the function of

photosynthesis is taken up by stems, and in such cases most of the cortex is chlorenchymatous (e.g., *Euphorbia tirucalli*). In many aquatics, the cortex develops an **aerenchyma** with a system of large intercellular spaces. Several structures, such as latex tubes, resin ducts, or reservoirs of waste products are frequently found in the cortex. Crystals of calcium oxalate and substances like tannins are also common in the cortex.

Cortex functions as food storage region. It also facilitates the lateral transport of inorganic and organic nutrients and water. In plants, where cortex contains chloroplasts, it also functions as a Photosynthetic region. It also provides mechanical Support to other tissues of the stem.

The innermost layer of the cortex in most of dicotyledons contains abundant starch grains.This layer is termed as **starch sheath**. In underground Stems, however, a typical endodermis with casparian strips is found. In certain herbaceous stems (e.g., *Senecio and Leonurus*) endodermis develops only when the plant reaches the flowering stage.

Tissue present in the centre of the axis, and enclosed by the vascular tissue, is known as **pith**. It is usually made up of loosely arranged parenchymatous cells. The pith cells contain starch forming leucoplasts and devoid of chloroplasts.Sometimes the cells of the pith become specialized repositories for waste products or may be modified into sclereids. Laticifers, secretory cells, sclereids, idioblasts, ergastic substances and crystals of various kinds are also found in the pith. The outer part of the pith is sometimes distinct in having smaller cells with thick walls. Such morphologically outer pith is called **perimedullary zone** or **medullary sheath**.

In contrast to dicotyledons in monccotyledons, the vascular bundles are scattered, the ground tissue is not differentiated into pith and cortex. But in some grasses, a part of parenchymatous ground tissue towards the centre is devoid of vascular bundles. This region is sometimes referred to as pith. In still other grasses (e.g., *Triticum*) the central region of the stem is hollow and this central cavity is referred to as **'pith cavity'**

The primary function of pith is to store food. But when it is fibrous or sclerenchymatous, it also provides mechanical support. Very often, pith dies or disintegrates as the plant matures and eventually loses all functions.

[III] Vascular tissue system

Vascular tissue system includes vascular cylinder and peri-vascular tissue. The former consists of **vascular bundles** and the latter, the pericycle.

In dicotyledonous stems pericycle occurs as a broad cylinder or patches of tissue that separates cortex from the vascular tissue. It is usually made up of closely packed fibres.

The vascular tissue is present in the form of strands, known as the **vascular bundles**. In a young stem the vascular tissue differentiates from the pro-cambium (pro-vascular meristem) in the - stem apex. In this differentiation process the primary xylem is formed centrifugally, i.e., the protoxylem which is formed first is towards the centre of the stem and the metaxylem which is formed later on, is towards the periphery. Such xylem is known as **endarch**.

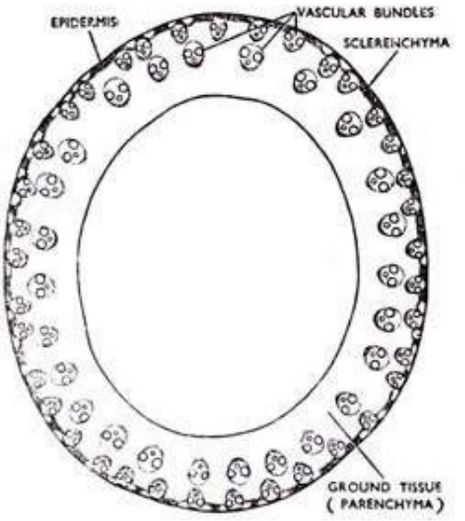
In dicotyledons all the cells of procambium are not differentiated into phloem and xylem. A residual meristem always remains between the Xylem and phloem and this residual meristem persists inside the vascular bundle as **intra-fascicular cambium**. The vascular bundles with intra-fascicular cambium are known as open **bundles**. They are characteristic of dicotyledonous stems and are capable of undergoing secondary growth. While In monocotyledons stem, no residual meristem is left after the differentiation of primaiy xylem and phloem. As such the vascular bundles are without cambium and are known as **closed**. The phloem is usually present on the outerside of the xylem, and such a vascular bundle is known as **collateral bundle**. However, in certain families, such as Solanaceae, Cucurbitaceae and Apocynaceae a phloem also differentiates on the inner side of the xylem and is known as **internal phloem**. A vascular bundle with both external and internal phloem is known as bi-collateral vascular bundle.

In Several members of Nymphaeaceae the rhizome has many steles. This condition is known as Polystely. Each polystele has two to twenty or“ten more vascular bundles. **Medullary** and Cortical bundles occur in several taxa of amaranthaceae, Chenopodiaceae Nyctaginaceae, verbenaceae, etc.

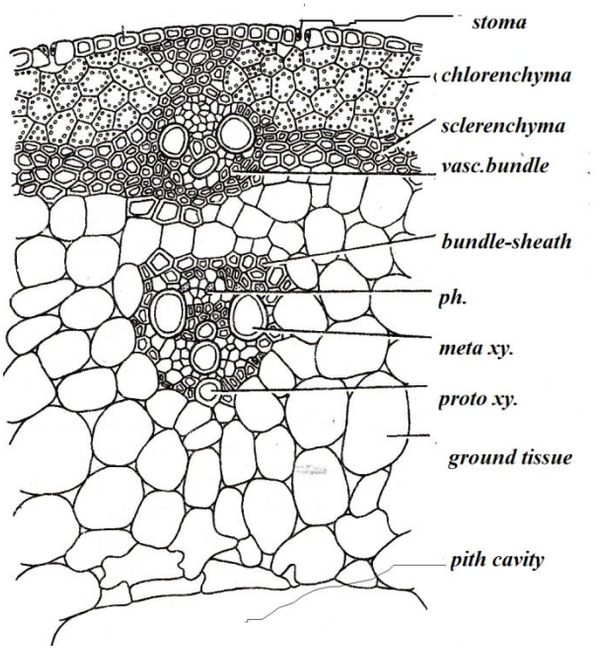
In Some dicotyledons (e.g., members of amaranthaceae, Chenopodiaceae etc.) vascular bundles are also found in the pith. Such bundles are known as medullary **vascular bundles**. They are definite (e.g., *Achyranthes, Boerhaavia*) or indefinite (e.g., *Bougainvillea*) in number. In some Nyctaginaceae, vascular bundles are also found in the cortex and then they are known as **cortical bundles**.

In the stems of monocotyledons many vascular bundles are present which are usually **scattered throughout the ground tissue.** All vascular bundles may be of uniform size (e.g., *Sorghum, Saccharum*) or the Outer bundles are smaller and become gradually larger towards the ceatre of the axis (e.g., *Triticum, Hordeum, and Oryza*). In monocots the vascular bundles are usually collateral and closed. The xylem is in the form of ‘Y’. It is composed of only four vessels, the two metaxylem vessels are situated at the tips of the arms of ‘Y’ and the two protoxylem vessels at the base of ‘Y’. Protoxylem often disintegrates, as in maize, due to rapid elongation of young stem, to form a lysogenous cavity. The phloem is situated in between the arms of ‘Y’, sometimes slightly above

towards the outside. It consists of sieve tubes with small companion cells and there is no phloem parenchyma A Conspicuous sclerenchymatous bundle cap is often present. Some monocotyledons have other type of vascular bundles, called **concentric bundle.** In a **concentric bundle** eitherthe Xylem completely surrounds the pholem (amphivasal bundle) or the phloem encircles the xylem tissue (amphicribal bundle).



A



B

Fig.10.10 : T.S. of a typical Monocot Stem *Triticum*
A: Outline diagram, B: Cellular diagram

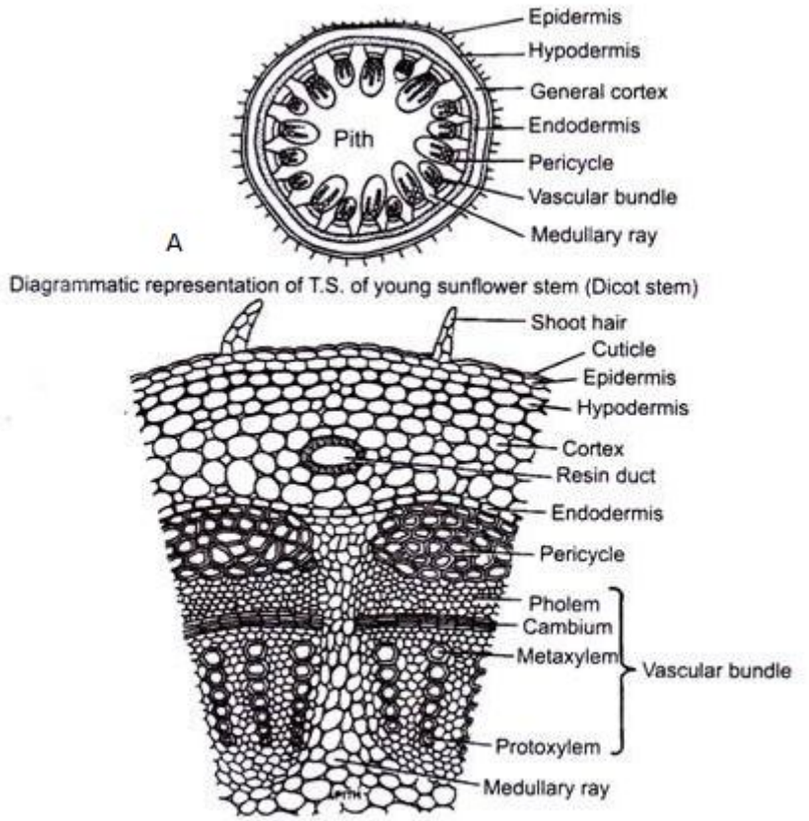


Fig. 10.11 : T.S. of a typical Dicot Stem *Helianthus* (Sunflower)
A: Outline diagram, B: Cellular diagram

There are two important organs of a vascular plant, the shoot and the root. The shoot consists of axis (stem) and its lateral appendages called leaves. The anatomical features of all these, i.e. stem, root and leaves are complicated and are discussed here in separate chapters. In this chapter, anatomical features of stem, as seen in its primary structure are accounted.

Both stem and roots have several characteristics and distinguishing features that help in identifying them. (Table 10.1)

In the stems of dicotyledons, vascular bundles are arranged in a ring. The space between the adjacent vascular bundles is occupied by parenchymatous cells which connect the pith with the cortex. These radially extending bands of parenchyma are called **medullary rays**.

In a transverse section they appear like the radiating spokes of a wheel. Medullary rays store food material and also help in radial translocation of food material.

Table 10.1 : Characteristics of Dicot and Monocot Stems

Dicot Stem	Monocot Stem
1. Several multicellular hairs usually arise from epidermis in dicot stem.	1. Epidermal hairs are usually not found.
2. Hypodermis consists of collenchy -matous cells.	2. Hypodermis is usually scleren -chymatous.
3. Cortex is distinct.	3. Hypodermis and cortex are not well distinguishable and ground tissue is present next tohypodermis.
4. Endodermis and pericycle are distinctly present.	4. Endodermis and pericycle are not present.
5. Vascular bundles are arrangedin a ring.	5. Vascular bundles are found scattered in the ground tissue.
6. Vascular bundles are conjoint, collateral and open type.	6. Vascular bundles are conjoint collateral and closed type.
7. Sc1erenchymatous bundle sheath is not found.	7. Each vascular bundle remains sur rounded by sclerenchymatous sheath.
8. In vascular bundles, several vessels are present and are linearly arranged.	8. Vessels in each vascular bundle are arranged in V-Shape and are fewer in number.
9. Phloem parenchyma is found.	9. Phloem parenchyma is absent.
10. Lysigenous cavity in vascular bundle is absent.	10. Lysigenous cavity in each vascular bundle is present.
11. Both pith (medulla) andmedullary rays are present.	11. Medulla and medullary rays are absent.
12. Secondary growth is found.	12. Secondary growth is not found.

Table 10.2 : Characteristics of Root and Stem

Root	Stem
A. Morphological Characters	***A. Morphological Characters***
1. Roots are misty in colour and of irregular shape.	1.Stems are usually green and straight.
2.Nodes and internodes are not distinct.	2.Nodes and internodes are well marked.
3.Spines or thorns are not found.	3.Spines, thorns and marks of leaf bases are present.
B. Anatomical Characters	***B. Anatomical Characters***
4. epidermal hairs uni- cellular	4. Hairs, if present are multi-cellular
5. Cuticle layer and stomata areabsent.	5. Cuticle layer is present and stomata may also be present.
6. Epidermis lakes the function of water absorption from soil.	6. Epidermis is only protective in nature.
7. Hypodermis is not differentiated.	7. Hypodermis is well marked.
8. Cortex is broad and well developed.	8. Cortex is relatively less developed.
9. Endodermis is quite distinct.Endodermal cells have radially thickened walls intermingled with few, thin walled Passage cell.	9. Endodermis is indistinct and passage cells are not found.
10. The type of side is actinostele	10. Side is polyfascicular siphonostele in dicots and atactostele in monocot
11. Pericycle is single layered and composed of parenchymatous cells.	11. Pericycle is several layered thick and usually consists of sclerenchymatous cells.
12. The vascular bundles are of radial type.	12. The vascular bundles are of collateral type.
13. Xylem is exarch in nature.	13. Xylem is endarch in nature.
14. Lateral branches arise endo genously, i.e., from pericycle.	14. Lateral branches arise exogenously.

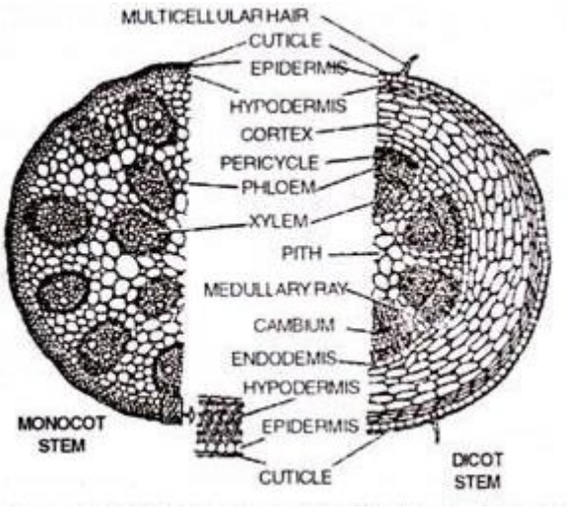


Fig. 10.12 : Comparison of the T.S. of Monocot and Dicot Stem

10.4 Secondary Growth in Dicotyecontous stem

The growth in thickness by the activity of Secondary tissue is called secondary thickening or secondary growth. Normal secondary growth ocuur in dictoylodons and it causes increase in thickness both in intrastelar and extrastelar region of stems.

In Intrastelar region growth is due to the activity of vascular cambium and in extrastealar region growth is due to cork cambium.

(A) Secondary growth in intrastealer region

Secondary Growth in Dicotyledonous Stems

The vascular cambiutu, responsible for the formation of the secondary xylem and the secondary phloem, is a portion of procambium which remains meristematic even after the differentiation of primary xylem and phloem. This residual meristem is in the form of a thin strip of meristematic cells between metaxylem and metaphloem of the vascular bundle. It is known as the **fascicular cambium**. Some of the living parenchyma cells of the medullary rays. Mostly in a line with the fascicular cambium, become meristematic and form new strips of meristem called **interfascicular cambium**. The fascicular (also called intra-fascicular) and inter-fascicular cambia join together forming a complete cylinder of cambial cells. In some plants like *Linum and Tilia*, where primary vascular bundles are very close to each other and interfascicular cambium is not formed, only the fascicular cambium forms the complete cylinder of the cambium.

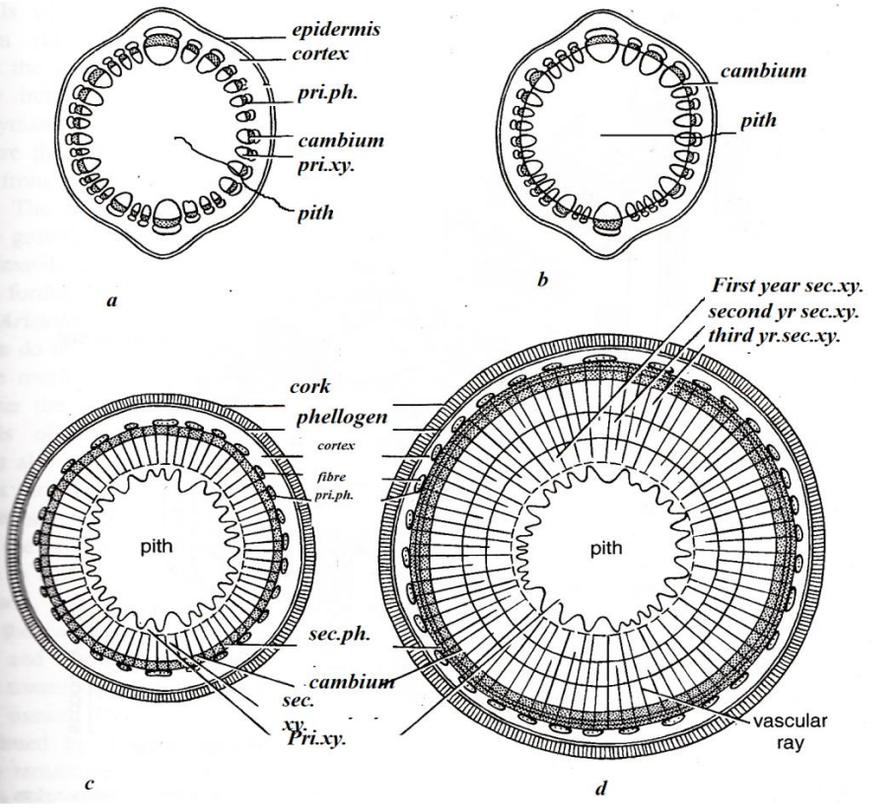


Fig. 10.13 : Various stages of Secondary growth in Typical Dicot stem

The vascular cambium is composed of two types of cells : (i) **fusiform initials** - which are long, vertically oriented cells with tapered ends and they produce secondary xylem and secondaryphloem, and (ii) **ray initials** - these are small isodiametric cells and they produce secondary medullary rays.

The initiation of secondary vascular tissue occurs after the maturation of primary xylem and phloem. At this stage the cambium is represent by a single layer of cells. The fusiform initials, the cambium divide periclinally, forming two daughter cells. One of these cells remark meristematic while the other forms the xylem and phloem mother cell which may direct differentiates into xylem or phloem element without any further division or it may divide one to several times to form two or more cell type.

The cells which are produced outwards by the cambium differentiate into phloem elements, whereas the cells inwards into xylem elements.

Ray initials of the cambium Cut plates of parenchymatous cells both outwards and inwards. These are the **secondary medullary rays** which extend from pith to the secondary xylem and pbloem. The activity of cambium results into a uniform growth around the whole circumference and ultimately a compact cylinder of secondary tissue is formed.

But in some plants (e.g. *Aristolochia*) interfascicular strips of cambium do not produce secondary vascular tissue with the result the vascular bundles are discrete even after the secondary growth.

Cells of the cambium, besides periclinal divisions also show some anticlinal and/or oblique divisions to keep pace with the increasing circumference of the stem. In most of the woody plants, the cambium shows periodic activity, whereas in herbaceous plants it is active for only one season.

As the cambium arises between the primary phloem and the primary xylem, the secondary vascular tissues are also interpolated between these primary tissues. The pith and the primary xylem are enclosed by the secondary vascular tissues, but they remain relatively unchanged as the stem increases in girth. The primary xylem is not distorted but sometimes the pith is deformed by an inward pressure due to the expansion of the Secondary tissues outside the pith.

Secondary Xylem

Secondary xylem forms the bulk of vascular tissue in woody plants. Its constituents are exactly similar to those of the primary xylem. The structure of both primary and secondary xylem is so similar that sometimes it becomes difficult to distinguish between the two tissues. However, the primary xylem has usually long tracheary elements in comparison to the secondary xylem.

Secondary xylem has two systems of tissues which show different orientation in the longitudinal axis of the plant.

1. Axial system. It consists of vertical files of tracheary elements, fibres and wood parenchyma. The elements of this system can be studied in transverse and radial longitudinal sections (R.L.S.). This system is also known as vertical or longitudinal system.

2. Radial system. It consists of rows of parenchymatous cells oriented at right angles to the longitudinal axis of the plant and forms **xylem rays** or **wood rays**. The height and thickness of xylem rays is revealed best in radial and tangential longitudinal sections respectively.

[I] Structure of xylem rays

As described earlier, the xylem rays are the sheets of parenchymatous cells, extending radially in the xylem. As the stem grows older, the rays become distant due to the expansion of the cambiutn along the increasing circumference of the axis.

Rays vary in their width and cellular composition. They may be **uniseriate** (when only one cell wide) or **multiseriate** (when several cells wide) . Usually both uniseriate and multiseriate medullary rays are present in a wood though one or the other type may be quite infrequent. The medullary rays consist of **erect** (upright or vertically elongated cells) and/or **procumbent cells** (radially elongated cells). The rays which are made up of only one type of cells are known as **homocellular rays** and the onesconsisting both the types of cells are called **heterocellular rays**. In primitive woods both uniseriate and multiseriate heterocellular rays of considerable vertical length are present, whereas in advanced woods they are multiseriate and tend to become homocellular with marked reduction in their vertical length.

[II] Growth rings

In woody plants of temperate regions, the cambium shows marked variations in its activity in different seasons. There is an annual alteration of growing and quiescent (inactive) periods. Thus the part of wood which is added in a single growing period can be easily distinguished fromthe preceding and succeeding years. The seasonal increase in the thickness of wood can be seen in a transverse section as concentric ring. These are usually referred to as **growth rings** or **annual** rings. Each growth ring denotes the growth of a single season but more than growth rings may be formed in one season under certain environment conditions. It is possible to determine the age of a plant by counting annual rings, and this study is known as *dendro-chronology*.

Each growth ring can be distinguished into **spring** and **autumn wood** on the basis of differences in shape, structure and distribution of homologous elements in the two regions.For example, in the spring wood the vessels are much wider and relatively thin walled than in the autumn wood. These differences in the structure of wood are to meet the varied requirements of the plant in spring and winter season. In spring season, for instance, the vegetative activity is at its peak and, moreover, there is an increase in the transpiring surface. Therefore, in this period of the year, additional water channels are needed to meet the requirements of the plant. Consequently the spring wood has broader vessels. In autumn, on the other hand, there is leaf fall and comparatively less vegetative growth, and there is no need of having an elaborate system for water Conduction. The autumn wood thus has relatively narrow vessels.

There may be variations in the thickness of successive annual rings, and these variations are due to the fluctuations in the general condition of nutrition as a result of serious injury, infection or unfavourable growth conditions to which the plant has been exposed. Thus, any such fluctuation is recorded permanently in the annual rings in the form of variations in their thickness. Such variations may be used in determining the meteorological

conditions which existed in the past. Growth rings of certain plants have also been analysed for Such Studies.

Sometimes there is a check in the development of secondary xylem, followed byresumption of growth in the same season. Such structural irregularity results in the formation of **false annual rings**.

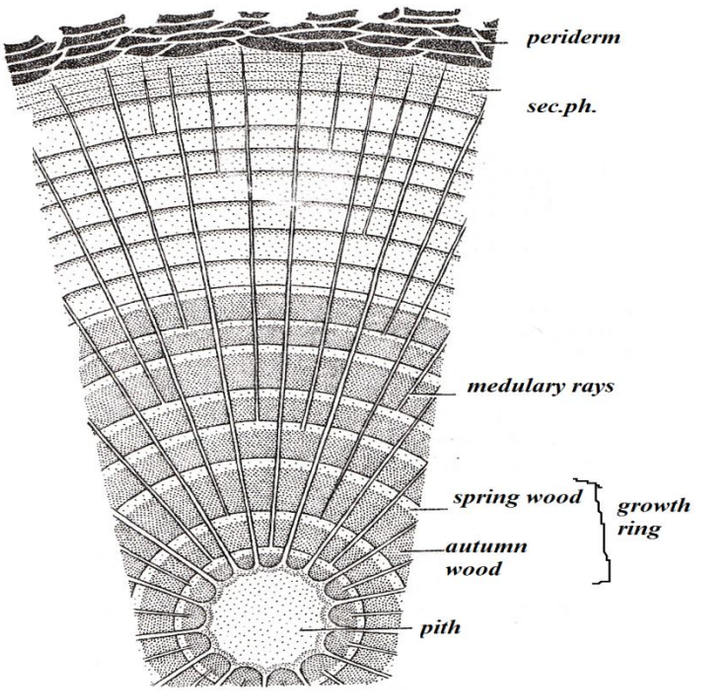


Fig. 10.14 : T.S. of old Dicot Stem Showing Growth Rings

[III] Heart wood and sap wood

In most of the woody plants with the exception of few deciduous species, the outer light coloured part of the wood is distinct from the inner darkcoloured region. The former constitutes the **sap wood** and the latter the **heart wood**. The **sap wood** consists of living cells which are still functional, but ordinarily only few outermost rings of sap wood are engaged in conduction of water and the inner rings function as storage organ.The **heart wood**, on the contrary, is composed of dead cells which become impregnated with various resins, gummy or tannin like substances and consequently are not suited for conduction. In some plants, heart wood contains important pigments of commercial importance, such as **haematoxylin** (*Hernaroxylon campechianim*) **brasilin** (*Caesalpinia soppan*) and **santalin** (*Plerocaipus santalinus*).

As the plant grows older, rings of sap wood bordering heart wood keep on converting intp heart wood. This is a gradual process and in this process all the living cells of sap wood lose their protoplast, water contents are reduced and the vessels are blocked by tylosis.

There is also impregnation of substanccs like gum, tannin suberin etc. The proportion of the heart and sap wood is variable in different species similarly heart wood and sap wood also vary in their colours and this feature sometimes is of diagnostic value. The colour distinction between heart wood and sap wood may be sharp (e.g., *pinus roxburghil, Dalbergia sissoo, A1bizzia lebbek*); somewhat gradual (e.g. *Shorea robusta, Adina cordifolia*) or there is no colour distinction at all (e.g., *Picea smithiana, Abies pindrow Holoptea integrifolia*).

[IV] Porous and non-porous wood

This categorisation of wood is based on the presence or absence of vessels (known as pores in commerce). The dicotyledonous wood which has vessels in the xylem is known as **porous wood**, and gymnospermous wood which lacks vessels is **non-porous wood**. The porous wood is technically referred to as **hard wood** and the non porous as **soft wood** This nomenclature has, however, nothing to do with the relative hardness or softness of the timber as porous wood of *Pterocymbium tinctorium and Bombax ceiba* although soft is classified as hard wood and the non-porous wood of *Pinus* and *Cedrus* although very hard is referred to as soft wood.

Structurally, the hard (porous) wood is more complex than the soft (non-porous) wood. In a transverse section of the hard wood, vessels appear in the form of small 'pores' which may be circular or oval in outline. The arrangement of pores is an important diagnostic feature of timbers. The wood can be classified into **ring-porous** and **diffuse-porous** on the basis of distributions of pores within a growth ring. In the **ring porous** wood, pores of the early wood aredistinctly larger than those of the late wood and they are arranged in the form of conspicuous ring or belt at the beginning of the growth ring (e.g. *Morus alba Toona ciliata*). In **diffuse porous** wood there is no appreciable difference in the size of early and late wood pores, and they are distributed more or less uniformly throughout the growth ring. Most of the Indian hard woods come under this category.

[V] Wood pardnchyama

The parenchyma associated with the secondary xylem can be differentiated into (i) **axial parenchyma**, initiating from the fusiform initials, and (ii) **ray parenchyma**, which initiates from the ray initials. The cells of the axial parenchyma are usually as long as the fusiform initials from which they are derived. But sometimes the fusiform initiais may divide transversely and then shorter axial parenchyma cells are differentiated. The ray parerichyma cells which are usually shorter than the axial parenchyma cells may sometimes develop secondary thickenings in their walls.

Many a times a small part of ray or axial Parenchyma adjoining the pits of vessels, Penetrates into the vessel in the form of a blunt and short protuberance. The nucleus and part of the cytoplasm of the parent cell migrate into the peg - like outgrowth. It gradually expands and assume the shape of a voluminous bladder into vessel lumen This structure is known as **tylosis**. As the tylosis grows further, it detached from the parenchyma cell from whichit has been derived. The separating wall is laid down at the point of its entrance into the vessel. Sometimes, the tylosis undergoes repeated divisions to form a multicellular structure which completely fills the lumen of the vessel. The tylosis may develop secondary walls in the woody plants and often has Corresponding pits on the surface of contact with other tylosis. In some plants tylosis, like other parenchyma cells, performs the function of storage of starch grains.

Secondary Phloem

The secondary phloem, like primary phloem, consists of sieve tube members, companion cells, phloem parenchyma and fibres. Secondary phloem usually resembles the primary phloem to such an extent that it is not possible to determine where one begins and the other ends.

As in secondary xylem, various elements of secondary phloem are arranged in two systems — the **axial** or **vertical** and the **radial** or **horizontal**. The axial system contains sieve tube members.

(B) Secondary growth in extrastealer region

Periderm

Due to continued formation of secondary tissues by vascular cambium in the intrastelar region a pressure is exerted on the epidermis and other extrastelar region. The epidermis considerably stretched and gets ruptured here and there. Now its protective function is replaced by periderm formed in extrastelar region to withstand the inner pressure of secondary tissues. The formation of periderm is a common phenomenon in woody stems and roots of dicotyledons and gymnosperms that increasein thickness by secondary growth

Structurally periderm consists of tissues, these are

1. a meristem known as Phellogen or cork cambium,
2. the layer of cells cut off by phellogen on the outerside of the phellem or cork,
3. cells cut off by phellogen towards inner side are the phelloderrn or secondary cortex.

1. Phellogen or Cork Cambium: (Phellos cork, gen - producing) - In contrast to the vascular cambium, the phellogen is simple and ,'composed of one type of cells. In transverse view the cells appear rectangular in shape and radially flattened.

Origin of Phellogen (Cork Cambium): a strip of secondary meristem, called the cork cambium (or phellogen) generally arises in the outer layers of cortex. !n a few plants the phellogen arises in the enidermal cell-; e.g .. Nerium- Pvrr Tn 1: it develops from sub-epidermal cells. Sometimes only a part of the phellogen is developed from epidermis, while the other part arises in sub-epidermal cells. In some stems the second or third cortical layer initiates the phellogen development. e.g., *Robina, Aristolochia, Boerhaavia*. In the roots of Gymnosperms and dicotyledons, phellogen arises in the pericycle

As soon as the phellogen layer develops, it divides tangentially and to a lesser extent radially in the same manner as the division takes place in vascular cambium. They produce new tissues both centrifugally and centripetally. The derivative cells are generally arranged in radial rows

2. Phellum (Cork): Generally, produces many cells towards the outside to form phellogen or cork. The phellogen produces more phellum or cork on the outerside than the parenchymatous phelloderm on the inner. The cells that constitute phellem are commonly known as cork cells. They are like phellogen cells, from which they are derived. In tangential section, they are polygonal and compactly arranged without inter-cellular spaces. They are dead cells at matu-rity and characterized by suberization of their walls. Because of suberin, cork cells become Impervious to both air and water and prevent dessication of the underlying parts. The cork cells are usually brown or yellowish due to some organic substances. Non-suberised cells of the phellem are called phelloids. After maturity, cork cells lose their living contents and die. They lose their turgidity. In Eucalyptus the cork cells are thick walled and cells are filled with resins and other substances. The cork is also resistant to acids and also serves as a good insulator for high temperature and electricity.

3. Phelloderm (Secondary Cortex): The cells produced towards inner side ofphellogen (cork cambium) are parenchymatous and form phelloderm or secondary cortex. The Phellodenn Cells are living, less isodiametric with intercellular spaces. Generally they contain chloroplast and carry on photosynthesis and store starch. The phellode~ could be identified from the pitmary cortex by their arrangement in radial rows.

Bark

The term bark is applied to all tissues outside the vascular cambium of the stem in secondary state of growth. It includes secondary phloem, primary phloem, peri cycle, endoder- mis primary and secondary cortex and periderm. Periderm is a part of the bark and thus, bark is made up of variety of tissues. However, many workers restrict the term bark to those tissues present outside the active phellogen. The term bark is loose and non-technical. The bark is protective in function. It is of two types:

(1) Ring bark (2) Scale bark

1. **Ring Bark:** Cork cambium develops a complete ring and the bark formed, gets peeled off in the form of a complete cylinder of bark around the stem e.g., Betula, grapevine, Eugenia.
2. **Scale Bark:** In many tropical trees, strips of cork cambium overlapping one another may be formed. They produce bark in the form of strips. Such a bark is known as scale bark e.g., Guava (*Psidium*), *Eucalyptus.*

Commercial Cork and Bark: Bottle cork is made from the bark of oak, Quercus suber (Fagaceae). The first phellogen in Oak arises in the epidermis. When the tree is twenty years old the first bark is removed. It is known as virgin cork. Commercially, it has no value. The process of removal of bark from the tree is known as stripping. A second phellogen arises in the deeper layers of the cortex, which also produces cork. After nine or ton years the second cork is stripped. By this time, it is sufficiently thick and has the commercial value and is of better quality than the virgin cork. The subsequently formed cork is best in quality. Cork is removed at intervals of nine to ten years. Cork oak yields cork upto 150 years. Good quality of cork is light, and impervious to air. The dots seen in the ccrk are the lenticels.

Bark of many frees is commercially important. Bank of Cinchona yields quinine and that cinnamum zeylamicum is used for the control of malaria and as spice respectively. The bark Acacia Arabica and Cassia auriculata is used for tanning.

Lenticels: By the formation of periderm, the stomata are closed and stomatal and also the cuticular transpirations are checked. The dead cork cells with subersied walls are impervious to gases. In lieu of this, some aerating pores which look like lens-shaped raised corky spots are formed in the bark called lenticels. They are present in most of the plants that exhibit secondary growth. Many climbers do not possess lenticels thought periderm is formed.

The lenticels generally appear beneath the old stomata or group of stomata. During secondary growth the periderm replaces the cpidemis. The dead cork cells with suberized walls are partly impervious to gases. Thus the gaseous interchange between the inner living cells and outer atmosphere becomes difficult. The lenticels are also called as air pore or cannal. They are slightly elevated than the surrounding tissue. The bottle cork lenticels are clearly seen.

The phellogen produce spherical, colourless, loosely arranged unsuberised thin walled cells on its outside, instead of giving rise to normal cork below the epidermis all the cells formed by the division of parenchyma in the substomatal region and those cells are formed by the phellogen, on its oter side are together knowns as complementary cells or filling cells. The new complementary cells, which press against the epidermis as a result the epidermis is ruptured. The outer most cells often die due to the exposure to atmosphere

and replaced by new complementary cells out off by the phellogen. In addition to the complementary cells, phellogen produces layers of compact cells known as closing layers in temperate plants. These are formed in the winter season and plug the lenticels. Temperate trees suffer from starts agin and phellogen produces new complementary cells. The spring secondary growth closing layers and ruptures them. A section of lenticels in spring season shows ruptured closing layers on the side of the complementary cells. Closing layers are not observed in the lenticels developed in tropical trees.

Sometimes, lenticels develop independent of the stomata. In such cases the phellogen devides cork cells for a while and then loose complementary cells which ultimately break the cork and give rise to a new lenticel. They may remain scattered or arranged in vertical rows.

Polyderm: It is a special type of protective tissue that develops in roots and under grounds, stems of c rtain families like Myrtaceae, Rosaceae and Onagraceae tec. It consist of alternating layers of one cell deep of partly submerged cells and many cells deep of non-suber- ized cells. Its outermost layer is the dead layer. This type of tissue is termed as polyderm.

Wound cork: When the living cells of the plant part is exposed to the air due to injury, wound cork is developed. Wound cork is restricted only to the injured areas. Soon after the injury, pre- existing cells of the phellogen form a fresh layer of suberized cells just below the injury part.

Later, a new layer of phellogen develops in living, undamaged layer of cells. This phellogen produces phellem (cork) and phelloderm (secondary cork) in the usual manner. The new layers of the cork seal the injury and protect the inner tissue from infections of bacteria and fungi.

Wound cork may develop in all parts of the plant including fruits and leav s. USI ally wound cork is more easily developed in woody plants as compared to herbaceous or monocotyledonous plants. Moist and warm climate favours the early development of wound cork than cold and dry climate.

10.5 Anomalous Secondary Growth of Dicot Stem & Monocot Stem

Anomalous Secondary Growth

In many dicotyledonous plants, the pattern of secondary growth shows a deviation from the normal type. The term anomalous secondary growth is given to this deviated pattern of secondary growth. In fact, the term anomalous simply serves to assemble growth patterns that appear to be less common (Esau, 1965). Anomalous type of growth may be more common than is known at present because the tropical flora in which it is frequently found has not been adequately studied anatomically.

Anomalous (or abnormal) secondary growth may be caused due to following reasons:

(i) The normal cambium behaves peculiarly or irregularly, resulting in abnormal arrangements of vascular tissues.

(ii) The normal cambium is situated in an abnormal position hence the tissues cut off are placed abnormally.

(iii) The normal cambium does not develop or if present, is replaced by additional or accessory cambial rings.

Anomalous Secondary Growth in Dicot Stem

Abnormal secondary growth in dicot plants may be studied under two headings -

(i) Plants in which cambium is of normal type but its activity is abnormal, resulting in abnormal arrangement of vascular tissues.

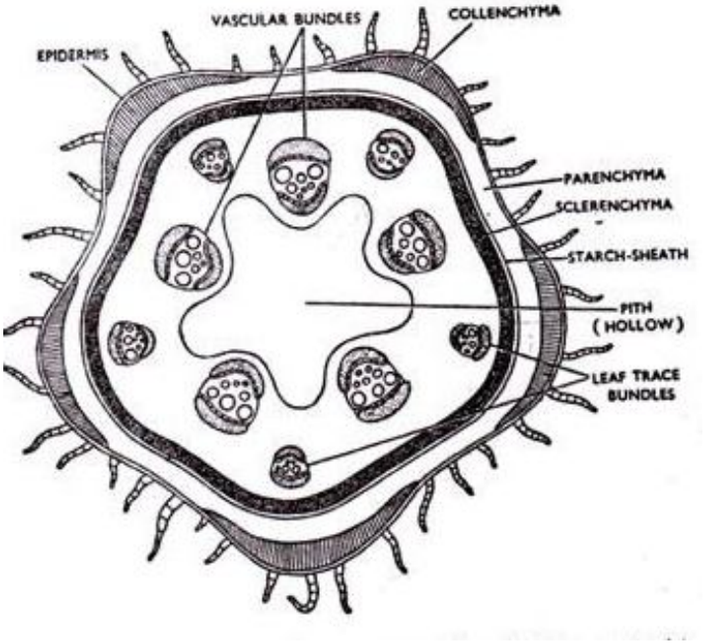
(ii) Plants in which the normal cambium is not formed or if formed, is soon replaced by accessory cambium. This accessory cambium may develop either from the cortex or pericycle region and exhibits abnormal activity. However, the abnormal secondary growth may be studied as follows:

I. Cambium normal but showing abnormal activity

The cambium ring is formed normally but behaves in an abnormal manner. The abnormal activity of the cambium may be categorised into the following types:

1. The cambium forms vascular tissues only in the region of vascular bundle. This type of anomaly is found typically in many woody climbers or lianas. The normal cambium ring is formed by the union of fascicular and interfascicular cambium, **but the activity is abnormal in the sense that either the fascicular cambium or the interfascicular cambium is active.** In case of the stems of *Vitis and Clematis*, the interfascicular cambium as active cutting off parenchyma only, whereas in case of *Cucurbita*, the fascicular cambium is active cutting off vascular tissue. However, in *Aristolochia*, the fascicular cambium cuts off secondary vascular tissue and the interfascicular cambium cuts off parenchyma cells.

***Cucurbita* stem** (Fam. - Cucurbitaceae) - The stem of *Cucurbita* has ten vascular bundles arranged in two rings of five each. Each vascular bundle is conjoint, open and bicollateral having and outer and inner cambium. The outer cambium of both inner and outer bundles becomes active along with the parenchymatous cells of the ground tissue and the two combine to form a more or less wavy ring of cambium. **This normal cambium behaves abnormally as it cuts off secondary xylem and phloem in vasc**
only, resulting in the increased size of the bundle.



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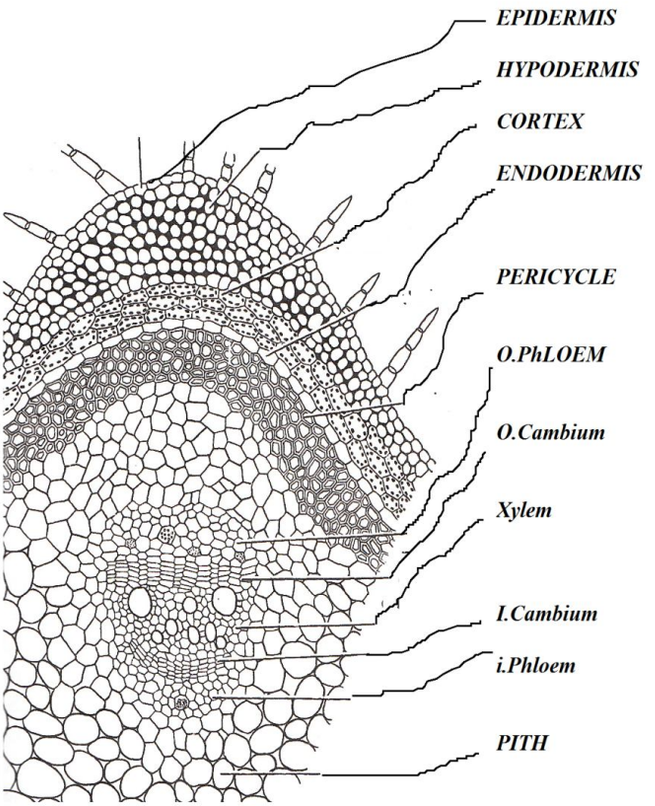


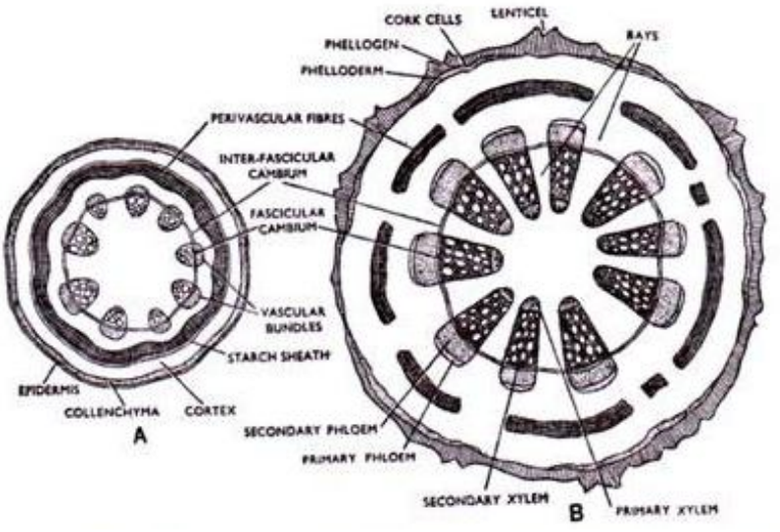
B

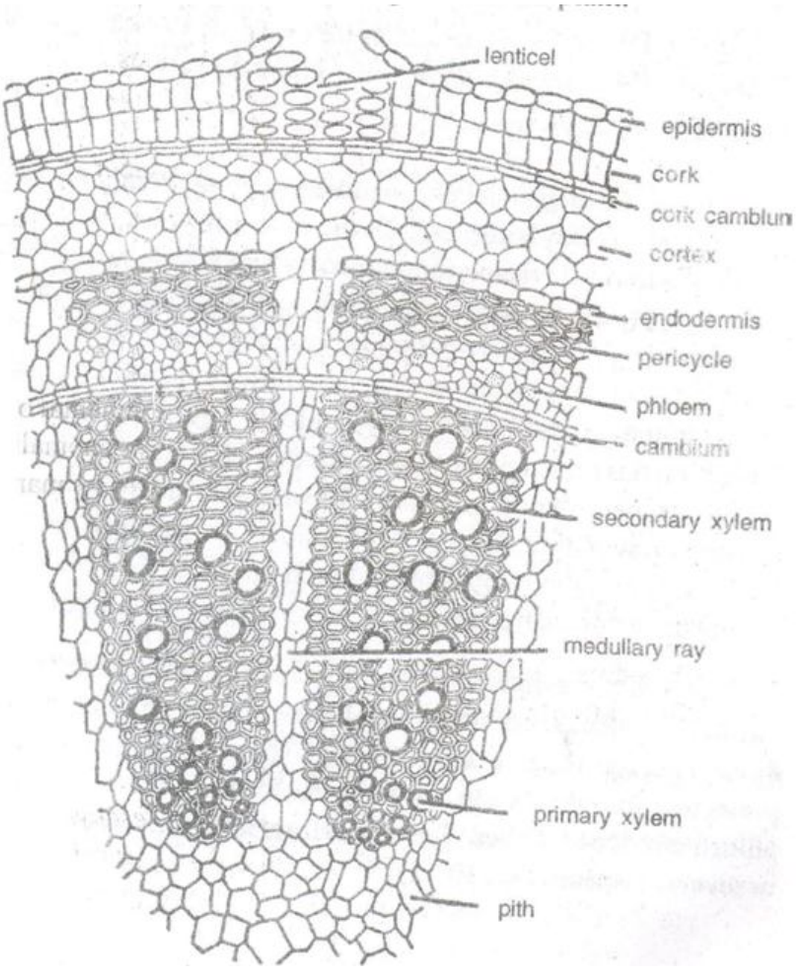
Fig. 10.15 : T.S. of Stem *Cucurbita*

A: Outline diagram, B: Cellular diagram

(b) *Aristolochia* stem (Fam. - Aristolochiaceae) - The stem in young conditions exhibits a typical dicotyledonous structure. The epidermis encloses the multilayered collenchymatous hypodermis, followed by the cortex made up of collenchyma and parenchyma. The endodermis known as starch sheath is followed by multilayered, sclerenchymatous pericycle. The vascular system consists of a ring of open, collateral vascular bundles separated by interfascicular areas. The median and two lateral leaf traces arise from in between the vascular bundles.

At the time of secondary growth, the inter and intra-fascicular cambium become active and form a complete normal cambium ring. However, **this normal cambium ring behaves abnormally, as it cuts off vascular tissue in the intra-fascicular region (within V.B. region) and parenchymatous cells in the interfascicular region**. As such the secondary vascular tissue is not prominent and the bundles remain discrete. In older stems, the pith and the rays become somewhat crushed due to increasing pressure of secondary tissues and partly due to the resistance offered by the pericycle to the expanding vascular system. After sometime, the cumulative pressure breaks up the continuity of the pericycle and the adjacent parenchyma cells invade the breaks by intrusive growth (Esau, 1965), and may differentiate into sclereids. The intra-fascicular cambium may at times behave abnormally and cuts off parenchyma cells, resulting in the xylem becoming fissured and the bifurcation of the bundles.





C

Fig.10.16 : T.S. of Stem *Aristolochia*

A &B : Outline diagram, C: Cellular diagram

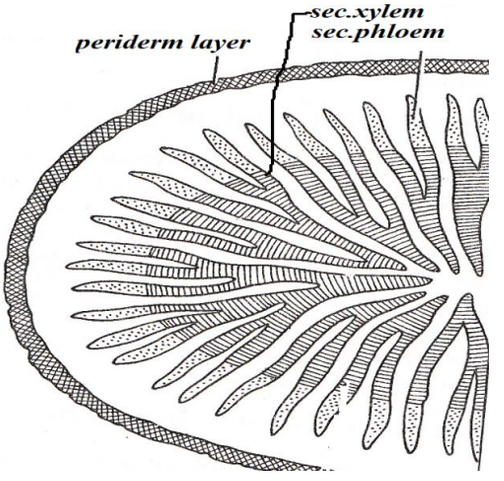


Fig.10.16 (D) : T.S. of stem *Aristolochia triangularis*

In case of *Aristolochia triangularis* the pith enlarges and the original vascular bundles assume a fan-like shape. The parenchyma present in between the bundles act like a shock absorber and also enables the stem to resist pulling and compression, resulting from pressure of high winds. **This specific trait arising as a result of anomalous activity of the cambium is thus an adaptation to the climbing habit of the plant.**

2. The cambium forms unusually large proportion of vascular tissues in the vascular region only. The cambium ring is formed normally by the union of the fascicular and inter-fascicular cambium. However**, the cambial ring behaves abnormally, in the sense that it is more active in certain regions and less active or inactive in other regions**. This peculiar growth pattern can be seen in *Bauhnia, Bignonia, Doxantha, Menispermum, Tinospora etc.*

(a) *Bauhinia* stem (Fam. - Caesalpinioideae) - In this plant, the cambial activity varies in different species. In B. *rubiginosa*, the cambium is more active at certain points but shows limited activity or no activity at other regions. In the regions of more activity, excessive secondary vascular tissue is produced, whereas in the regions of less activity, little or no vascular tissue is formed. **Such an abnormal growth pattern results in the formation of ridged secondary vascular cylinder and the mature stem exhibits a ridged appearance, externally**. In other species of *Bauhinia*, the activity of the cambium is limited to the two opposite poles only, resulting in an elongated flattened strap-like structure.

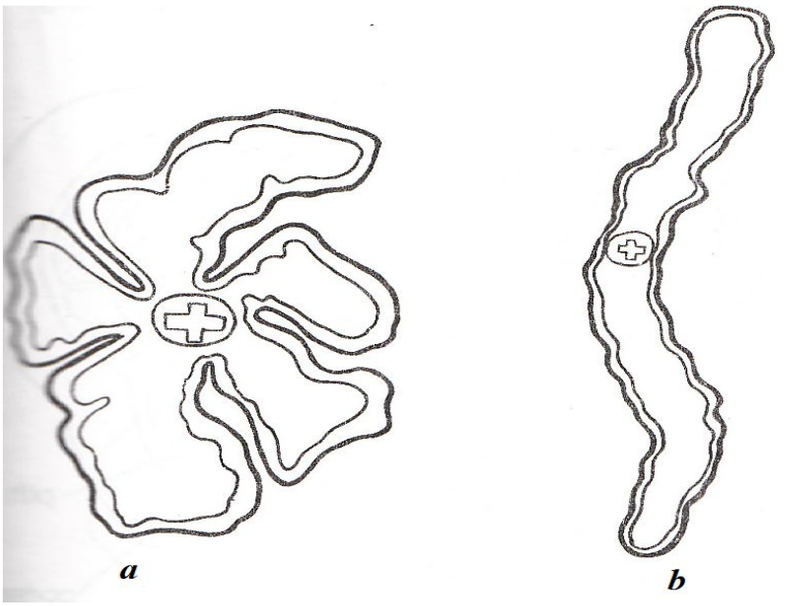
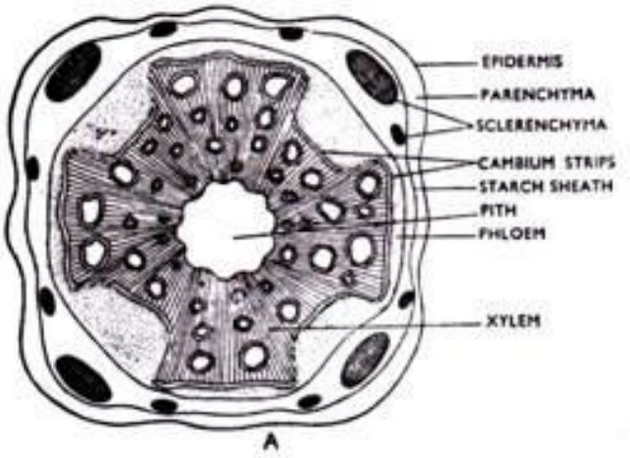
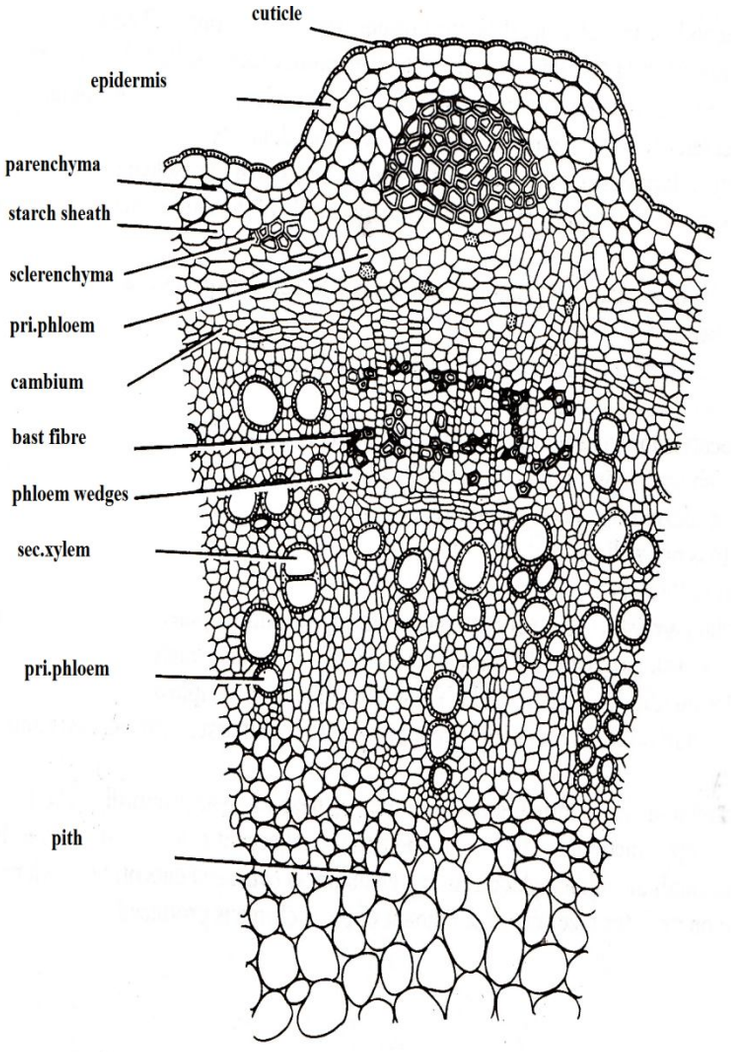


Fig. 10.17 : T.S. of Stem (a) *Bauhinia rubiginosa* (b) *B*auhinia

(b) *Bignonia* stem (Fam. - Bignoniaceae) - The young stem of *B.venusta*, a common garden climber, shows a wavy outline with prominent ridges and furrows. The thickly cuticularised epidermis is followed by the hypodermis which is collenchymatous or sometimes sclerenchymatous in the ridge region and sparsely chlorenchymatous beneath the furrows. The few layered parenchymatous cortex is followed by an ill - defined endodermis, supported by heterogenous pericycle. The vascular bundles are conjoint, collateral and arranged in a ring around prominent pith.

At the start of secondary growth, the fascicular and inter-fascicular cambium become active and join to form a cambium ring. This cambium ring behaves normally in the beginning, cutting more secondary xylem towards inner side and less secondary phloem on outer side. However, **after sometime the cambium ring develops unidirectional areas of cambium at four diagonal points**. Unidirectional area of cambium is that portion of the cambial cylinder that produces little or no secondary xylem and extensive amounts of secondary phloem. Hence at these four points lesser amounts of xylem are cut off internally, whereas massive amounts of secondary phloem are cut off externally. These phloem masses intrude inwards forming four deep wedges of irregular width and supported by transverse bands of sclerotic cells. After a short period, the central portion of the cambial strip, which was performing normal activity, develops unidirectional area of cambium and starts cutting off more phloem on the outer side as compared to the inner side. **This results in the formation of four or more wedges of phloem intruding into the xylem cylinders. The mature stem of *Bignonia* thus has four big - sized and four small-sized wedges of phloem projecting into the xylem.**





B

Fig. 18 : **T.S. of Stem *Bignonia***

A: Outline diagram, B: Cellular diagram

The cork cells are formed from the cork cambium which arises from the cortical cells. In later stages, the pericycle fibres are cast off after its activity and the stem assumes circular shape. These wedges of phloem are an adaptation to the plants mechanical requirement functioning as shock absorbers.

(c) *Doxantha* stem (Fam. - Bignoniaceae)—The anomalous pattern of secondary growth in D. *unguis - cati* was described by Dobbins (1971). A normal cambial ring develops in the earlier stages, which later develops unidirectional and bidirectional areas of cambium. Initially, the cambium behaves normally cutting off secondary xylem and phloem, that have a cylindrical configuration but later on four arcs of cambium are differentiated, one near each of the four major primary vascular bundles. These four arcs of cambium show unidirectional activity. i.e. little or no secondary xylem is produced but extensive masses of secondary phloem are produced, which project inwards in the form of four grooves or furrows. In the later stages, **portions of the cambial ring, except for these four arcs, exhibit bidirectional cambial activity i.e. they produce as much or more secondary xylem than secondary phloem.** The wedges of phloem produced by the unidirectional arcs of cambium dissect the secondary xylem and extend to the outer cylinder of the secondary phloem.

In older stems of *Doxantha*, additional furrows of phloem may be formed, either between the older existing wedges or directly adjacent to them. These phloem wedges function as shock absorbers allowing the stems of these lianas to bend under the strong action of winds.

3. The cambium forms irregular patches of parenchyma in xylem. This type of abnormal activity is seen in *Urtica dioica* stem.

(a) Urtica dioica stem (Fam. - Urticaceae) - The young stem of *Udioica* shows a typical dicotyledonous structure having a ring of conjoint, open and collateral vascular bundles. At an early stage, a normal cambial ring is formed which behaves normally cutting off secondary xylem towards inner side and secondary phloem towards outer side.

However, after sometime the cambium begins to behave abnormally at certain places only, cut off parenchymatous cells on the inner side, instead of secondary xylem. Alter cutting off a group of parenchymatous cells, the cambium again begins to behave normally and resumes its normal activity, forming secondary xylem above the parenchymatous group. The process is repeated again and again, resulting in the formation of *islands* of parenchyma which are embedded in the secondary xylem or wood. The parenchyma cells are irregular in shape, unlignified and elongated tangentially, giving a false appearance of

phloem. Usually, increased number and size of islands have been reported from spring wood. In later stages distintegration of parenchyma islands have been reported by Metcalfe and Chalk (1960) but no such disintegration was observed by Gupta (1960).

II. Abnormal cambium showing abnormal activity

In a number of dicot families such as Amaranthaceae, Chenopodiaceae, Nyctaginaceae etc. a new cambium ring or accessory cambium arises from the cortex or pericycle after the older, vascular cambium has ceased functioning or when the normal cambium ring does not develop at all. The cambial ring being placed abnormally functions in an abnormal manner, which may be of the following types:

(i) Formation of rings of vascular bundles embedded in parenchymatous tissue. This type of secondary growth has been observed in *Amaranthus, Mirabilis, Bougainvillea etc*. it occurs in the following manner.

(1) The first ring of cambium arises *de novo* in the pericycle region. Such a type of cambium, arising outside the vascular bundle is known as extra - stelar cambium.

(2) The extra - stelar cambium behaves abnormally, cutting off xylem patches alternating with parenchymatous patches on the inner side, whereas on the outer side, it cuts off continuous layers of parenchyma initially and later phloem patches over the xylem and parenchyma at the remaining places.

(3) After a complete ring of vascular bundle has been formed, the cambial ring becomes passive and ceases to function.

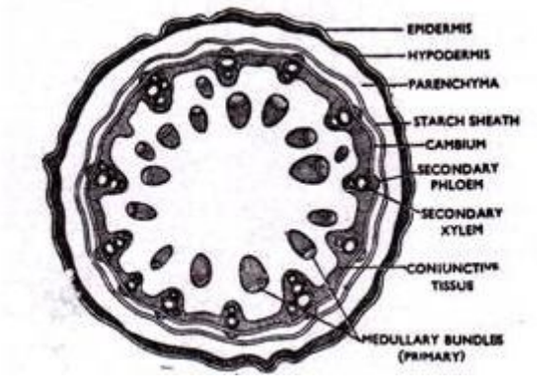
(4) A new ring of cambium (accessory cambium) is now formed from the parenchymatous cells cut off externally by the first cambium (extra - stelar).

(5) This cambium also behaves abnormally, similar to the previous one and cuts off a ring of vascular bundles separated by parenchyma. It then ceases to function and becomes passive.

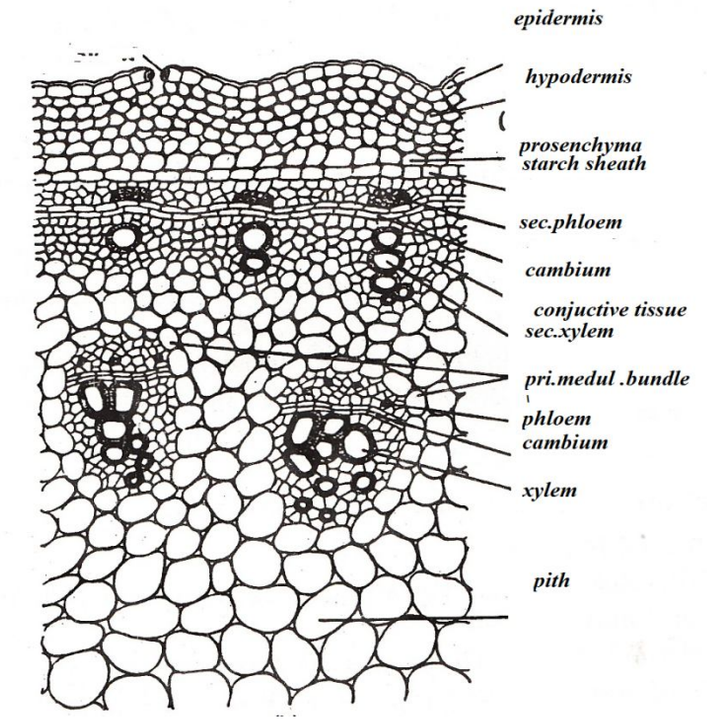
(6) Similarly, more accessory cambia are formed consecutively which give rise to consecutive rings of vascular bundles.

(a) *Amaranthus* stem (Fam. - Amaranthaceae) - The stem of *Amaranthus* exhibits abnormal activity by an abnormally placed cambial ring. The young stem has an outermost single layered epidermis followed by a few layered collenchymatous hypoderm is, parenchymatous cortex, endodermis and pericycle in small irregular sclerenchymatous patches. The stele is represented by two medullary bundles only. The normal ring of vascular bundles, typical of dicots, being absent, the cambial ring is not formed. However,

a layer of cells from the pericycle becomes active and forms a cambium ring (extra - stelar). This extrastelar cambium shows abnormal activity, as it initially cuts small amounts of parenchyma on the outer side and then starts cutting alternating patches of secondary xylem and parenchyma on the inner side. On the outer side, it cuts secondary phloem above the secondary xylem and parenchyma above the secondary parenchyma. These results in the formation of conjoint, collateral vascular bundles arranged in a ring and embedded in parenchyma.



A



B

Fig-19 : **T.S. of stem *Amaranthus***

A: Outline diagram, B: Cellular diagram

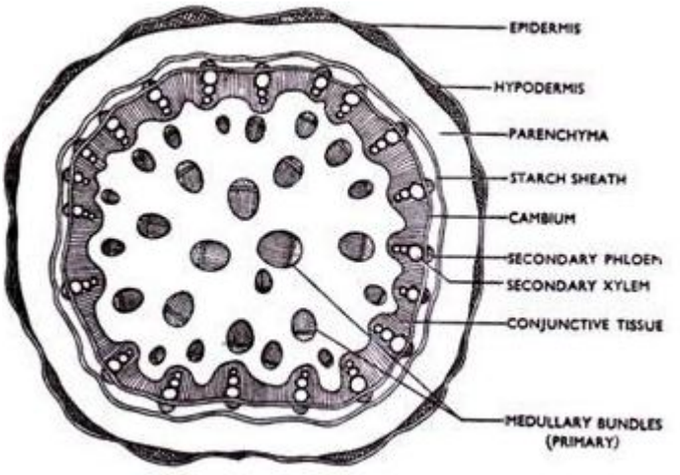
After the formation of these vascular bundles the cambium becomes inactive and stops functioning. However, a second accessory cambium develops from the secondary parenchymatous cells cut off by the first extra - stelar cambium. It behaves in a manner similar to the first cambium ring, i.e. cutting off vascular bundles embedded in parenchyma. The second ring of Vascular Bundle alternate in position to the first Vascular Bundle.

Similarly, more accessory cambia develop one after the other forming concentric rings of vascular bundles which appear as if scattered in ground tissue. The last cambial ring, however, behaves in a slightly different manner, as it cuts off secondary xylem alternating with sclerenchyma on the inner side. Hence, the last ring of vascular bundles appears to be embedded in sclerenchyma.

In mature stems, the two medullary bundles along with small amounts of parenchyma disintegrate forming a cavity. The cork cambium functions normally forming cork and secondary cortex.

(b) *Mirabilis* stem (Fam. - Nyctaginaceae) - The young stem of *Mirabilis* has two grooves opposite to each other, bearing numerous multicellular shoot hairs. Internally, the stem has a typical dicotyledonous structure with a single layered, cuticularised epidermis, a few layered collenchymatous hypodermis followed by multilayered parenchymatous cortex. The extra - stelar region is demarcated from the stelar region by a continuous layer of epidermis and a pericycle with patches of sclerenchyma. Numerous vascular bundles are scattered in the stelar region.

De Barry (1884) stated that a normal cambium ring is not formed but a completely new ring develops from the pericycle cells lying outside the vascular region. This view is supported by Mikesell and Popham (1977) who observed that the first cambium (extra-stelar) originates from the cells of the inner cortex or the pericycle. However, Maheshwari (1930) differs in his view and stated that the first cambium ring develops in the region of outermost vascular bundle.



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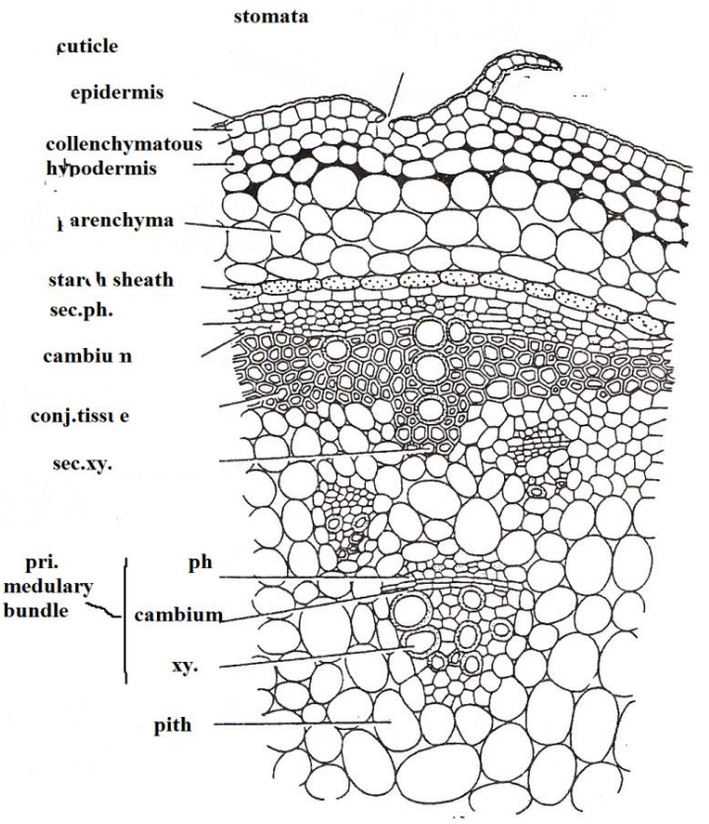


B

Fig-10.20 : T.S. of stem : *Mirabilis*

A: Outline diagram, B: Cellular diagram

This extra - stelar cambium forms secondary xylem alternating with parenchyma, on the inner side, and secondary phloem over the secondary xylem and outer parenchyma above

the inner parenchyma, on the outer side. A ring of conjoint, collateral vascular bundles is thus formed, separated by parenchymatous cells. The first cambium ceases its function after forming the ring of vascular bundles.

Successive rings of accessory cambia are formed, from the secondary parenchyma, cut off by the previous cambial ring and they behave in a manner similar to the first one. The last cambial ring cuts off sclerenchyma alternating with secondary xylem on the inner side and secondary phloem and parenchyma on the outer side. Hence the last rings of vascular bundles are embedded in lignified tissue and provide mechanical strength to the stem.

(ii) Formation of rings of vascular bundles embedded in conjunctive tissue. The cambial ring originates in the extra - stelar region and cuts off secondary xylem and thick - walled conjunctive tissue on the inner side and secondary phloem and conjunctive tissue on the outer side. The vascular bundles appear to be embedded in a mass of conjunctive tissue, e.g. *Bougainvillea, Boerhavia etc*.

(**a) *Bougainvillea* stem** (Fam. - Nyctaginaceae) The young stem of Bougainvillea has a circular outline formed of a single layer of thickly cuticularised epidermis, followed by a few layered collenchymatous hypodermis and a well developed parenchymatous cortex. The endodermis is indistinct and the pericycle is a heterogenous assemblage of parenchyma alternating with sclerenchymatous patches.

The primary vascular bundles, developing from the procambium strand, appear to be irregularly distributed in the central ground tissue. These bundles are actually the leaf trace bundles associated with the leaves, and may exhibit limited secondary growth accompanied by slight increase in size. The first cambial ring is extra - stelar in origin and arises from the pericycle (Esau and Cheadle, 1969 and Stevenson and Popham, 1973), followed by successive rings of cambia, however, Belfour (1965) stated that the entire secondary tissue is formed from a single cambium.

The cambial ring cuts off secondary xylem alternating with secondary parenchyma on the inner side and secondary phloem above secondary xylem and parenchyma above parenchyma on the outer side. The cells of the secondary parenchyma are fusiform, more or less radially arranged and usually develop lignified, thickened walls. These are referred to as conjunctive tissue or ground tissue (Metcalfe and Chalk, 1950).

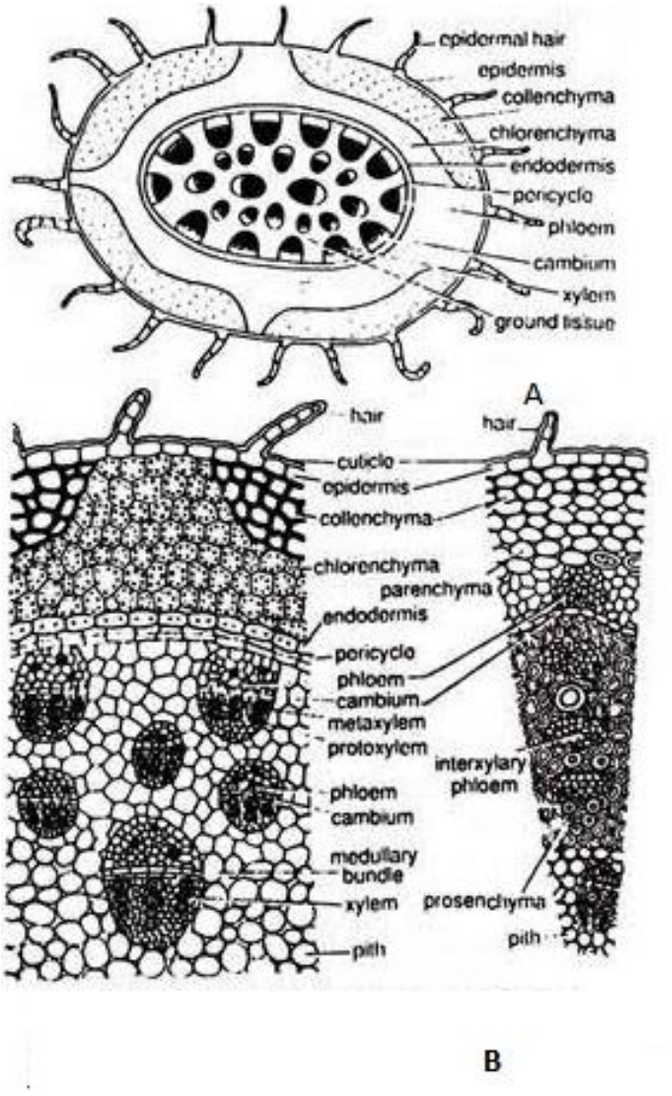


Fig. 10.21 : T.S. of stem : *Bougainvillea*

A: Outline diagram, B: Cellular diagram

Successive rings of cambia are formed which behave in a manner similar tothe first ring, as a result, concentric layers of vascular bundles are formed embedded in conjunctive tissue. This layering is conspicuous due to the prominent vessels in the xylem and the crushed cells in the oldest part of phloem . An interesting feature is that the new cambium does not arise as a complete ring around the periphery of the vascular cylinder but in the form of strips, which join with the older cambia. Thus, an ontogenetic continuity is maintained. In some species of *Bougainvillea*, the conjunctive tissue consists entirely of sclerenchyma and cannot be demarcated from the xylem of the embedded vascular bundles, hence the secondary phloem appears in the form of islands or isolated groups apparently surrounded by conjunctive tissue. They should not be mistaken as included phloem.

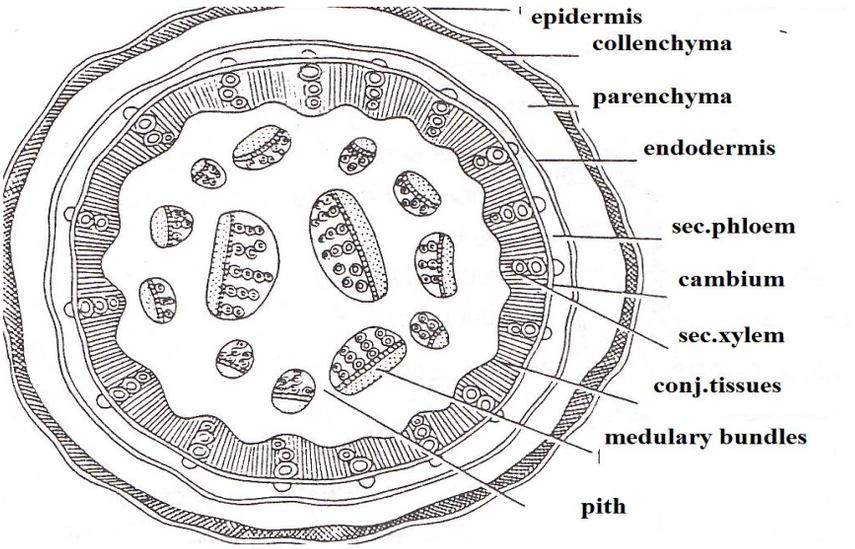
(b) *Boerhaavia* stem (Fam. - Nyctaginaceae) - The young stem of *Boerhaavia* is bounded by a single layered epidermis bearing numerous multicellular hairs. The hypodermis consists of 2 - 3 layers of collenchyma followed by parenchymatous cortex, interspersed with few chlorophyllous cells. An indistinct endodermis and 1 or 2 layered pericycle demarcates the well developed stelar region. In Boerhaavia, the vascular bundles are arranged in three rings (anomalous structure)—two large, centrally placed medullary bundles, middle ring of 6-14 loosely arranged bundles and an outer ring of small 15-20 bundles. The three rings of bundles originate from the pro cambial strands.

Secondary growth in the two medullary bundles and in the bundles of the middle ring is limited, resulting in slight increase in size only. The intra fascicular cambium of these bundles function normally, forming secondary xylem on the inner side and secondary phloem on the outer side, and as secondary growth progresses, the primary phloem gets crushed, forming a cap like structure of dead cells over the newly formed phloem.

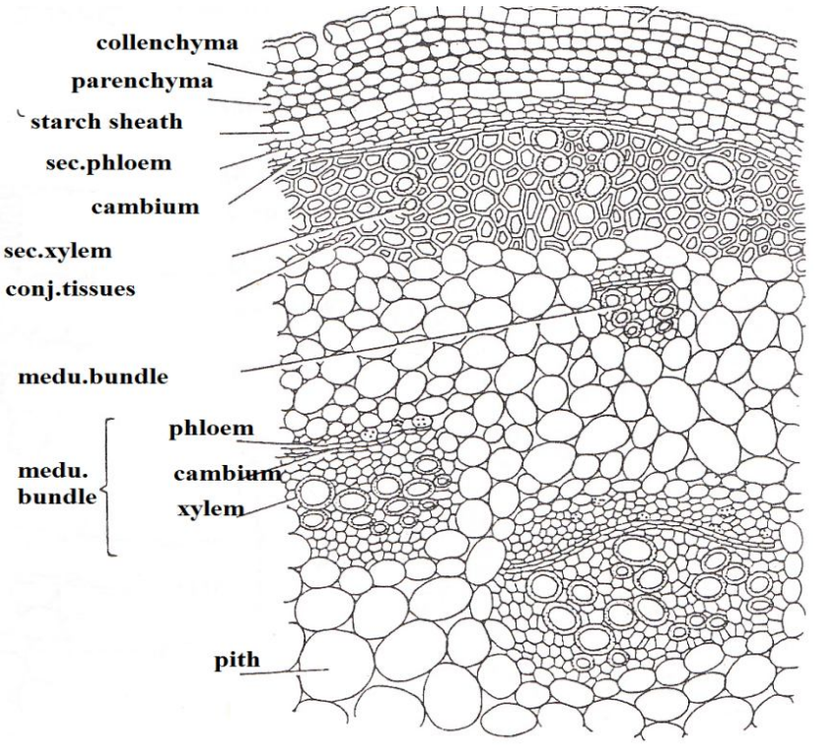
However, most of the secondary growth of the stem takes place from the cambial activity of the outer ring of bundles. The intra-fascicular and inter-fascicular cambiums join to form a cambium ring. Maheshwari (1930) observed that the ring forms secondary xylem in the intra-fascicular region and lignified, conjunctive tissue in the inter-fascicular region on the inner side, and secondary phloem above the secondary xylem and parenchyma opposite to conjunctive tissue, on the outer side.

After the formation of secondary tissue, the cambium ceases its activity and a new cambium ring arises by the joining of secondary parenchyma cells, opposite to the conjunctive tissue, and the pericycle cells outside the phloem. This first accessory cambium behaves similarly to the vascular cambium, cutting off secondary xylem alternating with lignified conjunctive tissue on the inner side and secondary phloem opposite to secondary xylem and parenchyma above the conjunctive tissue on the outer side. This results in the formation of a fourth ring of conjoint, collateral bundles of secondary origin.

Soon this supernumerary cambium ceases to function and a new accessory cambium develops which behaves in a similar manner. The process of formation of accessory cambium rings and secondary vascular bundles may be repeated till four or more concentric rings of vascular bundles are formed. The concentric rings of vascular bundles embedded in thick conjunctive tissue and separated by thin walled parenchymatous zone give the appearance of growth rings.



(A)



B

Fig. 10.22 : T.S. of stem : *Boerhaavia*

A: Outline diagram, B: Cellular diagram

III. Primary abnormal position of cambium showing abnormal activity

In various stems such as *Thinouia, Serjania, Paullinia etc*. the cambium has unusual position showing abnormal activity, and which results in the formation of anomalous structure during secondary growth.

(a) *Thinouia ventricosa* stem - In the young condition, the cambium is not in the form of a ring but appears as a folded structure. As the secondary growth starts, the cambium separates at the folds and forms individual groups of vascular tissues. These unusually positioned cambial rings form their separate vascular tissue resulting in a peculiar lobed structure of the stem.

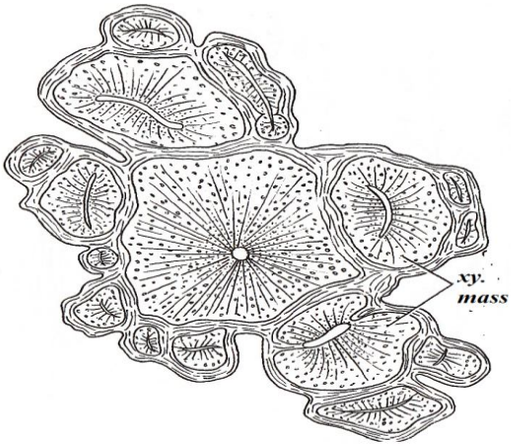


Fig- 10.23 (A) : T.S. of stem *Thinouia ventricosa*

(a) *Serjania corrugata* and *Paullinia* (Fam. - Sapindaceae)-_In these tendril climbers, specific groups of vascular tissue are constricted off from the main vascular cylinder. These constricted - off vascular tissues develop their individual cambium strips. With the initiation of secondary growth, each strip of cambium develops to form a complete and separate cambium ring, which functions normally forming secondary xylem on inner side and secondary phloem on outer side. Thus, the mature stem consists of a number of discrete vascular cylinders each of which develops its own periderm. In the mature condition, the stem seems to be made up of several smaller stems appressed to each other and giving the appearance of strands of rope.

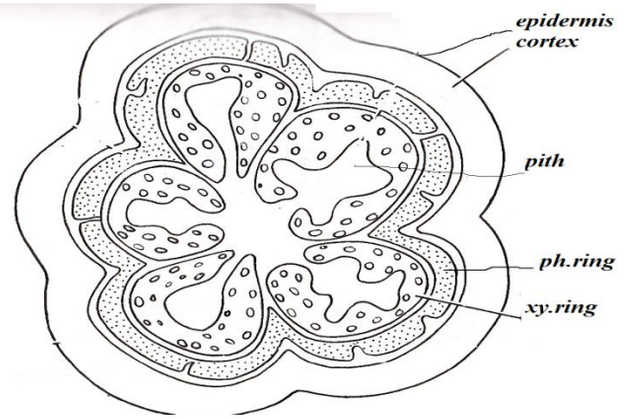


Fig-10.23 (B) : T.S . of stem *Serjania corrugate*

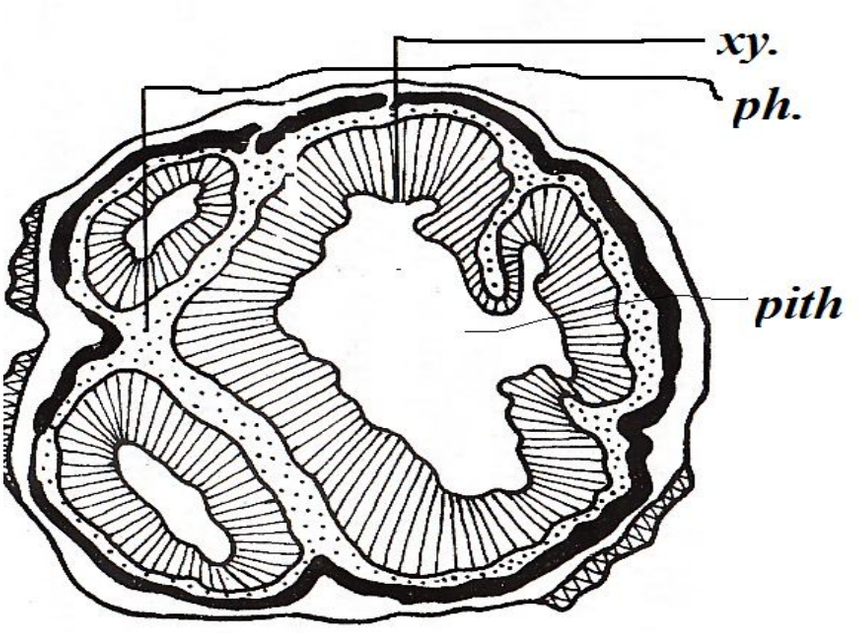


Fig. 10.23 (C) -T.S. of *Paullimia* stem

(C) *Bauhinia langsdorffiana* (Fam. - Caesalpiniodeae) - In this plant, the cambium is in the form of strips which do not unite but remains as such. At the time of secondary growth, these strips produce secondary xylem on inner side and secondary phloem on outer side, alternately mixed with secondary parenchyma. But the secondary parenchyma is produced in excessive amounts so much so that the vascular cylinder gets broken up into numerous parts resulting in fissured or small groups of xylem.

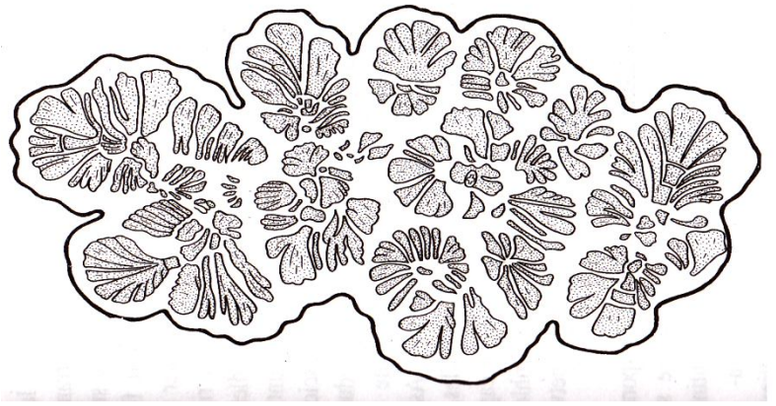


Fig – 10.23 (D) : T.S. of stem *Bauhinia langsdorffiana*

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IV. Formation of Included phloem

The abnormal activity of the cambium gives rise to groups of phloem cells embedded in the secondary xylem. This is known as included phloem or interxylary phloem. Formation of included phloem occurs by two methods -

(i) Formation and abnormal activity of arc cambium. The extra-stelar cambium formed, behaves abnormally forming secondary xylem and sclerenchyma on the inner side whereas on the outer side it forms secondary phloem above secondary xylem and parenchyma opposite to sclerenchyma. After sometime this cambium becomes inactive below the phloem region and new arcs of cambium are formed from the parenchyma lying above the phloem. This arc cambium also behaves abnormally, forming secondary xylem alternating with sclerenchyma on the inner side and secondary phloem and parenchyma on the outer side. As such, the secondary phloem formed by the first cambium gets enclosed by the xylem and sclerenchyma cut off by the arc of cambium. This enclosed phloem is known as included phloem or interxylary phloem. The remaining part of the cambium at rest of the places continues its original activity and finally comes to the level of the arcs of cambium. This type of formation of included phloem is termed as concentric type and has been observed in *Achyranthes, Chenopodium, Bougahwillea Celosia, Bosea, Pupalia, Strychnos* etc.

(a) *Achyranthes* stem (Fam.- Amaranthaceae) - The young stem has a wavy outline showing alternate ridges and furrows. The outermost layer is a single layered, cuticularised epidermis, followed by the hypodermis which is collenchymatous beneath the ridges but chlorenchymatous beneath the furrows. The endodermis is supported by the pericycle, which is multilayered with sclerenchymatous patches on the outer side. Two medullary vascular bundles lie opposite to each other in the pith

region and are decussately arranged in successive internodes. The position of medullary bundles was studied in *A aspera* by Srivastava (1960). He observed that the two bundles may be free only in the few upper internodes but fused in the lower internodes, or may remain free throughout the length of the plant. The two fused bundles present an amphixylic structure. In other species such as A. *coynei*, the number of medullary bundles may be four which after fusion forms two bundles. The normal cambium ring is not present but the accessory cambium develops from cells of the pericycle. It behaves abnormally, forming secondary xylem alternating with conjunctive tissue on the inner side and secondary phloem above xylem and parenchyma opposite to conjunctive tissue on outer side. After sometime, the cambium strip lying below the phloem becomes inactive and new arcs of cambium develop from the parenchyma lying above it. This newly formed arc of cambium similarly cuts off secondary xylem and conjunctive tissue on the inner side and secondary phloem and parenchyma on the outer side. As a result, the secondary phloem, formed by the first cambium gets enclosed by secondary xylem and conjunctive tissue and is known as included or interxylary phloem.

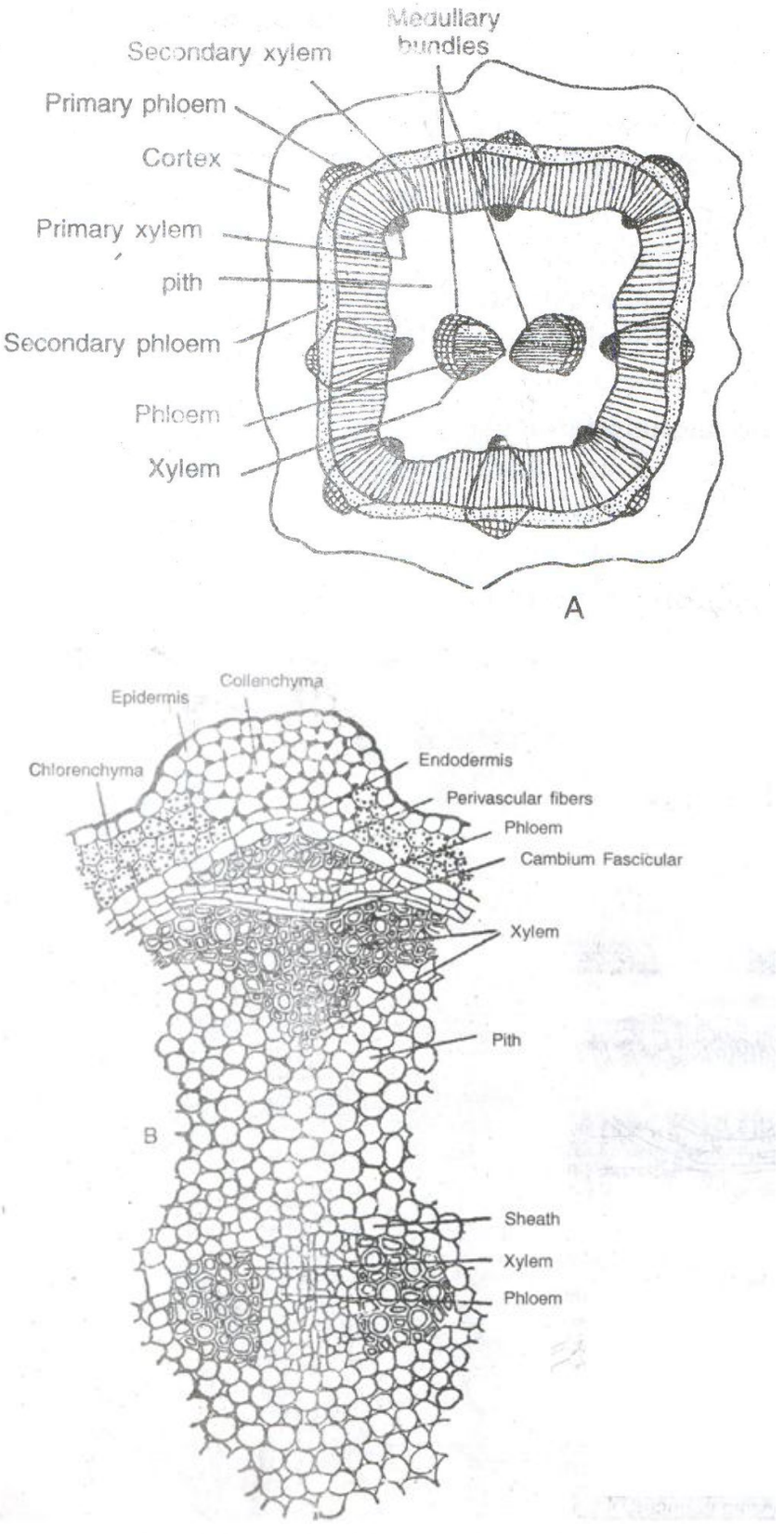


Fig. 10.24 : T.S. of Stem *Achyranthes*

A: Outline diagram, B: Cellular diagram

It has been observed that in certain species, the activity of cambium may change at intervals, forming secondary xylem in those regions where conjunctive tissue was being formed and conjunctive tissue where secondary xylem was being formed. Thus the secondary vascular bundles and the included phloem are found irregularly scattered.

(b) *Chenopodium* stem (Fam. - Chenopodiaceae)—The stem has a prominent ridged appearance with an outer, cuticularised single layered epidermis. The hypodermis consists of few collenchymatous cells beneath the ridges and chlorenchyma - containing cells beneath the furrows. The cortex is parenchymatous in nature followed by well defined endodermis and pericycle having patches of sclerenchyma. Vascular bundles are arranged in a ring around the parenchymatous pith. The normal cambium is not formed and secondary growth is initiated by the extra - stelar cambium originating from the pericycle cells. According to some workers this first ring of extra - stelar cambium first forms a layer of thin walled ground tissue and later the secondary vascular bundles. The primary vascular bundles, as a result of pressure from the ground tissue are pushed towards the central region and appear as medullary bundles. These primary bundles may have limited growth due to the formation of secondary tissue by their intrafascicular cambia.

The first extra - stelar cambium functions abnormally cutting off secondary xylem and conjunctive tissue on the inner side and secondary phloem and parenchyma on the outer side. The origin and function of the extra - stelar cambium is controversial. According to some workers this cambium remains active throughout the life of the plant and continuously forms secondary vascular tissue and conjunctive tissue; whereas other workers observed that the cambium is limited in its activity, hence resulting in the formation of successive arcs or rings of cambium which are responsible for forming secondary vascular tissue.

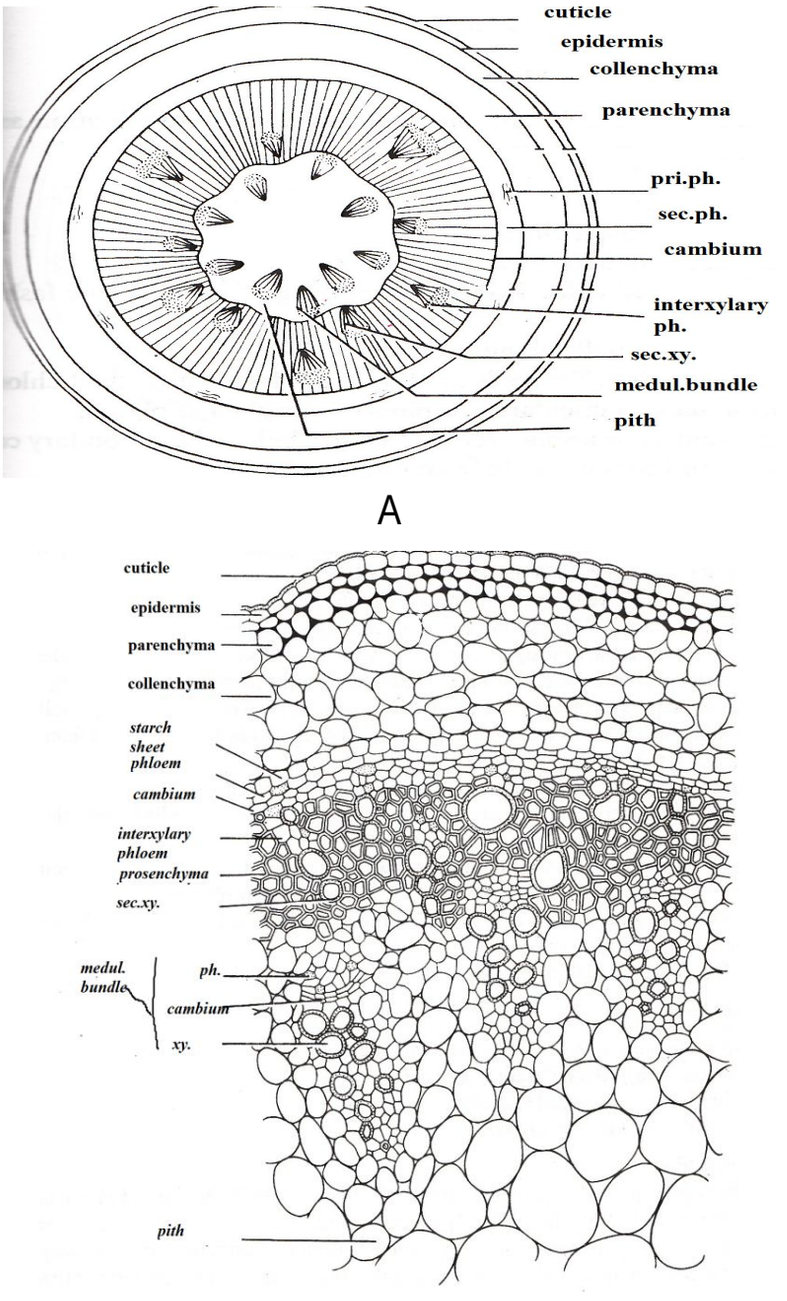


Fig. 10.25 : T.S. of stem *Chenopodium* (A: Outline diagram, B: Cellular diagram)

Balfour (1965) studied secondary growth in *Chenopodium* and observed that the inter - and intrafascicular arcs of cambium join to form a cambium ring, linking all the primary vascular bundles. This ring functions abnormally cutting off bands of fibres in the interfascicular region and xylem vessels and other xylem elements in the intrafascicular region towards the inner side, whereas externally the cambium forms a broad band of undifferentiated meristematic cells. This zone of undifferentiated meristematic cells increases in width, as result of which the primary phloem becomes embedded in it. Later, within the primary vascular bundle, a few layers of cambial cells, lying between the xylem

and phloem become non - functional and differentiate into parenchyma or sclerenchyma fibres. The outermost cambial cells now continue their secondary growth cutting off sclerenchyma fibres and xylem vessels internally. However, few cambial cells lying within the meristematic zone just external to the xylem vessels assume meristematic activity forming a group of phloem cells surrounded by lignified tissue. This meristematic zone functions indefinitely and it gradually moves outward, leaving behind isolated groups of phloem cells embedded in the conjunctive tissue. These isolated groups of Phloem cells are known as included phloem or interxylary phloem. The meristematic cells forming secondary vascular tissue are thus ontogenetically related to one another.

(c) *Bougainvillea* stem (Fam.- Nyctaginaceae)_Internal structure and formation of extra - stelar cambium and secondary vascular tissue has already been discussed .

Bougainvillea also shows the presence of included phloem, formed by the arc cambium method which takes place followingly.

The last ring of accessory cambium forms secondary xylem and conjunctive tissue on the inner side, but on the outer side, it becomes inactive, just below the phloem region. Hence arcs of cambium develop from the parenchyma cells lying externally. These arcs or strips of cambium behave in a manner similar to the accessory cambia, with the result, that the secondary phloem formed previously gets surrounded by the newly formed xylem and conjunctive tissue. This phloem is termed as included or interxylary phloem.

(d) *Strychnos* stem (Fam. - Nyctaginaceae) - The stem has a circular outline with an outermost layer of cuticularised epidermis, followed by one or two layers of collenchymatous hypodermis. The underlying cortex is distinguishableinto 2 regions, the outer composed of chlorenchymatous cells and the inner of parenchymatous cells. In the inner layers of the cortex, intercellular spaces are present. The endodermis is well defined supported by one or two layers of pericycle made of sclerenchymatous cells. The vascular bundles are arranged in a ring around the parenchymatous pith.

(ii) Formation of included phloem by a change in the activity of the cambium. In several families of dicots, such as Combretaceae, Malphigiaceae, Salvadoraceae, Asclepiadaceae, Acanthaceae, Gentianaceae etc. formation of included phloem occurs by a temporary change in the activity of the cambium. Under normal conditions, the accessory cambium forms secondary xylem on the inner side and secondary phloem on the outer side, but sometimes the cambium at certain regions behaves abnormally and a temporary change of activity occurs. These small strips of cambium instead of cutting off secondary xylem, now cut off thin walled parenchymatous tissue (*Leptadina and Salvadora*) or secondary phloem (*Combretum and Entada*) on the inner side. Later, they again resume their original activity, i.e., forming secondary xylem on inner side. This

deviation, from the normal function of forming secondary xylem, gives rise to the formation of phloem islands surrounded by secondary xylem and conjunctive tissue. These phloem islands are variously called included phloem or interxylary phloem

Included phloem has physiological significance in some xerophytic plants, since they retain and continue their function even during unfavourable conditions. These phloem islands serve to carry food to the axillary buds which on return of favourable conditions, develop into branches.

(a) *Leptadenia* stem (Fam. - Asclepiadaceae) - The young stem of *Leptadenia*, has a circular outline with an outer single layered cuticularised epidermis of compactly arranged cells. The epidermis is followed by hypodermis constituted of a few layers of collenchymatous cells. The underlying cortex is chlorenchymatous in nature followed by the layer of thin - walled endodermis. Lying beneath the endodermis, is the multilayered pericycle with patches of sclerenchyma. In the initial stage, conjoint, collateral vascular bundles are arranged in a ring around the parenchymatous pith. Secondary growth is initiated by the joining of inter and intrafascicular cambium to form the cambium ring. This cambium functions normally at first, cutting off secondary xylem towards inner side and secondary phloem towards outer side. After sometime, the cambium at certain points starts to behave abnormally cutting off parenchymatous cells towards inner side. This is, however, a temporary stage and soon the cambium reverts back to its original function of forming secondary xylem on inner side. This process is repeated several times resulting in the formation of a number of islands of thin walled tissue embedded in cells of secondary xylem. The cells of these islands, especially those in the centre, differentiate into sieve tubes and companion cells and the remaining cells form phloem parenchyma. Hence in the mature stem a number of phloem islands are formed.

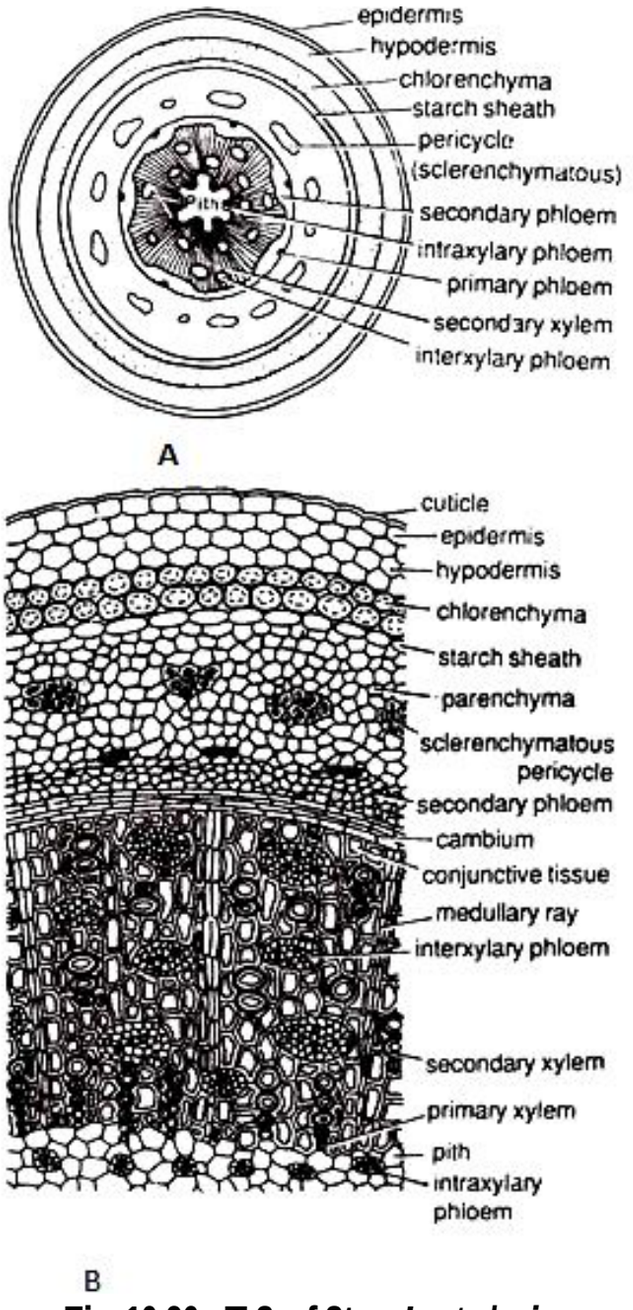


Fig. 10.26 : T.S. of Stem *Leptadenia*

A: Outline diagram, B: Cellular diagram

As interesting feature of *Leptadenia* is that the stem bears three types of phloem -(i) normal phloem (ii) interxylary phloem, and (iii) intraxylary phloem. The intraxylary phloem is present in the innermost region of the primary xylem. They may be formed by transformation of the parenchymatous cells of the pith or from xylem parenchyma

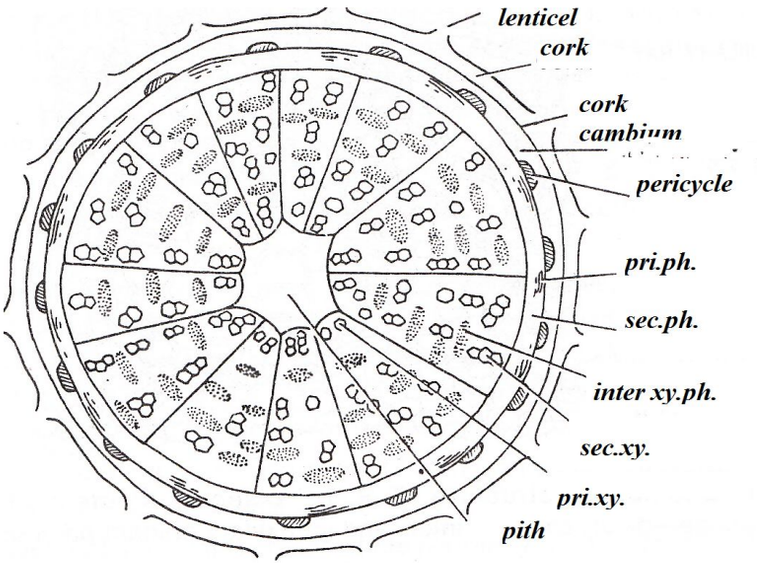
(b) ***Salvadora* stem** (Fam. - Salvadoraceae) - The stem is hard and smooth circular in outline. The outermost layer is the cuticularised epidermis followed by 2-3 layers of thin

walled cells of the hypodermis. The cortex lying beneath the hypodermis is constituted of parenchymatous cells having chloroplasts. The endodermis is a semi - distinct layer supported below by the heterogenous pericycle composed of parenchyma and groups of stone cells. The vascular bundles are typically arranged in a ring. The cambium ring is formed by the union of the interfascicular and intrafascicular cambium to form a single - layered cambium ring, which, however, appears multilayered due to formation of daughter cells.

The secondary growth in *S. persica* was described by Singh (1944). The cambium functions normally cutting off secondary xylem on inner side and secondary phloem on outer side. Alter sometime at certain points, the cambium begins to behave abnormally cutting off parenchymatous cells instead of secondary xylem on the inner side. This is, however, a temporary change and soon the abnormally behaving cambium strip reverts back to its original function of cutting secondary xylem on inner side. This abnormal behaving is repeated several times, resulting in the formation of a number of patches or islands of thin walled cells. The parenchymatous patches ultimately get converted to phloem parenchyma (peripheral cells) and sieve tubes and companion cells (central cells) giving rise to phloem patches termed as included phloem or interxylary phloem.

Secondary growth is initiated by the formation of the cambium ring, which at first functions normally cutting off secondary xylem on the inner side and secondary phloem on the outer side. However, after sometime small strips of cambium get detached from the main ring thus breaking up the continuity of the cambium cylinder. This breakage in continuity is, however, temporary as new complementary segments of cambium arise in the pericycle or in the peripheral region of the phloem above the non - functional segments of the cambium. These secondary formed cambium strips join the older, dissected cambium to form a complete ring. The cambium cylinder so formed again begins its normal function of forming secondary xylem on inner side and secondary phloem on outer side. Consequently, the phloem formed by the first ring gets enclosed by the secondary xylem formed by the newly joined cambium ring. This enclosed phloem surrounded by Secondary xylem is known as included phloem or interxylary phloem.

In each phloem group, a small segment of the original cambium remains embedded, which continues cutting off small amounts of secondary phloem on the outer side. As a result, the cells of (he older phloem or included phloem get crushed but remain in the form of a cap on the outer face of phloem islands. An additional feature of Strychnos is the presence of intraxylary phloem patches just below the primary xylem and which are capped by crescent shaped masses of sclerenchymatous cells. The intraxylary phloem may be formed by the transformation of parenchymatous cells of the pith or from the xylem parenchyma.



A

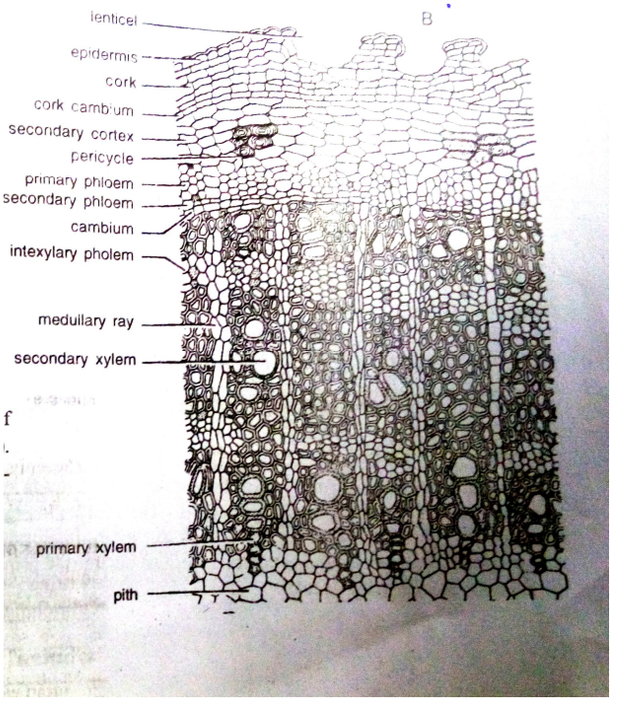


Fig- 10.27 : T.S. of stem *Salvadora*

A: Outline diagram, B: Cellular diagram

In certain plants, such as *Combretum and Entada*, included phloem is formed by a novel method. In these plants, a normal cambium ring is formedby the union of fascicular and interfascicular cambium which behaves in a normal manner, forming secondary xylem on inner side and Secondary phloem on outer side. After Sometime, small segments of the cambium behave abnormally cutting off small groups of phloem cells on the inner side instead of secondary xylem. These segments again assume their normal behaviour of

forming secondary xylem. These groups of phloem cells formed get surrounded by secondary xylem and are termed as included phloem or interxylaiy phloem.

Besides these, an interesting cast of abnormal secondary growth has been reported in *Adena palmata*, of family Passifloraceae (Swamy; 1975). The stem is tuberous and the cambium activity is normal giving rise to fan - shaped vascular tissue separated by parenchymatous tissue. However, after sometime, an additional inner cambium is formed on the inner side of the secondary xylem, which is active and forms medullary steles separated by parenchymatous tissue known as intermedullay stelar parenchyma.

V. Formation of interxylary cork

In several genera such as *Achillea fragmentissima, Artemesia tridentata and Epilobium angustifolium*, an interesting development occurs. The cambium ring cuts off secondary xylem and parenchyma towards the inner side. Soon it is observed that a layer of phellogen (cork cambium) develops in this parenchymatous zone between the secondary xylem. This layer of phellogen becomes active and cuts off cork cells which remain enclosed between two consecutive rings of secondary xylem, and is termed as *interxylary cork* In older stages, suberization of the medullary rays may occur or certain portions of the cambium may cease its activity resulting in the splitting of the stem.

Anomalous Secondary Growth in Monocot Stem

In monocotyledonous plants, secondary growth is usually absent as the vascular bundles are of closed type i.e. cambium is absent. But plants like *Dracaena, Yucca, Aloe arborescens, Sansevierja* (Fam. - Liliaceae), *Agave* (Amaryllidaceae) and *Lomandra, Xanthorrhoea and Kingia* (Xanthorrhoeacae) exhibit secondary growth which is anomalous in origin as well as in function. *Dracaena* plant may be considered as a typical example of anomalous secondary growth.

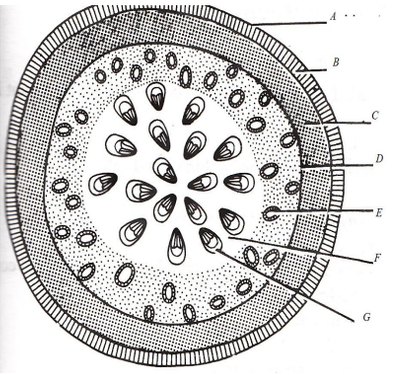
***Dracaena* stem** (Fam. - Liliaceae) - The young stem has a typical structure of an outer epidermis followed by sclerenchymatous hypodermis and a large number of closed, collateral vascular bundles embedded in parenchymatous ground tissue.

Secondary growth is initiated by the formation of a special meristematic zone which develops in the inner cortical region. The cortical cells lying external to the vascular bundle develop to form a multilayered secondary cambium. This cambium develops in that region of the stem which has ceased to elongate. The cells of the secondary cambium vary in shape and may be rectangular, fusiform or with one end flattened and the other narrow and pointed.

The activity of this cambium is abnormal as it cuts off a few cells on the outer side but a larger number of cells towards the inner side, i.e. towards the centre of the stem. The cells cut off towards outer side are parenchymatous in nature, whereas the cells cut off towards inner side are partly parenchymatous and partly vascular in nature. The parenchymatous cells cut off on the inner side develop lignification in their cell walls and are termed as *conjunctive tissue*.

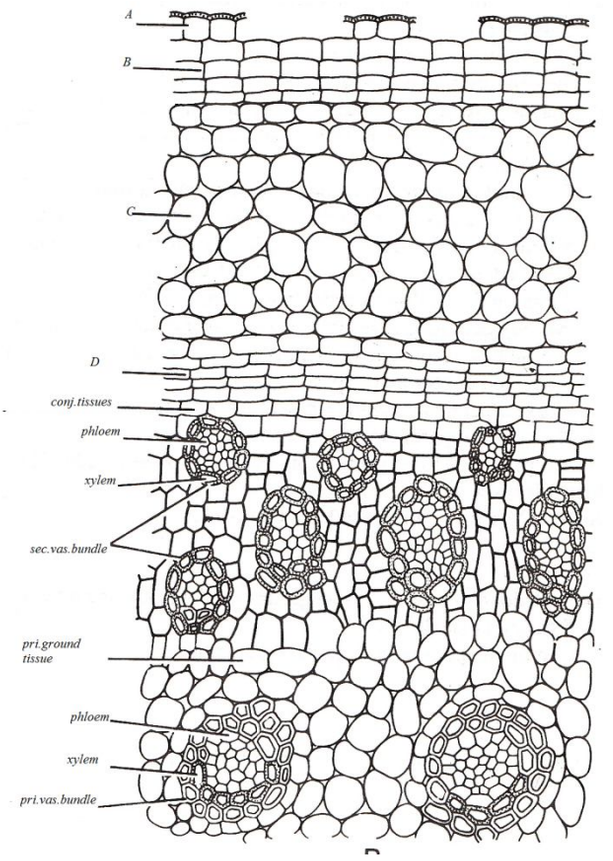
In the formation of secondary vascular bundle, usually cells from one or all the tiers of cambium participate. Sometimes, the median part of the bundle is formed from cells of one row and the lateral part from cells of the adjacent rows. However, it has been observed that secondary vascular bundle develops from a single cell, cut off by the cambial initial. This cell functions as the vascular bundle initial and divides by two anticlinal divisions 10 form a row of two or three colis. These cells further undergo periclinal divisions to form peripheral cells. Later divisions are irregular. The centrally situated cells of this group metamorphoses to form the phloem cells, whereas the peripheral cells differentiate into xylem element. The mature structure constituted of peripherally arranged xylem elements enclosing a central mass of phloem cells is act as amphivasal vascular bundle. The cambium then cuts of parenchyma cells on inner side which pushes the newly formed vascular bundle towards the central region.

After some time, the cambium again starts behaving abnormally, but this time the position of the vascular bundle cut off changes. The cambial initial which is to form the vascular bundle, now forms vascular elements, in these regions where it was forming parenchyma consequently the second row of vascular bundles is formed alternating in position with the first ring. Similarly, the activity and position of vascular bundles keep on changing resulting in rings of vascular bundles arranged in concentric rings by differing in position. Vascular bundles of the last ring are embedded in a mass of lignified, conjunctive tissue. The cork cambium may also develop normally and form cork cells on outer side.



A

A-Epidermis, B-Cork, C-Cortex,D-Cambium E- Pri.Vascular bundles
F-Groung tissues, G-Sec.Vas.bundle



B

Fig.10.28 : T.S. of Stem *Dracaena*

A: Outline diagram, B: Cellular diagram

In *Kingia*, development is more or less the same, except that the xylem is U-shaped, i.e. it surrounds the phloem on three sides only. The xylem of the vascular bundle is formed of tracheids and xylem parenchyma only. The vascular bundles are surrounded by a thin or thick sheath. In case of *Xanthorrhoea*, a lot ofresin is secreted in the parenchyma cells cut off on the outer side, as such the vascular bundle is surrounded by a resin sheath. In *Yucca brevifolia*, cambium develops from the cells of the pericycle which assumes meristematic activity. In some monocots, development of periderm has been reported as in *Aloe* and *Cocos*.

In some monocots, such as palms, rhizomes of *Musa* and bulbs of *Tulipa* and *Galanthus nivalis*, increase in thickness of the stem takes place by a special type of meristem known as primary thickening meristem. This meristem develops in the embryonic condition only from cells lying just beneath the leaf and sheath primordia. In the embryonic stage, it appears as a flat zone. In the initial seedling stage, it is concave and appears like a cane, in the later seedling stage it becomes flattened and finally in the young plant it again assumes a concave form. The meristem initials by periclinal divisions form a meristematic layer called primary thickening meristem which contributes to the height and girth of the stem. In the palms and rhizome of *Musa*, theprocambial strands are derived usually from this meristen. The procambial strands forni the vascular bundles which add to the thickness of the stem.

Another method of thickening of the stem n palms has been reported. In *Roystonea*, the parenchymatous cells in the centre of the stem as well as the undifferentiated cells of the bundle sheath constantly divide and increase in size. The intercellular spaces also grow in size resulting in the increase in thickness of the stem. This type of growth and increase in thickness of the stem is known as diffuse secondary growth.

10.6 Wood Development in relation to environmental factors

Wood is an important porous and fibres structure tissue found in woody plants dicot as well as monocots.This type of ood is used by humankind for fuel and construction purpose.Basically it is a natural composite of cellulose fibre embedded in matrix of lignin.here cellulose fibre provide strong tension while lignin resist compression.Anatomically wood mostly consist of secondary xylem and form during secondary growth.In a living tree it is supportive in function and enabling woody plants to grow largely and stand themselves in a properway.

It has been found that in temperate region, the cambium shows marked variation in its activity in different season. It has been found that in spring wood the vessels are much wider and relatively thin walled than in autmn wood.It has been observed that such

differences in the structure of wood is able to meet the varied requirement of the plant in spring and winter season.

For example, in the spring season the vegetative activity is at peak and so there is increase in the transpiring surface. Therefore in this period there is additional water channels are required to meet the requirement.As a result the spring wood has broader vessels. Whereas in autumn season there is just opposite environment, like no vegetatative growth and so no requirement of additional elaborate water conduction system so relatively narrow vessels are found.

10.7 Summary

Shoot is an important part of plant system .It bears leaf, branches, inflorescence, flower and fruit and seed in later stage. When leaf originates from the shoot apex some changes occurs in related tissues. After some times plant require strength ful tissue to help its increase weight so role of secondary growth is very important. In plants, the pattern of secondary growth shows a deviation from the normal type. The term anomalous secondary growth is given to this deviated pattern of secondary growth. Monocotyledonous plants, secondary growth is usually absent But plants like *Dracaena, Yucca,* and *Lomandra, Xanthorrhoea and Kingia* (Xanthorrhoeacae) exhibit secondary growth which is anomalous in origin as well as in function. In temperate region, the cambium shows marked variation in its activity in different season. It has been found that in spring wood the vessels are much wider and relatively thin walled than in autmn wood.It has been observed that such differences in the structure of wood is able to meet the varied requirement of the plant in spring and winter seas

10.8 Glossary

- **Abaxial :** Directed away from the axis. Opposite of *adaxial.* With regard to a leaf, the lower, or “dorsal,” surface
- **Adaxial :** Directed toward the axis. Opposite of *abaxial.* With regard to a leaf, the upper, or “ventral,”surface.
- **Annual ring :** In secondary xylem; growth ring formed during one season. The term is deprecated because more than one growth ring may be formed during a single year.
- **Anomalous secondary growth :** A term of convenience referring to types of secondary growth that differ from the more familiar ones.
- **Anomalous secondary growth :** A term of convenience referring to types of secondary growth that differ from the more familiar ones.

- **Apical cell :** Single cell that occupies the distal position in an apical meristem of root or shoot and is usually interpreted as the initial cell in the apical meristem; typical of seedless vascular plants.
- **Bark :** A nontechnical term applied to all tissues outside the vascular cambium or the xylem; in older trees may be divided into dead outer bark and living inner bark, which consists of secondary phloem. See also *rhytidome.*
- **Cambium :** A meristem with products of periclinal divisions commonly contributed in two directions and arranged in radial fi les. Term preferably applied only to the two lateral meristems, the *vascular cambium* and the *cork cambium*, or *phellogen.*
- **Diffuse-porous wood :** Secondary xylem in which the pores (vessels) are distributed fairly uniformly throughout a growth layer or change in size gradually from earlywood to latewood.
- **Earlywood :** Wood formed in fi rst part of a growth layer and characterized by a lower density and larger cells than the latewood. Term replaces *spring wood.*
- **Growth ring :** A growth layer of secondary xylem or secondary phloem as seen in transverse section of stem or root; may be an *annual ring* or a *false annual ring.*
- **Heartwood :** Inner layers of secondary xylem that have ceased to function in storage and conduction and in which reserve materials have been removed or converted into heartwood substances; generally darker colored than the functioning *sapwood.*
- **Histogen concept :** Hanstein's concept stating that the three primary tissue systems in the plant—the epidermis, the cortex, and the vascular system with the associated ground tissue—originate from distinct meristems, the histogens, in the apical meristems. See *histogen.*
- **Latewood :** The secondary xylem formed in the later part of a growth layer; denser and composed of smaller cells than the earlywood. Term replaces *summer wood.*
- living cells and reserves; it may or may not function in the conduction of water. Generally lighter colored that the *heartwood*.
- **Meristem :** Embryonic tissue region, primarily concerned with formation of new cells.
- **Meristematic cell :** A cel synthesizing protoplasm and producing new cells by division; varies in form, size, wall thickness, and degree of vacuolation, but has only a primary cell wall.

- **Periderm :** Secondary protective tissue that replaces the epidermis in stems and roots, rarely in other organs.Consists of *phellem* (cork), *phellogen* (cork cambium),and *phelloderm*.
- **Phellem (cork) :** Protective tissue composed of nonliving cells with suberized walls and formed centrifugally by the phellogen (cork cambium) as part of the periderm. Replaces the epidermis in older stems and roots of many seed plants.
- **Phelloderm :** A tissue resembling cortical parenchyma produced centripetally by the phellogen (cork cambium) as part of the periderm of stems and roots in seed plants.
- **Phellogen** (**cork cambium**) **:** A lateral meristem forming the periderm, a secondary protective tissue common in stems and roots of seed plants. Produces phellem (cork) centrifugally, phelloderm centripetally by periclinal divisions.
- **Phyllotaxy** (or **phyllotaxis**) **:** Mode in which the leaves are arranged on the axis of a shoot.
- **Primary phloem :** Phloem tissue differentiating from procambium during primary growth and differentiation of a vascular plant. Commonly divided into the earlier *protophloem* and the later *metaphloem*. Not differentiated into axial and ray systems.
- **Quiescent center :** Initial region in the apical meristem that has reached a state of relative inactivity; common in roots.
- **Sapwood :** Outer part of the wood of stem or root containing
- **Vascular bundle :** A strand-like part of the vascular system composed of xylem and phloem.
- **Vascular cambium :** Lateral meristem that forms the secondary vascular tissues, secondary phloem and secondary xylem, in stem and root. Is located between those two tissues and, by periclinal divisions, gives off cells toward both tissues.
- **Xylem :** Principal water-conducting tissue in vascular plants characterized by the presence of tracheary elements. The xylem may also serve as a supporting tissue, especially the secondary xylem (wood).

10.9 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. What do you understand by porou wood?
2. What hi Bar?
3. Differeent is amphibribal and amphivosal vascular bundles.

Section C : (Long Answer Type Questions)

1. Decribe the internal structure of a dicot stem with suitable diagrams.

2 What is Anomalous Secondary Growth describe with suitable diagrams

3 Write short notes on the followings----(a)Shoot apex organization (b) Growth Rings (c) inter xylary pholem

Section B : (Short Answer Type Questions)

1 What is Medulary bundles?

2 What is medulary rays?

3. What is rib meristem?

4. What is Heart Wood ?

5. What is SapWood ?

10.10 References

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Unit-11

Root Development

Structure of the Unit:

11.0 Objectives

After going through this unit you will be able to understand about:

- structural variation in root of various groups of plant
- structural modification in anatomy of root in response to habitat
- secondary growth in root of plants and its utility

11.1 Introduction

Root as important part of plant usually found in the soil ,grow towards gravitational force and grow against light .Its main role is to absorb water ,minerals and whatever is present in soil like fertizer provide artificially. Its surface is non-green. During germination radical is the first organ to grow in the seedling and give rise to primary or tap root system later on. It s important is anchoring the plant also, from small seedling level to became of large tree. To helpful in all activity of such type plant showing many anatomical modifications describe in following paragraphs.

11.2 Root Apical Meristem

Root Apex Organisation

According to old concepts that apices of shoot and root represent the earliest self perpetuating promeristem made up of homogenous maristematic tissues. Later on modern workers give the opinion that a distinct zonation can be seen by the immediate derivatives of the promeristem.According to them that there is distinct zonation of distinct region differing one another by nature of cells, plane of cell division ,position of initiating cells. Usually the root apex organization is less complex than the shoot because of absence of node, internodes and lateral appendages.

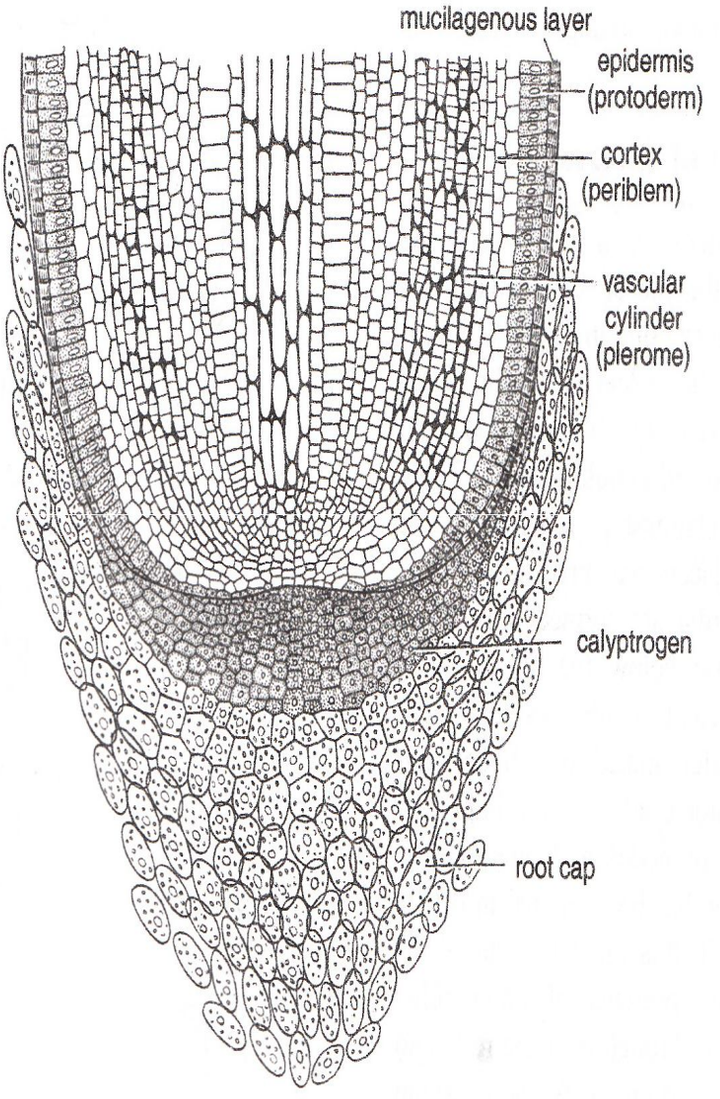


Fig. 11.1: L.S. of Root Apex

Since the nineteenth century was following important theories have been proposed to describe the root apex organisation.

1. Apical cell theory. Nageli (1958) proposed this concept and according to him that a single tetrahedral apical cell in the root apices brings about growth. It has three cutting faces for giving rise to the tissues of the body of the root. The root cap is derived from its base. Such a proposition was not found true to angiosperms. However, different types of root apices have been found in angiosperms which are schematically shown in figure

2. Histogen theory. Hanstein (1868) proposed this concept and according to him that in root apex regions, three distinct histogens are found. The **dermatogen** gives rise to epidermis, the **periblem** to the cortex and the **plerome** to the vascular cylinder. Later on, Haberlandt (1914) proposed the terms *protoderm, ground meristem* and *procambium*, respectively for these histogens

3. Körper-Kappe theory. Schuepp (1917) proposed this concept and according to them that the cells at the root apex divide in two planes. The zone with *inverted 'T'* type of division was referred to as Körper (cap) whereas other with *straight 'T* as Kappe (body). The theory basically recalls the tunica corpus concept of shoot apex and similarly, it also fails to explain the differences in behaviour in different species.

4. Concept of quiscent zone

Clowes (1958) in his autoradiographic studies of DNA synthesis in the root tip of *Zea mays* noted a central cup-like region, the cells of which had fewer mitochondria and ER, very small nuclei and low rate of DNA and protein synthesis. He referred this inactive region as **quiscent centre**. Since then such regions have been observed in many plants. It consists of arround 500 cells in *Zea mays*, 600 in *Sinapsis alba* and 1100 in *Vicia faba*.

In general, cells of quiscent region do not divide, but may become active when active initials are damaged. Thus, this region acts as reservoir of cells. These undividing cells offer more resistance to damages and irradiations than actively dividing cells and under certain conditions may divide actively. Clowes (1961) proposed that the cells of quiscent centre are inactive because of their position in the apical meristem, and not due to any inherent inability to undergo divisions.

In general, the differentiation of different tissues is schematically shown in figure.

11.3 Primary Structure of Root- Dicot & Monocot

Primary Structure of Root

Root having almost uniform environment under soil so it shows a simple internal organisation than stem, absence of nodes, and internodes, and lateral appendages.

The following three zones can be distinguished in root in the primary state of growth –

(1) **Epidermis**

(2) C**ortex**

(3) **Vascular System**

(1) **Epidermis—**It is also known as epiblema it is a continuous layer composed of tightly placed, thin walled non-cutinised cells. In some herbaceous perennials, where epidermis is long lived and acts as a chief layer, the outer tangential walls of epidermal cells are cutinised.

It is usually **uniseriate**, except aerial roots of some orchids and epiphytic aroids where it is multiseriate and called **velamen** .The velamen is composed of compactly arranged non-living cells which often have secondary wall thickenings. It is primarily an absorptive tissue and also acts as mechanical tissue which protects and checks loss of water from the cortex. The cells of velamen are filled with air in dry season, but in rainy season they are filled with water. Some specialized groups of cells with dense spiral thickenings are also present in the velamen. These are known as **pneumatodes** and they help in gaseous exchange.

In young roots Epidermis bears many unicellular **root hairs** .The root hairs are initiated a few millimetres from the root apex. As the root grows, hairs present on the more mature portions usually die and dry out and new hairs continue to develop in relatively young parts of the root. Sometimes, root hairs are persistent e.g. *Eupatorium purpun*.

All the epidermal cells of the root are capable of initiating root hairs but only a few form root hairs. The cells of the epidermis, which form root hairs, are sometimes morphologically distinct from the other cells. These cells are known as **trichoblasts**. A root hair originates bump on the epidermal cell and this bump has cytoplasm and nucleus towards its tip.

2. Cortex. It is situated just internal to epidermis, simple and homogenous type and made up of thin walled, is diametric, parenchymatous cells having several intercellular spaces. In aquatic plants e.g. *Hydrilla*, it is largely aerenchymatous enclosing air cavities. Sometimes, air cavities develop in the root by breaking down of some cortical cells.

Cortical cells usually store starch. Chloroplast is not found but it may be found in cortical cells of some aquatic plants e.g. *Trapa*. Some of the aerial climbers such as *Tinospora* have got photosynthetic roots, and the chloroplasts are found in their cortical cells.. The arrangement of cells may also vary, e.g. *Avena sativa*, cortical cells are arranged in distinct radial rows. In *Commelina*, the cells of inner cortex are arranged in the form of concentric rings. The cortical cells in roots of several gymnosperms and some members of

families like Cruciferae, Rosaceae have lignified reticulate thickenings. In most monocotyledons, some dicotyledons and gymnosperms, the sub epidermal cortical layers differentiate into protective tissue, called exodermis which consists of suberised wall. Some of the exodermal cells show the presence of Casparian strip. Exodermis is distinctly developed in different species of *Iris*.

3. Endodermis, It is the innermost layer of cortex and is quite distinctive in roots. Endodermis is of universal occurrence in roots and is composed of closely packed living cells. Two types of cells are found, one which have characteristic casparian thickening in their radial and transverse walls and other which are thin walled and found opposite to the protoxylem points. These thin walled cells are also called passage cells. The Casparian thickening is formed due to suberin and lignin deposition.

Endodermis together with cortex is cast off with secondary growth in dicot roots.

4. Stele. It includes pericycle, vascular bundles and pith.

(a) Pericycle - It occurs on the inner side of endodermis and forms the outer boundary of primary vascular bundles. It is usually uniseriate but multiseriate pericycle is found in *Salix* and *Smilax*. However, it may be even absent in certain cases such as in some parasites and few hydrophytes. In most cases it is parenchymatous in nature but In *Smilax* this layer consists of thick walled cells.

Schizogenous secretory canals are also reported in pericycle of members of Umbelliferae.

The pericycle originates quite early from procambial strands which retain meristematic properties. It functions as site of origin of lateral roots, especially before secondary growth. During secondary growth, it forms a part of fascicular cambium and plays important role in completing the cambial ring, particularly opposite the protoxylem points, in addition to that, the entire initial cork cambium (phellogen) originates from it.

(b) Vascular system - The vascular bundles found in roots show radial arrangement. Xylem elements show exarch condition. The development of xylem is centripeta1 type. The number of vascular bundles ranges from one to eight in dicotyledonous roots and eight to twenty in monocotyledonous roots. Depending upon number of xylem bundles or patches roots may be called *monarch*, *diarch*, *triarch*, *tetrarch* or *polyarch.* The number of protoxylem elements in each bundle is very few, usually; there is a single protoxylem vessel with annular or spiral thickening. The metaxylem is composed of polygonal vessels. There may be some variation in number of protoxylem and metaxylem vessels, e.g. *Sisyrinchium* has a single large central metaxylem vessel surrounded by five to seven strands of protoxylem vessels. In *Gladiolus* and *Freesia*, there is present a single metaxylem vessel at the base of each protoxylem strand.

Phloem consists of sieve tubes, companion cells and phloem parenchyma.The vascular bundles are separated from each other by conjunctive tissues composed of parenchymatous cells.

(c) Pith - Pith occupies a small central area of stele and consists of thin walled parenchymatous cells with intercellular spaces. In older roots of dicots, the pith is altogether absent. It is well developed in monocotyledonous roots which may also contain a few tannin cells. Sclerenchymatous pith is found in certain members of iridaceae.

Anatomy of Dicot Root

For studying anatomical features of a typical dicot root, young root of Cicer, sunflower (*Helianthus annuus*) etc material are generally selected.

The following important anatomical features are noted in the typical dicot root.

1. **Epidermis**. It is the outermost uniseriate layer composed of thin walled, compactly packed, parenchymatous cells without intercellular spaces. Here and there, the ridace view epidermal cells give rise to unicellular root hairs. Epiblema of roots is characterised by absence of cuticle and stomata. Root hairs absorb water and minerals from soil.
2. **Cortex.** It lies below the epiblema and is homogenous and well developed. It is composed of thin walled, loosely arranged, parenchymatous cells with large number of intercellular spaces. However, outer few layers may be compactly arranged. These cells contain amyloplasts (leucoplasts) and store the starch in the form of starch grains.
3. **Endodermis.** It is the innermost layer of cortex which surrounds, the stele. Endodermis is composed of barrel-shaped, parenchymatous cells without intercellular spaces. Most of the cells, except those lying opposite the protoxylem point, possess strip-like thickenings of suberin and lignin on their radial and tangential walls which are known as Casparian strips. Cells lying opposite the protoxylem elements are thin walled and are called passage cells because these allow the passage of water from cortex to xylem.
4. **Stele.** It is tetrarch actinostele (protostele) composed of pericycle and vascular bundles. In general following parts are observed.

   (i) Pericycle -The layer present on inner side of endodermis and outer most of stele is the pericycle. It consists of a single uniseriate layer of thin walled parenchymatous cells containing dense protoplasm. It is usually two to three

layered opposite the protoxylem elements it shows the point of origin of lateral roots.

(ii) Vascular bundles - These are four in number which are arranged in the form of a ring. Vascular bundles are radial type, i.e. xylem and phloem are placed on different radii and alternate in their position.

Xylem is conical is shape with metaxylem vessels towards centre and protoxylem towards periphery, i.e. exarch in nature. In mature and well developed roots, the metaxylem elements meet in the centre and as result, the pith gets obliterated. Fibres and xylem parenchyma are absent. Xylem tracheids are found present around the vessels.

Phloem lies alternate to xylem patches on separate radii, It Consists of sieve tubes, companion cells and pholem parenchyma. The phloem fibres are absent. Phloem is present in the form of small patches, the outer part (towards pericycle) being *protophloem* and inner part (towards metaxylem) *metaphloem.*

(iii) Conjunctive tissue - These are parenchymatous in nature which lies in between patches of xylem and phloem.

(i) *Pith* - In young stege the central part of stele is the pith which consists of few parenchymatous cells compactly arranged without intercellular spaces. In older root, pith may be altogether absent.

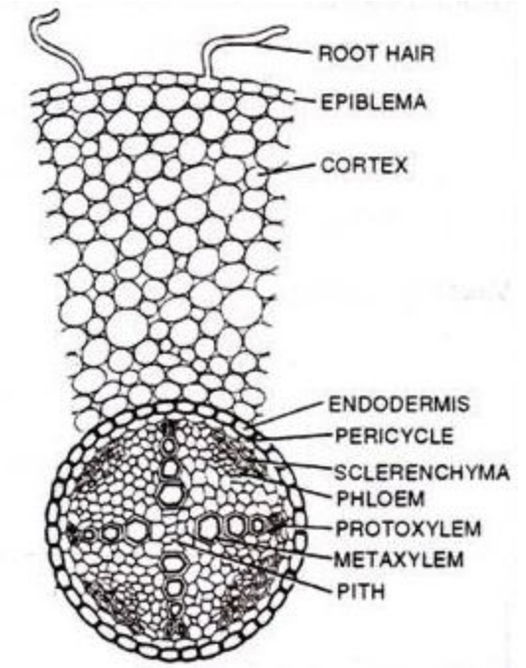


Fig.11.2 : T.S. of a typical Dicot Root *Cicer*

Anatomy of Monocot Root

The root of *Zea mays* may be selected to study the anatomical features of typical monocotyledonous roots. Sections are cut and double stained for study.

The internal structure of maize root shows following parts-

1. **Epiblema, Cortex** and **Endodermis** of monocot roots are structurally similar to those found in dicot roots. in maize, endodermal cells are thick walled and casparian strips are indistinct.
2. **Stele:** It is actinostele (protostele) which shows following features.

   (i) Pericycle - It is single layered lying on the inner side of endodermis. It is partly composed of parenchymatous cells and partly of sclerenchymatous cells.

   (ii) Vascular bundles - These are found in large number (more than eight) and each one is radial type (as described in dicot root). These are arranged in a ring around the central pith.

   Xylem is composed of single rounded metaxylem vessel towards the pith and one or sometimes two small, polygonal or rounded thick-walled protoxylem vessels towards the periphery. So the xylem shows exarch arrangement.

   Phloem lies in form of small patches alternate to xylem and is composed ofsieve tubes, companion cells and phloem parenchyma.

   Parenchyma cells associated with xylem undergo sclerosis and thus become thick walled.

   (iii) Pith - Pith occupies the central portion of stele. it is composed of thin walled parenchymatous cells containing abundant starch grains. Cells are loosely arranged.

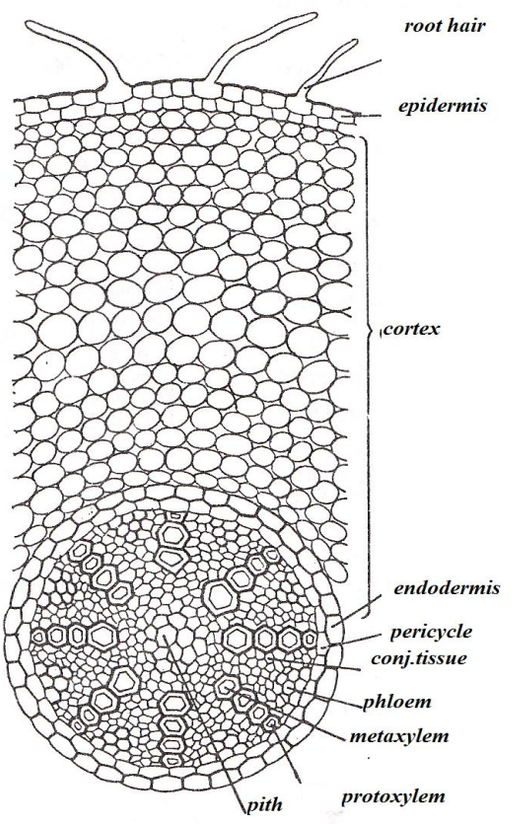


Fig.11.3 : T.S. of Root *Zea mays*

11.4 Lateral Roots and Root hairs

Development of Lateral Roots

Lateral roots are initiated behind the apical meristem, typically in the region of maturation. However, in some aquatic plants, such as *Eichhornia crassipes*, the lateral root primordia are formed close to the root apex.

The lateral roots of both gymnosperms and angiosperms are usually initiated in the pericycle opposite to the xylem ray and subsequently grow through the root cortex. The position of the lateral root varies depending upon the position of vascular strands in the parent root. In diarch root, it arises between the phloem and xylem strands in tri-, tetra-, and pentarch roots, opposite the protoxylem and in polyarch roots opposite the protophlocm strands.

Primordia of lateral roots are formed just behind the region of root hairs. At the site of the future primordium cells of the pericycle become meristematic and divide periclinally. These initial divisions are followed by divisions in both periclinal and anticlinal planes. These divisions establish a primordium of the lateral root.

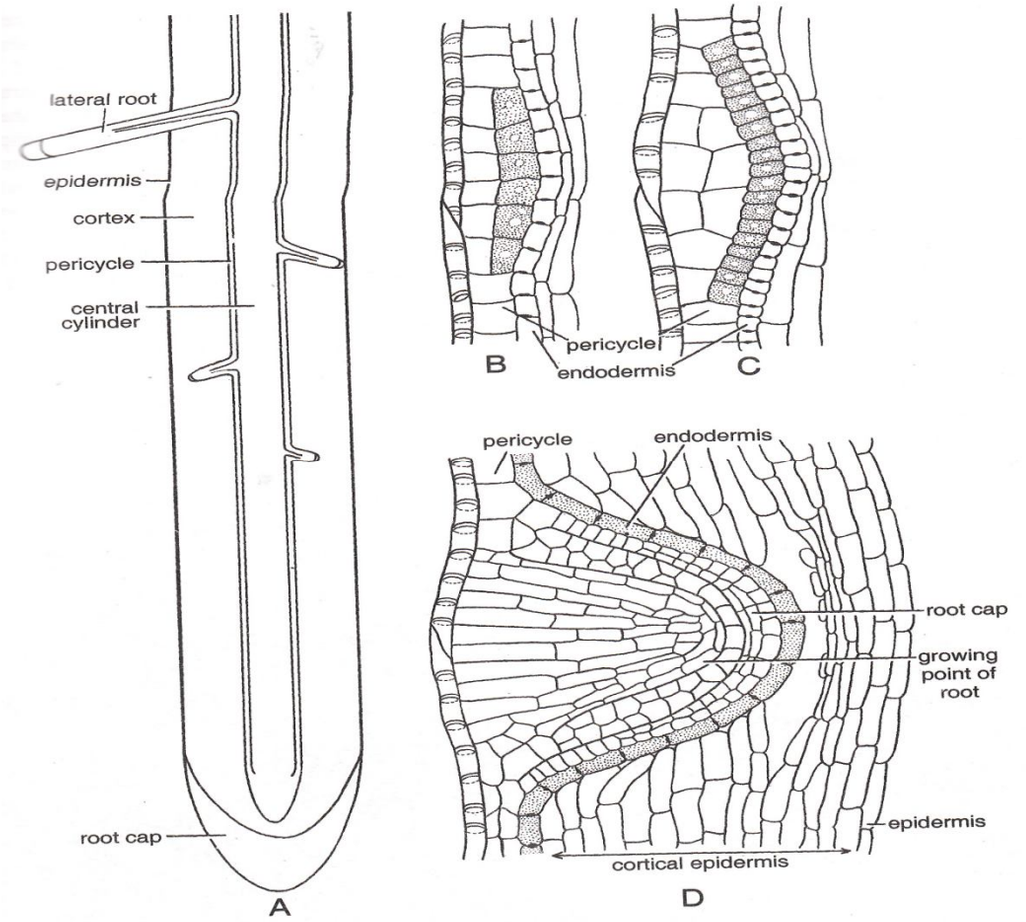


Fig.11.4 : Various stages of initiation and development of lateral roots in L.S. view

Sometimes, endodermis also takes part in the formation of lateral root primordia, e.g *Daucus carota* and *Zea mays*. In such cases the endodermis divides predominantly anticlinally but some periclinal divisions may also occur. Young lateral root grows, through the cortex and epiblema and passes into the soil. During this process the cells of the cortex are crushed and pushed aside. The passage of lateral roots through the cortex is mainly by mechanical penetration but enzymes may also facilitate the growth of lateral roots through the cortex. The vascular elements differentiating in the lateral roots become connected with that of parent root through the cells of the pericycle. Root cap and promeristem of the growing lateral root develop when it is still within the cortex.

Usually, the lateral roots usually develop in aero- petal order, i.e. older at the base and younger towards the apex.

In mature roots, where pericycle has been sloughed off, lateral roots commonly initiate in the cambium.

11.5 Root-Microbe Interactions

In many plant species it has been found that microorganisms are responsible for Nitrogen fixation in roots. In some tropical trees these are blue green algae while in other species actinomycetes like organism (filamentous bacteria) carry out this process.In Legumes, bacterial species of the Rhizobium are responsible.

The Rhizobia are aerobic bacteria that persist saprophytically in the soil until they infect a root hair.Root hair usually respond to invasion by surrounding the bacterium with athread like structure called infection thread. This infection thread consists of infolded and extended plasma membrane of cell being invaded, along with new cellulose formed on the inside of this membrane. The bacteria multiply extensively inside the tread, which extends inwardly and penetrate through and in between the cortex cells.

In the inner cortex cells the bacteria are released into the cytoplasm and stimulate some cells to divide.these division lead to mature nodule, called root nodule which is made up of tetraploid cells containing bacteria and some diploid cells.Each enlarged, nonmotile bacterium is reffered to as bacteroid.the bacteroid usually occurs in cytoplasmin groups and each group surrounded by membrane called peribacteroid membrane.between the peribacteroid membrane and bacterial group is a region called the peribacteroid space. Outside the peribacroroid space in the plant cytoplasm is aprotein called Leghhaemoglobin.this pigment gives legume nodule a pink colour.It is thought to transport O2 into the bacteroid.Too much O_2 inactivates the enzyme that catalyze nitrogen fixation.

Nitrogen fixation occurs directly with in bacteroids. The plant provides bacteroid with carbohydrate, which they oxidize and obtain energy.

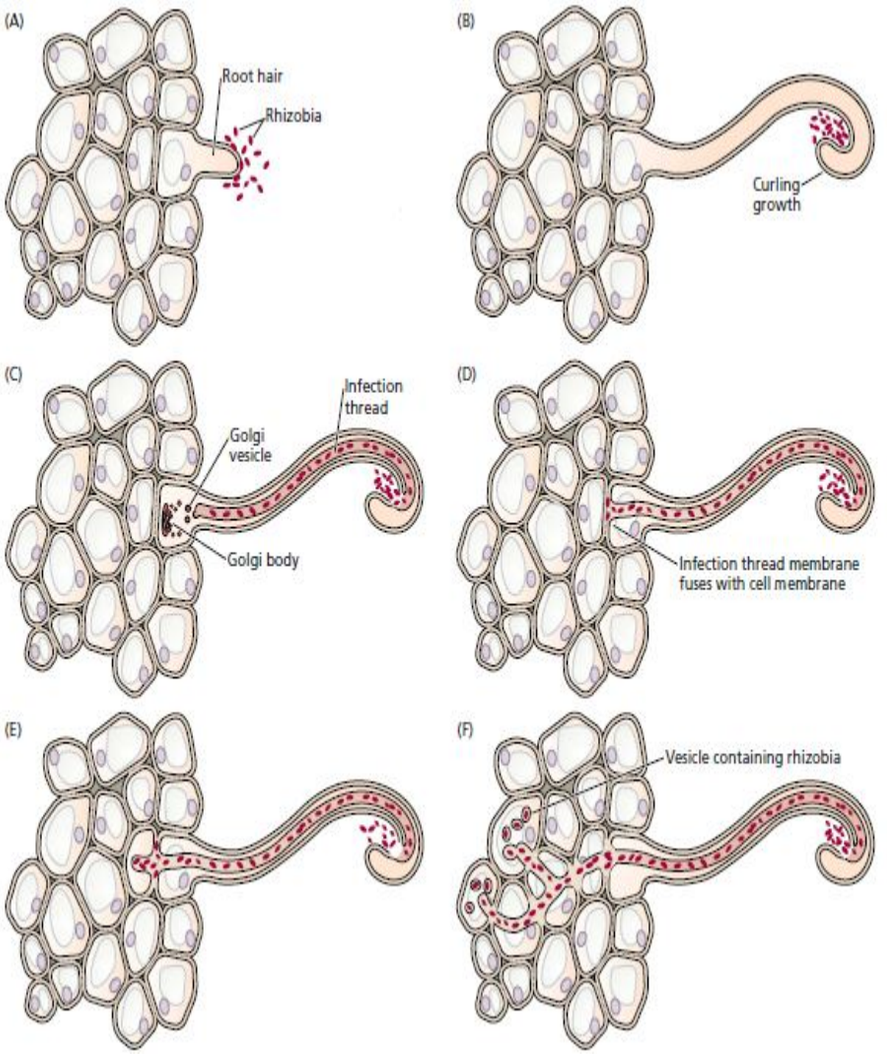


Fig. 11.5 : The infection process during nodule organogenesis

(A) Rhizobia bind to an emerging root hair in response to chemical attractants sent by the plant.

(B) In response to factors produced by the bacteria, the root hair exhibits abnormal curling growth, and rhizobia cells proliferate within the coils.

(C) Localized degradation of the root hair wall leads to infection and formation of the infection thread from Golgi secretory vesicles of root cells.

(D) The infection thread reaches the end of the cell, and its membrane fuses with the plasma membrane of the root hair cell.

(E) Rhizobia are released into the apoplast and penetrate the compound middle lamella to the subepidermal cell plasma membrane, leading to the initiation of a new infection thread, which forms an open channel with the first.

(F) The infection thread extends and branches until it reaches target cells, where vesicles composed of plant membrane that enclose bacterial cells are released into the cytosol.

11.6 Anomalous Secondary Growth of Dicot Root

Secondary growth occurs normally in the roots of gymnosperms and dicotytedons to support the spreading shoot system and to meet the growing needs of absorption and conduction of food materials. **Roots of most monocotyledons do not show secondary growth and remain in primary state throughout their life**. Some monocots however, do show secondary thickenings in roots (e.g. *Dracaena*).

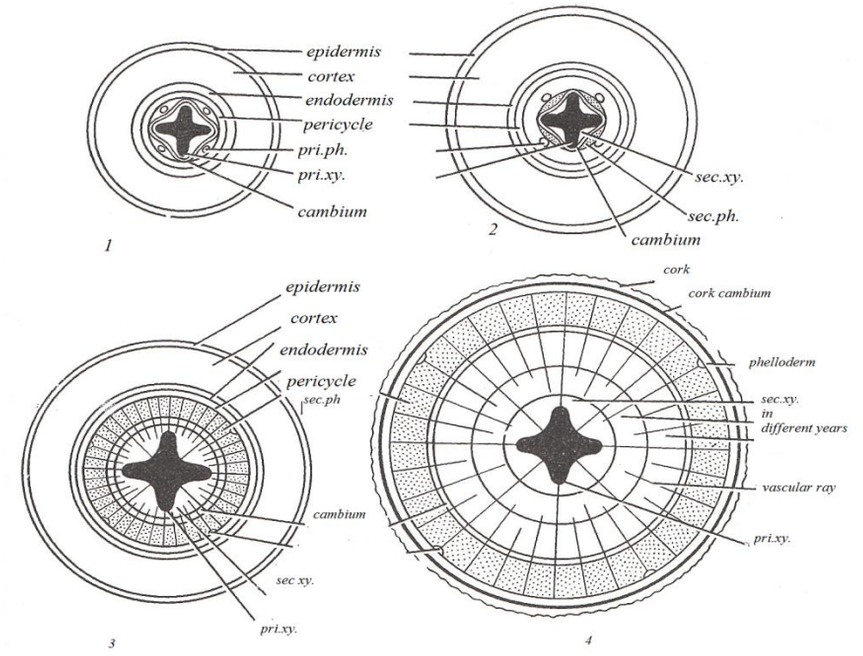


Fig. 11.6 : Secondary growth in typical Dicot Root

Though the process of secondary growth in the root is initiates in a different manner. The fundamental structure of secondary tissues formed is similar to that of the stem. The two **secondary meristems** responsible for secondary growth in roots are:

1. **Fascicular or vascular cambium.** It lies in the stelar region and its activity results in the formation of **secondary xylem** and **secondary phloem**.
2. **Phellogen or cork cambium.** It develops in the extra-stelar region and its activity results in the formation of **periderm**.

The initiation and activity of vascular cambium starts earlier than that of cork cambium.

[I] Initiation and activity of vascular cambium

The root is characterized by radial arrangement of vascular strands. A few parenchyma cells situated along the inner edges of the phloem groups become meristematic and form crescent shaped strips of cambium .These strips of cambium produce secondary xylem centripetally and secondary phloem centrifugally . Consequently, collateral strands of vascular tissue are formed in between the primary xylem bundles. As the activity of these cambial strips proceeds, the cells of the pericycle lying against the protoxylem also become meristematic and thus small strips of cambium are also formed outside the xylem Strands the first formed cambial strips below the phloem strands join with the newly formed strips outside the xylem strands. As a result, a complete but wavy ring of cambium is formed which runs outside the xylem and inside the phloem strands. The first formed cambial strips located along the inner face of the phloem start functioning earlier than the later formed cambium from the pericycle. As a result of the formation of secondary xylem opposite the phloem the cambium is pushed outwards. Thus the wavy cambium ring becomes circular in outline; this cambium ring produces secondary phloem on the outer side and secondary xylem on the outer side and secondary xylem on the inner side.

The secondary vascular tissues produced in the root are fundamentally similar to those of the stem and are in the form of a continuous cylinder. The primary xylem bundles remain in their original position on inside the cylinder of secondary tissue. At the secondary stage, therefore, the root can be distinguished from the stem on the basis of exarch primary xylem strands. The primary phloem, endodermis and cortex usually become crushed due to enormous increase in the volume of the secondary tissue.

In many roots, the cells of the cambium which arise in the pericycle, opposite the primary xylem poles do not give rise to secondary vascular tissues.

They function as **ray initials** and produce wide primary vascular rays, radiating outwards through the secondary xylem and phloem from the tips of the protoxylem. In addition, short and narrow secondary vascular rays are also formed similarly in the secondary vascular tissue.

Although the xylem of- the root and stem is made up of the same kinds of elements, in **relative proportions of these elements differ the two.** For example, as compared to stems, roots have more parenchyma, fewer fibres, larger or more abundant rays and vessels with thinner walls. Similarly, the phloem in roots has less sclerenchyma and more storage parenchyma.

[II] Initiation and activity of cork cambium

In the roots of herbaceous dicotyledons, there is very little secondary growth and, therefore, the tissues outside the stele remain almost intact. In such plants, cortical layers beneath the epidermis become suberized and from a thick walled **exodermis** which is protective in function.However, in woody roots, as a result of extensive activity of the vascular cambium, enormous amount of secondary tissues is formed consequently, a pressure is exerted and eventually the peripheral tissues of the root are sloughed off. They are replaced by a more efficient protective covering, the **periderm**, formed by the activity of the **cork cambium** or **phellogen.**

Formation of periderm starts soon after the production of secondary vascular tissue has begun. The **phellogen** originates from the outer layers of pericycle. It produces **phellem** or **cork** toward the outer side and **phelloderm** or **secondarycortex** towards the inner side. The tissue from endodermis to exodermis is sloughed off and consequently, the cork or the phellem becomes the outermost protective layer. Lenticels may also be formed in roots during secondary growth. The first formed phellogen may persist for a considerable period of time and new phellogen layers may be formed in the deeper layers after the older ones have been shed.

In the roots of certain angiosperms, such as members of Myrtaceae, Onagraceae and Rosaceac, the periderm is made up of two types of cells, **suberized** and **non-suberized** cell. One cell deep layer of suberized cells alternates with many cells deep layers of non-suberized cells. The latter perform the storage function. Such periderm is known as **polyderm**.

Abnormal Secondary Growth in Dicot Roots

In a number of dicot plants, the roots are modified to function as storage roots.These organ develop considerable parenchyma storage in nature. **Secondary growth in these roots is usually of anomalous type as an adaptation to their storage function.** Members of Umbelliferae, Cruciferae, Chenopodiaceae, Compositae, Convolvulaceae, etc. are good examples of anomalous secondary growth exhibited, by the roots. The aerial root of *Tinospora* shows normal nature of secondary growth but medullary rays are very conspicuous and large and store reserve food material

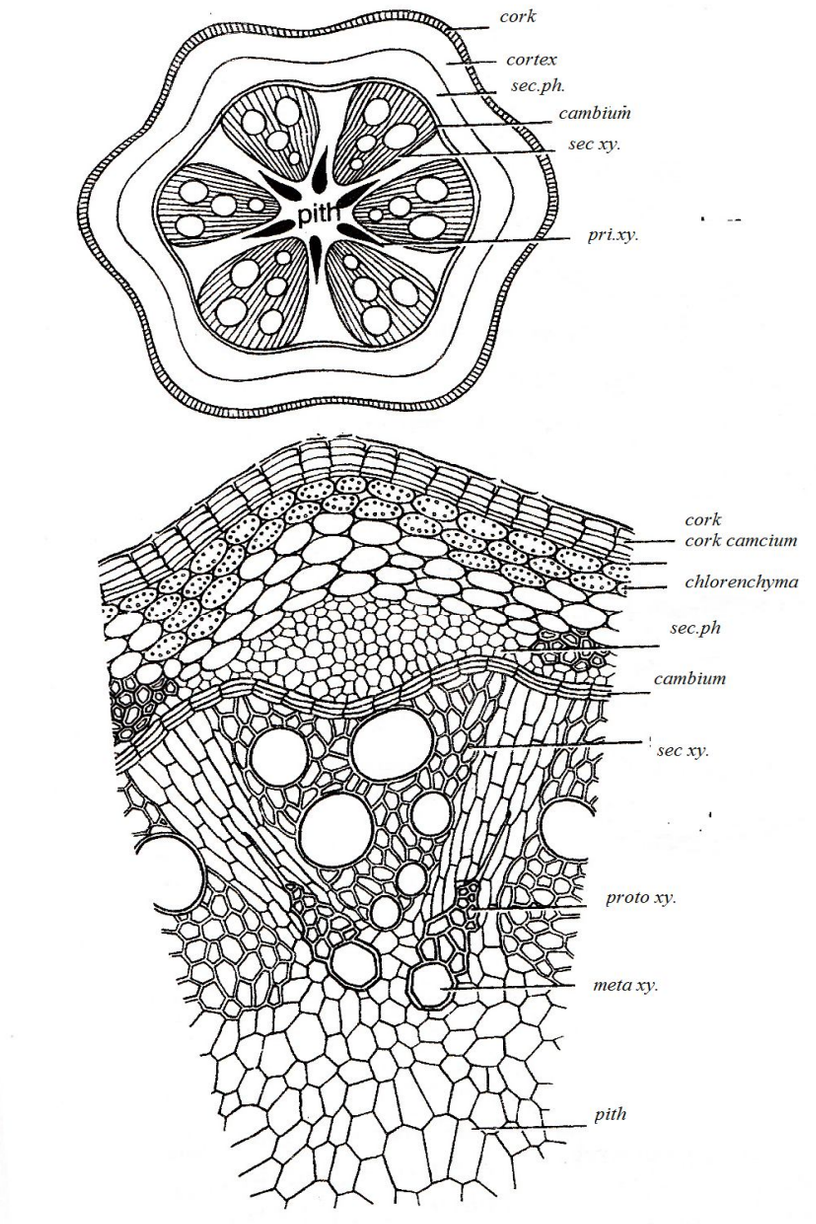


Fig. 11.7 : TS of Root *Tinospora*

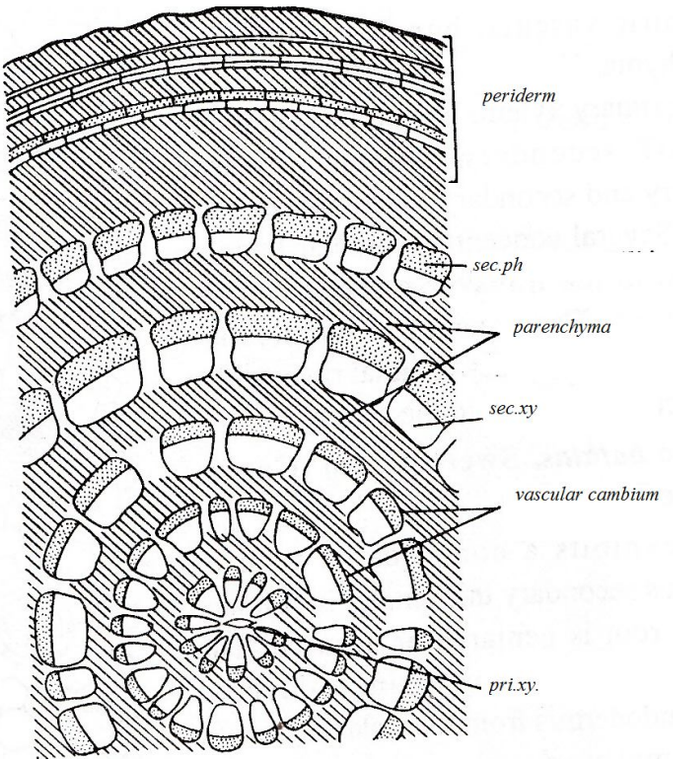
A : Outline Diagram B : Cellular Diagram

Beta vulgaris root (Fam. - Chenopodiaceae) –Its root has a typical dicot structure showing radial and diarch condition of stele. Because it is a storage root, there occurs an increase in girth of the root to accommodate the increase in number of cells. Secondary growth, however, is of anomalous type due to abnormal activity as well as position of cambium.

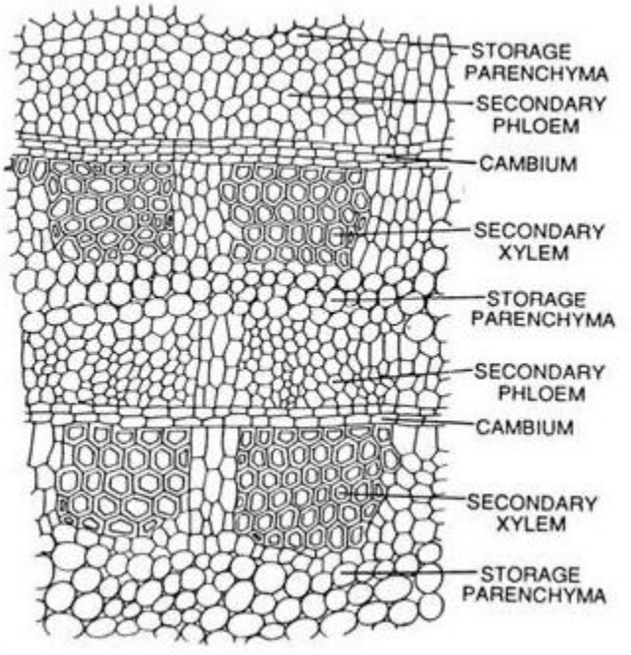
Secondary growth is initiated by the formation of the primary cambium ring, which is formed by the joining of the cells of the pericycle lying outside the protoxylem and the parenchyma cells lying between xylem and phloem.

This primary cambium ring exhibits abnormal activity, forms secondary xylem and parenchyma cells in alternate groups, towards the inner side, whereas on the outer side, it forms secondary phloem. This result in the formation of a ring of conjoint, collateral vascular bundles embedded in parenchymatous tissue. The parenchymatous cells function as storage tissue. The cambium after forming the ring of vascular bundles ceases its activity. Soon a second ring of cambium arises outside the first cambium ring. This accessory or secondary cambium develops from the phloem parenchyma cells or sometimes from the inner cells cut off by the pericycle. This secondary cambium also behaves in an abnormal manner, similar to the first cambium ring, resulting in the formation of another ring of conjoint, collateral vascular bundles arranged external to the first ring of bundles. These vascular bundles are separated by wide gaps occupied by parenchymatous cells. This first ring of accessory cambium also ceases its function and a third cambium ring or second accessory cambium now develops outside this second ring of vascular bundles. This cambium ring develops from the cells of the pericycle and behaves in a similar manner. At later stages, the developing cambial ring encloses a few layers of pericyclic cells which are meristematic in nature and rapidly divide forming layers of parenchyma cells. These cells get filled with reserve food material and function as storage tissue.

Later on as more and more proliferated parenchymatous cells are formed; the vascular bundles get separated further apart. The subsequent cambial rings produce alternate layers of vascular bundles and proliferated parenchymatous cells filled with reserve food. In a transverse section, the rings of parenchyma appear dark red (due to anthocyanin pigments) while those of vascular bundles appear lighter in colour. The root of *Beta vulgaris* thus increases in size by the formation of vascular bundles and proliferated parenchyma cells, rich in sugar. In mature roots, a cork cambium or phellogen develops which forms cork cells on the outer side and secondary parenchyma towards inner side.



A



B

Fig.11.8 : T.S. of Root *Beta vulgaris* showing growth rings during Secondary Growth
A : Outline Diagram B : Cellular Diagram

***Ipomoea batatas* root** (Fam.- Convolvulaceae) - The anomalous secondary growth in *Ipomoea* was described by Artschwager (1926). In young conditions, the cortex is well developed with large intercellular spaces. A well defined endodermis demarcates the stellar region from the cortex. The stele typically shows radial and pentarch or hexarch type.

Secondary growth of the storage root takes place by the formation of a cambium ring which is normal in function initially; however, at later stages it begins to behave abnormally forming separate strands of secondary xylem and phloem embedded in the parenchymatous ground tissue. These strands of xylem get surrounded by individual rings of secondary cambium which originate from the parenchymatous cells of the ground tissue surrounding the xylem vessels.

This secondary cambium surrounding the xylem vessel behaves abnormally forming few elements of secondary xylem towards the centre and a few sieve tubes and laticiferous elements away from the centre. The cambium also forms considerable amounts of parenchymatous cells on both inner and outer side. These cells get filled with reserve food material (sugar) and function as storage tissue. In later stages the larger vessels often develop tyloses. From the pericycle develops the cork cambium which forms the periderm.

***Raphanus sativus* root** (Fam. - Cruciferae) - It's roots are fleshy, having a typical diarch primary xylem. At the beginning of secondary growth, a normal cambium is formed which cuts off secondary xylem towards inner side and secondary phloem towards outer side. The xylem cells are constituted of xylem parenchyma which repeatedly divide and proliferate in all directions there by greatly increasing the secondary tissues. Some of these parenchymatous cells become meristematic and develop to form the secondary cambium. These cambia partly or completely surround the strands of secondary xylem scattered in the ground tissue. The activity of the secondary cambium is abnormal as it forms few xylem elements but large amounts of parenchyma both internal and external to the cambium ring. As the root matures, numerous patches of secondary xylem surrounded by secondary cambium lie scattered in the parenchymatous ground tissue. These parenchyma cells accumulate reserve food and function as storage tissue. Thus the increase in girth of the root is due to the formation of circular patches of secondary xylem and the mass of proliferating parenchymatous storage cells.

The original cambium is persistent and is visible in the peripheral region with sufficient amount of secondary phloem attached externally. The cork cambium forms a few layers of thin walled cork cells.

11.7 Summary

Root develops from the radical of the embryo.Its tip is covered by a cap, apical meristem is sub-terminally situated .Meristematic tissues gradually differentiated and becomes mature later on by a set procedure .The primary body consist of three tissue systems. Structurally monocot and dicot roots shows some differences.The root of herbaceous dicots,all gymnosperms and woody dicots show secondary growth as a result thicknesss of root increase.most of the monocot roots are entirely primary in nature

11.8 Glossary

- **Abaxial :** Directed away from the axis. Opposite of *adaxial.* With regard to a leaf, the lower, or "dorsal," surface
- **Adaxial :** Directed toward the axis. Opposite of *abaxial.* With regard to a leaf, the upper, or "ventral,"surface.
- **Annual ring :** In secondary xylem; growth ring formed during one season. The term is deprecated because more than one growth ring may be formed during a single year.
- **Anomalous secondary growth :** A term of convenience referring to types of secondary growth that differ from the more familiar ones.
- **Anomalous secondary growth :** A term of convenience referring to types of secondary growth that differ from the more familiar ones.
- **Apical cell :** Single cell that occupies the distal position in an apical meristem of root or shoot and is usually interpreted as the initial cell in the apical meristem; typical of seedless vascular plants.
- **Bark :** A nontechnical term applied to all tissues outside the vascular cambium or the xylem; in older trees may be divided into dead outer bark and living inner bark, which consists of secondary phloem. See also *rhytidome.*
- **Cambium :** A meristem with products of periclinal divisions commonly contributed in two directions and arranged in radial fi les. Term preferably applied only to the two lateral meristems, the *vascular cambium* and the *cork cambium*, or *phellogen.*
- **Diffuse-porous wood :** Secondary xylem in which the pores (vessels) are distributed fairly uniformly throughout a growth layer or change in size gradually from earlywood to latewood.
- **Earlywood :** Wood formed in fi rst part of a growth layer and characterized by a lower density and larger cells than the latewood. Term replaces *spring wood.*

- **Growth ring :** A growth layer of secondary xylem or secondary phloem as seen in transverse section of stem or root; may be an *annual ring* or a *false annual ring.*
- **Heartwood :** Inner layers of secondary xylem that have ceased to function in storage and conduction and in which reserve materials have been removed or converted into heartwood substances; generally darker colored than the functioning *sapwood.*
- **Histogen concept :** Hanstein's concept stating that the three primary tissue systems in the plant—the epidermis, the cortex, and the vascular system with the associated ground tissue—originate from distinct meristems, the histogens, in the apical meristems. See *histogen.*
- **Latewood :** The secondary xylem formed in the later part of a growth layer; denser and composed of smaller cells than the earlywood. Term replaces *summer wood.*
- living cells and reserves; it may or may not function in the conduction of water. Generally lighter colored that the *heartwood*.
- **Meristem :** Embryonic tissue region, primarily concerned with formation of new cells.
- **Meristematic cell :** A cel synthesizing protoplasm and producing new cells by division; varies in form, size, wall thickness, and degree of vacuolation, but has only a primary cell wall.
- **Periderm :** Secondary protective tissue that replaces the epidermis in stems and roots, rarely in other organs.Consists of *phellem* (cork), *phellogen* (cork cambium),and *phelloderm*.
- **Phellem (cork) :** Protective tissue composed of nonliving cells with suberized walls and formed centrifugally by the phellogen (cork cambium) as part of the periderm. Replaces the epidermis in older stems and roots of many seed plants.
- **Phelloderm :** A tissue resembling cortical parenchyma produced centripetally by the phellogen (cork cambium) as part of the periderm of stems and roots in seed plants.
- **Phellogen** (**cork cambium**) **:** A lateral meristem forming the periderm, a secondary protective tissue common in stems and roots of seed plants. Produces phellem (cork) centrifugally, phelloderm centripetally by periclinal divisions.
- **Phyllotaxy** (or **phyllotaxis**) **:** Mode in which the leaves are arranged on the axis of a shoot.
- **Primary phloem :** Phloem tissue differentiating from procambium during primary growth and differentiation of a vascular plant. Commonly divided into the earlier *protophloem* and the later *metaphloem*. Not differentiated into axial and ray systems.

- **Quiescent center :** Initial region in the apical meristem that has reached a state of relative inactivity; common in roots.
- **sapwood** Outer part of the wood of stem or root containing
- **Vascular bundle :** A strand-like part of the vascular system composed of xylem and phloem.
- **Vascular cambium :** Lateral meristem that forms the secondary vascular tissues, secondary phloem and secondary xylem, in stem and root. Is located between those two tissues and, by periclinal divisions, gives off cells toward both tissues.
- **Xylem :** Principal water-conducting tissue in vascular plants characterized by the presence of tracheary elements. The xylem may also serve as a supporting tissue, especially the secondary xylem (wood).

11.9 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1 In the roots,root hair arise from ------

a) epiblema (b) cortex (c) endodermis (d) pericycle

2 Lateral branch of the root arise out which of the following -------

(a) epiblema (b) cortex (c) endodermis (d) pericycle

Section B : (Short Answer Type Questions)

1 What are trichoblast?

2 What is Casparian strips?

3. What are velamen ?

Section C : (Long Answer Type Questions)

1. Decribe the primary structure of a dicot root with suitable diagrams.

2 Write short notes on the followings----

(a)Root-Microbe interaction

(b) Anomalous sec.growth

11.10 References

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Unit -12

Leaf Growth and Differentiation

Structure of the Unit:

12.0 Objectives

Leaf is an important part of plant, with the help of this unit you will be understand the character, ecological utilities of the leaf in terms of-

- Leaf structure (morphological & anatomical) and its arrangement on axis
- Xerophytic leaf structure
- Hydrophytic leaf structure

12.1 Introduction

Leaf is the most important organ of plant system. It is the lateral appendage of the stem borne at the node and subtending a bud at its axil.These are develop in acropetal order on the stem and are usually flat, thin expanded structure. The growth of leaf is limited type.

12.2 Primary Structure of Leaf

Leaf is an important part of plant. These are very important vegetative organs mainly concerned with photosynthesis and transpiration activities. Like stem and roots,

Leaves also have the three types of tissue systems –

- dermal
- ground
- vascular

The dermal tissue system consists of an upper epidermis and lower epidermis. Stomata occur in both the epidermis but more frequently in the lower epidermis. The ground tissue system that lies between the both epidermal layers of leaf, is called as mesophyll tissue. Usually it is differentiated into palisade parenchyma on the adaxial side and spongy parenchyma on the abaxial side.

The leaf showing this differentiation in mesophyll is designated as dorsiventral. It is common in dicot leaves.

When mesophyll is not differentiated and made up of only spongy or palisade parenchyma (as in monocots), it is called isobilateral.

The mesophyll tissue, especially spongy parenchyma cells encloses air spaces. The presence of air spaces is a special feature of spongy cells. This space facilitates the gaseous exchange between the internal photosynthetic tissue and the external atmosphere through the stomata.

The vascular tissue system is composed of vascular bundles. These are collateral and closed. In C_4 plant Kranz anatomy in leaf is also observed .The vascular tissue forms the skeleton of the leaf and they are known as veins. The veins supply water and minerals to the photosynthetic tissue.

The morphological and anatomical features of the leaf help in its physiological functions.

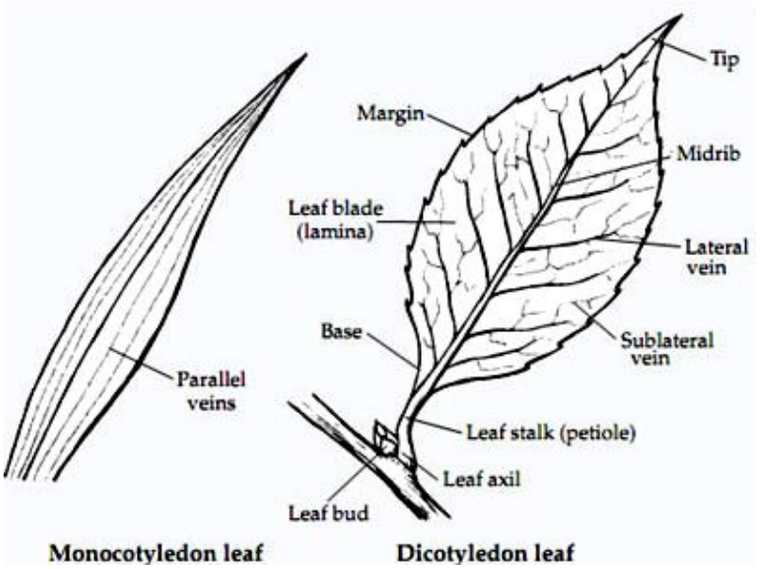


Fig. 12.1 : A typical Monocot and Dicot Leaf

Structure of Petiole

As described earlier, petiole is that part of the leaf with which it remains attached to the stem. In its internal structure, it is very much similar to the primary Stem but for some variations in the arrangement of vascular tissue.

In a transverse section, the petiole may be circular (e.g., *Nymphaea* and *Eriobotrya*), triangular (e.g., *Mangifera*) or flattened (e.g., *Mahonia*) in outline. A distinct groove is present on the adaxial (upper) side of the petiole in *Prunus, Citrus,* etc. In *Cucurbita*, the petiole has ridges and grooves just like the structure of stems.

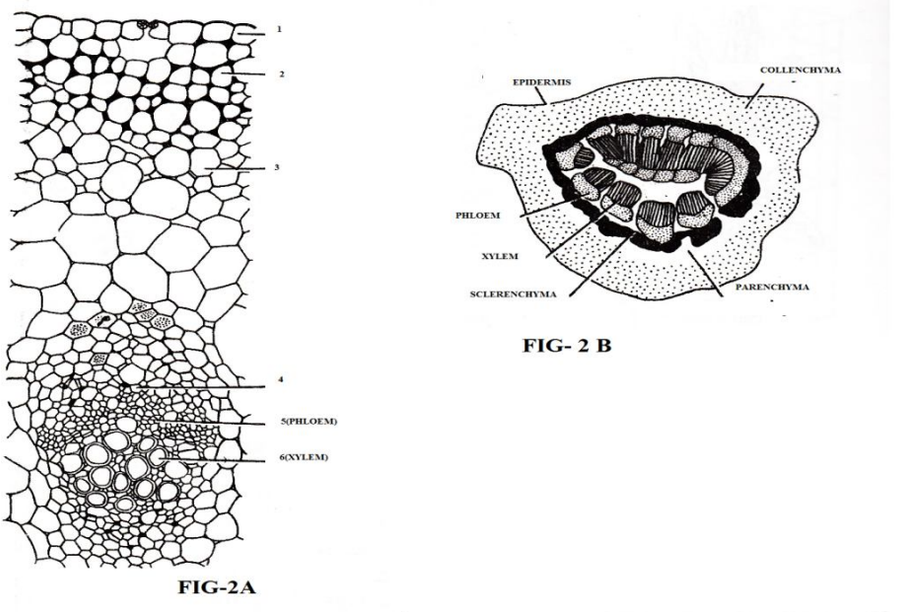


Fig. 12.2 : T.S. of Petiole

[I] Epidermis

Epidermis is made up of a single layer of compactly arranged barrel shaped cells. In some aquatic plants, such as *Nymphaea* and *Eichhornia* the epidermal cells are radially elongated. The epidermis is usally covered with a thick layer of cuticle and the thickness may extend to the radial walls of the cells. The epidermis is sometimes covered by multicellular hairs as in *Cucurbita* and *Banksia*.

[II] Hypodermis

Hypodermis is represented by many layers of collenchymatous cells situated just below the epidermis .In some plants the hypodermis consists of Patches of collenchyma and palisade parenchyma (e.g., *Mirabilis*). In some plants sclerenchymatous hypodermis is present on the adaxial side (e.g., *Banksia*).

[III] Ground tissue

Ground tissue is made up of parenchymatous cells with distinct intercellular spaces. Some times as in *Mangifera*, sclerenchymatous cells are also scattered in the ground tissue. Laticifers and resin canals are frequently present in the ground tissue of the petiole. In some aquatic plants (e.g., *Nyphaea* and *Nuphar*) air chambers are also present in the ground tissue.

[IV] Vascular bundles

There is considerable variation in the arrangement of vascular tissue in the petiole. It is arranged in a ring (e.g., *Ricinus*, *Aquilegia*, *Platanus*, *Quercus)* or in the form of a girdle with adaxial xylem and abaxial phloem (e.g., *Euonymus*, *Ligustrum, Prunus*, *Galphimia*;). In *Oxalis corniculata*, there is a ring of discrete vascular bundles with their xylem facing towards the pith. The petiole of *Cucurbita* has bicollateral vascular bundle of two different sizes, the smaller bundles lie below the grooves and the larger opposite the ridges.

In the petiole of many Leguminosae and Oxalidaceae a joint like thickening is present at the base which is known as *pulvinus* .Anatomically, the pulvinus is largely made up of thin walled parenchymatous tissue with intercellular spaces. in this region the vascular tissue is represented by only one concentric strand, although several bundles are present above and below the pulvinus The pulvinus is involved in the leaf movements through changes in the turgor and Concomitant changes in the size and shape of the ground Parenchyma cells.

B. Anatomy of Lamina

The surface of the leaf facing the axis is referred to as adaxial Surface and morphologically this is the upper Surface of the leaf. The surface of the away from the axis is known as abaxial surface (lower surface). Histologically leaves may be distinguised, into the following three categories.

I. Bifacial or dorsiventral leaves. They are extended at right angles to the direction of the most intense diffuse light and consequenty possess distinct upper and lower (dark and light) faces.The palisade tissue is mostly confined below the upper epidermis and spongy parenchyma occurs just inner to the lower epidermis Some times the palisade may also occur next to the lower epidermis. The latter types of leaves are known as **inverted bifacial.**

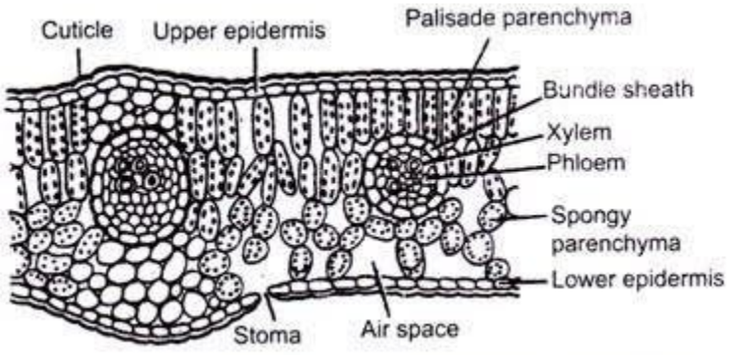


Fig. 12.3 : V.T.S. of Dicot Leaf : *Mangifera*

2. Unifacial leaves. Such leaves have both the sides equally illuminated, and there is no distinction between the upper and lower epidermis. To this category belongs the cylindrical leaf of *Allium, Juncus*, *Trig1ochin*, *Sisyrinchium* etc.

3. Equifacial or isobilateral leaves. In these leaves palisade is present next to both the upper and lower epidermis or there is no distinction of mesophyll into palisade and spongy tissue as in grasses.

Like the root and stem, the leaf consists of three tissue system : (i) **dermal system** - consisting of the upper and lower epidermis, (ii) **ground tissue system** - the main photosynthetic tissue consisting of mesophyll, and (iii) **vascular system** - comprising of veins of various degrees.

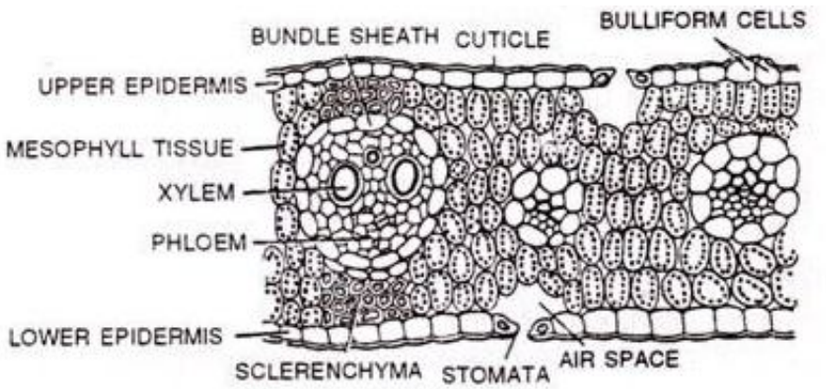


Fig. 12.4 : T.S. of Monocot leaf : *Zea mays*

[I] Epidermis

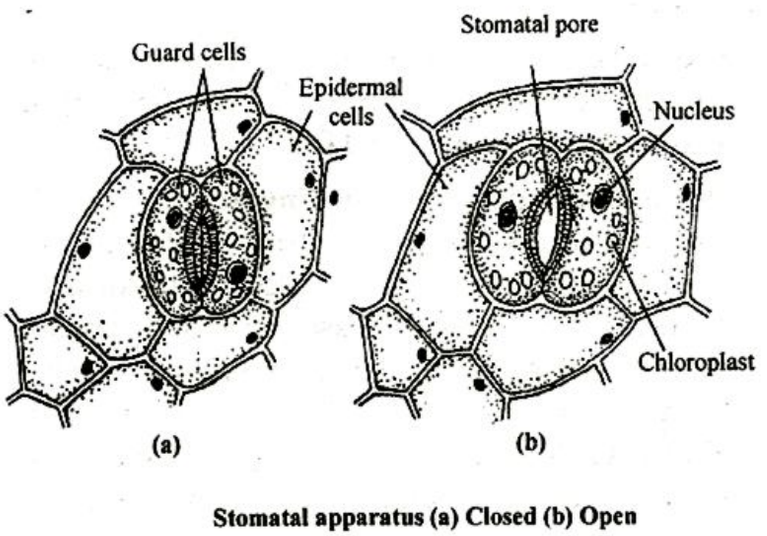
The upper as well as the lower epidermis of the leaf is usually made up of a **single layer** of compactly arranged cells. However, a **multiseriate** adaxial epidermis occurs in some plants, such as *Ficus, Piper* and *Nerium* It is formed as a result of periclinal divisions of protodermal cells. In succulent species of *Begonia* the upper epidermis is three-layered,

whereas the lower epidermis is two layered. The radial walls of the epidermal cells may be straight or sinuous. The epidermal cells may sometimes have simple convex protrusions, called **papillae** (e.g. *Gladiolus*). All the epidermal cells of a leaf are alike or those of the coastal area are narrow. In some xerophytic leaves, especially those of grasses, epidermal cells situated in longitudinal furrows are large with thin flexible walls. These cells, known as **motor cells** or **bulliform cells** are considered to help in rolling of leaves in dry weather. The epidermal cells are cutinized and thickness of the cuticle varies from species to species. Xerophytic species, as a rule have a comparatively thick cuticle.

I. Stomata. An important feature of leaf epidermis is the presence of stomata which occur either on both sides of the leaf or only on one side of the leaf. When stomata occur on both sides, the leaf is known as **amphistomatic**, when they are confined to the upper side, the leaf is known as **epistomatic**; and when to the lower side, the leaf is called **hypostomatic**.

(a) Structure

Each **stoma** consists of an opening (pore) bounded by two specialized, usually kidney shaped, epidermal cells known as **guard cells**. The guard cells have unevenly thickened walls. The inner wall facing the aperture is highly thickened while the one away from the aperture is thin and extensible. The guard cells are also covered with cuticle which extends to the inner wall forming the boundary of the pore and the sub-stomatal chamber. Mitochondria, dictyosomes ribosomes and endoplasmic reticulum are present in the gaurd cells. They also contain starch grains. Unlike other epidermal cells, guard cells contain chloroplasts. Electron microscopic studies have revealed the chloroplasts of guard cells have fewer and less well organised lamellae than (those of mesophyll cells. Due to the presence of chloroplast, active carbon assimilation takes place: in guard cells.



Stomatal apparatus (a) Closed (b) Open

Fig. 12.5 : Struture of Stomata

Consequently, increased turgor pressure after photosynthesis results in the opening of stoma, while a decrease in turgor pressure results in its closing. The strands of cellulose in the guard cell walls are arranged radially allowing more elongation than lateral expansion of the cells.

The guard cells are surrounded by a variable number of epidermal cells which are called **subsidiary** or **accessory** cells. These cells must be morphologically similar to the other epidermis cells or very different from them. **The two guard cells of a stoma, its aperture and adjoining subsidiary cells together constitute the stomatal apparatus.**

Stomata occupy roughly 1-12% of the total leaf area. There is a great variation in stomatal frequency of leaves. In dicotyledon leaves, their number varies from 1,000 - 10, 00,000 per cm^2. The average size of stomata ranges between 6.7 and 17.7 microns.

(b) Position

Stomata may occupy three different positions in relation to epidermal cells.

(1) They may be at the same level as the adjoining epidermal cells as in most of the mesophytic plants.

(2) In xerophytic plants, as in *Hakea*, the stomata are sunken as they are located in a cup shaped depression

(3) Sometimes as in *Cucurbita, Prunus* and *Solanum*, the stomata are slightly raised abovethe surface of epidermis

(c) Types of stomata

The first classification of stomata was presented by **Vesque (1889).** He distinguished four stomatal types, viz.,

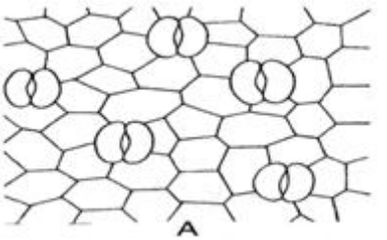
(i) Ranunculaceous type, (ii) Cruciferous type,

(iii) Rubiaceous type, and (iv) Labiateous or Caryophyllaceous type.

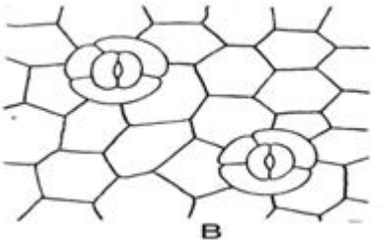
However, the terminology of Vesque was found faulty since these types also occur in many other families.

Metcalfe and Chalk (1950), on the basis of the number and orientation of subsidiary cells recognised the following four types of stomata in dicotyledons:

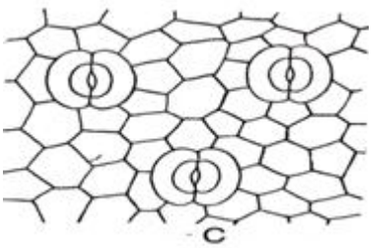
(i) *Anomocytic (irregular-celled) type*. In this type, the stomata are surrounded by a limited number of epidermal cells which are indistinguishable from other epidermal cells. The stomata of this type occur in the members of Ranunculaceae, Papaveraceae, Capparidaceae and Nyctaginaceae.



(ii) *Anisocytic (unequal-celled) type*. This type is characterised by the presence of three subsidiarycells of which one is distinctly smaller than the other two. This type is common in the members of Cruciferae. Umbelliferae. Solanaceae and Convolvulaceae.



(iii) *Paracytic (parallel-celled) type*. In this type, the stoma is accompanied on eitherside by one or more subsidiary cells which lie parallel to the long axis of the pore and guard cells , This type occurs in the members of Magnoliaceae, Schizandraceae and Rubiaceae.



(iv) *Diacytic (cross-celled) type*. In this type the stoma is enclosed by a pair of subsidiary cells whose common wall is at right angles to the lone axis of the guard cells. This type is represented be some members of Caryophyllaceae Labiatae and Acanthaceae.

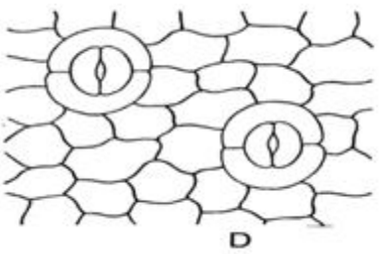
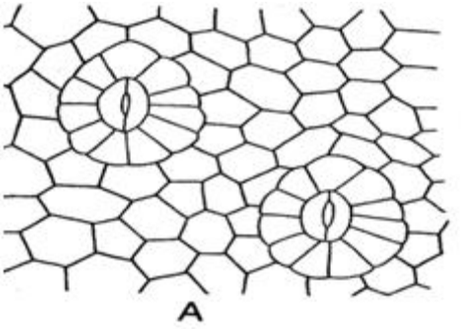


Fig. 12.6 : Stomal Types by Metcalfe and Chalk (1950): (A), (B), (C), & (D)

In addition to these four types, two others are also **recognised by Stace (1965)**

Actinocytic type with four or more subsidiary cells elongated radially to the stoma



Cyclocytic type with four or more subsidiary cells arranged in a close ring around the stoma

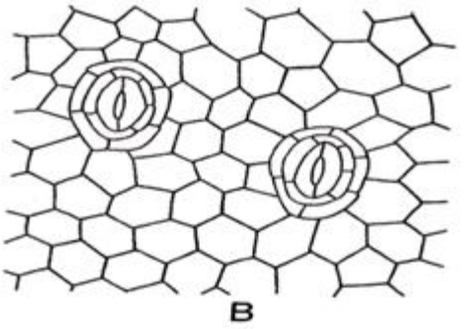
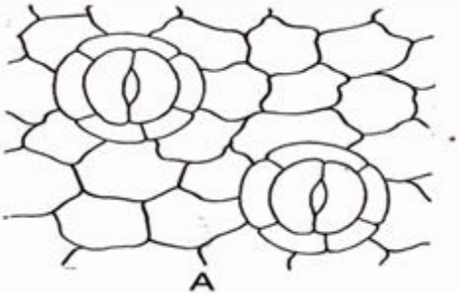


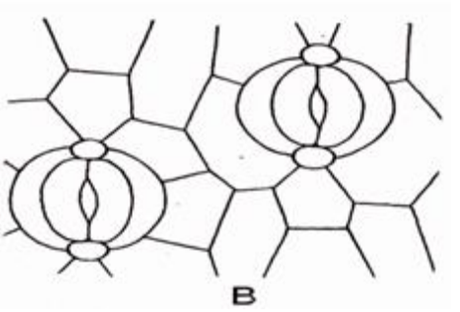
Fig. 12.7 Stomatal Types by Stace (1965) (A -actinocytic,B-cyclocytic)

Stebbins and Khush (1961) recognised the following four types of stomata in monocotyledons:

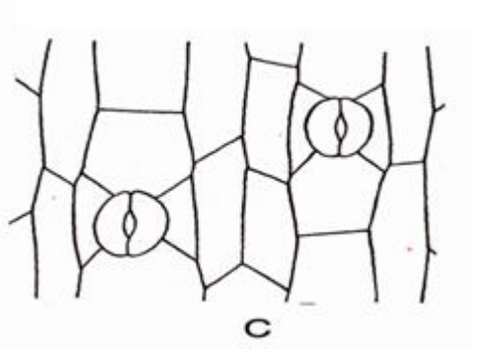
In **Type 1** the stoma is surrounded by 4-6 subsidiary cells around the guard cells in all four directions. This type occurs in Commelinaceae, Araceae, Musaceae, Zingiberaceae and Cannaceae.



In **Type 2** the stoma is also surrounded by 4-6 subsidiary cells, but the two subsidiary cells at the ends of the guard cells are roundish and smaller than the others which are placed lateral to the guard cells. This type is recorded in Palonse and Pandanaceae.



In **Type 3** the stoma is surrounded by two Subsidiary cells placed lateral to the guard cells. This is the most common type in monocotyledons and found in a large number of famalies belonging to Butomales, Alismatales, juncales, Graminales and Cyperales.



In **Type 4** the guard cells are without any subsidiary cells. It is the second mostcommon type in monocotyledons and occurs in various families of Liliales, Dioscoriales, Amaryllidales, Iridales, etc.

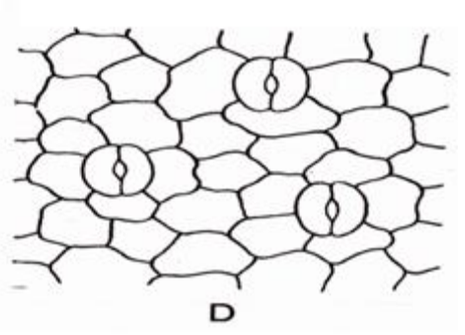


Fig. 12.8 : Stomatal Types by Stebbins and Khush (1961)

(A : Type – 1, B : Type 2, C : Type 3 & D : Type 4)

Pant (1965) proposed a classification of stomata on the basis of ontogeny. He divided the stomata in the following three types :

(i) *Mesogenous*. In this type, the guard cells and the subsidiary cells are derived by consecutive divisions of the same meristemoid. This type of stomatal development is found in the members of Cruciferae, Solanaceae, etc.

(ii) *Perigenous.* In this type, there is an independent origin of the guard cells and the subsidiary cells. The meristemoid directly functions as guard cell mother cell, which divides once by a straight wall to form a pair of guard cells. The subsidiary cells are derived by oblique divisions in the epidermal cells neighbouring the guard cells. This type of development may acquire anomocyctic (eg. Nymphaeaceae, Cucurbitaceae) or tetracytic (e.g., Poaceae, Cyperaceae) condition.

(iii) Perimesogenous. In this type, the subsidiary cells have a dual origin. At least one of the subsidiary cells has a common origin with the guard cells, whereas the others differentiate in the neighbouring cells on the other side of the meristemoid. This type of development is found in the members of Ranunculaceae Caryophyllaceae, etc.

(d) Functions. Stomata are the main portals of gaseous exchange. Hence they are of great importance in processes like photosynthesis and respiration. They help in maintaining the temperature of plant body by evaporating excess amount of water absorbed by roots.

2. Trichomes. The leaf epidermis usually has trichomes which are outgrowths of epidermal cells. They are highly variable in form, structure and function. They may be classified into two categories: **non-glandular** and **glandular trichomes**

(a) Non-glandular trichomes. These are living or dead and are chaxacterjscd by the absence ofsecretion. They are **unicellular** (straight, e.g. *Nerium*, ; Y- shaped, e.g. *Buddleja*; T- shaped, e.g., *Lobularia*,) or **multicellular** (ramulose, e.g., *Platanus. Verbascum*,; *tufted*, e.g., *Dombeya*, *stellate*, e.g.. *Sida*, : *peltate*. e.g *Olea*,).

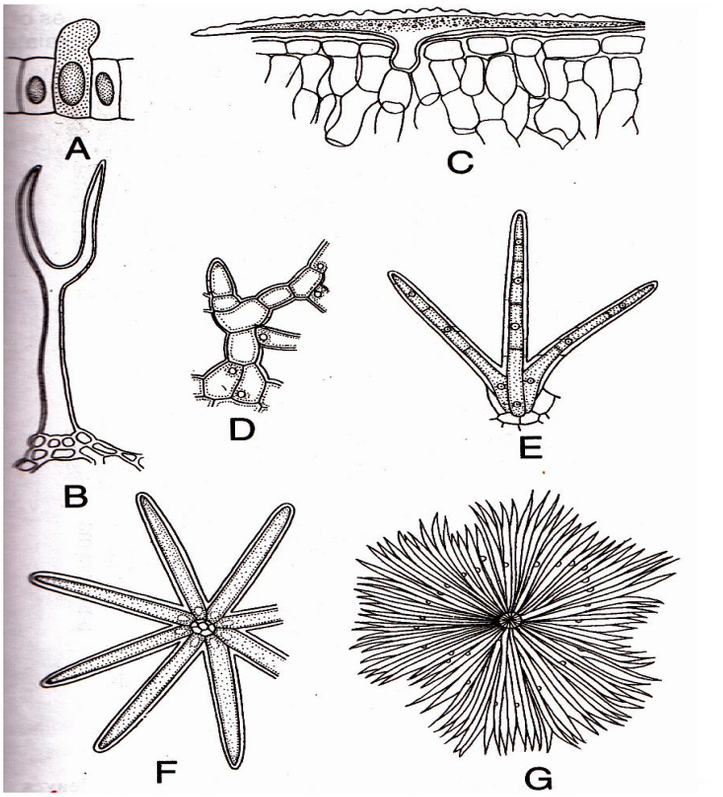


Fig.12.9 : Non-glandular trichomes

A:Unicellular straight;B:Yshaped; C:TShaped; D:Ramlose; E:Tufted; F:Stellate; G:Peltate

(b) Glandular trichomes. These are livieg and are characterised by the secretion of salts, sugars gums, resins, acids, etc. They may be unicellular (e.g., stinging hairs of *Urtica*, A; stigmatic papillae or multicellular (e.g., salt glands of *Avicennia*, ; digestive gland of *Pinguicula* and *Nepenthes*,

(c) Functions. Trichomes present on the plant surface reduce transpiration. Thus they are anadapation to xerophytic habitat. They also serve as water storage organ. Besides, glandular trichomes Secreting Poisonous substanc protect plant from animal attack Nectar secreting trichomes help in Pollination by attracting insects and digestivetrichomes help in the capture and digestion of insects.

[II] Mesophyll

The bulk of the internal tissue of the leaf enclosed by the Upper and lower epidermis, forms mesophyll. It is composed of thin walled parenchymatous cells containing numerous chloroplasts. Mesophyll may bedifferentiated into **palisade** and **spongy** parenchyma. The colunmar palisade cells are arranged compactly with their long axis at

right angles to the leaf epidermis. Such an arrangement of palisade cells protects chloroplasts from excessive heat of sun rays. The spongy parenchyma cells are of irregular shape and are loosely arranged enclosing large intercellular spaces. These air spaces are connected with the sub-stomatal chambers and maintain gas exchange with the outside through stomata. In a **dorsiventral leaf** the palisade tissue occurs on the adaxial surface and the abaxial surface is occupied by spongy parenchyma whereas in a **isobilateral leaf** the palisade tissue occurs on both the abaxial and adaxial surfaces. The amount of palisade varies in different plants, for example only a single layer of palisade is present in *Quercus*, *Rosa* and *Lawsonia*; it is two to three layered in *Eucalyptus*, *Malus* and *Musa* and many layered in *Nerium.* In *Alliun*, *Iris* and Juncus, where the leaves are cylindrical, the palisade occurs all around the periphery of the leaf. In some other monocotyledonous leaves, such as grasses, there is no differentiation of mesophyll into palisade and spongy parenchyma and the entire ground tissue is occupied by one type of cells which appear somewhat rounded in a transverse section. These cells contain chloroplasts and enclose large intercellular spaces.

Typical palisade cells are tubular or cylindrical in shape. But in some dicotyledons (e.g., *Paeonia, Anemone and Aconitum*) and monocotyledons (e.g., *Bambusa*) these cells possess tangential extensions which project into the cell cavity. Such palisade cells are known as **arm-palisade.** The projections in the cell wall of palisade cells provide maximum exposure area to the cell wall along which chloroplasts are arranged. In some members of Gesneraceae and Pipcraceae, inhabiting moist localities, the palisade cells become cone-like with a wide upper end and relatively narrow lower end. These cells are known as **funnel cells**.

In aquatic plants large air chambers or air canals are found in the mesophyll tissue. The cells forming the air chambers contain comparatively few chloroplasts. **Stellate cells** are present in the partition walls of air chambers and these cells provide mechanical support. Various types of idioblasts are also present in mesophyll, such as lithocysts (e.g. *Ficus*), oil glands (e.g. *Citrus*) and laticifers (e.g.*Calotropis*).

[III] Vascular tissue

Leaves receive their vascular supply from the vascular cylinder of the stem on which they are borne. Each leaf receives one, two, three or many vascular traces which may continue unbrached throughout the entire length of the leaf or may branch and anastomose. Vascular bundl of the leaf are called **veins** and these veins by further ramification form a network which is known as **venation.** In dicotyledonous leaves, usually there is a single main vein which passes through the centre of the lamina and gives off successively thinner branches that form a netted pattern throughout the lamina. This venation is called

reticulate venation. InMonocotyledonous leaves veins of relatively Uniform size are arranged longitudinally and conaverge and join at the apex of the leaf. Such venation is termed as **parallel** venation. However, in some monocotyledonous leaves as those of Orchidaceae the venation is reticulate. Parallel venation may also occur in some dicotyledons, such as *Plantago* and *Trgopogon.*

In dicotyledonous leaves which have reticulate venation veins can be differentiated into a number of size classes or 'orders'. The veins of primary order are the thickest veins of the leaf and they occur either singly (occupying median position) or form lateral pairs of roughly equal thickness. The **primary vein** (1^0) gives out lateral branches which form the veins of secondary order. The **secondaries** (2^0) can be easily differentiated as they are slightly thinner than the primaries. The secondaries ramify further to form veins of higher orders (tertiary, quatemary and so on). The veins of higher order may further divide many times and may merge into the reticulum or remain distinct.

Along the margins of leaf, the ultimate veins may show various patterns. The following types of marginal ultimate venations may be found in dicotyledonous leaves:

1. Incomplete. When the vein endings are free directly adjacent to the margin

2. Looped. When the ultimate veins along the margin are recurved to form loops

3. Fimbriate. When several high order veins are fused into a vein running just inside the leaf margin to form fimbrial vein

The smallest area of the leaf tissue surrounded by veins is known as **areole**. Areoles are of various shapes and sizes. They may be **well developed** withmeshes of almost uniform size and shape; **imperfect,** with meshes of irregular shape and size or **incomplete**, when one or more sides of the mesh are not bounded by a vein. The leaves of certain succulents, however, do not form areoles al all.

In monocotyledons where leaves have parallel venation, the vascular bundles, apart from those in the midrib, are arranged in a single row on either side of the midvein.

The midrib or midvein in most dicotyledons consists of a typical collateral bundle with an adaxial xylem and abaxial phloem. A thin strip of cambium is also present between the xylem and phloem. Sometimes, the midrib hasseveral vascular bundles which are arranged in a ring (e.g., *Liriodendron Vitis*), or in a semi-circle (e.g.. *Abrosia*) or irregularly distributed (e.g., *Helianthus*). The vascular tissue of midvein and larger veins has vessels in the xylem and sieve tube in the phloem. In smaller veins the xylem is represented by tracheids and the phloem by some sieve elements only. The larger veins of the leaves of some woody dicotyledons (e.g., *Ficus religiosa*) show a limited amount of secondary growth as a result of the activity of cambium occurring in between xylem and phloem.

In monocotyledonous leaves also the vascular bundles are Collateral but great variation occurs in the size and disposition of the components. On the basis of the shape of xylem and phloem groups and fibre characters the following major types of vascular bundles are recognized in the leaves of monocotyledons, : (i) elongated bundles With sclerenchymatous cap, somewhat linear Phloem and two to three groups of xylem elements of moderate size , (ii) oval bundles with fibrous cap and one large metaxylem elements Surrounded by few comparatively small xylem elements meeting with phloem along aslightly convex surface , (iii) oval bundle with a cap of stone cells, xylem and phloem meeting along a straight line and (iv) typical grass type bundle with xylem and phloem meeting along a distinct V or U shaped curve ,

[IV] Bundle sheath

In most of the angiosperm the cells surrounding the vascular tissue of leaves are morphologically distinct from mesophyll cells. These cells constitute the **bundle sheath**, In dicotyledons, the vascular bundles are surrounded by thin walled parenchymatous cells extending in the direction parallel to the veins. These cells may or may not contain chloroplasts.

The parenchymatous bundle sheath of dicotyledons usually extends up to the epidermis on one or both sides of the leaf. It probably plays an important role in the conduction of food material between the vascular bundle and epidermis.

In monocotyledons the vascular bundles are completely or partially surrounded by one or two bundle sheaths, each consisting of a single layerof cells. The outer parenchymatos sheath is translucent or may contain chloroplasts. The cells of the inner sheath are smaller, have thicker walls and do not contain chloroplast.

The chloroplast containing bundle sheath cells of some monocotyledons (e.g., maize, sugarcane and tropical grasses) have attracted the attention of many physiologists because of theirparticipation in alternate pathway of photosynthesis Hatch and Slack (1966) Observed that in all grasses the chloroplast of bundle sheath cells is morphologically distinct and devoid of grana. In such plants the first stable product of photosynthesis is a 4-carbon atom compound in place of 3-carbon atom compound of Calvin cycle.

12.3 Phyllotaxy

It is the mode of arrangement of leaves on the stem. It is very regular and mathematical. The leaves are arranged on the stem of a particular species in a definite manner. The arrangement is usually regular and leaves are never placed on the stem in haphazard manner.

In most of the leaves the foliage leaves are spread about on the stem with long or short internode between them, this arrangement is called **cauline**. In some of the plants the leaves are arise as a **cluster** or a **rosette** from the stem just on the top of the roor, such arrangement is called **radical**.

In cauline leaves there may be one, two, three or more leaves at each node.When there is only one leaf arrangement it is called **spiral** or **alternate** or **acyclic** (it is commonest type). If there are two or more leaves at each node, the phyllotaxy is called **Cyclic**.

Fig 12.10 : Phyllotaxy : Alternate

I Cyclic Phyllotaxy

In this type of phylotaxy the leaves at each node form a whorl with the leaves placed on a circle in which the angles between adjacent leaves are the same. Thus, if there be two leaves in a whorl, the two will be placed opposite one another. If there be three leaves, the angle between leaves in the same whorl is 120^0 (i.e.one third of a circle), if four, it is 90^0 and so on .when are more than two leaves in a whorl the phyllotaxy is called **verticillate**.

(i) Opposite Phyllotaxy

In this type the two leaves at each node are opposite to one another when the successive pairs of leaves placed at right angle to one another, the arrangement is called opposite decussate. This type is found in *Calotropis*, *Ixora* etc. When the successive pairs are placed exactly on the top of one another and as a result all the leaves lies in one plane and if we view from above all the leaves are found to lie in two vertical rows such type is called opposite superposed phyllotaxy and found in *Guava,Quisqualis*.

(ii) Verticillate Phyllotaxy

This type of phyllotaxy can be seen in *Nerium odorum* in which three leaves forming a whorl at each node in the case of *Alstonia scholaris* 3 or more leaves forming a whorl at each node. This type is also called **Whorled phyllotaxy**.

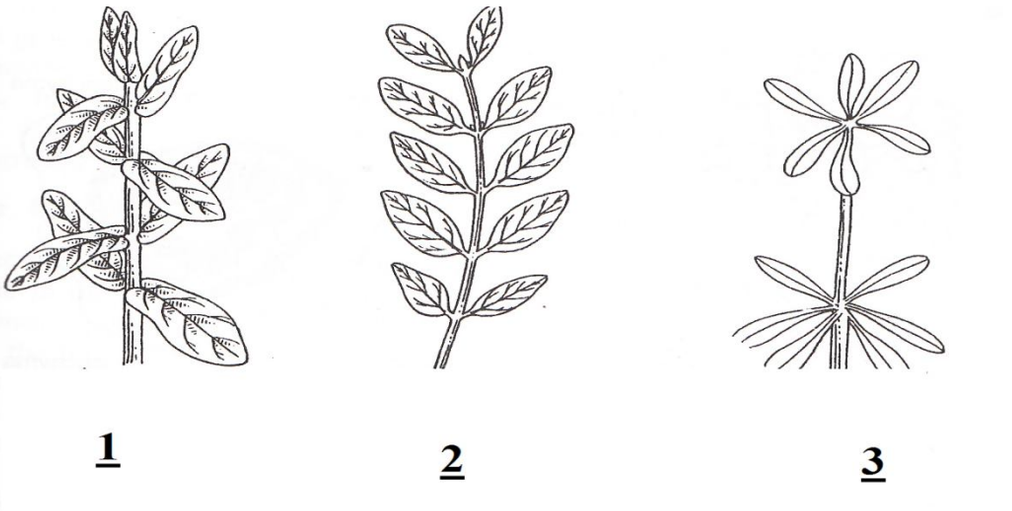


Fig. 12.11 : Phyllotaxy :1&2 Opposite, 3 Whorled

II Acyclic or Spiral phyllotaxy

This is the most common type of phyllotaxy in which one leaf arrangement. Here the mathematical regularity of the arrangement is astonishing. In this type we can seen the regular distance between any two consecutive leaves is constant; when looked from the top all the leaves found to lie in a fixed number of vertical rows:close examination shows that the leaves are evenly dispersed on all side of the stem.

12.4 Differentiation of Epidermis and Mesophyll

Leaf is made up of three type of tissue system---Epidermis, Mesophyll and Vascular tissues.The epidermis of leaves shows variation in number of layers, structure, arrangement of stomata, and arrangement of trichome and occurrence of specialized cells if any.

In the leaf mesophyll cells comprises the parenchymatous tissues internal to the epidermis.It undergoes differentiation to form the photosynthetic tissues. The various forms of mesophyll tissues were reviewed by Meyer (1962). In most of the dicot plants two type of parenchyma can be distighuished in the mesophyll---palisade parenchyma and spongy parenchyma. The cells of typical palisade parenchyma are elongated and to be arranged in rows. While in certain plants the palisade cells differ in shape from the typical ones.in certain species of Xanthorrhoeaceae small papilla-like projections and constriction which run arroud the cells are found (Fahn, 1954). In Lilium large lobes are present on the palisade cells which appear branched.

The palisade cells are found immediately below the uni or multiseriate epidermis but, sometimes a hypodermis may be present between the epidermis and palisade tissues.The

cells of palisade parenchyma may be arranged in one or more layers. The palisade tissues is usually found on the adaxial surface of the leaf.in some plants,e.g.Tamarix and species of Frankenia the adxial surface of the small layer is depressed to the stem,the palisade parenchyma is found only on the abaxial side of the leaf.In certain plants of xeromorphic species like Dianthus caryophyllus,species of Artemisia palisade parenchyma is present on both the side of leaf with the result that only a small strip of spongy parenchyma is present in the central portion of the lamina. A leaf in which the palisade parenchyma occurs on one side of the leaf and spongy parenchyma on other side of the leaf is termed as dorsiventral of bifacial leaf .Whereas if the palisade parenchyma occurs on both side of the leaf; the leaf is termed as isobilateral or isolateral leaf.

In certain plants like Zeamays thev mesophyll cells are more or less uniform in shape.In the certain species of Eucalyptus and Atriplex, it is not possible to distinguish between the two type of parenchyma, and the mesophyll is entirely made up of palisade cells.

The palisade tissue has become specialized in such a way that the efficiency of photosynthesis has been increased.Because of the shape and arrangement of the palisade cells the chloroplast can be placed so as to enable the maximum utilization of light.another factor that responsible for increasing photosynthetic efficiency is the presence of well-developed system of intercellular spaces which is present in the mesophyll tissues.The ratio of the internal surface area to the external surface area is of ecological importance(Turrell,1936,1939,1942,1944).The internal surface area of Styrax officinalis, is eight times larger than the external surface area.The specialization of the palisade tissues also result in more efficient photosynthesis.In the case of Styrax officinalis ,the free surface area of the palisade tissue is abou twise as large as that of the spongy tissue.Usually the intercellular spaces of the mesophyll develops schizogenously,but in certain plants the development may also be lysigenous by disintegration of group of cells(e.g. Water and marshy plants,Banana Leaf)

12.5 Ecological Anatomy of Xerophytic and Hydrophytic Leaf

Ecological Anatomy

Warming (1909) classified plant communities on the basis of dependence and relation of plants to water and referred to them as hydrophytes, mesophytes, xerophytes etc. **Hydrophytes** (*Gr. hudor - water; phyton - plant*) are those plants that live wholly or partly submerged in water or in very wet places. **Mesophytes** (*Gr. mesos-middle;phyron-plant*) are those plants that live in an environment with average or optimum water supply. These are, therefore, intermediate between hydrophytes and xerophytes. **Xerophytes** (Gr. Xeros-dry; phyton plant) include those plants which live in dry places where sufficient

water is not available to them. These can survive under long dry conditions and are, therefore, drought resistant plants. Between hydrophytes and mesophytes and extreme xerophytes and mesophytes, several plants showing different gradations are found. Beside these three forms, some other forms are also found in nature such as epiphytes, saprophytes, parasites, halophytes etc. **Epiphytes** grow upon other plants but they do not absorb food or minerals from them. **Saprophytes** grow upon dead organic matter while parasites grow with hostplants (living) and obtain their food from them. Halophytes grow in saline soil or water. Plants growing under different conditions become adapted structurally and physiologically to habitats in order to maintain a balance for their better growth. These adaptations are of special interest to study. Some of the anatomical features of plants belonging to these groups are discussed here.

Hydrophytes

Term *hydrophyte* refers to those plants which live in water or in a condition where water is available much more than their requirement. With respect to their relation to water and air, the hydrophytes are broadly divided into three groups, (i) submerged, (ii) floating, and (iii) amphibious.

I. Submerged plants

These plants grow entirely submerged in water either as attached submerged plants or submerged suspended plants. Common examples of plants belonging to this category are *Hydrilla, Valtisnaria, Zostera, Ceratophyllum, Myriophyllum, Elodea, Najas and Potamogeton*.

These plants show several morphological and anatomical adaptations in response to their environment.

Leaves: The epidermis of leaves consists of a single layer of tangentially elongated thin walled chlorophyllous cells lacking cuticle. Stomata are altogether absent, rarely non - functional and vestigeal ones may be found in some cases. The cells are capable of absorption of water and minerals and synthesis of food (photosynthesis). The mesophyll tissues remain undifferentiated but have large air chambers in continuation with stem. The air chambers are often separated by one or two celled thick diaphragms. Mesophyll cells have abundant chloroplasts. Vascular tissues mainly consist of phloem, xylem being represented by few thin walled elements. Leaves insome cases are membranous e.g. in Elodea these are only two cell thick except for the mid - rib region.

II. Floating plants.

The floating hydrophytes may be of three types: (i) free floating, e.g. *Spirodela, Lemna, Wolffia, Wolffiella, Azolla etc.;* (ii) rooted in mud with only flowers and leaves floating on the surface of water, e.g. *Nymphaea, Nelumbium,Victoria, Limnanthemum etc*.; and (iii) roots fixed and plants float freely in water, e.g. *Pistia, Trapa, Eichhornia etc. Eichhornia* may be found free floating also.

These plants show various adaptations in different parts.

Leaves: The floating leaves are usually characterised by having a large and specialised petiole and broader lamina. Leaves are not found in some floating forms such as *Spirodela, Lemna, Woiffia etc*. Here stem takes the function of a leaf.

Petiole: Petiole is usually a well developed part of the floating leaves. In *Nymphaea*, it is very long and cylindrical while in *Eichhornia*, it is swollen and spongy. The epidermis is single layered made up of thin walled cells. Cells are compactly arranged and in most cases lack cuticle. Hypodermis is two to three layered and made up of collenchyma. The cortex is well developed and has abundant air chambers, smaller towards periphery and larger towards the centre. Trichosclereids are commonly found which provide mechanical support to the thin and delicate walls of air chambers. Vascular bundles are found scattered in between the air chambers. The xylem is poorly developed and it is usually reduced to a single large element in each bundle but phloem is normally developed.

Lamina: In Lamina, the epidermis is well developed and consists of a single layer of thin walled cells. The cells have a thick cuticle or wax on upper surface. Leaves are often amphistomatic with functional stomata only on upper side. In *Nymphaea*, the leaves are, however, epistomatic type. The mesophyll tissues are often differentiated into palisade and spongy parenchyma. The palisade cells are arranged in a single layer *Trapa* but in several layers in *Nymphaea*. Spongy cells have large air chambers and several scattered trichosclereids (stellate cells). Air chambers provide buoyancy and trichosclereids provide mechanical support to the leaves. The vascular bundles have few xylem elements and relatively more developed phloem.

III. Amphibious plants

Amphibious plants usually grow in muddy soils or in soils saturated with water. These can withstand waterlogged conditions during rainy season or floods. Common examples of amphibious plants are *Eclipta, Lippia, Ranunculus, Sagittaria, Typha, Marsilea etc*. These plants grow under two diverse situations, the lower part of plant lives in an atmosphere of water rich conditions while upper part remains exposed to direct sun and hence live in hot conditions.

Accordingly different structural adaptations are found in the following manner.

Leaves: Since leaves of plants usually live in two different types of environments, heterophylly is found in several plants, e.g. *Ranunculus aquatilis*, *R sceleratus*, *Sagittaria sagittifolia* and *Limnophylla* sp.

Epidermis: It is single layered. The epidermis of aerial leaves remains covered with cuticle and has functional Stomata. Their mesophyll cells show differentiation into palisade and spongy parenchyma but such differentiation is not found in submerged leaves. Vascular tissues of aerial leaves are well developed. It remains Surrounded by sclerenchymatous sheath in *Sagittaria* and *Typha*. In several cases, sclerenchymatous patches along the margins are found.

General characters of hydrophytes

Adaptations of hydrophytic characters can be studied dividing into two broad categories, the external and internal features.

External Features

1. Roots are poorly developed, short; less branched and some times may be absent. Roots are well developed in *Nymphaea, Cyperus* and *Typha*, poorly developed in *Eichhornia, Spirodela* and *Lemna* and absent in *Woiffia, Wolffiella* and *Ceratophyllum*. In *Utricularia*, leaves take up the function of roots. In *Eichhornia* and *Pistia*, adventitious roots develop to provide buoyancy. InJussiaea, negatively geotropic spongy respiratory roots are found. Root hairs and root caps are completely absent.

2. Stem of the hydrophytes is delicate and flexible exhibiting sponginess.

3. Leaves are either highly dissected or narrow. In submerged plants, they are often provided with spongy covering. Emerged floating leaves possess waxy coating (e.g. *Nymphaea*), leaf hairs (e.g. *Salvinia*) or leathery texture. Amphibious hydrophytes show heterophylly, i.e. morphologically different kinds of leaves. Submerged leaves are furcated but emerged leaves have broad lamina (e.g. *Ranunculus sceleratus*).

4. **Petioles** of the leaves are very long and delicate in the rooted plants (e.g. *Nymphaea, Sagittaria*) which help the lamina to float on water surface. In floating plants, petioles may be reduced or altogether absent. *Eichhornia* develops bulbous petiole providing buoyancy.

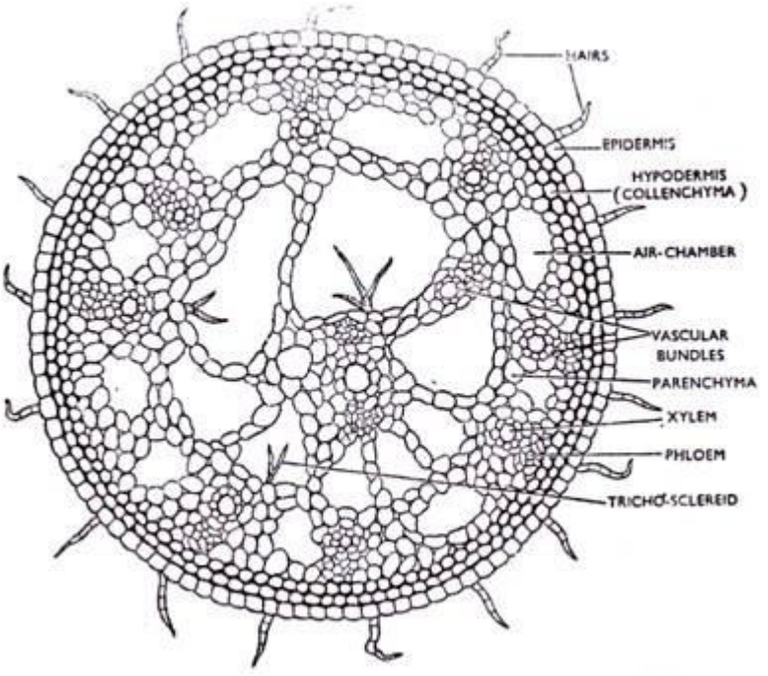


Fig. 10.12 : T.S. of Petiole *Nymphea*

5. Hydrophytes commonly reproduce vegetatively by forming runners, offset, stolon or by fragmentation.

Internal Features

1. Epidermis. The cells of the epidermis are thin - walled and often contain chloroplasts (e.g. *Hydrilla*). Epidermis of submerged organs lacks cuticle, epidermal hairs and stomata but that of emerged organs bears waxy coating and sotmata.

2. Cortex. Cortex is quite well developed and parenchymatous possessing large air chambers. Few outer layers of the cortex possess chloroplasts.

3. In leaves: mesophyll cells are present but are not differentiated into palisade and spongy parenchyma. Spongy parenchyma with large air cavities constitutes most of its part.

4. Vascular tissues are poorly developed. Commonly there is no distinction between xylem and phloem. Xylem is much reduced and may be represented by single element. Xylem cells are unlignified. Phloem is comparatively well developed.

5. Mechanical tissues are less developed. In certain cases, scattered raphides are present.

Some other characters are:

(i) Mucilaginous cells and sclereids are present.

(ii) Occasionally diaphragms form partition of air chambers.

(iii) Water is absorbed by the general surface of the submerged parts.

An amphibious plant may show hydrophytic characters in the submerged portion and xerophytic (or mesophytic) characters in the emerged portions. e.g. *Cyperus*, *Typha* etc.

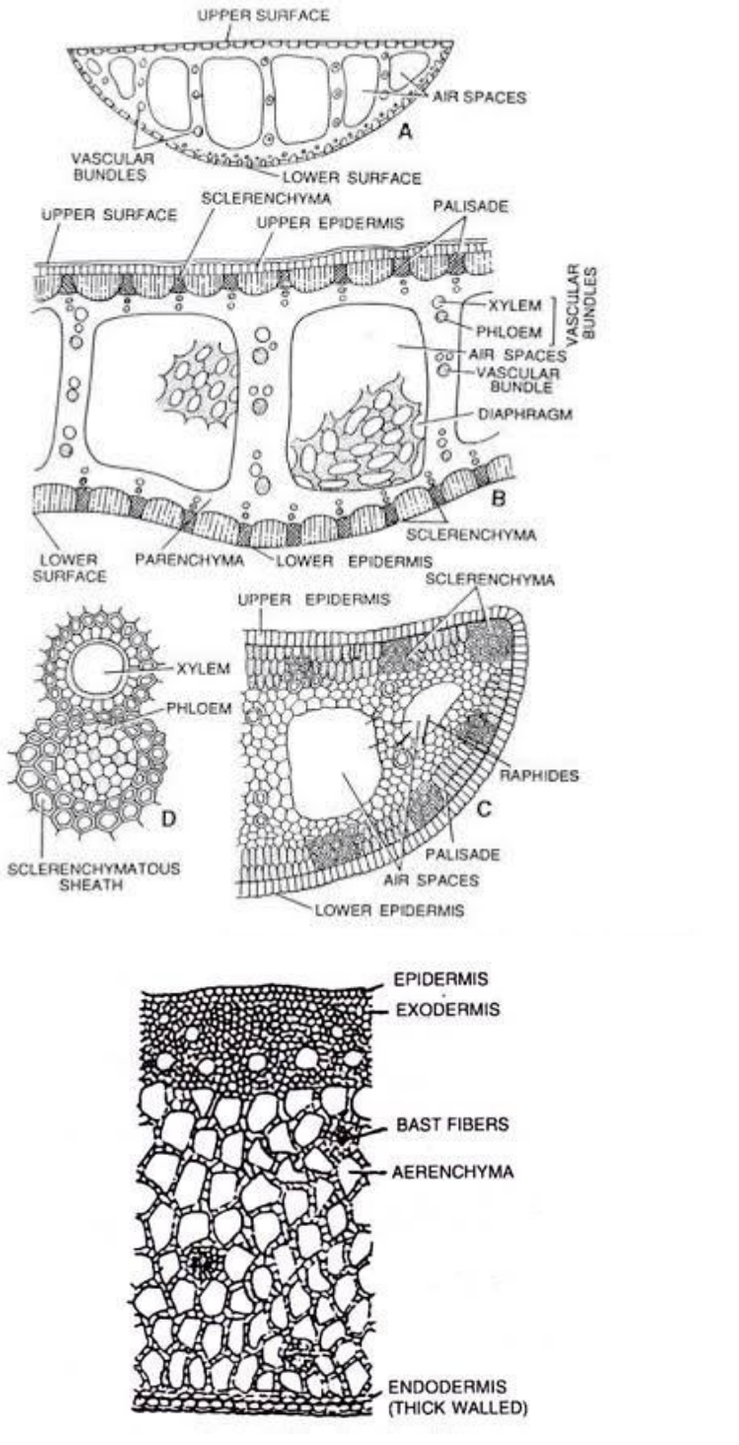


Fig12.13 V.T.S. of Hydropytic Leaf : *Typha*

A: T.S. of Leaf, B:Detailed structure of A, C:Detail of Corner of Leaf, D: Single Vascular Bundle, E: Cellular details

Xerophytes

Plants growing in dry habitats (physical dryness) or in habitats where water is unfit for absorption such as saline water (physiological dryness) are called xerophytes. Because of shortage of water, these plants develop certain characters to minimise transpiration. Xerophytes in general show following important characters.

External features

Roots: Root system of xerophytes is well developed, profusely branched and mechanically strong. Such plants develop a long tap root growing deep into the soil in search of water. Occasionally, roots may grow near the surface of soil to absorb the rain water.

Stem: Stems are generally hard and woody. A few xerophytes possess succulent (fleshy) stem due to abundance of mucilage which allows them to store water. Occasionally, the stem takes the function of leaf and adapts leaf - like structures to reduce the transpiring surface. When such leaf - like structure consists of one or two internodes, it is called **cladode** as found in *Asparagus* and if several internodes, it is called phylloclade as found in *Ruscus, Opuntia, Muehlenbeckia, Euphorbia, Casuarina* etc.

Leaves: **Leaves show variation in xerophytes and accordingly these areclassified into following categories.**

(i) Sclerophyllous: Leaves are strongly cutinised and sclerifled and appear strong and leathery, e.g. *Banksia.*

(ii) Trichophyllous: Leaves have hairy covering on the surface, e.g. *Nerium* and *Calotropis*.

(iii) Microphyllous: Leaves are reduced in size to avoid excessive transpiring surface, e.g. scale leaves in *Casuarina,* and *Equisetum*, spiny in *Cactus* and needle - like in *Pinus*. In *Capparis aphylla*, leaves are not seen.

(iv) Malacophyllous: Leaves, are fleshy due. to excessive mucilage to store maximum water, e.g. *Bryophyllum*, and *Kalanchoe*.

Certain xerophytes develop caducous leaves as in *Euphorluatirucalli* and *Capparis decidua*.

II. Anatomical features

Epidermis: Cells of epidermis are radially elongated and thick walled. The thick cutin gets deposited on the outer surface of cells. Deposition is sometimes mamillate and multilayered as in *Capparis* stem. The thickness of deposition is directly proportional to the xeric conditions. Sometimes, cuticle is supplemented by additional waxy coatings as in

Ricinus and *Calotropis*. Occasionally, the epidermis is multilayered which in case of a leaf may be on the dorsal surface as in Ficus or on both the surfaces as in *Nerium*. Presence of waxy coating on epidermal surfaces and multilayered epidermis help in reducing evaporation of water through epidermis.

Leaves are **hypostomatic** and stomata are usually present in cavity, i.e. stomata are **sunken type**. The stomated cavities are often provided with a dense covering of stomatal hairs so that the air in the cavity does not come in direct contact of the wind currents. Stomata in certain desert plants such as *Capparis spinosa* and *Aristida ciliata* may sometimes get blocked due to deposition of resinous matter or wax. All these adaptations are found to reduce the loss of water during transpiration.

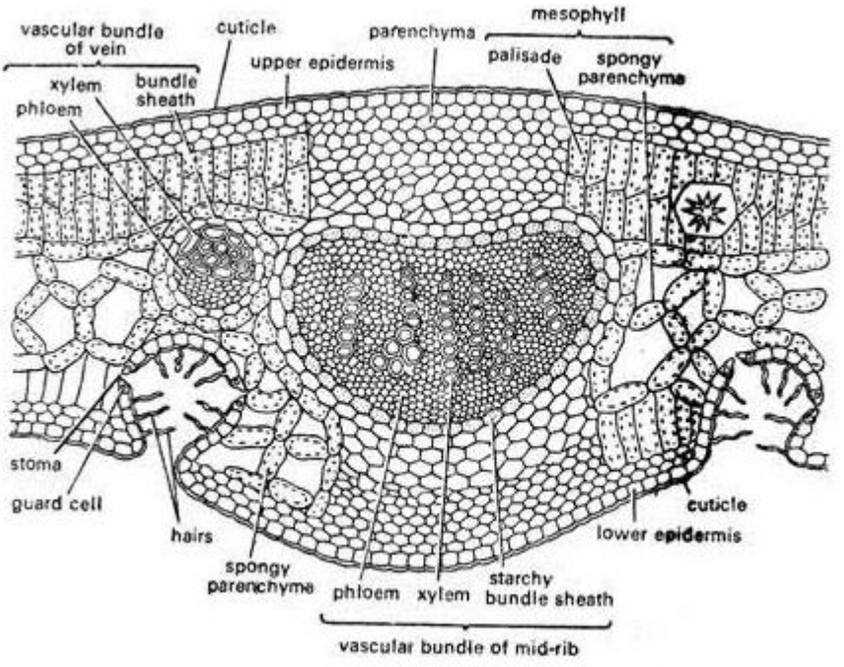
Epidermis of certain grass leaves have **bulliform cells** or **motor cells** which play an important role in rolling of the leaves during the period of dryness. These cells are thin walleo, geatly enlarged and sensitive to turgor changes. When these are turgid, leaf remains flattened and when flattened, the lamina rolls or minimise the exposure of transpiring surface.

Hypodermis: In most xerophytes hypodermis consists of mechanical tissues, also called sclerenchyma. There may one to many cell thick sclerenchymatous hypodermis both in stem as well as leaves. in certain cases, sclerenchyma are found in patches, e.g. in *Casuarina* stem, T-shaped sclerenchymatous hypodermis is found below the ridges. Besides providing mechnical support, sclerenchyma also protects the internal tissues from high light intensities, thus minimising the damage.

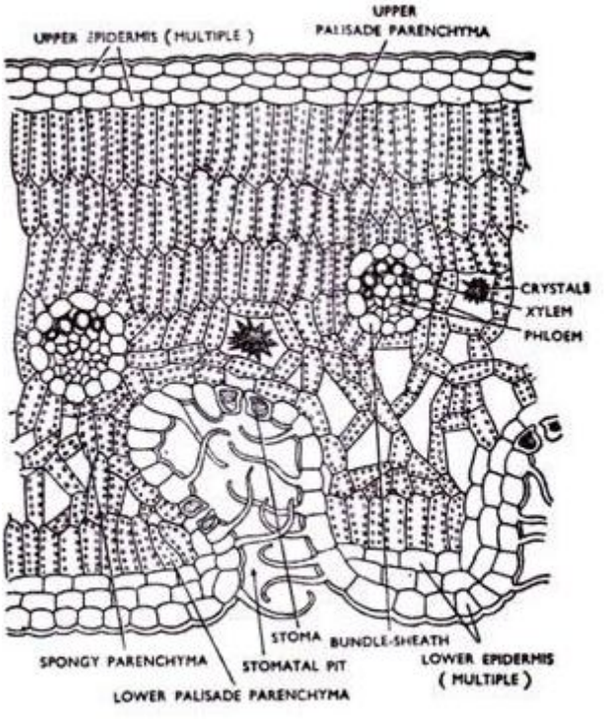
Mesophyll tissues: Mesophyll tissues are well developed and differentiated in xerophytes. These show a well - marked differentiation into well developed compactly arranged palisade tissues and loosely arranged spongy tissues. Palisade cells remain radially elongated to minimise the direct penetration of light. These are usually present towards dorsal surface and are arranged in a single layer. However, in *Nerium*, *Ficus* and *Atriplex*, the compactly arranged palisade tissues are present on both adaxial (dorsal) and abaxial (ventral) surfaces of a leaf and spongy tissues lie in between the palisade layers. In certain xerophytes, leaves are highly reduced and stem takes their function as in case of *Capparis*, *Salvadora* and *Casuarina*. In these cases, compactly arranged palisade cells are found present in the outer cortex of the stem.

The chloroplasts often show a relational distribution in palisade cells. These remain linearly arranged in a cell which has closer contact with the light. Chloroplasts can change their position in a palisade cell in response to intensities of light.

Pericycle: In most cases, pericycle consists of either sclerenchymatous



A



B

Fig. 12.14 : T.S. of Xerpotytic leaf : *Nerium*

A: Section through Mid-Rib; B: A Cellular Portion

12.6 Summary

Morphologically and antomically leaf is the most variable plant organ.It plays a very important role in photosynthesis reactions .Stomata found on the leaf surface is showing variation in structure and distribution and also help in plant classification . Its structure also shows various type of variation from ecological point of view. Xerophytic leaves are more adapted for least water available conditions

12.7 Glossary

- **Abaxial :** Directed away from the axis. Opposite of *adaxial.* With regard to a leaf, the lower, or "dorsal," surface
- **Adaxial :** Directed toward the axis. Opposite of *abaxial.* With regard to a leaf, the upper, or "ventral,"surface.
- **Annual ring :** In secondary xylem; growth ring formed during one season. The term is deprecated because more than one growth ring may be formed during a single year.
- **Apical cell :** Single cell that occupies the distal position in an apical meristem of root or shoot and is usually interpreted as the initial cell in the apical meristem; typical of seedless vascular plants.
- **Cambium :** A meristem with products of periclinal divisions commonly contributed in two directions and arranged in radial fi les. Term preferably applied only to the two lateral meristems, the *vascular cambium* and the *cork cambium*, or *phellogen.*
- **Meristem :** Embryonic tissue region, primarily concerned with formation of new cells.
- **Meristematic cell :** A cel synthesizing protoplasm and producing new cells by division; varies in form, size, wall thickness, and degree of vacuolation, but has only a primary cell wall.
- **Periderm :** Secondary protective tissue that replaces the epidermis in stems and roots, rarely in other organs.Consists of *phellem* (cork), *phellogen* (cork cambium),and *phelloderm*.
- **Phellem (cork) :** Protective tissue composed of nonliving cells with suberized walls and formed centrifugally by the phellogen (cork cambium) as part of the periderm. Replaces the epidermis in older stems and roots of many seed plants.
- **Phelloderm :** A tissue resembling cortical parenchyma produced centripetally by the phellogen (cork cambium) as part of the periderm of stems and roots in seed plants.

- **Phellogen** (**cork cambium**) **:** A lateral meristem forming the periderm, a secondary protective tissue common in stems and roots of seed plants. Produces phellem (cork) centrifugally, phelloderm centripetally by periclinal divisions.
- **Phyllotaxy** (or **phyllotaxis**) **:** Mode in which the leaves are arranged on the axis of a shoot.
- **Primary phloem :** Phloem tissue differentiating from procambium during primary growth and differentiation of a vascular plant. Commonly divided into the earlier *protophloem* and the later *metaphloem*. Not differentiated into axial and ray systems..
- **Sapwood** Outer part of the wood of stem or root containing
- **Vascular bundle :** A strand-like part of the vascular system composed of xylem and phloem.
- **Vascular cambium :** Lateral meristem that forms the secondary vascular tissues, secondary phloem and secondary xylem, in stem and root. Is located between those two tissues and, by periclinal divisions, gives off cells toward both tissues.
- **Xylem :** Principal water-conducting tissue in vascular plants characterized by the presence of tracheary elements. The xylem may also serve as a supporting tissue, especially the secondary xylem (wood).

12.8 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1 In the flowering plants the leaf gap found in which place-----

(a)Internode (b) Node (c) Shoot apex(d) Root apex

2 Multiple Epidermis found in in which of the following -------

(a) leaf of Grass (b) leaf of Nerium(c) leaf of Guava (d) leaf of maize

Section B : (Short Answer Type Questions)

1 What is Leaf Gap?

2 What is Abscission?

3. What are paracytic stomata?

Section C : (Long Answer Type Questions)

1. Decribe the internal structure of a dicot leaf with suitable diagrams.

2 Write short notes on the followings----(a)Leaf Abscission (b) Bundle sheath (c) Anatomy of Hydrophytic Petiole

12.9 References

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Unit-13

Introduction to Plant Reproduction

Structure of the Unit:

13.0 Objectives

After studying this unit you will be able to understand following concepts about reproduction in angiosperms:-

- Introductory aspects about reproduction in flowering plants.
- Artificial vegetative reproduction and its importance in agriculture.
- Development of flower and its morphological aspects.
- Sex determination and its genetic and evolutionary aspects
- Homeotic mutants in *Arbidopsis* and theirs current status of study.

13.1 Introduction

Most of the plants we see daily are angiosperms. The 250,000 species of flowering plants range in size from almost microscopic herbs to giant *Eucalyptus* trees, and their form varies from cacti, grasses, and daisies to aquatic pondweeds. Most shrubs and trees (other than conifers and *Ginkgo*) are also in this phylum. This unit focuses on reproduction in angiosperms because of the many uses by humans. Virtually all of our food is derived, directly or indirectly, from flowering plants; in fact, more than 90% of the calories we consume come from just over 100 species. Angiosperms are also sources of medicine, clothing, and building materials. While the other plant phyla also provide resources, they are outnumbered seven to one by the angiosperms. For example, there are only about 750 extant gymnosperm species! When flowering plants originated, Africa and South America were still connected to each other, as well as to Antarctica and India, and, via Antarctica, to Australia and New Zealand. These landmasses formed the great continent known as Gondwanaland. In the north, Eurasia and North America were united, forming another supercontinent called Laurasia. The huge landmass formed by the union of South America and Africa spanned the equator and probably had a climate characterized by extreme temperatures and aridity in its interior. Similar climates occur in the interiors of major continents at present. Much of the early evolution of angiosperms may have taken place in patches of drier and less favorable habitat found in the interior of Gondwanaland.

Many features of flowering plants seem to correlate with successful growth under arid and semiarid conditions. The transfer of pollen between flowers of separate plants, sometimes over long distances, ensures *out crossing* (cross-pollination between individuals of the same species) and may have been important in the early success of angiosperms. The various means of effective fruit dispersal that evolved in the group were also significant in the success of angiosperms. The rapid life cycle of some of the angiosperms *Arabidopsis* can go from seed to adult flowering plant in 24 days) was another factor. Asexual reproduction has given many invasive species a competitive edge. Xylem vessels and other anatomical and morphological features of the angiosperms correlate with their biological success. As early angiosperms evolved, all of these advantageous features became further elaborated and developed, and the pace of their diversification accelerated. Many features of flowering plants seem to correlate with successful growth under arid and semiarid conditions.

Human beings have had an intimate relationship with flowers since time immemorial. Flowers are objects of aesthetic, ornamental, social, religious and cultural value – they have always been used as symbols for conveying important human feelings such as love, affection, happiness, grief, mourning, etc. To a biologist, flowers are morphological and embryological marvels and the sites of sexual reproduction. The remarkable evolutionary success of flowering plants can be linked to their reproductive strategies. Here we explore some aspects from reproductive strategies in the angiosperms and how their unique features, flowers and fruits, have contributed to their success. Vegetative reproduction is a strategy to clonally propagate individuals. An unusual twist to sexual reproduction in some flowering plants is that senescence and death of the parent plant follows.

13.2 Reproduction in Plants (Angiosperms)

13.2.1 Vegetative Reproduction

While self-pollination reduces genetic variability, asexual reproduction results in genetically identical individuals because only mitotic cell divisions occur. In the absence of meiosis, individuals that are highly adapted to a relatively unchanging environment persist for the same reasons that self-pollination is favored. Most roses and potatoes for example, are vegetatively propagated. In a very common form of asexual reproduction called vegetative reproduction, new plant individuals are simply cloned from parts of adults. The forms of vegetative reproduction in plants are many and varied.

13.2.1.1 Natural Vegetative Reproduction

1. **Stolons:** Some plants reproduce by means of runners, or stolons are long, slender stems that grow along the surface of the soil. In the cultivated strawberry, for example, leaves, flowers, and roots are produced at every other node on the runner. Just beyond each second node, the tip of the runner turns up and becomes thickened. This thickened portion first produces adventitious roots and then a new shoot that continues the runner.
2. **Rhizomes:** Corms, bulbs, and tubers are rhizomes specialized for storage and reproduction. Rhizomes invade areas near the parent plant, and each node can give rise to a new flowering shoot. e. g. White potatoes are propagated artificially from tuber segments, each with one or more "eyes." The eyes, or "seed pieces," of potato give rise to the new plant.
3. **Suckers:** The roots of some plants—for example, cherry, apple, raspberry, and blackberry—produce "suckers," or sprouts, which give rise to new plants. Commercial varieties of banana do not produce seeds and are propagated by suckers that develop from buds on underground stems. When the root of a dandelion is broken, as it may be if one attempts to pull it from the ground, each root fragment may give rise to a new plant.
4. **Adventitious Leaves:** In a few species, even the leaves are reproductive. One example is the house plant *Kalanchoe daigremontiana,* familiar to many people as the "maternity plant," or "mother of thousands." The common names of this plant are based on the fact that numerous plantlets arise from meristematic tissue located in notches along the leaves. The maternity plant is ordinarily propagated by means of these small plants, which, when they mature, drop to the soil and take root.
5. **Apomixis:** In certain plants, including some citruses, certain grasses (such as Kentucky bluegrass), and dandelions, the embryos in the seeds may be produced asexually from the parent plant. This kind of asexual reproduction is known as *apomixis.* The seeds produced in this way give rise to individuals that are genetically identical to their parents. Thus, although these plants reproduce asexually by cloning diploid cells in the ovule, they also gain the advantage of seed dispersal, an adaptation usually associated with sexual reproduction. In general, vegetative reproduction, apomixis, and other forms of asexual reproduction promote the exact reproduction of individuals that are particularly well suited to a certain environment or habitat. Asexual reproduction among plants is far more common in harsh or marginal environments, where there is little margin for

variation. There are a greater proportion of asexual plants in the arctic, for example, than in temperate regions

13.2.1.2 Artificial Vegetative Reproduction

The methods to propagate asexually fruit plants are classified in two main and six sub categories: (Fig. 13.1 to 13.3)

I. Vegetative Propagation by Rooting

1. Stock Division

This method is used when the plant produces custard of rooted stems. In the late growing season and at beginning of the dormant stage, we can divide the plant for several new specimens according to the number of rooted stems. During this process, the old part of the plant should be removed. This technique is traditionally used for banana propagation.

2. Runner Division

The most widely known example of propagating plants with runner division is the strawberry. After fruiting, the strawberry begins to grow several runners. Wherever the runner has contact with the surface, it will root and form a new plant. After cutting off the new plant from the mother plant, we can transplant it before the cold season begins. The new plant will then produce fruit with its highest potential yield in the next year.

3. Layering

The main purpose of layering is to provide rooting for the stem of the mother plant. The new growing plant will keep the union with the mother plant until it is able to survive on its own. When this happens, the new plant will be cut off from the mother plant.

(a) Banking Up : This is the most common method to propagate pear, quince and apple rootstock (M type clones). This technique needs some preparative work. For about 2-3 years, we cut back the mother plant up to the surface level (or close to it), which will then result in a thicker root neck. This thick root neck will grow custard of stems. We then have to bank up the plant to 10-15 cm high (when the stems have an average 20-25 cm length). We can subsequently harvest the rooted stems at the end of the growing season.

(b) Simple Layering : This technique is commonly used for hazel-nut propagation. During the dormant season, stems are bent down into a 20-25 cm deep trench and covered with soil. The top parts of the stems, which usually have 2-3 buds on them, remain above the surface.

(c) Radial or Chinese Layering : In this case, the whole stem is bent down into a 10 cm deep trench and covered with soil. We then have to bank up the suckers to 2/3 of their

height on regular bases. This occurs when the sucker grows 10cm above the surface leaving only the top 1/3 of the plant free. This method is used mostly to propagate Gooseberry, Currant and Hazel-nut.

(d) Air Layering : This method is used on the tip of the branch, when stems are usually younger then one year old. A strip of bark is cut approximately 2 cm wide on the stem about 20 cm from the tip (just below a leaf stalk, or join). Once cut, a rooting hormone is applied and rooting material is placed under the strip. Finally, the cut is covered with a thin plastic bag, which is opened at both ends.

(e) Propagation by Rooted Cuttings

(i) Semi-Wooded Cuttings

- **Rooted Cuttings**: This method is one of the most popular vegetative propagation techniques, due to its use on both fruit and vegetable plants (such as cassava). The stems, which are used for cloning, have to be harvested during the dormant stage from the mother plant. This method always uses cuttings from the previous season's growth (more than one year old).
- **Hardwood Cuttings:** There are two types of hardwood cuttings. Those are taken from deciduous plants (such as mulberry, grape, apple, plum, peach, pomegranate and figs and those that are taken from evergreen plants (such as olive and granadilla). Hardwood cuttings are taken from deciduous plants in early winter after the plants have dropped their leaves.
- **Simple Cuttings:** This simple cutting is done on a stem, which usually contains 4-6 buds. The top part of the stem is cut off at an angle. If the cutting originated from an evergreen plant, the bottom two leaves should be removed and planted immediately after being cut. Typical examples for the use of this method are the Gooseberry, Currant, Quince, Fig and Olive.
- **Torn Cuttings:** This cutting is performed at the bottom portion of the stem where there is a union with the mother plant. This is a very old technique and it is rarely used now a days. **Hammer Cuttings:**
- In this case, a piece of twig is cut together with the stem. Some plant cuttings, like Gooseberry cuttings are difficult to root and the additional piece of twig helps to develop root system.

(ii)Truncheons

Truncheons are branches, about as thick as a human arm that we can grow into new plants. The branches are about 170-180 cm long. Cut the top of the branch at a slant, which prevents water from rotting the truncheon. Before planting the truncheon, it should first be kept under shade for a few days to develop a hard layer over the cut end. If the cut end is not covered with this hard layer, the truncheon may not root. The truncheon should be planted into a narrow hole about 60 cm deep. The best time for this method is the end of the dormant season when the plant still grows slowly. This method can be used with most trees which drip a white sap when they are cut.

(iii) Root Cuttings

Take root cuttings about 1 meter away from the tree trunk. These cuttings should be 20-25 cm long and 1-2 cm thick. Place these cuttings horizontally into the soil about 10 cm deep until they shoot. This technique is useful for propagation of guava, breadfruit, apple, blackberry and raspberry

II Vegetative Propagation by Grafting

1. Vegetative Propagation by Bud Grafting or Budding:

The method of budding is the most common technique for plant propagation in commercial nurseries. A "T" cut on the stock is done about 20-25 cm above the surface with a 2 cm long vertical cut and a 7-8 mm long horizontal cut on the stock. A slight twist with the budding knife may open the two flaps of bark. After that, the bud should be inserted under the two flaps of bark by pushing downward. Inverted T budding and Chip budding are also related buddings.

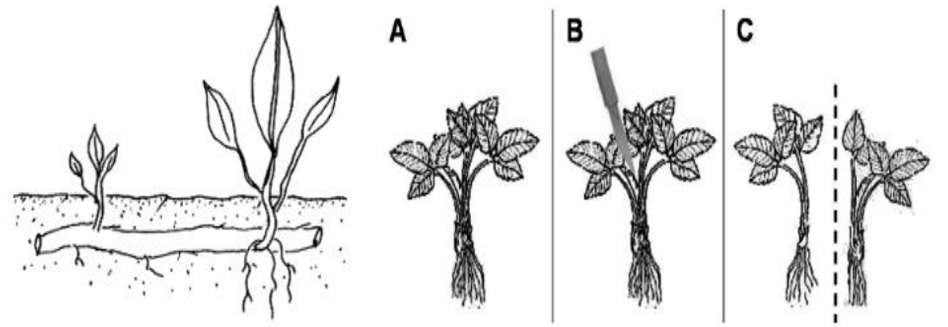
2. Vegetative Propagation by Grafting:

Two types of rootstock can be used for grafting: the cultivar and the seedling rootstock. The cultivar rootstock is produced by vegetative methods, generally by layering and cuttings. Seedling rootstocks grow from seed. One of the best examples for cultivar rootstock is the apple and for the seedling rootstock, the mango.

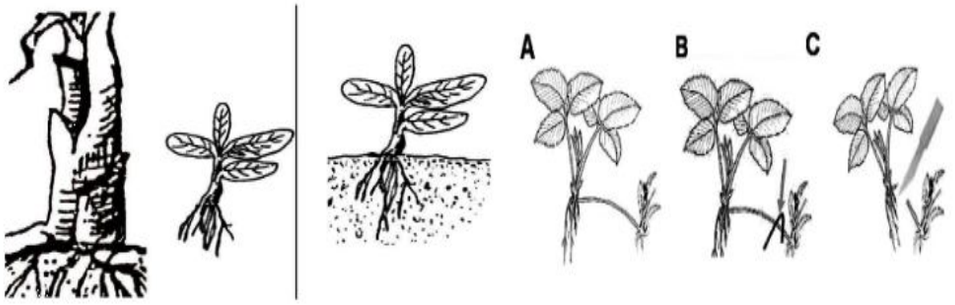
(a) Whip Grafting: The whip graft is useful for plants that unite easily. This method is useful for apples, mangos and pears. It can be used to graft root, stem or top graft. The diameter of the scion and rootstock should be the same, from the size of a pencil to 10-15 mm. It is further classified into Simple Whip grafting, Tongue Whip Grafting and root tongue grafting.

(b) Cleft Grafting: It is suited for apple and pears, but, in tropical areas, in can also be used for propagation of mango and avocado trees. Citrus and guava trees also use this method. In the case of top and side work, the scaffold limb is usually wider than the scion. In the case of tree propagation, both parts, the rootstock and scion, should be the same size. It is sub divided into Top cleft Grafting and Side Cleft Grafting.

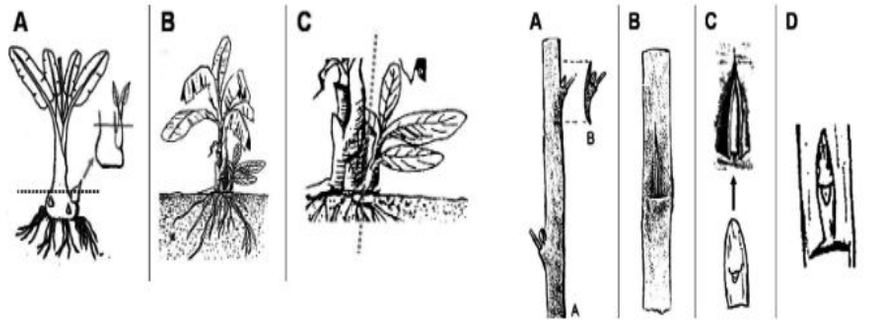
(c) Bark Grafting: Bark grafting is used when the stock is too large for whip grafting. It is one of the most difficult grafting techniques. Perfect application of this method requires much practice and experience. The use of this technique is common for pear, apple and different nuts grafting. There are different types of bark grafting like Top Bark Grafting, Side Bark Grafting, Wedge grafting, Bridge grafting and green grafting.



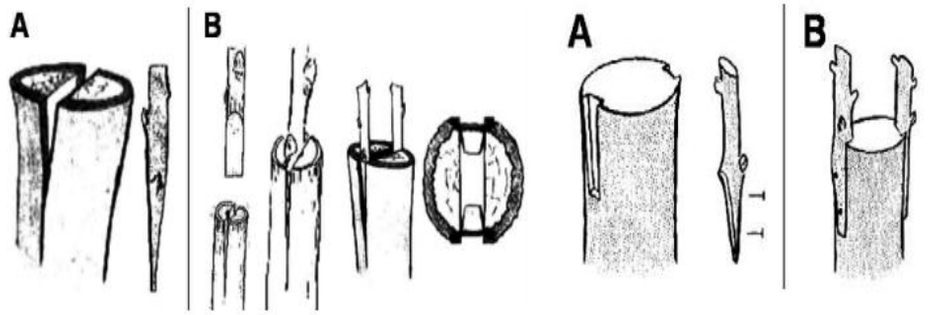
a. Root division b. Stock division



c. Runner Division d. Runner division-2

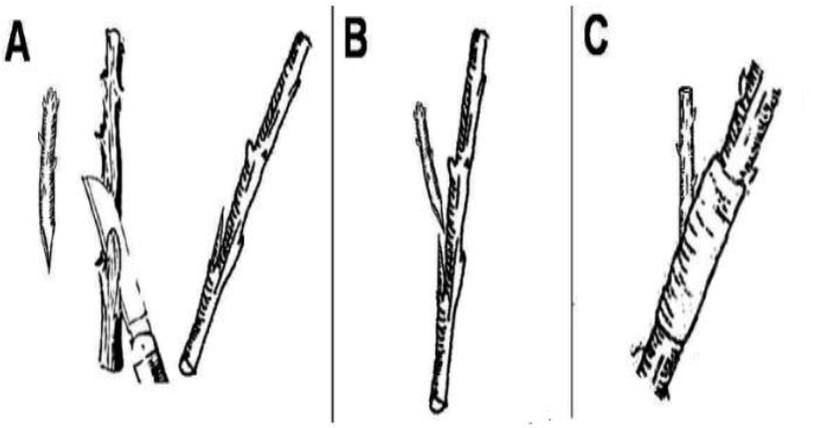


e. Sucker grafting f. Chip budding

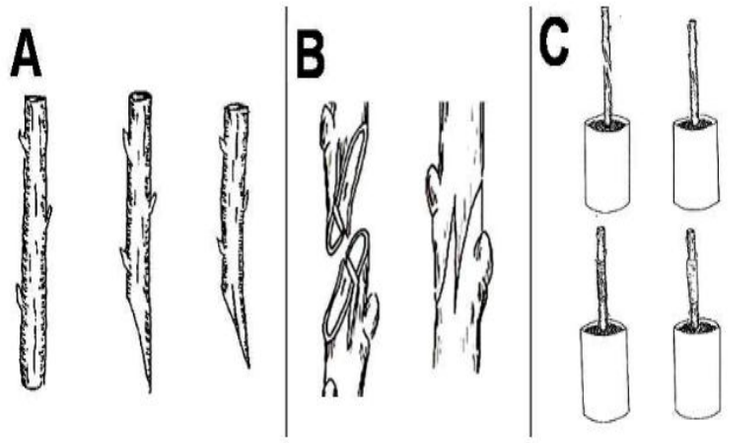


g. Top cleft grafting h. Top bark grafting

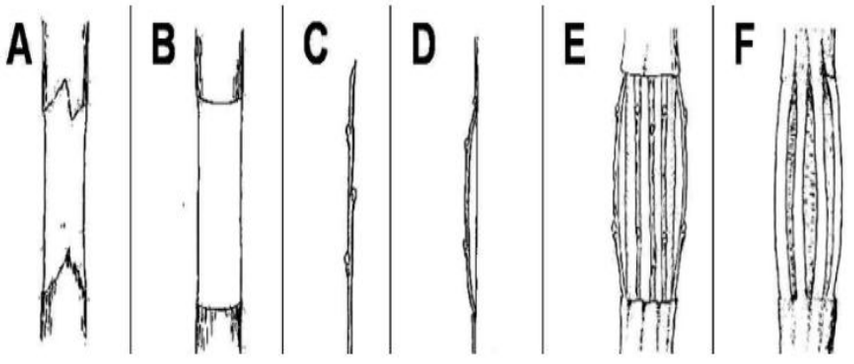
Fig. 13.1: Angiosperms: Various methods of vegetative reproduction



i. Side cleft grafting

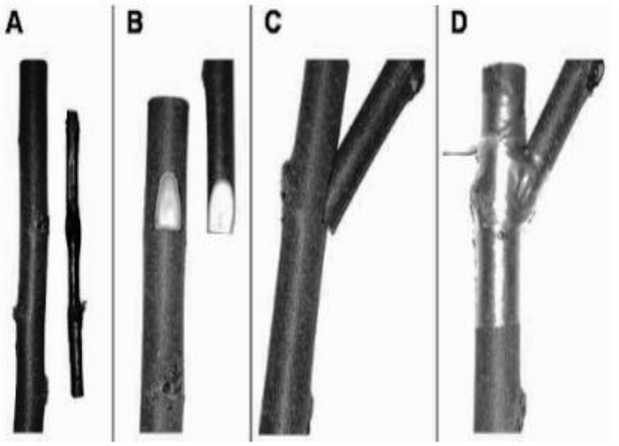


j.Tomgue chip grafting

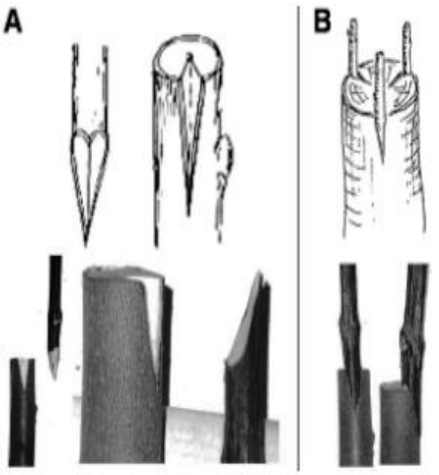


k. Bridge grafting

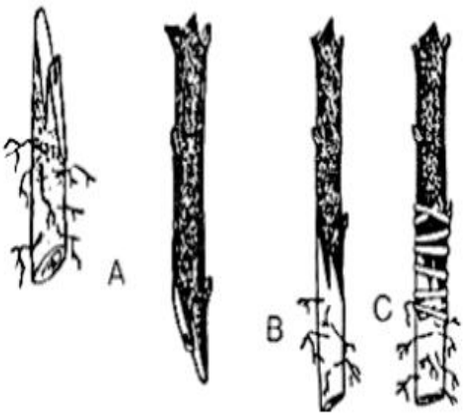
Fig. 13.2: Angiosperms : Various methods of vegetative reproduction



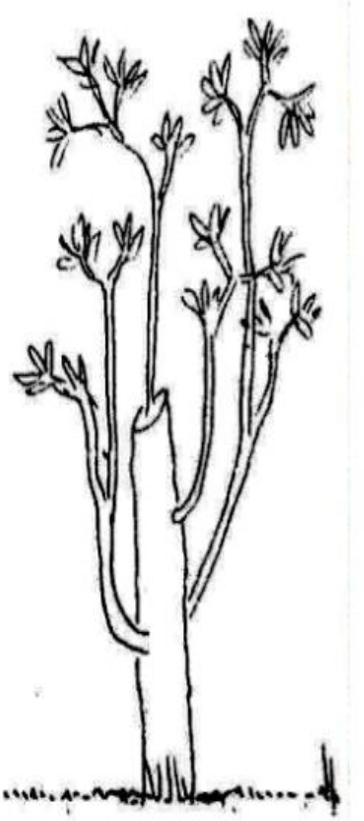
l. Side bark grafting



m. Wedge graftting.



n. Root tongue grafting



o. Truncheon

Fig. 13.3: Angiosperms : Various methods of vegetative reproduction

13.2.2 Sexual Reproduction:

The life cycle of plants is constituted by two generations: sporophytic and gametophytic. Gametophytic generation is the sexual generation. In angiosperms, when meiosis occurs in anther sporangia, the spores are called androspores. When it occurs in seminal rudiment sporangia, they are called gynospores. Sexuality is present in the gametophytes i.e. male gametophyte (Germinated bi or tri-nucleate microspore) and female gametophyte (generally 8-nucleate embryo sac), a generation that produces the male and female gametes, in individuals that are separate and show unisexuality. They form, through double fertilization, the new sporophytic generation (embryo) and the xenophytic generation (endosperm). Life cycle of angiosperms is given in Fig 13.4.

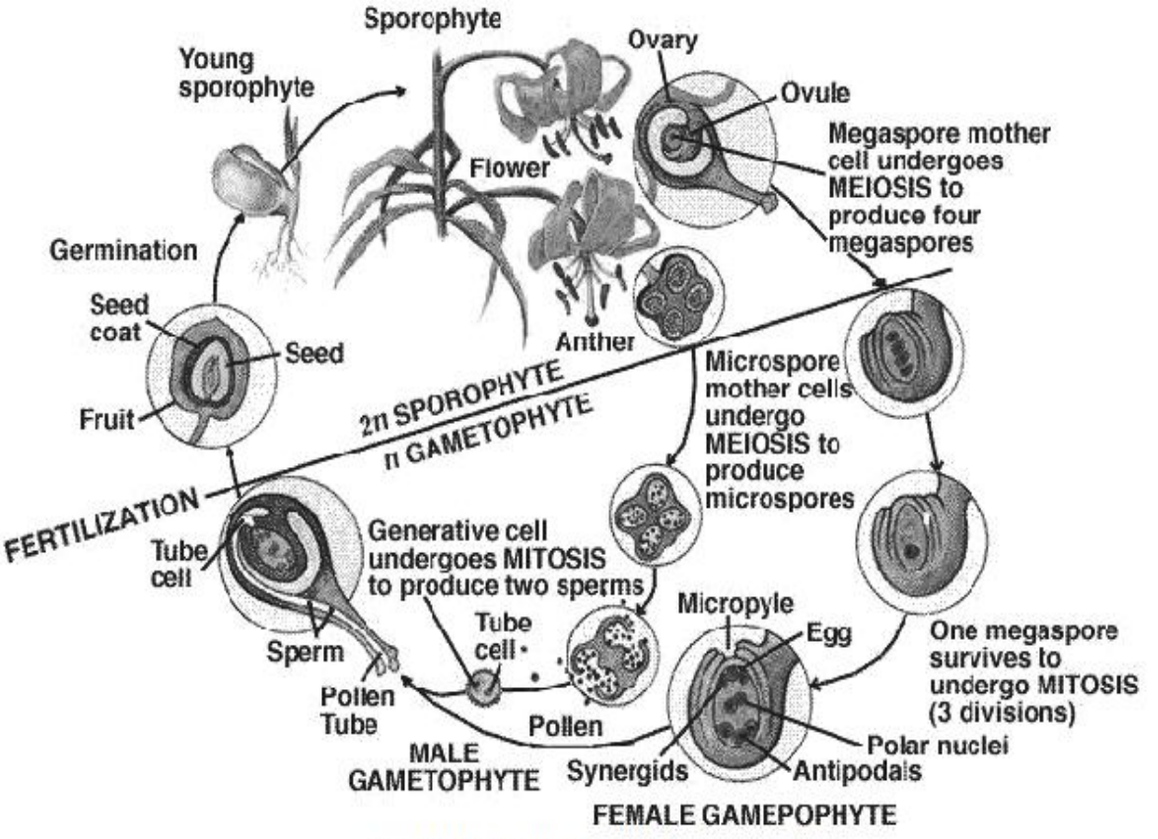


Fig.13.4 Life Cycle of Typical Angiosperm Plant

13.3 Flower Development

13.3.1 Rise of Flowering plants

Much of the early evolution of angiosperms may have taken place in patches of drier and less favorable habitat found in the interior of Gondwanaland. Many features of flowering plants seem to correlate with successful growth under arid and semiarid conditions. The

transfer of pollen between flowers of separate plants, sometimes over long distances, ensures *outcrossing* (cross-pollination between individuals of the same species) and may have been important in the early success of angiosperms. The various means of effective fruit dispersal that evolved in the group were also significant in the success of angiosperms. The rapid life cycle of some of the angiosperms (*Arabidopsis* can go from seed to adult flowering plant in 24 days) was another factor. Asexual reproduction has given many invasive species a competitive edge. Xylem vessels and other anatomical and morphological features of the angiosperms correlate with their biological success. As early angiosperms evolved, all of these advantageous features became further elaborated and developed, and the pace of their diversification accelerated. Angiosperms began to dominate temperate and tropical terrestrial communities about 80 to 90 million years ago, during the second half of the Cretaceous Period. Angiosperms arose in temperate and tropical terrestrial communities in a relatively short time. At about the time those angiosperms became abundant in the fossil record, pollen, leaves, flowers, and fruits of some families that still survive began to appear.

13.3.2 Evolution of Flower

Pollination in angiosperms does not involve direct contact between the pollen grain and the ovule. Pollen matures within the anthers and is transported, often by insects, birds, or other animals, to the stigma of another flower. When pollen reaches the stigma, it germinates, and a pollen tube grows down, carrying the sperm nuclei to the embryo sac. After double fertilization takes place, development of the embryo and endosperm begins. The seed matures within the ripening fruit; the germination of the seed initiates another life cycle. Successful pollination in many angiosperms depends on the regular attraction of **pollinators** such as insects, birds and other animals, so that pollen is transferred between plants of the same species. When animals disperse pollen, they perform the same functions for flowering plants that they do for themselves when they actively search out mates. The relationship between plant and pollinator can be quite intricate. Mutations in either partner can block reproduction. If a plant flowers at the "wrong" time, the pollinator may not be available. If the morphology of the flower or pollinator is altered, there may be physical barriers to pollination. Clearly floral morphology has coevolved with pollinators and the result is much more complex and diverse than the initiation of four distinct whorls of organs.

Characteristics of Floral Evolution

Diversity of angiosperms is partly due to the evolution of a great variety of floral phenotypes that may enhance the effectiveness of pollination. All floral organs are thought to have evolved from leaves. In early angiosperms, these organs maintain the spiral

phyllotaxy often found in leaves. The trend has been toward four distinct whorls. A complete flower has four whorls of parts (calyx, corolla, androecium, and gynoecium), while an incomplete flower lacks one or more of the whorls. In both complete and incomplete flowers, the **calyx** usually constitutes the outermost whorl; it consists of flattened appendages, called **sepals,** which protect the flower in the bud. The petals collectively make up the **corolla** and may be fused. Petals function to attract pollinators. While these two outer whorls of floral organs are sterile, they can enhance reproductive success. **Androecium** is a collective term for all the **stamens** of a flower. Stamens are specialized structures that bear the angiosperm microsporangia. There are similar structures bearing the microsporangia in the pollen cones of gymnosperms. Most living angiosperms have stamens whose **filaments** are slender and often threadlike, and whose four microsporangia are evident at the apex in a swollen portion, the **anther.** Some of the more primitive angiosperms have stamens that are flattened and leaflike, with the sporangia producing from the upper or lower surface. The **gynoecium** (from the Greek *gyne,* "woman," + *oikos,* "house") is a collective term for all the female parts of a flower. In most flowers, the gynoecium, which is unique to angiosperms, consists of a single **carpel** or two or more fused carpels. Single or fused carpels are often referred to as the simple or compound pistils, respectively. There may be several to many separate pistils, each formed from a single carpel. **Ovules** (which develop into seeds) are produced in the pistil's swollen lower portion, the **ovary,** which usually narrows at the top into a slender, neck like **style** with a pollen-receptive **stigma** at its apex. Sometimes the stigma is divided, with the number of stigma branches indicating how many carpels are in the particular pistil. Carpels are essentially in rolled floral leaves with ovules along the margins. It is possible that the first carpels were leaf blades that folded longitudinally; the margins, which had hairs, did not actually fuse until the fruit developed, but the hairs interlocked and were receptive to pollen. In the course of evolution, there is evidence the hairs became localized into a stigma, a style was formed, and the fusing of the carpel margins ultimately resulted in a pistil. In many modern flowering plants, the carpels have become highly modified and are not visually distinguishable from one another unless the pistil is cut open.

Trends of Floral Specialization

Two major evolutionary trends led to the wide diversity of modern flowering plants:

(1) separate floral parts have grouped together, or fused, and

(2) floral parts have been lost or reduced

In the more advanced angiosperms, the number of parts in each whorl has often been reduced from many to few. The spiral patterns of attachment of all floral parts in primitive

angiosperms have, in the course of evolution, given way to a single whorl at each level. The central axis of many flowers has shortened, and the whorls are close to one another. In some evolutionary lines, the members of one or more whorls have fused with one another, sometimes joining into a tube. In other kinds of flowering plants, different whorls may be fused together. Whole whorls may even be lost from the flower, which may lack sepals, petals, stamens, carpels, or various combinations of these structures. Modifications often relate to pollination mechanisms and, in some cases like the grasses, wind has replaced animals for pollen dispersal. While much floral diversity is the result of natural selection related to pollination, it is important to recognize the impact breeding (artificial selection) has had on flower morphology.

Trends in Floral Symmetry

Other trends in floral evolution have affected the symmetry of the flower. Primitive flowers such as those of buttercups are radically symmetrical; that is one could draw a line anywhere through the center and have two roughly equal halves. Flowers of many advanced groups are bilaterally symmetrical; that is, they are divisible into two equal parts along only a single plane. Examples of such flowers are snapdragons, mints, and orchids. Such bilaterally symmetrical flowers are also common among violets and peas. In these groups, they are often associated with advanced and highly precise pollination systems. Bilateral symmetry has arisen independently many times. In snapdragons, the *Cyclodia* gene regulates floral symmetry, and in its absence flowers are more radial. Here the evolutionary introduction of a single gene is sufficient to cause a dramatic change in morphology. Whether the same gene or functionally similar genes arose in parallel in other species is an open question.

13.3.3 Development of Flower in Angiosperms

Starting from primitive flower angiosperms have been evolved up to modern dicotyledons and monocotyledons. Development of flower in angiosperms can be studied under following heads.

The Primitive Flower

The type of flower now generally recognized as morphologically simple is one that shows the least change, under adaptive evolutionary modification, from the original primitive flower. Evidence now seems to support strongly the theory that the ancestral flower was bisexual, with numerous stamens and carpels, without a perianth, or with a uniseriate perianth of simple, bractlike organs. (This theory differs greatly from that still held by some botanists that the primitive flower was unisexual.) All the appendages were spirally arranged, and the flower was symmetrical and without fusion among its parts. From this

theoretically basic flower essentially the "pattern flower" of pre evolutionaiy taxonomy have developed many lines of modification, with reduction producing types more simple in kinds and number of organs, and with elaboration producing complexity in form. The major principles of evolutionary modification, upon wliich is based the acceptance of this type of flower as morphologically simple and primitive among living forms, are the following advances:

1. From many parts, indefinite in number, to few, definite in number
2. From three or four sets of appendages-perianth, androecium, and gynoecium to one
3. From spiral to whorled arrangement of appendages
4. From freedom of floral parts to fusion—connation and adnation
5. From radial symmetry (actinomorphy) to bilateral symmetry (zygomorphy)

Reduction in the Flower

Reduction in the flower may occur in many or all parts, simultaneously in several parts, or progressively from part to part. Loss may be of individual organs or of entire whorls of organs; loss may be minor, as loss of one petal or one stamen, or may occur in all parts of the flower, so that there remain only the receptacle and one or few sporophylls of one kind. All stages in loss of function and reduction in size can be seen in closely related taxa. Organs in process of evolutionary reduction, "vestigial organs," can be recognized by abortive form and structure and by position in the flower; vascular anatomy mav aid in the interpretation of vestigial structures where identity is uncertain. The vascular traces of lost organs are usually present when external form is reduced, and may persist in the receptacle after the organ itself has disappeared. Under reduction, organs may be reduced in form and structure and changed in function. Transformations of petals and stamens into glands and of stamens into staminodia are probably the commonest changes. Evidence of the change of petals and stamens into glands is usually apparent in position of the glands and type and origin of vascular supply. (Some glands and glandular surfaces represent merely secretory areas, not modified organs.) Plants with nonfunctioning petals are sometimes called apetalous when the petals are still present in vestigial or greatly reduced form, as in the Proteaceae

Reduction in the Stamen

Reduction in the stamen occurs in all stages, from abortion of sporangia only, to complete disappearance of the organ. The abortion of two sporangia one of each pair is frequent; abortion of three of the four is rare. Loss of the entire anther is frequent; the stamen survives as a sterile, laminar, or filamentous organ, which may be petaloid. The petals of the majority of families seem to represent completely petaloid stamens. Stages in the loss

of the stamen by gradual reduction in size of a staminodium are well shown in the genera of the Scrophulariaceae. In genera where no external remnant of the lost stamen survives, the vascular trace of the stamen is still present in the receptacle.

Reduction in the Carpel

Reduction in the carpel is primarily in size and in number of ovules. Primitively, the carpel contained many ovules. Reduction in ovules has been too few, and to one in the achene type. Loss of ovules may take place progressively from either end; the surviving ovules may be either the proximal or distal ones. The persistence of a median ovule appears to be rare. The position of the single ovule distal or proximal may form a good generic character, as in the Proteaceae. The sequence in ovule loss is demonstrated by the position of abortive ovules and presence of ovule traces where no ovule vestiges remain. This sequence in ovule loss is well shown in carpels of the Ranunculaceae and Rosaceae. Where there is but one ovule surviving, the position of that ovule in the ancestral follicle can often be shown only by vestigial ovule traces.

13.3.4 Basic structure of Flower

The morphological basis of the flower is rarely emphasized in definitions; often the flower is "the reproductive structure of the angiosperms." Morphologically, it is a determinate stem tip bearing sporophylls and, commonly, other appendages that are sterile but this definition applies equally well to many cones, those of the gymnosperms and even of some of the lower vascular plants. If the term flower is to be restricted to the fertile stem tip of the angiosperms and its appendages, it is necessary to compare these structures and their sporangia with those of other groups. The looseness of use of the term flower by botanists is, in part, responsible for the difficulty in defining the structure. By "the flowering plants" is commonly meant the angiosperms, but such phrases as "the flowering of the conifers" (referring to the period of pollination), "the flowers of the gymnosperms," and "the flowers of the seed plants" are frequently seen.

The Receptacle

The flower is, first of all, a stem tip, the receptacle, resembling in ontogeny and fundamental structure a vegetative tip. It consists of nodes and internodes and bears appendages. The nodes are usually closely crowded by shortening and often brought together by suppression of internodes. Apical growth is limited early in development, but other growth may continue until the fruit is mature. The receptacle is often greatly modified and unstem like in form, size, and structure; and, as it matures with the fruit, it may become still less stem like. On the receptacle are borne, typically, both fertile and

sterile appendages. The shortening and suppression of internodes bring the appendages close together, either in spirals or whorls.

The Sterile whorls

The sterile appendages are typically of two kinds: sepals, which together form the calyx; and petals, which make up the corolla. These appendages are below the fertile appendages, the calyx below the corolla. (In *Eupomatia,* petaloid staminodia form a pseudocorolla between the stamens and the carpels.) Sepals and petals commonly differ in form, size, and other characters. In some families, they may be closely alike, as in most of the Liliaceae; in others, transitional forms occur, as in the Magnoliaceae. Interpretation of the perianth as calyx or corolla may be difficult and unimportant where the organs, ranging from two to several, are spirally placed; and where the perianth is represented by only one or few appendages, which serve as a bud-scale-like cap. Commonly, sepals are more or less leaflike or bractlike in form and structure, especially in their vascular relations to the stem. Morphologically, they are modified leaves.

The Fertile whorls

The fertile appendages, also of leaf rank, are of two types: microsporophylls (stamens), which bear microsporangia; and megasporophylls (carpels), which bear megasporangia. The stamens constitute the androecium; the carpels, the gynoecium. (The term pistil is applied to a unit of the gynoecium: to a single carpel when the carpels are free from one another; to a group of carpels when they are fused to one another.) Where the flower has only one kind of sporophyll, it is unisexual; where both are present, it is bisexual. If flowers of both unisexual kinds are borne on the same plant, the taxon is monoecious; if staminate and pistillate flowers are borne on separate plants, the taxon is dioecious.

13.4 Floral Organ Differentiation

13.4.1 Differentiation of Calyx

Flower development began with initiation of sepals covered by a thin uniseriate protoderm. Differentiation of sepals starts at the margin of the flattened floral apex. Sepal primordial initiate simultaneously. The sepal's procambial strands discernible, and on the vascular bundles with tracheary elements arise. When buds enter into the state of winter dormancy, the sepals are covered by uniseriate protoderm with dense cytoplasm, and 4–5 layers of mesophyll cells also filled with dense cytoplasm.

13.4.2 Differentiation of Corolla

Following sepal initiation, four petal primordia initiate simultaneously at the edge of the floral meristem, alternating with sepals. Petal primordial formation starts after sepal

initiation and petals are produced. In early petal development, frequent mitotic divisions take place at the basal part of the petal primordium. Initially the basal part of the petals is narrow and contained procambial strands. At this time the petals enclose the developing androecium and gynoecium also. Gradually provascular bundles than vascular bundles with tracheary elements and petal enlargement happens.

13.4.3 Differentiation of Androecium

In flowering season, it starts from development of stamen primordial that contains dense cytoplasmic meristematic cells. Then procambial strands are formed, and the stamens are differentiated into anthers and filaments. After it uniseriate protoderm is discernible, provascular bundle are formed within the connective tissue, and layers of sporogenous tissue are visible. On the parietal side of the anther, anticlinal and periclinal mitotic divisions of sporogenous cells take place. The sporogenous cells (Group of pollen mother cells) contain dense cytoplasm undergoes meiosis. Anthers continue differentiation throughout the season. At the time of pollen maturity sporogenous tissue differentiate into pollen mother cells and these pollen mother cells form microspore tetrads by meiosis and Mitotic cytokinesis. The middle layer and tapetum began to degenerate at the same time. In this period the endothecium develope secondary cell wall thickenings. Changes are also seen in the stomium cells. The space between stomium cells enlarge and the cells become irregular inshape. During the following days, dehiscence occurred at the stomium.

Primitive Stamen

The primitive stamen has no distinction between filament and anther. The most primitive stamens in living angiosperms are probably those of some of the woody Ranales. These are broad, more or less leaflike organs, without, or with weak, distinction between fertile and sterile parts. The sporangia are borne near the center of the sporophyll on the abaxial side in Degeneriaceae, Annonaceae, and Himantandraceae; on the adaxial side in Austrobaileya and Magnolia, and other primitive taxa. Stamens with semi laminar form occur in other primitive or fairly primitive families Ceratophyllaceae, Lardizabalaceae, Eupomatiaceae, Lactoridaceae, and Nymphaeaceae. In the specialization of tilts simple stamen, there has been reduction of the sterile tissues, with retraction of the marginal areas. The lamina has been progressively narrowed. The proximal part became the filament; the median section with the sporangia and the distal part became the anther. In the anther, the midvein region formed the connective and the sterile, distal part, the appendage.

Anther Development

The anther shows great variety in form and in its relation to the filament. Its morphological details are difficult to determine, so great has been the reduction and so elaborate the form of the greatly reduced types. In form, the anther ranges from linear to arrow shaped, sub globose, and strongly four-angled. Typically, it is two-lobed, deeply so in the more specialized types. The lobes are called anther lobes and anther sacs. With lateral enlargement of the connective, the anther lobes may be widely separated, and the lobes simulate anthers, as in the Commelinaceae, Melastomaceae, and many Labiatae.

Development of Filament

The filament ranges in form from broad and winged to terete and thread like, and from short to long. The broader and shorter types are, in general, the more primitive. Sessile anthers occur occasionally eg. *Juglans*. Absence of the filament may be the result of reduction in adaptation to special habitats or methods of pollination, as in many aquatic genera *Najas, Zostera* or the result of adnation of the filament to the perianth, where the filament is absent superficially, though not lost morphologically, and is often represented by its vascular supply some of the Proteaceae and Loranthaceae. It may be much enlarged and may have appendages at the top, *Mahonia*, at the base, *Viola*. The morphological nature of the appendages is difficult to determine and is clearly various. Some appendages are glandular excrescences or modifications of form correlated with pollination methods. Lateral appendages may be prominent, as in some Amaryllidaceae, where they become petaloid and connate, forming a corona. Anatomical structure and comparison with related taxa are necessary to determine the nature of these appendages.

Reduction of Anther

Reduction in the stamen occurs in all stages, from abortion of sporangia only, to complete disappearance of the organ. The abortion of two sporangia one of each pair is frequent; abortion of three of the four is rare. Loss of the entire anther is frequent; the stamen survives as a sterile, laminar, or filamentous organ, which may be petaloid. The petals of the majority of families seem to represent completely petaloid stamens. Stages in the loss of the stamen by gradual reduction in size of a staminodium are well shown in the genera of the Scrophulariaceae. In genera where no external remnant of the lost stamen survives, the vascular trace of the stamen is still present in the receptacle. In reduction in the androecium, individual organs or entire whorls may be lost. In related taxa, the inner whorl may be lost in some families, as in the Iridaceae; the outer whorl in others, as in the Burmanniaceae and Haemodoraceae. The Orchidaceae are an example of members lost from both ancestral whorls.

Fusion in the Androecium

Connation of members of a whorlontog enetic or phylogenetic is frequent in both whorled and fasciculate androecia. An androecium with connation by filaments is monadelphous where all stamens form a single cluster; diadelphous where two clusters are formed; polyadelphous where there are more than two clusters. It is adelphous where there is no connation. An androecium with anthers united is termed syngenesious. Connate anthers are infrequent or rare the Typhaceae, some Cucurbitaceae. The anthers of the Lobelioideae seem to show stages from cohesion to connation. Fusion by filaments monadelphous or diadelphous may involve one, two, or perhaps more whorls. Union of two whorls to form an apparent one is frequent, as in some legumes and the Thymelaeaceae.

Adnation of Anther

Adnation of the Androecium Fusion of stamens to other organs of the flower, especially to the corolla (this is termed epipetaly), is common. Fusion to the calyx is less common than to the corolla Proteaceae. Fusion to the carpels, where all the outer organs are together fused to the gynoecium in perigyny and epigyny, is common. Fusion to carpels alone is rare *Sarcandra*, Monimiaceae. Fusion to styles and stigma, with the formation of a gunostemium, is characteristic of the Orchidaceae and Stylidiaceae. Adnation may be by filaments for part or for all their length, where the anthers are sessile on other organs. (The term sessile is unfortunately applied both to anthers where free stamens consist of anthers only, and to those where filaments merge with the uniting organ.) Adnation of the anther to other organs varies in extent, from attachment near the base only to union by the entire dorsal or ventral surface. The dorsal surface to a sepal in many Proteaceae, the entire ventral surface to the gynoecium in the orchids.

Cohesion of Anther

Connation of members of a whorl ontogenetic or phylogenetic is frequent in both whorled and fasciculate androecia. An androecium with connation by filaments is monadelphous where all stamens form a single cluster; diadelphous where two clusters are formed; polyadelphous where there are more than two clusters. It is adelphous where there is no connation. An androecium with anthers united is termed syngenesious. Fusion by filaments monadelphous or diadelphous may involve one, two, or perhaps more whorls. Union of two whorls to form an apparent one is frequent, as in some legumes and the Thymelaeaceae.

13.4.4 Differentiation of Gynoecium

The ovule-bearing organs of the flower, the carpels or megasporophylls, make up the gynoecium. They range in number from many to one and, in arrangement, from spiral to

whorled. The carpels may be free from fusion with one another the gynoecium apocarpous or connate in various degrees the gynoecium syncarpous. A line between apocarpy and syncarpy is difficult to draw, because fusion may be slight and even developed late in ontogeny. The gynoecium perhaps shows more simply than the androecium and the perianth the major changes in the evolution of the flower. Especially prominent are the advances from spiral to whorled arrangement, from free to fused members, and from many to one which is pseudoterminal.

Primitive Carpel

The primitive carpel (Fig. 13.5) arises on the floral meristem as a more or less crescent-shaped primordium, which soon becomes broadened by lateral extension. Apical and marginal meristems appear early and increase its length and width. The presence of marginal meristems in the carpel was recognized only in the early twentieth century. The apparent absence of marginal meristems had earlier been considered evidence that the carpel was unlike the leaf. Differential growth brings about an upturning and bringing together of the lateral "wings," with, ultimately, their appression and more or less complete histological union. Comparative studies of the general method of closure of the carpel seem not to have been made. Earlier descriptions of carpel closure as an upfolding or inrolling appear to be generally accurate, though little attention has been given to details of contact from the stand point of position of the marginal meristems. Histological studies of the development of some primitive carpels especially *Degeneria* show a simple closing by folding; the conduplicate carpel so formed has the ventral surfaces of the lamina halves in contact, with the edges side by side. The frequent occurrence of the conduplicate type in primitive taxa and its simplicity have been by reduction of the marginal strips and a withdrawal of the edges, together with the ventral, half-inverted, placental bundles. But carpels with edges and margins deeply inrolled are complex and seem not to have been derived from the conduplicate type.

The Complex Carpel

The differentiation of ovary, style, and stigma from the simple, primitive carpel was a gradual one, which took place in many lines, with elaboration of style and stigma proceeding at different rates. The stigma is sometimes sessile, the style undeveloped in some of the Winteraceae and Euptelea; the style may be well developed, the stigma still a primitive stigmatic crest, decurrent on the style, as in Cercidiphylium. Morphologically, the style is usually the distal part of the primitive carpel. with its ovules lost; several families show gradual transition from ovary to style, with vestigial ovules and traces for lost ovules in the transitional region. Elongation of the style is characteristic of many taxa. The nature of the stigma is evident in the stigmatic crests of primitive carpels, especially

those of the woody Ranales, such as *Degeneria* and the Winteraceae. The pollen-receiving surface of the stigma is characteristically papillose and often secretory. Where this area extends the full length of the carpel, forming an undifferentiated stigma, a stigmatic crest, the papillae cover the margins of the carpel and narrow adjacent bands of the lamina surface. Where the marginal areas are merely approximated or appressed, the carpel still open, the papillae fill the slit loosely or compactly.

Apocarpous Gynoecium

An apocarpous gynoecium is made up of many carpels that are separate from each other. When we look at an apocarpous flower, such as a rose, we can see many small carpels clustered together in the center. The gynoecia in which carpels are free from fusion with one another show a single ovarian chamber, with placentation parietal, free central or basal in apocarpus gynoecia. e. g. Ranunculaceae, Asclepidiaceae, Rosaceae.

Syncarpous Gynoecium or Coenocarpous Gynoecia

Attempts have been made to supplant "syncarpous" by introducing a term, coenocarpous, reviving an old term, paracarpous, and redefining syncarpous. In the new treatment, gynoecia made up of united carpels are coenocarpous; those only those with two or more separate chambers and longitudinal fusion incomplete are syncarpous. Syncarpous gynoecia, under this definition, may show three structural zones: a fertile base, the ovary, which is syncarpous; a median part, the style, which is paracarpous; and a stigma, apocarpous. These terms, so used, are not good, because coenocarpous and syncarpous have much the same meaning, and the classification is valueless, because "paracarpous" and "syncarpous" forms are considered morphologically distinct types. The term false coenocarpy has been proposed to describe gynoecia where adnation of carpels to a cup-shaped receptacle ties together carpels otherwise free Hydrocharitaceae, Pomoideae, *Butomus.*

13.5 Sex Determination

13.5.1 Bisexual and Unisexual Flowers

In most species of flowering plants, cross-pollination (allogamy) is a common breeding mechanism. Outcrossing avoids the deleterious effects of inbreeding depression and promotes heterozygosity, genetic variability, and genetic exchange, the consequences of which are advantageous to the long-term surviva1 and adaptation of a species. Plants have evolved various mechanisms to promote allogamy, including the production of unisexual staminate or pistillate flowers on the same (monoecious) or different (dioecious) plants.

13.5.2 Regulation of Unisexuality

Production of unisexual flowers can be controlled by selectively activating or inactivating homeotic gene function. The available data, based on mutational analysis of the bisexual flowers of *Arabidopsis* and *Antirrhinum*, do not seem to support this idea. Basically, homeotic genes control organ formation in two or more whorls. Phenotypes conditioned by mutant alleles of these genes often result in homeotic transformation of the floral organs of two adjacent whorls into different structures. For instance, mutations in homeotic genes acting in region B cause the transformation of petals into sepals and stamens into carpels. These patterns are a typical of unisexual flowers found in natural plant populations, in which a single whorl (i.e., stamens or carpels) is usually affected. It is possible that sex determination genes might selectively affect the action of homeotic genes in one whorl, such that stamen development is altered, for example, without secondary effects on carpel formation. Moreover, there are examples of homeotic genes acting in a single whorl: the *Arabidopsis* homeotic mutation, **flo70,** also known as superman, replaces stamens with carpels; the heptandra mutants of *Digitalis* selectively affect whorl2, replacing petals with stamens; and certain petunia mutants, such as green peta/ and ***ph3,*** also show defects in just one whorl. To our knowledge, however, the attainment of unisexuality in flowers by means of homeotic transformation has not been reported as a mechanism of sex determination in natural populations. Unisexuality in plants is usually caused by the reduction or abortion of sex organ primordia; given the available data, a more plausible explanation is that sex determination genes act down stream or independently of homeotic functions. Consistent with this model are detailed morphological studies of severa1 unisexual plants, which have shown that unisexual flowers often pass through a "bisexual stage" in which all floral organs are initiated. Only in *Mercurialis* (mercury) and *Cannebis* (hemp) do the floral primordia lack any vestiges of inappropriate sex organs (see below). The formation of unisexual flowers from this bisexual meristem requires the action of sex determination genes. These genes have been identified in maize by the analysis of mutants that misregulate the normal program of unisexuality.

13.5.3 Genetics of Sexuality in Dioecious Plants

Active-Y System of Sex Determination

Heteromorphic sex chromosomes are rarely found in angiosperms but have been reported in a number of plant species including *Rumex, Cannabis*, *Humulus,* and *Silene*. In *dioecious Silene,* males are the heterogametic sex (XY) and females are homogametic (XX). As is the case in mammals, *Silene* has an active Y system of sex determination, with dominant male factors and female suppressing factors mapping to the Y

chromosome. The X chromosome appears to be essential in both males and females because only monoploid females can be obtained by in vitro techniques. Application of hormones, including GA, auxins, and cytokinins, does not result in sex conversion. However, the presence of a single Y chromosome can suppress female development when three X chromosomes are present. Higher X copy number overcomes the **Y** chromosome masculinization effect. Autosome ratios have no profound effects on the sex determining factors present on the **Y** chromosome. This suggests that the Y chromosome is decisive in determining sex in *Silene*. Three different regions of the Y chromosome have been identified as having separate functions in sex determination. One end contains a genetic factor (or factors) that suppresses formation of the gynoecium, the opposite end contains a male fertility factor (or factors), and the middle region includes a gene or genes needed for anther initiation. Therefore, the Y chromosome of *Silene* contains complete linkage between female-suppressor and essential male sex genes. *Asparagus* is generally a dioecious plant, with sex determined by homomorphic sex chromosomes in which the males (XY) are the heterogametic sex. Genetic evidence suggests that asparagus is "male dominant" and contains male-activator-female-suppressor genetic determinants similar to those postulated for *Silene*. In addition to these major sex determination genes, genetic modifiers can influence the stage of stylar degeneracy. In the dioecious populations, male plants with a few perfect flowers are occasionally found. These flowers can self-pollinate and produce homogametic males (YY). Because males are the desired sex in commercial applications, due to their increased vigor, selected YY male and XX female plants are used as parents for producing all male F1 hybrid seed. In summary, the absence of heteromorphic chromosomes and the viability of the YY genotype suggest that dioecy in asparagus may have been derived relatively recently.

X-to- Autosome Balance system of sex determination

Approximately 10 dioecious species exist in the genus *Rumex,* sub genus *Acefosa*, in which, in contrast to *Silene,* the X-to autosome ratio appears to control sex determination. Females are XX and males XY_1Y_2 (2n = 14 and 2n = 15, respectively); however, diploid plants with XXY and XXY_1Y_2 genotypes are fertile females. The Y chromosomes are late replicating and heterochromatic. In polyploids, an X-to-autosome ratio of 1.0 or higher is female; X-to-autosome ratios of 0.5 or lower are males. Intersexes (partia1 male/female) or hermaphrodites result from ratios of between 0.5 and 1.0. Sex is determined by X-to-autosome ratios even in plants that are trisomic for single autosomes. The Y chromosomes in *Rumex* are required for pollen fertility but do not seem to be required for stamen development. Both Y1 and Y2 appear to be required for normal progression of microspore mother cells through meiosis. In contrast to *Silene*, Y chromosomes of *Rumex* do not

inhibit female gynoecium development. Thus, the situation in *Rumex* is remarkably similar to that in *Drosophila* and *Caenorhabdifis elegans,* in which the primary determinant of sex is the X-to autosome ratio. Two species of the genus *Humulus* (hops) are dioecious, with a sex determination system similar to that of *Rumex*. The sex chromosomes of two species (*H. lupulus* and *H. japonicus*) are heteromorphic, and, as with Rumex, females (2n = 14 + XX) and males (XY_1Yp) are determined by X-to-autosome ratios rather than by the presence or absence of the Y chromosome. In cultivated hops, an XX female-XY male system is found, and multiple X systems (X_1X_1XpXp females, $X_1Y_1X_2Y_2$ males) are found in Japanese varieties (*H. jupulus* cv *cordifolius*). An unusual case of sex determination is found in the genus *Fragaria.* This is one of the plant species in which sex chromosomes are heteromorphic and the heterogametic sex is female. *Fragaria* species form a polyploid Series S with 2n = 14,28,42, and 56. All diploid species are hermaphrodites, and wild polyploid species are dioecious. Sex is determined late in floral development, after microspore or megaspore mother cell formation but prior to meiosis.

In summary, sex determination in plants can be controlled genetically by mechanisms also found in the animal kingdom. In some dioecious species, such as *Silene* and *Asparagus*, the sex determining mechanism resembles that of mammals in that the Y chromosome plays an active role in female suppression male activation. In other dioecious species, such as *Rumex* and *Humulus***,** the X-to-autosome ratio determines the sexual fate of floral primordia, similar to the situation found in *Drosophila* and *C. elegans.* It should be noted, however, that even though both *Drosophila* and *C. elegans* share overall genetic similarity in having an X-to-autosome determination of sexuality, the underlying molecular mechanisms that regulate sexual dimorphism are quite different. Therefore, we can assume the mechanistic basis of sex determination in plants will also be species specific. The variations in underlying mechanism are reflected in the physiological control of sex determination in plants.

13.6 Homeotic Mutants in *Arabidopsis*

13.6.1 Systematics of *Arabidopsis*

Arabidopsis thaliana is a member of the mustard family (Brassicaceae) with a broad natural distribution throughout Europe, Asia, and North America. Many different ecotypes have been collected from natural populations and are available for experimental analysis. The Columbia and Landsberg ecotypes are the accepted standards for genetic and molecular studies. The entire life cycle, including seed germination, formation of a rosette plant, bolting of the main stem, flowering, and maturation of the first seeds, is completed in 6 weeks. When it comes to size, almost everything about *Arabidopsis* is small. Flowers

are 2 mm long, self-pollinate as the bud opens, and can be crossed by applying pollen to the stigma surface. Seeds are 0.5 mm in length at maturity and are produced in slender fruits known as siliques. Seedlings develop into rosette plants that range from 2 to 10 cm in diameter, depending on growth conditions. Leaves are covered with small unicellular hairs known as trichomes that are convenient models for studying morphogenesis and cellular differentiation. Plants can be grown in petri plates or maintained in pots located either in a greenhouse or under fluorescent lights in the laboratory. Bolting starts about 3 weeks after planting, and the resulting inflorescence forms a linear progression of flowers and siliques for several weeks before the onset of senescence. Flowers are composed of an outer whorl of four green sepals and inner whorls containing four white petals, six stamens bearing pollen, and a central gynoecium that forms the silique. Mature plants reach 15 to 20 cm in height and often produce several hundred siliques with more than 5000 total seeds. The roots are simple in structure, easy to study in culture, and do not establish symbiotic relationships with nitrogen-fixing bacteria. Natural pathogens include a variety of insects, bacteria, fungi, and viruses.

Recent molecular and morphological studies have outlined systematic relationships across the Brassicaceae, and across the closest relatives of *Arabidopsis*. The genus *Arabidopsis* contains about ten species that are native to Eurasia, North Africa and North America. The closest wild relatives of *A. thaliana* include *A. lyrata* and *A. halleri*, which are self-incompatible diploids with eight chromosome pairs.

Species are interfertile within this group of closely related *N*= 8 taxa. Chromosome number is reduced to *N*= 5 in *A. thaliana*, so diploid mapping crosses with wild relatives are impossible. However, *Arabidopsis suecica* is believed to be an allotetraploid derived from *A. thaliana* and *A. arenosa*.

Fig. 13.5 : *Arbidopis thaliana*

13.6.2 Wild relatives of *Arabidopsis*

A recent study of molecular markers in 142 accessions sampled from the native range of *A. thaliana* found statistically significant isolation

by distance, probably reflecting Pleistocene migrations across Europe and Asia. Although human disturbance clearly influences the biogeography of *A. thaliana*, there is evidence for post-Pleistocene colonization of Europe from glacial refugia near the Mediterranean and in Asia. Only a few ecotypes of *A. thaliana* are available from central Asia, and these show tantalizing patterns of genetic diversity for molecular and phenotypic traits. There is no evidence for different subspecies among the *Arabidopsis* ecotypes that have been examined. Further more, studies of amplified fragment length polymorphisms (AFLPs) and nucleotide variation find evidence for modest, but important historical recombination within and among genes, hence ecotype evolution does not proceed in a bifurcating, tree-like manner. Instead, evidence for historical outcrossing and recombination is apparent in nucleotide polymorphisms segregating among individuals. Species of *Arabidopsis* and *Arabis* have evolved a broad range of life-history, developmental and physiological traits that adapt them to a wide variety of habitats. Combined with molecular tools and biological information from *A. thaliana*, researchers can address many ecological questions in these species. Although *A. thaliana* is common in agricultural fields and disturbed sites in the temperate zones, it is also found in undisturbed rocky sites and forest openings. Significant isolation by distance across its native range suggests that populations of *A. thaliana* in Europe and Asia predate human agricultural disturbance, hence they have long ecological and evolutionary histories in association with insects, pathogens, competitors and abiotic environmental factors.

13.6.3 Plant Transformation Methods in *Arabidopsis*

Genetic transformation of plants occurs naturally. Scientists have been able to carry out controlled plant transformation with specific genes since the mid-1970s. The most common methods for introduction of DNA into plant cells use *Agrobacterium tumefaciens* bacteria or rapidly propelled tungsten microprojectiles that have been coated with DNA. Other methods such as electroporation, microinjection, or delivery by virus have also been exploited. To allow physiological selection of cells that have been successfully transformed, the DNA of interest is typically cloned adjacent to DNA for a selectable marker gene such as *nptII* (encoding kanamycin antibiotic resistance). Genetic transformation can be transient or stable, and transformed cells may or may not give rise to gametes that pass genetic material on to subsequent generations. Transformation of protoplasts, callus culture cells, or other isolated plant cells is usually straightforward and can be used for short-term studies of gene function. Transformation of leaf mesophyll

cells or other cells within intact plants may in some cases broaden the utility of single-cell assays. Exciting new approaches such as virus-induced gene silencing may also be applicable for some studies. In the era of genomics these short term assays will become increasingly important. However, in many cases it is desirable or necessary to produce a uniformly transformed plant that carries the transgene in the nuclear genome as a single Mendelian locus. The generation of genetically homogeneous plants carrying the same transformation event in all cells has typically presented two separate hurdles: transformation of plant cells and regeneration of intact, reproductively competent plants from those transformed cells. Although many successful plant regeneration methods have been developed, these methods often require a great deal of protocol refinement and the focused effort of expert practitioners. It is unfortunate that plant regeneration from single transformed cells often produces mutations ranging from single base changes or small rearrangements to the loss of entire chromosomes. In addition, significant epigenetic changes (for example, in DNA methylation) can also occur. It is often necessary to generate and screen a dozen or more independent plant lines transformed with the same construct to find lines that have suffered minimal genetic damage and that carry a simple insertion event. Transformation is feasible 1 Plant transformation research in the author's laboratory was supported by the North Central Soybean Research Program in many plant species, but has required acceptance of the above limitations.

Mechanism of Transformation Procedure in Arabidopsis

In *Arabidopsis* seed transformation and vacuum infiltration methods, it was shown early on that most primary transformants carry hemizygous T-DNA insertion events. The presence of the T-DNA on only one of two homologous chromosomes implies that productive transformation occurs late in floral development, after the divergence of male and female germ lines (*Arabidopsis* self-pollinates within individual flowers, and if transformation occurred earlier self. It's really that simple. For floral dip transformation of Arabidopsis, plants are grown to a stage when they have just started to flower (A), plants are dipped briefly in a suspension of *Agrobacterium*, Suc, and surfactant (B), plants are maintained for a few more weeks until mature and then progeny seeds are harvested (C), and seeds are germinated on selective medium (e.g. containing kanamycin) to identify successfully transformed progeny (D) fertilization would be expected to give rise to some homozygous transformants due to presence of the same T-DNA insert in pollen and embryo sac cells). The transformation target is further defined in that transformants obtained from a given plant usually carry independent T-DNA insertion events. This suggests that transformation occurs after the divergence of individual pollen or egg cell lineages within a flower. A developmental end point for the typical target of

transformation can also be postulated. Although the result is not as well established, typical primary transformants apparently carry the transgene in all parts of the plant, suggesting that transformation occurred before the cell divisions in a fertilized embryo that establish independent meristems and other distinct adult plant cell lineages. Hence, transformation seems to occur in developing flowers after individual gametophyte cell lineages form, but before extensive development of the embryo. *Arabidopsis* researchers in the mid-1990s focused on empirical transformation protocol improvement. Practical motivation to proceed with the generation of transformants was understandably paramount, and overall satisfaction with the new transformation method delayed efforts to understand how it worked. Nevertheless, protocol modifications, ideas, and anecdotal observations were shared widely through meetings, word-of-mouth, and the *Arabidopsis* electronic newsgroup. Significant findings resulting from this community effort included the discoveries that (a) Plants did not need to be uprooted, treated with *Agrobacterium*, and re-planted. Transformants could be obtained by treating only the protruding inflorescences; (b) inclusion of Silwet L-77, a strong surfactant that shows relatively low toxicity to plants, often enhanced transformation reliability; and (c) many different *Arabidopsis* ecotypes were transformable and many different *Agrobacterium* strains could be used, although notable differences in efficiency were observed. Most important, the popular name "vacuum infiltration" was superseded when a number of groups found that plants could be transformed when dipped in *Agrobacterium* solution with no vacuum infiltration. Some workers subsequently moved to spray application of *Agrobacterium* rather than dipping. A number of other mechanistic clues and procedural tips were shared.

Ovules are the Primary Target for Transformation

Returning to the question of the cellular target of transformation, three research groups worked in parallel to address this issue and have now published their results. Given that transformation can occur by mere dipping of flowers in *Agrobacterium* solution and that anthers and pollen are exposed whereas ovules are not, it seemed likely that the male germ-line would be the target of transformation. However, all three groups found that the female germ-line is the primary target of transformation. In one set of experiments, transformants were produced by outcrossing after Agro-inoculation of only the pollen donor or pollen recipient. No transformants were observed among more than 14,000 seeds produced following inoculation of the pollen donor, but 71 transformants were recovered out of roughly 14,800 seeds produced following inoculation of the pollen recipient. Ye and colleagues observed zero and 15 transformants, respectively, in a similar study. These findings seemingly to rule out transformation of pollen as it develops within anthers, but do not preclude the possibility that pollen is transformed after it germinates on the

stigmatic surface of the pollen recipient. Ovule transformation was convincingly demonstrated when constructs containing a GUS marker gene were used to document sites of delivery of T-DNA. 35S and other standard promoters are poorly expressed in gametophyte tissues, so additional promoters used for GUS fusions were *Arabidopsis* ACT11, an oilseed rape Skp1- like promoter, or a Figwort mosaic virus promoter. Staining was observed in ovules in mature flowers and in younger flowers that had not yet reached pollination.

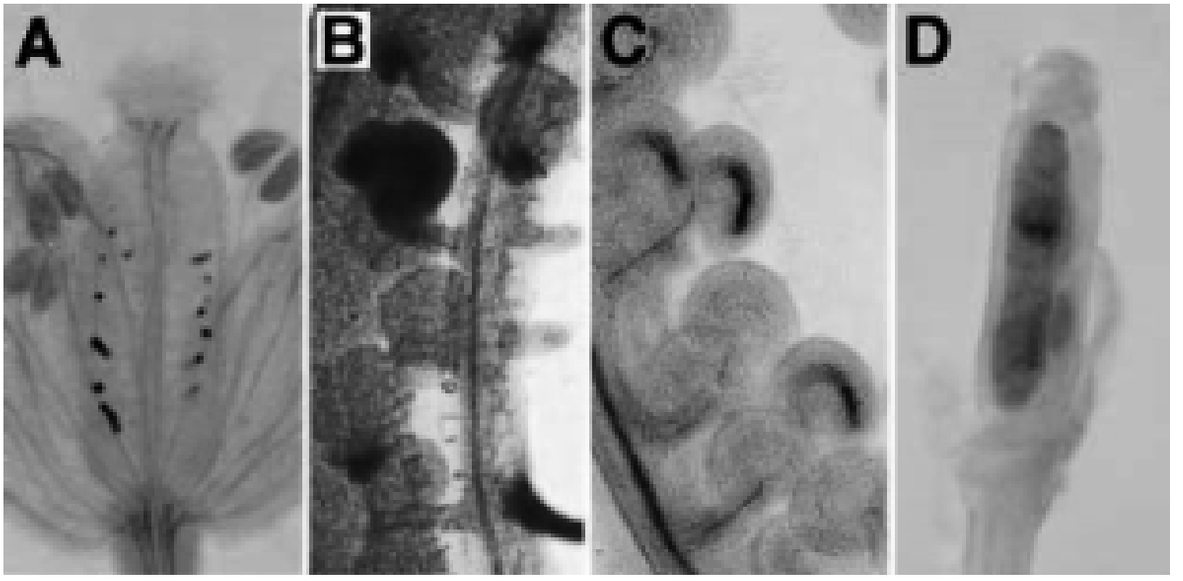


Fig.13.6: *Arbidopsis thaliana*: GUS staining of anthers and ovules

stigmatic surface and in various crevices of the flower (not shown), but also produced examples in which closed locules were filled with blue stain suggesting that locules can harbor substantial colonies of *Agrobacterium*. Transformants have been obtained from siliques located at multiple sites across the inflorescence. However, transformants in one study were not randomly (Poisson) distributed on per silique or per plant basis. In another study roughly one-half of the transformant-bearing siliques contained more than one and up to seven transformants.

13.6.4 Homeotic Mutants

Integuments are converted to carpels in bell mutants. A mature wild-type flower of *Arabidopsis thaliana* contains approximately 50 ovules within the bicarpelloid gynoecium. Ovules are initiated as small finger-like primordia from the internal surface of carpels (the placentas). An inner and an outer integument arise from the surface of each primordium, their site of origin demarcating the boundary between the apical nucellus and

the supporting stalk (funiculus) of the ovule. The integuments grow to cover the nucellus and meet at the tip, leaving a small opening (the micropyle). Within the nucellus a megasporocyte differentiates and undergoes meiosis, and one meiotic product develops into a seven-celled megagametophyte. The nucellus then degenerates, and the innermost cell layer of the integuments differentiate into the endothelium that surrounds the embryo sac. After fertilization the ovule will develop into a seed, with the integuments forming the seed coat.

The correlation between the site on AG expression and the eventual appearance of carpels or carpelloid structures led us to postulate that aberrant AG expression could be responsible for the homeotic transformation of bell mutant ovules. We tested this hypothesis by in situ hybridization of sections of ovules with 35S-labeled anti-AG cDNA. In late stage (after anthesis) wild-type ovules, AG expression is high in the endothelial cells and low in other integument layers, confirming the results of Bowman et al. By contrast, a significant level of AG message is found distributed uniformly within the bell-1 ovules in flowers at this same developmental stage. We quantitated the in situ hybridization signal (the number of autoradiographic grains per unit area) on sections of several wild-type and bell-i ovules.

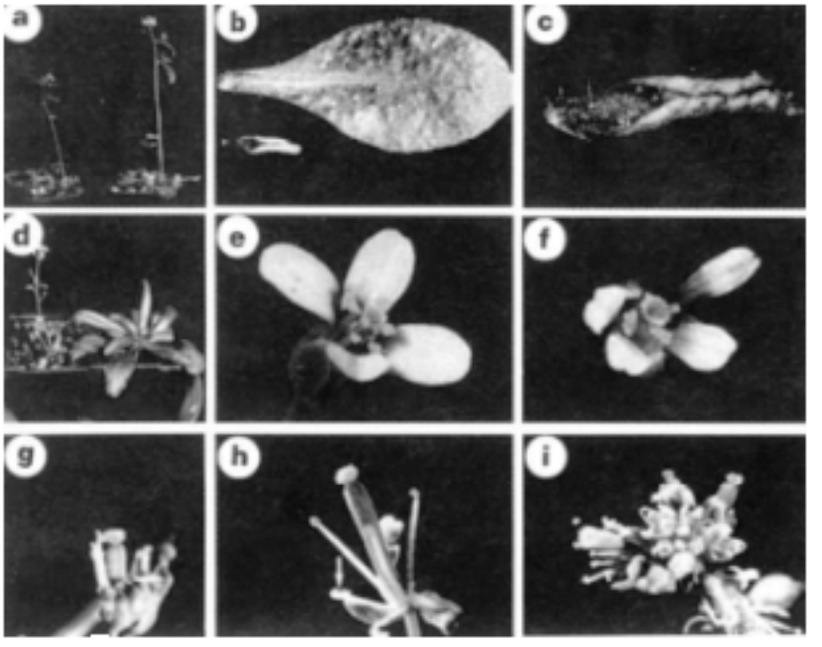


Fig.13.7 : Comparision of mutants (c,f,g,h) and wild type

The level of AG mRNA in the collar of tissue that replaces the integuments in mutant ovules was significantly higher than that observed in the wild-type integuments, and this difference was even more pronounced when corrected for background. We conclude that the cells in the collar of tissue that replaces the integuments in a mature bell ovule actively express AG and propose that one role of Bell is to suppress AG expression directly or indirectly in developing integuments. Ectopic AG expression results in a Phenocopy of the bell Mutant. While the above experiments demonstrate an association between the bell mutation and ectopic expression of AG in the integuments, they do not imply a causal relationship between this expression and the homeotic transformation of the ovules. The role of AG expression in the Bell phenotype can be directly tested through induced overexpression of AG in otherwise wild-type plants. A T-DNA construct in which expression of the *Brassica napus* homolog of AG is driven by the strong "constitutive" 35S promoter of cauliflower mosaic virus was introduced into wild-type *Arabidopsis* plants. While ovules of the primary transgenic lines were wild type, progeny of two independent lines produced only bell-shaped ovules or ovules in which the integuments had undergone a complete conversion to carpels (data not shown).

The transgenic plants were normal in all other respects. Thus, AG overexpression is sufficient to duplicate the effects of loss of BELL gene function, producing an exact phenocopy of the bell mutant.

13.7 Summary

Plants offer unique systems to study sex determination. Because the production of unisexual flowers has evolved independently in many plant species, different and nove1 mechanisms may be operational. Hence, there is probably not one unifying mechanism that explains sex determination in plants. Advances in our understanding of sex determination will come from the analysis of the genetics, molecular biology, and biochemistry of genes controlling sexual determination in plants. Severa1 excellent model systems for bisexual floral development (*Arabidopsis* and *Antirrhinum*), monoecy (maize), and dioecy (*Silene, Asparagus*) are available for such analyses. The important questions that remain concern the mechanism of action of sex determination genes and their interrelationship, if any, with homeotic genes that determine the sexual identity of floral organ primordia. At the physiological level, the connection between hormone signaling and sexuality is not well understood, although significant correlations have been discovered. Finally, once the genes that regulate these processes are identified, cloned, and studied, new strategies for the manipulation of sexuality in plants should be forthcoming.

13.8 Glossary

- **Bud stick:** It is the current season's shoot growth, which contains the buds for budding purposes.
- **Budding:** It consists of inserting a single leaf bud (scion), with or without attached bark and wood piece, into the stock by specific techniques.
- **Callus:** A mass of parenchyma cells, which are able to regenerate tissue. It grows from and around the wounded tissue.
- **Canopy:** The part of the tree composed of leaves and small twigs.
- **Clone:** A specific cultivar propagated asexually (vegetative propagation).
- **Cultivar:** It is the variety, which was originated from a controlled cross under cultivated conditions.
- **Graftage:** Vegetative propagation, which uses budding and grafting techniques.
- **Grafting:** They are various techniques to insert a piece of stem with buds (scion) into the stock.
- **Latent bud:** A dormant bud that is more than 2 years old but has grown enough each year so that its growing point remains at or near the surface of the bark.
- **Leader:** A dominant upright branch. The central leader is the trunk that extends from the root to the top of a tree.
- **Mega:** Suffix that originally denoted large, but has, in a botanical sense, taken on the meaning "female"
- **Megagametophyte:** Female gametophyte: In the seed plants is retained on the sporophyte in the nucellus. In the gymnosperms, the megagametophye is present in the mature ovule (seed) where it functions as the food storage tissue for the embryo. In the angiosperms this generation is destroyed by double fertilization.
- **Megaspore mother cell/Megasporocyte:** Diploid cell destined to undergo meiosis to produce megaspores. In seed plants this cell is buried in the tissue of the megasporangium.
- **Megaspore:** Haploid cells resulting from the meiosis of a megasporocyte. It develops into the megagametophyte. In seed plants the megaspore is not released but is retained in the nucellus.

- **Microgametophyte:** In seed plants this is the pollen grain.
- **Microsporangiate cone/male cone:** terminal clusters of microsporophylls such as the pollen cones of conifers.
- **Microsporangium:** Sporangium that bears microspores. In seed plants synonymous to a pollen sac.
- **Microspore Mother Cell/Microsporocyte:** Diploid cell destined to undergo meiosis to produce microspores in the seed plants.
- **Microspore:** Spore that develops into a microgametophyte.
- **Microsporophyll:** Modified leaf that bears microsporangia. Examples from the seed plants include the stamens of flowers and the subunits of the pollen cones of conifers.
- **Ovule:** The integuments together with the nucellus form the ovule. Later stages include the megagametophyte and embryo.
- **Primary scaffold limb:** One of the major limbs arising from a tree trunk.
- **Rootstock: It** is the part of the grafted or budded tree, which will be theroot system of the plant.
- **Sapling:** Refers to a plant grown from a vegetative part of theoriginal plant asexually.
- **Scaffold:** Main branch that forms the structure of an open center tree.
- **Scion:** A short piece of twig or bud with attached section barkinserted into the stock.
- **Stock:** It is a plant or root system to which a scion is grafted or budded.
- **Sucker:** A shoot grown from the crown or roots of the tree below the graft union or surface.
- **Waterspout:** It is a vigorous, current season shoot, which is growing unbranched from a primary scaffold or smaller branch.

13.9 Self-Learning Excercise

Section – A : (Very Short Answer Type Questions)

1. What is stolon?
2. Name two species which reproduce vegetatively in nature.
3. Give examples of two families with apocarpous ovary.
4. Define cultivar.

5. What are stocks and scion?
6. Define homeotic mutant.

Section – B : (Short Answer type)

1. Write general characters of *Arbidopsis.*
2. Differentiate between ovule of Apocarpy and Syncarpy.
3. Give a note on Sexual reproduction in angiosperms.
4. Briefly describe Grafting.
5. Briefly explain budding in angiosperms.

Section – C : (Long Answer type)

1. Describe various Homeotic mutants in *Arbidopsis*.
2. Give a detailed account on Vegetative propagation in angiosperms.
3. Describe Evolution of Flower in angiosperms.
4. Describe sex determination in angiosperms.

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Unit-14

Structure and Development of Male Gametophyte

Structure of the Unit:

14.0 Objectives

After studing this unit you will able to understand following concepts about reproduction in angiosperms:

- Detailed account on microsporangium and microsporogenesis.
- Various stages of microgametogenesis.
- Ultrastructure of pollen grain / microspore.
- Various causes of pollen allergy.
- Development of embryo sac like pollens

14.1 Introduction

Plant sexual life cycles are characterized by an alternation of generations, in which a diploid sporophyte generation gives rise to a haploid gametophyte generation. In angiosperms, the gametophyte generation is very small and is completely enclosed within the tissues of the parent sporophyte. The male gametophytes, or microgametophytes, are pollen grains. The female gametophyte or megagametophyte is the embryo sac. Pollen grains and the embryo sac both are produced in separate, specialized structures of the angiosperm flower. Both male and female structures usually occur together in the same individual flower. Flowers and reproductive organs develop seasonally at times of the year most favourable for pollination. Pollen grains form in the two pollen sacs located in the anther. Each pollen sac contains specialized chambers in which the microspore mother cells are enclosed and protected. The microspore mother cells undergo meiosis to form four haploid microspores. Subsequently, mitotic divisions form four pollen grains. Each pollen grain contains a vegetative nucleus and a generative nucleus. Later generative nucleus divides mitotically and forms two male gametes.

During flowering season pollen grains are formed in large amount and these flew into air and become a cause of pollen allergy in human beings and animals that is drawback of pollen development. On the other hand pollen grains are haploid and are used to develop

haploid plants using tissue culture technique; Haploid plants are one of the most useful equipment for the development of improved as well as new varieties for human welfare.

14.2 Structure of Anther (Microsporangium)

In typical angiospermic flower androecium is male essential whorl. Each unit of this whorl is stamen that shows anther at top terminus and anthers bear anther lobes (microporangia). Microsporangium produces the microspores and eventually the male gametophyte. Similarly the megasporangium or ovule is the place of formation of the megaspores and the female gametophyte. A typical anther comprises four elongated microsporangia but at maturity the two sporangia of each side become confluent owing to the breaking down of the partition between them. Very young anther shows a mass of homogeneous meristematic cells surrounded by the epidermis It soon becomes slightly four-lobed and rows of hypodermal cells become differentiated in each lobe by their larger size, radial elongation and more conspicuous nuclei these cells are archesporium. The extent of the archesporial tissue varies considerably both lengthwise and breadthwise. Either a single archesporial cell may be seen in each lobe in a cross section of the anther and or a plate of such cells. The archesporial cells divide to form a primary parietal layer toward the outside and a primary sporogenous layer toward the inside. The cells of the former divide by periclinal and anticlinal walls to give rise to a series of concentric layers, usually three to five composing the wall of the anther. The primary sporogenous cells either function directly as the spore mother cells or undergo further divisions to form a larger number of cells.

14.2.1 Monothecous and Dithecous anthers

Stamen is the male reproductive organ or microsporophyll of a flower. It consists of two parts, filament and anther. Filament is long and slender stalk. It is attached proximally to thalamus, petal or tepal and distally it bears an anther. Anther is broader knob- like fertile part of the stamen. It consists of one or two anther lobes and accordingly, anthers are called **monothecous** or **dithecous**. The two anther lobes are separated in the anterior region by a deep groove but are attached to each other on the back side by a sterile parenchymatous tissue called connective. Connective possesses a vascular strand. Connective is absent in monothecous stamen. A dithecous anther is tetrasporangiate while monothecous stamen is bisporangiate.

14.2.2 Epidermis

All the microsporangia are covered on the outside by a well-defined common epidermis of the anther. Epidermal cells often become stretched and shrivel off at maturity. The epidermis is the outermost layer of the anther, undergoes only anticlinal divisions. Its cells

become greatly stretched and flattened in order to keep pace with the enlargement of the anther, and in many plants, especially those of dry habitats, they eventually lose contact with each other so that only their withering remains can be seen at maturity.

14.2.3 Endothecium

The layer of cells lying immediately beneath the epidermis is the endothecium. Its maximum development is attained at the time when the pollen grains are about to be shed. The cells become radially elongated, and from their inner tangential walls fibrous bands run upward, ending near the outer wall of each cell.

In aquatics with aerial flowers like *Utricularia* and *Wolffia* the fibrous thickenings occur as usual, but in several members of the Hydrocharitaceae and in some cleistogamous forms whose flowers never open, they fail to develop and there is no special mode of dehiscence. In those plants, also, whose anthers open by apical pores, the endothecium may not develop any fibrous thickenings and dehiscence takes place here by the dissolution of certain cells at the apex of the anther.

In *Erica*, (Fig:) which is an example of this kind; there is a further peculiarity in that the "apical" pores are in fact basal. Among other exceptions may be cited *Musa sesamum, Anona* and *Ipomoea* in which the fibrous thickening are absent but the walls of the epidermal cells undergo a general cutinization and lignification over the entire surface.

In *Oryza, Ditepalanthus* and *Balanophora* are peculiar in that the parietal layers, one or two in number, become crushed and disorganized during the development of the anther so that a fibrous layer is absent and the epidermis abuts directly on the tapetum.

14.2.4 Middle layers

Just below to endothecium, generally one to three layers of meristematic tissue present. "middle" layers. These are flattened and crushed at the time of the meiotic divisions in the microspore mother cells. In *Holoptelea* there are three to four middle layers, of which the outermost persists for a long time. In *Ranunculus* there are two middle layers, of which the inner soon disappears but the outer persists; occasionally its cells become densely protoplasmic and simulate those of the tapetum. In *Lilium* there are several middle layers, of which those lying adjacent to the endothecium persist for a long time, and in *Gloriosa* the outermost middle layer develops fibrous thickenings similar to these of the endothecium. In *Wolffia* middle layer in anther are absent

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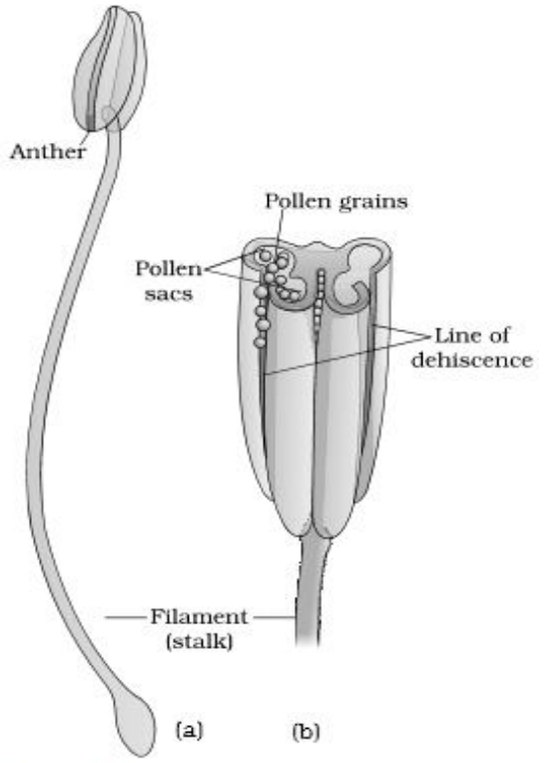

Fig.: 14.1 a. A typical stemen
b. Cut section of an anther

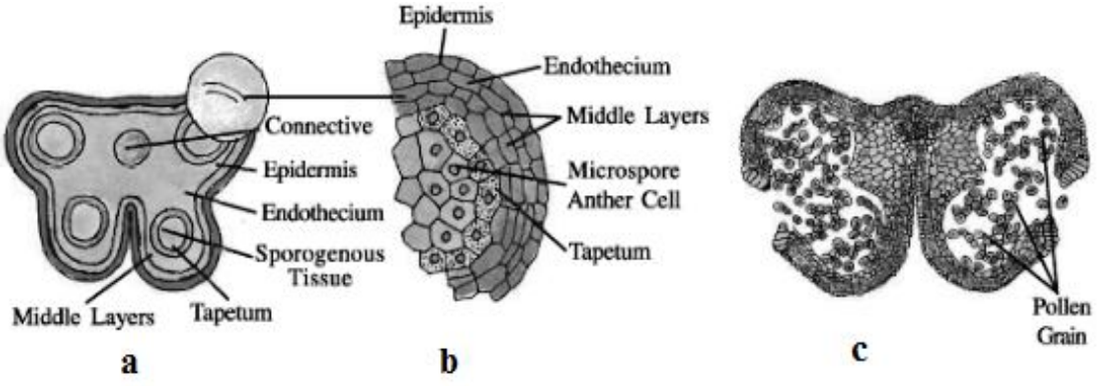

Fig. 14.2 Struture of Anther (a) Anther T.S. (b)Enlaged view of microsprangium
(c) Mature dehisced anther

14.2.5 Pollen chambers

A typical angiosperm anther is bilobed with each lobe having two theca, i.e., they are dithecous. A longitudinal groove runs lengthwise separating the theca. The bilobed nature of an anther is very distinct in the transverse section of the anther. The anther is a four-sided (tetragonal) structure consisting of four pollen chamber or microsporangia covered by a nursing layer of cells i.e. **Tapetum**, located at the corners, two in each lobe. These extend longitudinally all through the length of an anther and are packed with pollen grains. (Fig.14.1 and Fig. 14.2)

14.2.6 Tapetum: Structure and Function

The innermost wall layer or tapetum is of considerable physiological significance, for all the food materials entering into the sporogenous cells must pass through it. Tapetal cells are full of dense cytoplasm, and at the beginning of meiosis the tapetal nuclei may also undergo some divisions. Because of these similarities of appearance and behavior between the cells of the tapetum and the micrcsporogenous tissue, earlier botanists supposed that the former is derived by a sterilization of the outer sporogenous cells. Developmental studies of a precise nature have, however, nearly always confirmed its parietal origin. The nuclear divisions in the tapetum were formerly believed to be amitotic, but further studies have shown that this is incorrect and that appearances suggesting amitosis are really caused by mitotic irregularities andnuclear fusions.

According to present conceptions, the nucleus of a tapetal cell may divide in any of the following ways:

1. **By free nuclear division**: The Mitotic division takes place in the ordinary way, but no cell plate is laid down. The two daughter nuclei, which are diploid, remain inside the cell.
2. **By forming restitution nucleus**: Here the chromosomes behave normally up to the early anaphase stage. After this, one or more of them fail to separate, forming chromosome bridges which persist during the telophase as well as the resting stage. As a result a single dumbbell-shaped tetraploid nucleus is formed whose middle portion may be broad or narrow depending on the number of chromosome bridges present.
3. **By endomitosis:** Here the nucleolus and the nuclear membrane remain intact and there is no spindle formation. The chromosomes contract and split longitudinally, but all of them remain within the same nucleus, which becomes tetraploid. The first nuclear division in a tapetal cell is often followed by further divisions. Some of the divisions may be accompanied by nuclear fusions, resulting in one or more

large polyploid nuclei. The latter may, however, divide again and give rise to smaller nuclei. Since this type of behavior is very frequent in tapetal cells, it is unnecessary to give specific instances.

An interesting condition has been reported in certain haploid and diploid plants of *Oenothera rubricalyx*. In the former the tapetal cells are uninucleate and in the later they are binucleate a fact which is no doubt related to the general reduction of tissues in haploid individuals. More difficult to explain is the marked difference in shape and structure of the tapetal cells belonging to the same anther. In *Lathraea*, *Salvia, Moringa* the tapetal cells on the inner side of the loculus show a marked radial elongation and are much larger than those on the outer side. Further, in *Lathraea* the cells on the outer side are uninucleate while those adjacent to the connective are binucleate. In *Lactuca sativa* the tapetal cells lying on one side of the loculus may be quadrinucleate while those on the other are binucleate. The binucleate cells are nearly always shorter and broader than the quadrinucleate. Possibly these differences are related to the varying amounts of nutritive materials passing into the cells. (Fig. 14.3 and Fig. 14.4)

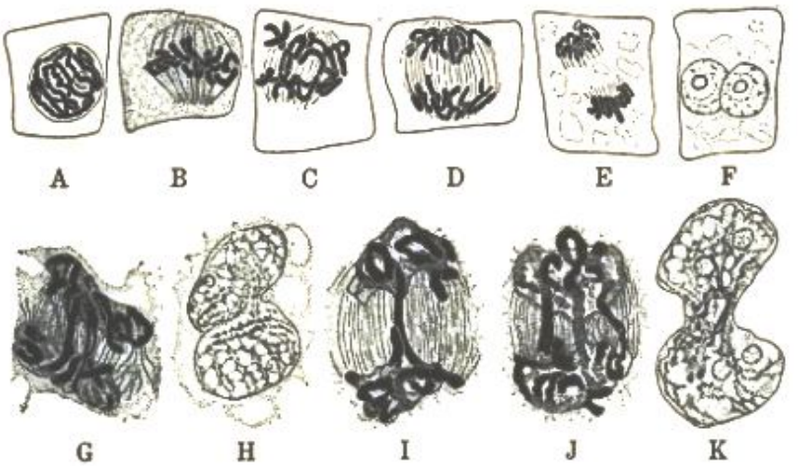

Fig. 14.3 : Nuclear division in tapetal cells of *Zea mays* (A - F)
Nuclear division in tapetal cells of *Lilium canadense* (G - H)
Nuclear division in tapetal cells of *Podophyllum sps.*(I - K)

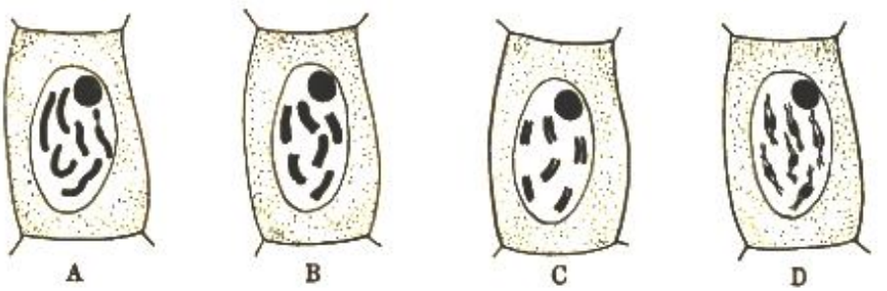

Fig. 14.4 Endomitosis in *Spinacia oleracea*

A. Endoprophase B. Endometaphase C. Endoanaphase D. Endotelophase

14.2.7 Sporogenous tissue

The primary sporogenous cells give rise to the microspore mother cells. In some plants the sporogenous cells undergo several divisions, in others only a few divisions, and rarely there are no divisions at all, so that the primary sporogenous cells function directly as the microspore mother cells. In some members of the Loranthaceae, *Dendrophthoe, Elytranthe,* and *Amyema*, the microsporangia become vertically partitioned by the formation of sterile septa, and in *Viscum* such partitions arise not only in the vertical plane but also in the horizontal one. Formation of sterile septa is also known in a few other plants. In *Quamoclit* there is a single row of sporogenous cells in each lobe of the anther but one or two of these fail to keep pace with the others and become nonfunctional. These give rise to sterile partitions separating the loculus into two or three parts. In some plants there are fewer than four groups of sporogenous cells. In the family Malvaceae the anthers are uniformly bisporangiate and the two loculi eventually fuse to form a single loculus. In *Elodea, Styphelia, Wolffia,* and *Moringa* also, there are two microsporangia which may later become confluent by the breaking down of the intervening cell layers. The anthers of *Najas* are said to be unilocular, but the developmental stages have not been traced satisfactorily. In *Vallisneria* there are all gradations from a unilocular to a tetralocular condition. Typically two loculi are formed, owing to the appearance of a sterile septum in the sporogenous tissue, but sometimes the septum is incomplete, resulting in a unilocular condition, and frequently each of the two loculi becomes bisected so as to form four loculi. The stamens of *Piper betle* are peculiar in that the number of microsporangia in an anther may be four, three, two, or one, and it remains constant from the time of initiation of the sporangia to the maturation of the anther. There is no secondary fusion of the sporogenous tissue. In *Korthalsella* there are three stamens, each of which consists of two microsporangia, but since all the anthers fuse to form a synandrium, a cross section of the flower shows six microsporangia arranged in a ring.

14.3 Microsporogenesis (Pollen Development)

14.3.1 Microsporogenesis

Pollen grains/ Microspores form in the two pollen sacs located in the anther. Each pollen sac contains specialized chambers in which the microspore mother cells are enclosed and protected. The microspore mother cells undergo meiosis to form four haploid microspores. Subsequently, mitotic divisions form four pollen grains. Inside each pollen grain there are two nuclei namely vegetative nucleus and generative nucleus. Generative nucleus divides later mitotically to produce two sperms/ male gametes. Pollen grain shapes are specialized for specific flower species.

During meiosis in the microspore mother cells cytokinesis may be of the successive or the simultaneous type. In the former a cell plate is laid down immediately after the first meiotic division and another in each of the two daughter cells after the second meiotic division. In the simultaneous type, on the other hand, no wall is laid down after the first division and the mother cell becomes separated all at once into four parts after both the meiotic divisions are over.

The investigations of C. H. Farr (1916) and others have shown that there is also another difference in the mechanism of cytokinesis. In the successive type the cell plate is laid down in the center and then extends centrifugally on both sides, dividing the cell into two equal halves. In the simultaneous type, on the other hand, the division usually occurs by centripetally advancing constriction furrows, which meet in the center and divide the mother cell into four parts.

Farr (1916) studied *Nicotiana tabacum* in special detail. At first there is an enlargement of the nucleus of the microspore mother cell, accompanied by a thickening of the mother cell wall. No cell plate is laid down after Meiosis I, and the spindle fibers of this division disappear during the metaphases of Meiosis II. After the four daughter nuclei have become organized, they assume a tetrahedral arrangement and a spindle is re-formed between every two nuclei, for an account of the nuclear changes in meiosis. However, spindles have nothing to do with the quadripartition of the mother cell, and there is no laying down of centrifugally growing cell plates such as are characteristic of other dividing cells. Instead, constriction furrows now start at the periphery and proceed inward until they meet at the center, so that there is a simultaneous division of the protoplast into four cells, i.e., the microspores.

In *Melilotus alba* (Castetter, 1925) vacuoles seem to play a conspicuous part in cytokinesis. After Meiosis II, hyaline areas develop between the four nuclei, apparently as the result of a migration of the denser cytoplasm toward the nuclei and an extrusion of sap into the regions between them. The small vacuoles arising in this manner soon fuse to form larger ones which virtually split the cytoplasm into four masses. Furrows originating at the surface now grow inward and soon meet the vacuoles. Meanwhile, the mother cell round up and secretes a thick layer of callose or some other gelatinous material, which extends inward with the cleavage furrows and eventually completes the division of the cell into the four microspores.

Reeves (1928) studied *Zea mays* as an example of the successive type of microspore formation. Here, at the end of Meiosis I, thickenings are formed on the spindle fibers at the equatorial region of the cell. They gradually increase in size, coming in contact with each other and fusing to form the cell plate. Additional spindle fibers continue to appear

just beyond the periphery of the plate so as to increase the diameter of the spindle. At the same time the cell plate extends centrifugally and joins the wall of the mother cell, so as to complete the division of the protoplast into two halves. Now the second meiotic division follows, and a new partition wall develops in each cell in the same way as after Meiosis I, resulting in a tetrad showing the bilateral arrangement of microspores.

In general, simultaneous type is prevalent in the majority of dicotyledons and the successive type in the majority of monocotyledons. There is no hard and fast rule, however, and exceptions are frequent. Thus the successive type is found in a few dicotyledonous families like the Asclepiadaceae, Podostemonaceae, and Apocynaceae, and the simultaneous type in a few monocotyledonous families, viz., the Iridaceae, Taccaceae, Juncaceae, and Dioscoreaceae, and in several genera of the Liliaceae, Palmaceae, and Orchidaceae.

In *Magnolia* (Farr, 1918) there are isobilateral tetrads formed by furrowing instead of by cell plates. A cleavage furrow starts after Meiosis I, but its development is arrested during the second meiotic division. It resumes growth at the end of Meiosis II and forms a partition through the equatorial region of the mother cell. At the same time additional furrows originate at the periphery, and the two dyad cells now become subdivided to give rise to the four microspores.

14.3.2 Microspore (Pollen) tetrads

Usually, Microspores are arranged in a tetrahedral or isobilateral fashion, but there are exceptions also. (Fig 14.5) A decussate arrangement of the cells has been recorded in *Magnoli*a, *Atriplex*, *Comas* and many other plants. In some genera of the Asclepiadaceae and Hydrocharitaceae the mother cells divide transversely so as to give rise tolinear tetrads. T-shaped tetrads also occur sometimes as in *Aristolochia* and *Butomopsis*. In *Zostera*) the elongated microspore mother cells, measuring 5 by 60 microns at the time of meiosis, divide in a plane parallel to the longitudinal axis of the cell, resulting in a group of four filiform cells which undergo further elongation and become approximately 2000 microns long when mature. Considerable interest are *Musa, Neottia , Agave, Habenaria* and *Ottelia* in which two or three types of dispositions may be found in one and the same species.

Occasionally there are either fewer than four spores resulting from the divisions of the microspore mother cell, or more than four. The former condition originates as the result of a failure of one division, or the formation of a "restitution nucleus" after the first division, or an irregular wall formation giving rise to one binucleate and two uninucleate spores. The later condition, i.e., the formation of more than four spores (polyspory), usually results from the occurrence of lagging chromosomes which organize into micronuclei.

In general, however, such abnormalities in the number of microspores are found only in hybrids characterized by a high degree of sterility and the pollen grains arising in this way are nonfunctional. Usually the microspores soon separate from one another but in some plants they adhere in tetrads to form the so-called "compound" pollen grains.As examples may be cited *Drimys, Anona, Drosera, Elodea, Typha, Furcraea*, and several members of the Ericaceae, Apocynaceae, Asclepiadaceae, Juncaceae, and Orchidaceae.

In the Mimosaceae there are larger units composed of 8 to 64 cells, and in a number of genera belonging to the Asclepiadaceae all the microspores in a sporangium remain together to form a single mass called the pollinium. The family Orchidaceae is especially interesting in this connection.

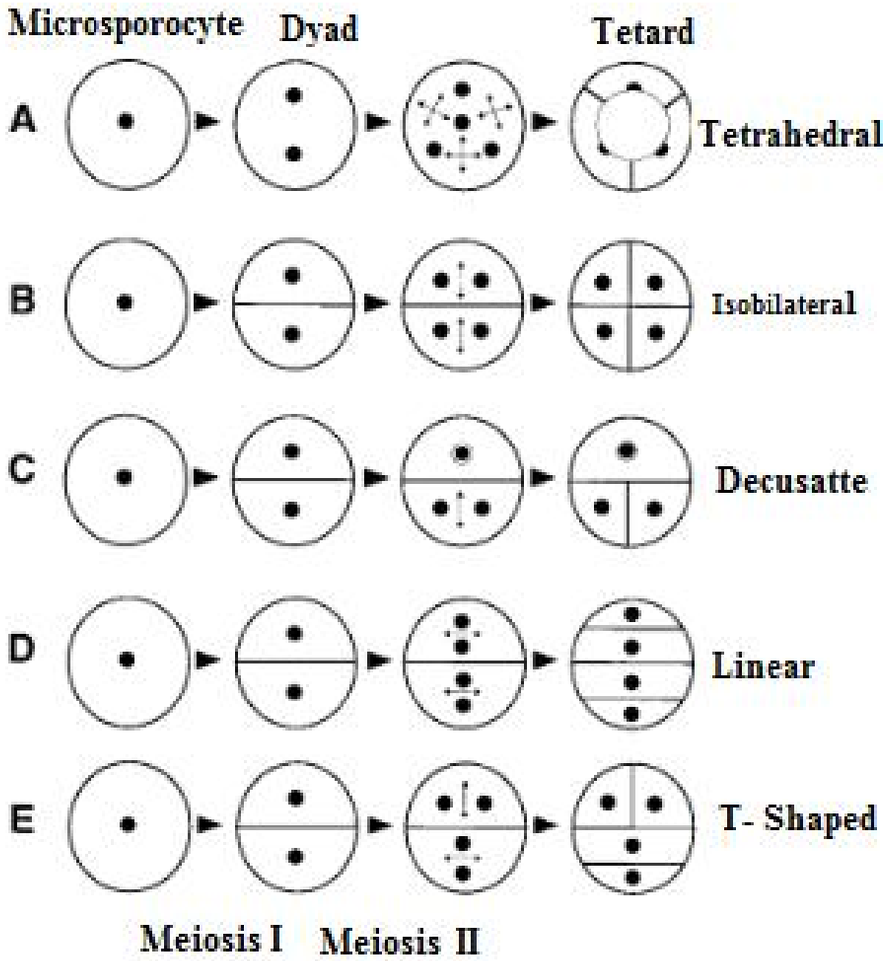

Fig.14.5 Diagram of the development of common tetrad types. *A*, Simultaneous division. *B–E*, Successive division. Arrowheads indicate equatorial plane of spindle; solid circle indicates nucleus; solid circle inside a small circle indicates nucleus directly below the one above.

In some genera, such as *Cypripedium* and *Vanilla*, the microspores separate from one another and become free. In *Pogonia* the four cells of a tetrad adhere and form a compound pollen grain. In the tribes Ophrydeae and Neottieae this tendency is carried further and the compound grains are themselves held together in small units known as massulae. Finally, in *Coelogyne* and *Pholidota* all the microspore mother cells and their derivatives remain together and continue their development as a single unit.

14.4 Development of Male Gametophyte (Microgametogenesis)

The microspore is the first cell of the gametophytic generation. During gametogenesis, the nucleus of microspore divides mitotically to produce a bigger vegetative cell and a smaller generative cell. The generative cell is initially attached to the wall of the pollen grain but later comes to lie freely in the cytoplasm of the vegetative cell. Before the start of pollen mitosis, the nucleus of microspore is displaced from the centre toward one side of the cell. At this stage, the cytoplasm between the nucleus and the wall, on the side where vegetative cell is to be cut becomes highly vacuolated. Initially, the cytoplasm of the vegetative cell and that of the generative cell are separated by two plasma-membranes. The wall of the generative cell is soon formed in between the two cell membranes and adjoins the intine (innermost layer of pollen wall) on either side of the generative cell. The wall of the generative cell grows inwards between the plasmalemma of the generative cell and the intine until the two ends of the wall meet and fuse and the cell is finally pinched of. Soon the wall of the generative cell disappears and the cytoplasm of the generative cell remains enclosed in two plasm membranes, its own and the detached invagination of the plasmalemma of the vegetative cell. (Fig.:14.6)

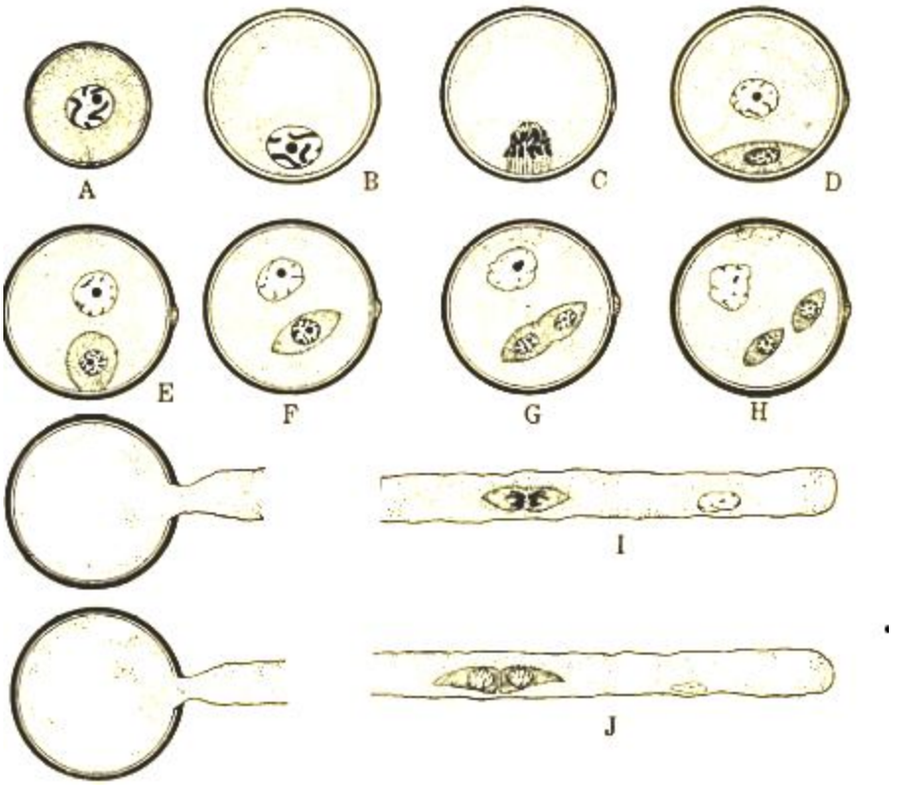

Fig. 14.6 : Development of Male Gametophyte

After its formation, the generative cell separates from the intine and moves to a position where it is completely enclosed by the vegetative wall. The sperm cells are formed by mitotic division of the generative cell. In a mature anther, the tapetum degenerates while the outer endothecial cells become fibrous. At this stage, the dehiscence of anther takes place and the pollen grains are released. The pollen grains are shed either at bicelled or tri-celled stage.

14.4.1 Formation of Vegetative and Generative Cells

The first division of the microspore gives rise to the vegetative and generative cells. Geitler (1935) noted that the metaphase spindle usually shows a pronounced asymmetry, the wallward pole being blunt and the free pole acute. Studies seem to indicate that this asymmetry is associated with the form of the prophase nucleus. In *Allium*, where the nucleus is strongly flattened on the wallward side, the asymmetry is extreme; in *Pancratium*, where it is only slightly flattened, the asymmetry is much less pronounced; and *Tradescantia* shows an intermediate condition. The direct cause of the asymmetry has

been attributed to a difference in the time of development of the two spindle poles, the wallward or generative pole developing more slowly than the vegetative, presumably because of the smaller amount of cytoplasm associated with the former. With the onset of the anaphase, the asymmetry becomes less pronounced. In the telophase the generative chromosomes are arranged in a plane surface parallel to the inner wall of the microspore, while the vegetative ones form a somewhat hemispherical pattern. Symmetrical spindles have been observed only occasionally. To mention a few examples, in *Asclepias* and *Anthericum* both the poles of the spindle are blunt; in *Adoxa* both are more or less pointed; and in *Podophyllum* both symmetrical and asymmetrical spindles are said to occur in the same loculus. Further, in *Adoxa*, *Myricaria*, *Sambucus*, *Colylanthera*, and *Uvularia* the spindle is not situated near the wall of the pollen grain but occupies almost the entire width of the latter. In any case the cells formed by the division are always unequal, although the conditions which bring about this result are not clearly understood. In *Cuscuta* and *Slrychnos* where the daughter cells are sometimes of the same size, this is clearly an abnormality leading to the formation of double microspores, each of which is destined to divide again to give rise to the vegetative and generative cells. Double pollen grains comprising two units, each with its own generative and vegetative cells, have also been studied in *Podostemon subulalus.*

The separating wall between the two pollen grains is pitted and only one of them produces a pollen tube, the other presumably serving as a source of food material. It may be noted that unlike the reduction divisions, which occur more or less simultaneously in all the microspore mother cells of an anther, the microspres usually divide without any such synchronization, and the same loculus may show different although not widely separated stages of division and development. In those plants in which the microspores remain together in a tetrad, all four cells in a tetrad are usually in the same stage of division, but not all the tetrads of an anther.

A complete synchronization may perhaps be expected only where the microspores are united into pollinia (Mimosaceae, Asclepiadaceae, and Orchidaceae), for here the cells probably exercise some influence over one another through the uncuticularized walls which lie between them. Exceptions do occur, however, even in such cases. The pollinium of *Acacia baileyana* shows one of the microspores in prophase, another with the tube and generative cells already formed, and the rest in various intermediate stages. Goebel (1933) thought that in the angiosperms the generative cell is always cut off on the ventral side of the microspore.

There is considerable variation in the form of the generative cell. Usually it is elliptical, lenticular, or spindle-shaped, but in *Cuscuta* and *Ottelia* it becomes long enough to

occupy the entire width of the pollen grain, coming quite close to the inner wall of the latter on either side. In *Monochoria* it is one and a half times as long as the diameter of the pollen grain and is accommodated in the latter only by the incurving of its whip-like ends. In *Campanula ranunculoides* the two ends are dissimilar, one being pointed and the other more or less blunt and swollen so as to look like a "head." There are also occasional reports of changes in the form of the generative cell. More frequently, however, such appearances are merely due to the plane of sectioning. A spindle-shaped cell appears round when cut across and oval when cut obliquely. In fixed material the cytoplasm of the generative cell is usually distinguishable from that of the vegetative by its hyaline appearance and general lack of food materials. Plastids and chondriosomes have, however, been demonstrated in a few cases, and some studies on living pollen grains and pollen tubes have confirmed the presence of a vacuome and mitochondria in the generative as well as the sperm cells of *Narcissus, Asclepias, Vinca, Crinum,* and *Lilium*. Mention may also be made of the "colored bodies" described by Kostriukova (19396) in living pollen tubes of *Lilium martagon*. He saw two structures of a pale greenish color, one at each end of the generative nucleus. In older stages these bodies were found to divide and occupy similar positions in the sperm cells. They were not recognizable in fixed material, but in their places small areolae were seen which stained black with osmic acid. It is also concludes that they probably correspond with the structures described as Golgi bodies, but a further study is of course necessary to confirm this.

Regarding the contents of the vegetative cell, starch and fat are the most conspicuous substances. The distinction between starchy and fatty pollen has been recognized for a long time and their possible ecological significance has been a subject of much interest. Luxemburg (1927) traced the origin of the starch grains and fat bodies from plastids in the pollen grains of several members of the Malvaceae, and believes that theplastids in turn arise either from preexisting plastids or from chondriosomes.

In very young pollen grains the reserve food consists almost entirely of droplets of fat, and starch formation begins only after the pollen grains have increased in size. Certain proteinaceous bodies have also been reported in pollen grains and pollen tubes but their exact origin remains unknown. They probably arise in plastids but soon become liberated in the general cytoplasm of the pollen grain and pollen tube.

14.4.2 Division of Generative Cell (Male "Cells" or "Nuclei"Formation)

The generative cell may divide either in the pollen grain or in the pollen tube. Formerly the second condition was believed to be the more frequent, but during recent years three-celled pollen grains have been reported in several genera and it seems certain that many of the older records were based on a study of immature pollen. There is also considerable

evidence to indicate that even in those plants in which the pollen grains are shed in the two-celled condition, the generative nucleus is already in the prophase stage and the process of division is merely continued in the pollen tube. Sometimes the nucleus may even show a pro-metaphase stage which is distinguishable from a typical metaphase only by the delay in the dissolution of the nuclear membrane and the organization of the spindle. Occasionally both two- and three-celled pollen grains have been reported in the same plant, as in the cleistogamous flowers of *Viola*, in *Dionaea, Circaeaster, Nicotiana*, and *Iris*, but this is probably due to environmental influences. In *Holoptelea integrifolia* the pollen grains are shed at the two-celled stage, but the generative cell divides on the surface of the stigma before the pollen tube has started to grow. Details of the division of the generative cell vary depending on whether it takes place in the pollen grain or in the pollen tube. In the former case, spindle fibers and a normal metaphase plate have been regularly observed, and the process does not seem to differ essential way from a normal mitosis. Cytokinesis, resulting in a bipartitioning of the cell, may take place either by a process of furrowing as in *Juncus* or by the laying down of a cell plate as in *Asclepias* and *Portulaca*. In some pollen grains a definite cell plate was laid down in the beginning, but it soon faded away, leaving the final separation of the sperms to a constriction furrow which arose soon afterwards. In others the cell plate persisted, and the progress of the constriction furrow was arrested in this region although evident on either side of it; here the splitting of the cell plate divided the generative cell before the constriction could make much progress. It has proved more difficult to understand the mechanism of the division when it occurs in the pollen tube.

14.4.3 Vegetative Nucleus

The vegetative nucleus or Tube nucleus had an important role in directing the growth of the pollen tube. Present evidence seems to indicate, however, that its functional importance had been greatly exaggerated. The vegetative nucleus is not always in the distal end of the pollen tube (where it would be most expected if it had any important function in directing the growth of the tube) but frequently lies considerably behind the male gametes. When the tube becomes branched as in *Aconitum, Cucurbita,* and *Papaver* the individual branches continue their growth for an appreciable period, although only one of them contains the vegetative nucleus. In *Ulmus*, *Senecio, Crepis*, and *Secale* it degenerates even before the pollen grains begin to germinate and does not enter the tube at all; nevertheless the tube continues to function normally. In *Chenopodium, Atriplex,* and *Salsola* it seems to break up and diffuse into the surrounding cytoplasm and in *Musa* and Senecio it fragments into small bits which seem to be quite functionless. In some other plants also the vegetative nucleus assumes a very abnormal appearance. For instance, in

the pollen tubes of *Viola odorata* it becomes 4 times, in *Cymbidium bicolor* 18 times, and in *Vallisneria Americana* 27 times longer than broad. In a few members of the Labiatae and in *Nicotiana* the elongation is sufficiently pronounced to give it a filamentous outline. On the basis of these and other data regards the vegetative nucleus as a vestigial structure without any important function in the growth of the pollen tube. This view was studied in pollen grains of *Crinum*. It is stated that soon after its formation the vegetative nucleus increases in size and becomes amoeboid. Later it indicates a decomposition of the chromatin. Therefore, that it is a degenerating structure without any important function in the life of the pollen tube. While further evidence would be welcome, it seems safe to conclude that the old view attributing a leading role to the vegetative nucleus in the growth and direction of the pollen tube now needs modification. It is likely that these functions are really discharged by the nucleus of the generative cell itself and later by the nuclei of the two male cells formed by its division.

14.4.4 Development of Pollen in Cyperaceae

The course of development described above is generally characteristic of all angiosperms, dicotyledons as well as monocotyledons but the family Cyperaceae being the only notable exception. Juel (1900), Stout (1912), Piech (1928) and others have shown that, of the four microspore nuclei produced after meiosis, only one develops further, while the other three become pushed toward one end of the mother cell. The functional nucleus, which lies in the center, divides with its spindle oriented in the direction of the long axis of the cell. The cell plate, which is laid down between the vegetative nucleus and generative nucleus, extends around the latter so as to give rise to a continuous plasma membrane. The generative cell soon becomes spindle-shaped and divides to form the two sperm cells. A few doubtful points, which need further clarification, are the following:

(1) Whether the functioning microspore nucleus is separated from the three nonfunctioning nuclei by a wall.

(2) Whether the nonfunctioning nuclei are separated from one another by walls.

(3) What the fate of the nonfunctioning nuclei may be.

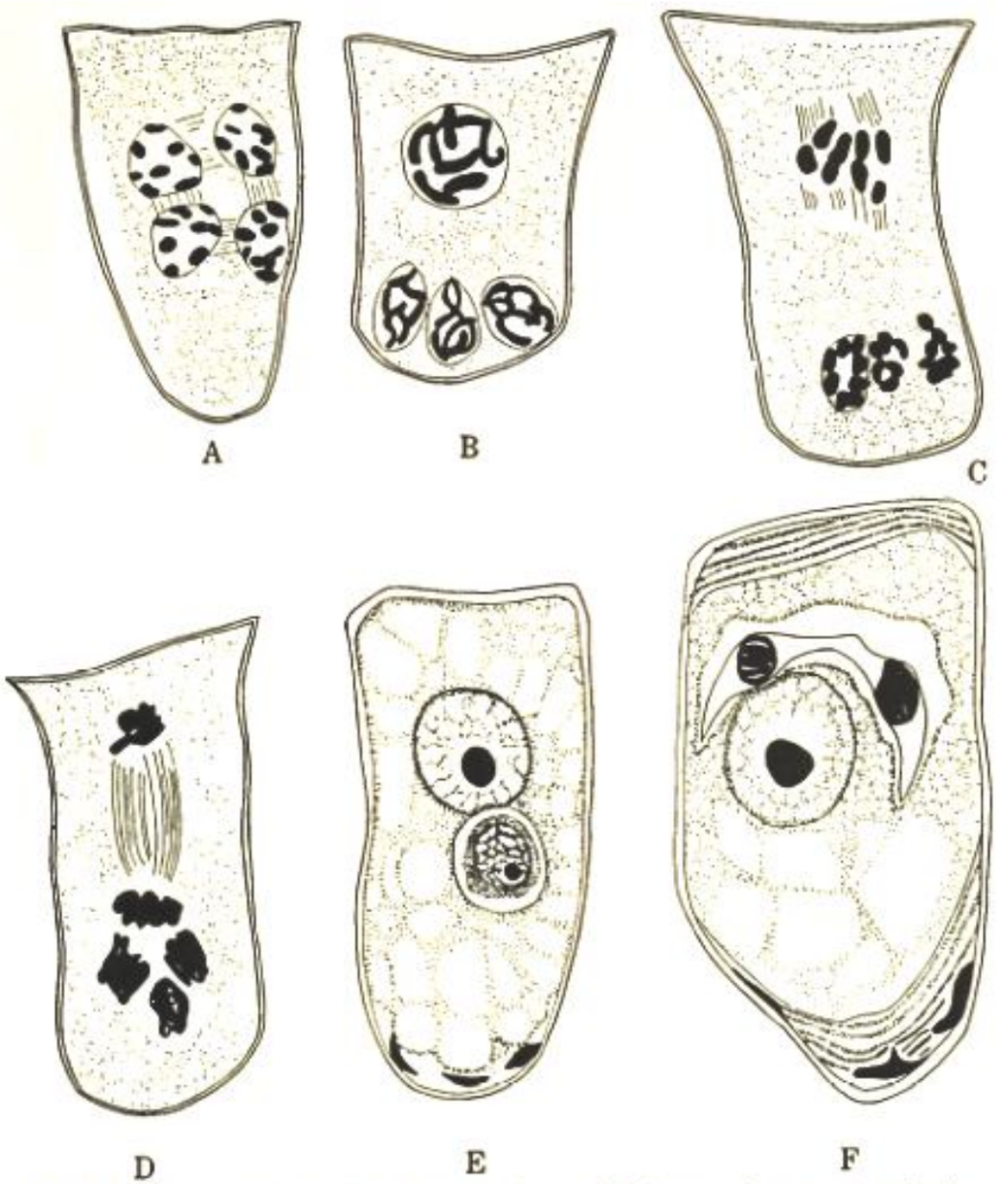

Fig.:14.7 Development of male gametophyte of *Scirpus paluster*. *A*, telophase of Meiosis II resulting in formation of four microspore nuclei. *B*, three of microspore nuclei pushed to one end of pollen grain; functioning nucleus in center. *C*, *D*, functioning nucleus dividing; remaining three nuclei in process of degeneration. *E*, pollen grain, showing vegetative and generative cells. *F*, generative cell dividing to form two male cells.

Tanaka (1940, 1941), who has discussed these questions, believes that normally a plasma membrane separates the functioning microspore nucleus from the three nonfunctioning nuclei and that subsequently similar membranes arise between the later. The nonfunctioning nuclei sometimes undergo one division, resulting in a pair of daughter

nuclei in each of the three cells. No separating wall is formed between them, however, and they are soon absorbed.

14.5 Pollen Structure (Structure of Microspore)

14.5.1 Pollen walls and Protoplasm

It is commonly globular in outline, though several other shapes are also found. The diameter is 25-50 µm. There is a highly resistant wall on the outside and cellular contents inside. Wall or covering of pollen grain is called sporoderm. It has two layers, outer exine and inner intine. Intine is pecto-cellulosic in nature. At places it contains enzymatic proteins (Knox and Heslop-Harrison, 1971). Exine is made of a highly resistant fatty substance called sporopollenin (Zelisch, 1932). Sporopollenin is not degraded by any enzyme. It is not affected by high temperature, strong acid or strong alkali. Because of the sporopollenin, pollen grains are well preserved as microfossils. At places, exine possesses proteins for enzymatic and compatibility reactions. Exine is differentiated into outer ektexine (sexine) and inner endexine (nexine). Ektexine is further made up of an inner continuous foot layer, a middle discontinuous baculate layer and outermost discontinuous tectum. Tectum provides a characteristic sculpturing or designs over the surface of pollen grain, e.g., ridges, tubercles, spines, reticulations. It can help experts to identify the pollen grains and refer them to their family, genus or species. The study of external morphology of mature pollen grains is called palynology.

In insect pollinated pollen grains the exine is spiny as well as covered over by a yellowish, viscous sticky and oily layer called pollenkit. **Pollenkit** is made up of lipids and carotenoids. At certain places the exine is thin or absent. The areas may have thickened intine or deposition of callose. They are called germ pores (if rounded) or germinal furrows (if elongated). Pollen grains are generally tricolpate (with three germ pores) in dicots and monocolpate (with single germinal furrow) in monocots.

Its cytoplasm is rich in starch and unsaturated oils. The latter protect the chromo-somes from radiation damage. Pollen grain protoplast is uni-nucleate in the beginning but at the time of libera-tion it becomes 2-3 celled. (Fig: 14.7 and Fig 14.8)

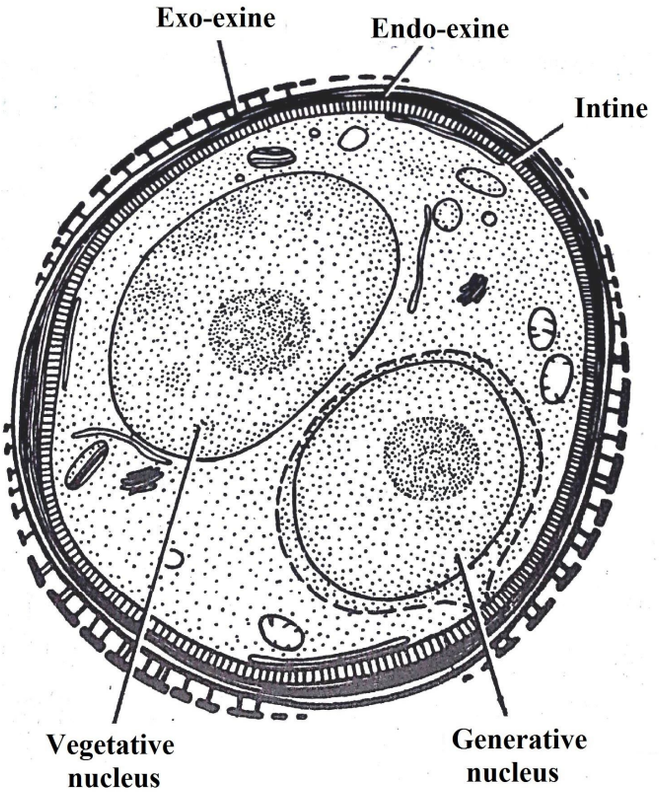

Fig. 14.8 : Structure of Pollen Grain(Microspore)

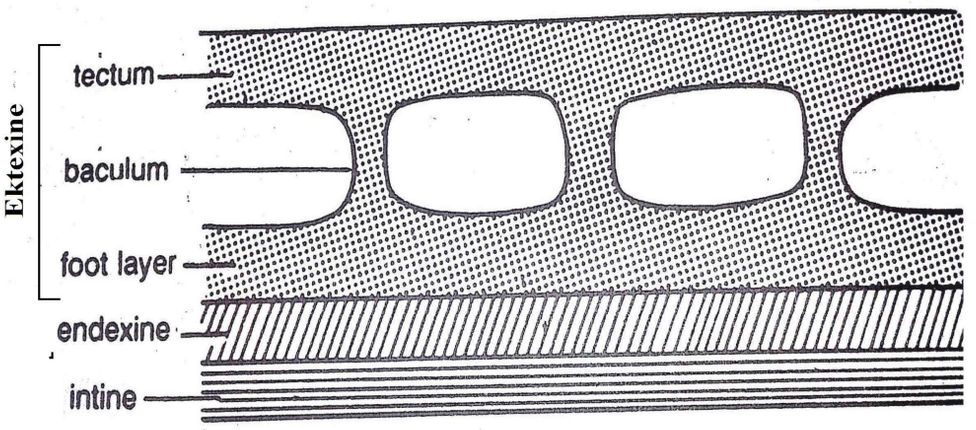

Fig.14.9 : Pollen wall-Different sub layers

14.5.2 Pollen Viability

It is the period for which pollen grains retain the ability to germinate. Pollen viability is little in flowers which are pollinated in bud condition. It is 30 minutes in Rice and Wheat. In others the period of viability is long, even months in some mem-bers of family rosaceae, leguminosae and solanaceae. It, however, depends upon envi-ronmental conditions of temperature and hu-midity. It is possible to store pollen grains for years in liquid nitrogen (- 196°C) in pollen banks for later use in plant breeding programmes.

14.6 Pollen Allergy and Pollen Embryo

An allergy is a condition which manifests as an exaggerated defense reaction of the body to allergens. Pollen grains may act as allergen that is a foreign substance that can cause an allergic response in the body but is only harmful to some people. Some other common allergens are, animal dander, house dust, feathers, and varied foods. In the case of pollen allergies, these allergens are from a wide variety of plants such as trees, bushes, grasses, grains, herbs and less frequently flowers. Airborne pollen grains are a major trigger of a variety of allergic symptoms. Most pollens of allergic importance are wind-borne and too small to be seen by the unaided eye. If the pollen is easily seen, it is usually too large as well as too heavy to be an important allergen. Most plants with windborne pollen are drab with inconspicuous flowers.

14.6.1 Causes of Pollen Allergy

Generally, the heavy pollens of showy plants with colorful, fragrant flowers require insect transport. The first pollens appearing in the spring in the Northeast are tree pollens. They begin to appear in mid-March and are present into June. Maple, poplar, and ash are the earliest pollens beginning in April. In late April, May, and early June, birch and oak pollens are present in the air. Each tree species produces pollen differently from other species, with a variation in the intensity, duration and seasonal pattern of pollination.

In the spring time, people often notice their cars are covered with yellow pollen. This large, waxy pollen released from pine trees is not particularly problematic from an allergy standpoint. Likewise, the fuzzy seed balls seen in the air in May and June do not cause allergy problems.

Throughout the pollen season, even mild winds can carry pollen for many miles and produce high concentrations in urban areas, far from their rural and suburban sources. Air cleansing by rainfall has been found to correlate well with duration of precipitation.

Utilizing air conditioning can markedly cut down on the amount of pollen in room air, as well as, the car. Over-the counter antihistamines can be helpful, however, they may cause

drowsiness. There are several non-sedating antihistamines available by prescription. There are anti-inflammatory nasal sprays; an antihistamine nasal spray, decongestant products, and eye drop preparations that can help alleviate symptoms. Allergy shots can be very helpful in reducing sensitivity to pollen.

Grass pollens are major allergens begin to appear in late May. This season tends to peak in June and then low levels of pollen are present throughout the summer months. In the northeastern states, the majority of grass pollen comes from blue grass, orchard grass, timothy grass, and red top. At this time of year, many people will speak of having "rose fever", a term that has been passed down through the generations. Actually, the pollen of the rose is too heavy to cause symptoms; it's the grass pollen causing problems at this particular time of the year. Ragweed pollen becomes airborne from mid-August through September, which is also hay baling season. "Hay fever" is not associated with hay but with ragweed that is pollinating in late summer and early fall. In our region,

14.6.2 Symptoms

Pollen allergy can cause symptoms such as a runny nose and watery eyes. Reactions to allergens often also play an important role in asthma. Common symptoms of allergic rhinitis and asthma include:

- Runny nose and mucus production
- Sneezing
- Itchy nose, eyes, ears, and mouth
- Stuffy nose
- Red and watery eyes
- Swelling around the eyes
- Coughing
- Wheezing
- Chest tightness
- Shortness of breath

However, not all of these seasonal symptoms are due to pollen. Rhinovirus, the cause of the common cold, also can cause runny noses in the fall and spring. It is not always easy to figure out whether an allergy or a common cold is the cause of these symptoms, although some clues can help distinguish between the two. For example, a fever suggests a cold rather than an allergy, and symptoms lastin gmore than 2 weeks suggest allergies rather than a cold.

The correct name for hay fever is seasonal allergic rhinitis. Even though it was known that pollen rather than hay was the cause as far back as the early 1800's, the term hay fever is still frequently used. Allergic rhinitis symptoms are caused by the body's immune response to inhaled pollen, resulting in chronic inflammation of the eyes and nasal passages.

Allergic rhinitis is a common and debilitating disease it includes the followings:

1. Allergic rhinitis predisposes people to more frequent sinus infections

   People with allergic rhinitis often suffer from fatigue due to poor quality sleep.
2. Moderate or severe allergic rhinitis impairs learning and performance in children, results in more frequent absenteeism in adults and reduced productivity, and therefore can cause considerable impairment in quality of life.
3. Around 8 in 10 people with asthma have allergic rhinitis, and having allergic rhinitis can make asthma more difficult to control
4. Pollen can also trigger asthma Pollen can also trigger asthma.
5. Some people with moderate or severe allergic rhinitis believe that their allergic rhinitis 'turns' into asthma or that it makes them tight in the chest or wheeze. However, pollen can directly trigger asthma as well as allergic rhinitis. Small particles of allergens can penetrate deep into the airways of the lung.

Thunderstorms can also contribute to this:

1. When pollen granules come into contact with water, starch granules are released that are small enough to be breathed into the airways, causing allergic rhinitis and asthma in some people.
2. If anyone wheeze mostly during Spring and/or Summer, should contact to doctor for appropriate advice.

Pollen seasons can last for several months. Pollination times vary with the plant variety and its location. For example, trees pollinate in late winter and early spring. Grasses flower next, and the weed flowers from August through to May. Grass pollen numbers are also higher in in land areas, where there are no natural barriers to wind dispersal.

14.6.3 Control Measures

a. Medications:

Certain over the counter and prescription medications may help reduce the severity of pollen allergy symptoms.

Antihistamines

Antihistamines, which are taken by mouth or as a nasal spray, can relieve sneezing and itching in the nose and eyes. They also reduce runny nose and, to a lesser extent, nasal stuffiness. Some older antihistamines can cause side effects such as drowsiness and loss of alertness and coordination. Effective, newer antihistamines cause fewer or no side effects.

Nasal Corticosteroids

Nasal corticosteroid sprays are anti-inflammatory medicines that help block allergic reactions. They are widely considered to be the most effective medication type for allergic rhinitis and can reduce all symptoms, including nasal congestion. Unlike corticosteroids taken by mouth or as an injection, nasal corticosteroids have few side effects. Combining a nasal antihistamine with a nasal corticosteroid appears to be more effective than using either of the sprays alone. However, it is not clear if taking an oral antihistamine with a nasal corticosteroid is helpful.

Decongestants

Oral and nasal decongestants help shrink the lining of the nasal passages, relieving nasal stuffiness. Decongestant nose drops and sprays are intended for short-term use. When used for more than a few days, these medicines may lead to even more congestion and swelling inside the nose. Doctors may recommend using decongestants along with an antihistamine because antihistamines do not have a strong decongestant effect.

Leukotriene Receptor Antagonists

Leukotriene receptor antagonists, such as the prescription drug montelukast,

block the action of important chemical messengers other than histamine that are

involved in allergic reactions.

Cromolyn Sodium

Cromolyn sodium is a nasal spray that blocks the release of chemicals that cause allergy symptoms, including histamine and leukotrienes. The drug causes few side effects but must be taken four times a day.

b. Allergen Immunotherapy

Many people with pollen allergy do not get complete relief from medications and may be candidates for immunotherapy. Immunotherapy is a long-term treatment that can help prevent or reduce the severity of allergic reactions and change the course of allergic disease by modifying the body's immune response to allergens.

Allergy Shots (Subcutaneous Immunotherapy-SCIT)

Allergy shots, also known as Subcutaneous Immunotherapy (SCIT), have been used for more than 100 years and can provide long lasting symptom relief. SCIT involves a series of shots containing small amounts of allergen into the fat under the skin. SCIT includes two phases: a buildup phase and a maintenance phase.

During the buildup phase, doctors administer injections containing gradually increasing amounts of allergen once or twice per week. This phase generally lasts from 3 to 6 months, depending on how often the shots are given and the body's response. The aim is to reach a target dose that has been shown to be effective.

Once the target dose is reached, the maintenance phase begins. Shots are given less frequently during the maintenance phase, typically every 2 to 4 weeks. Some people begin experiencing a decrease in symptoms during the buildup phase, but others may not notice an improvement until the maintenance phase. Maintenance therapy generally lasts 3 to 5 years. The decision about how long to continue SCIT is based on how well it is working and how well a person tolerates the shots. Many people continue to experience benefits for several years after the shots are stopped. Side effects from SCIT are usually minor and may include swelling or redness at the injection site.

However, there is a small risk of serious allergic reactions such as anaphylaxis, a potentially life threatening reaction that can develop very rapidly. Because most severe reactions occur shortly after injection, it is recommended that

patients remain under medical supervision for at least 30 minutes after receiving a shot.

Sublingual Immunotherapy (SLIT)

There are some approved types of under-the-tongue tablets to treat allergies to grass and ragweed. The treatments, called sublingual immunotherapy (SLIT), offer people with these allergies a potential alternative to allergy shots. People taking SLIT place a tablet containing allergen under the tongue for 1 to 2 minutes and then swallow it. SLIT tablets are taken daily before and during grass or ragweed season.

Studies show that there are fewer allergic reactions to SLIT compared with SCIT. After the first SLIT dose is given at the doctor's office, patients can take subsequent doses at home. Side effects of SLIT are usually minor and may include itching of the mouth, lips, or throat. Although severe allergic reactions to SLIT are extremely rare, because SLIT treatment takes place at home, doctors usually prescribe an epinephrine auto-injector (EpiPen) for use in the event of a serious reaction. Effective treatments are available Seek advice from your pharmacist or doctor about medications or treatments that will relieve your symptoms. Although medications do not cure allergies, they are much more effective with fewer side effects than medications available 20 years ago. You just need to know the best way to use them, and to avoid medicines that can cause more problems than they solve, like frequent decongestant (unblocking) nose sprays or tablets.

d. Precaution against Pollen Allergy for Pollen Sensitive Individuals:

1. During the pollen season keep all windows in your home and car closed. This is particularly useful during the night hours to prevent pollens or molds from drifting into your home. Instead, use air conditioning, which cools, cleans and dries the air. Avoid the use of fans, which will increase the amount of airborne pollen and result in increased symptoms.
2. HEPA air cleaners may be helpful when air conditioning is unavailable.
3. Stay indoors on high pollen days and especially on windy days. Avoid early morning activity when pollens are usually emitted (between 5am – 10am).
4. When vacationing during the height of the pollen season, choose places such as the beach or seacoast, which are more pollen-free.

5. Remember that pollen is sticky and will adhere to your clothes and person until washed off. Showering after long exposure and rinsing your hair before going to bed to remove pollens that have accumulated during outdoor exposure is helpful. When working outdoors, wear a mask when raking leaves or mowing the lawn.
6. Take your medications prescribed by your allergist regularly, in the recommended doses. Don't take more medication than is recommended to relieve your symptoms.
7. Don't hang your clothing outside to dry. Pollens and molds will collect on Sleep with windows closed.
8. Limit the time you spend in the open air you cannot live pollen-free. Adapt walking and sporting activities to suit the prevailing conditions.
9. Wash or rinse your hair before you go to bed to get rid of pollen which would otherwise be deposited in your bed and breathed in.
10. Remove clothes on which pollen has collected and keep them outside your bedroom.
11. Do not dry laundry outdoors.
12. Avoid working in the garden, or only work in appropriate weather conditions.
13. Avoid additional irritations to the mucous membranes, e.g. eye cosmetics, cleaning agents, smoking, frying fumes etc.
14. Choose high mountain areas or the seaside when planning your holiday.

14.6.4 Pollen Embryo/ Embryo sac like Pollen Grains

In 1898 Nemec noted that in the petaloid anthers of *Hyacinthus orientalis* the pollen grains sometimes form large eight-nucleate structures showing a surprising resemblance to embryo sacs. He believed that they arose as the result of a degeneration of the generative nucleus and three divisions of the vegetative nucleus. De Mol (1923) observed this so-called "Nemec-phenomenon" in the anthers of other varieties of *Hyacinthus orientalis* which had been subjected to certain special conditions in order to obtain early flowering. He attributed the origin of the abnormality to a duplication of the generative nuclei. Stow (1930, 1934) found similar embryo-sac-like pollen grains or "pollen-embryo sacs" in the anthers of a variety called "La Victor" whose bulbs had been subjected to a temperature of 20°C. at the time of meiosis and were further "forced" in a

greenhouse. He traced their development more fully than either Nemec or De Mol.

At first the microspores increase in size to form large sac-like bodies, after which the nucleus undergoes three successive divisions to form 8 daughter nuclei. Of these, 3 lie at the end where the exine is still intact, 3 at the opposite end, and 2 in the middle. The 6 nuclei at the two poles organize into cells, while the remaining two fuse in the center. Since the three cells at the exine end were found to remain healthy for a much longer time than those at the opposite end, Stow regards the former as corresponding to the egg and synergids, and the latter to the antipodals. In addition certain abnormal pollen-embryo sacs were also seen, showing the following types of organization:

1. 8 nuclei forming an egg, two polars, and five antipodal cells;
2. 4 nuclei forming an egg, two polars, and one antipodal cell;
3. 4 nuclei forming a polar and three antipodal cells but no egg;
4. 16 nuclei forming a 5- to 10-celled egg apparatus, one or two polars, and a few antipodal cells; and
5. More than 16 nuclei without any definite arrangement.

According to Stow, it is not the divisions of the vegetative or generative nucleus which give rise to the pollen-embryo sacs but those of the microspore nucleus itself. Once the vegetative and generative cells have been differentiated, further development is quite normal and no pollen-embryo sacs are formed. Further, the pollen-embryo sacs were always accompanied by a large number of dead pollen grains, leading Stow to suggest that the later secrete a "necrohormone" which causes an abnormal growth of the surviving pollen grains. Stow also observed that when the pollen-embryo sacs were placed on an agar medium, together with some normal pollen grains of another variety, the pollen tubes formed from the latter coiled around the former. Once a sperm nucleus was observed to be in process of entering the pollen-embryo sac; and in another case the pollen-embryo sac showed 16 nuclei, believed to have been derived from the divisions of a triple fusion nucleus. In conclusion Stow says that all pollen grains are potentially capable of assuming either the male or the female form. Under normal conditions the "male potency" is dominant over the "female potency" leading to the formation of the generative cell and the male gametes; but under abnormal conditions, when there is a

release of necrohormones, the female potency gets the upper hand resulting in the formation of embryo-sac-like structures.

After Stow, Naithani (1937) found embryo-sac-like pollen grains in the variety "Yellow Hammer" whose bulbs had been treated for early flowering. He confirms Stow's observations regarding the mode of development of these abnormal pollen grains, but believes their formation to be a temperature effect and not the result of liberation of necrohormones. According to him, the degeneration of the other pollen grains is not the cause but the effect of a hypertrophied growth of the more favored ones, which use up all the available food for their own growth. More recently, pollen-embryo sacs with 8 and 16 nuclei have also been observed in another plant, *Orniihogalum nutans* (Geitler, 1941). Those with 8 nuclei showed the typical embryo-sac-like organization, but, contrary to Stow, Geitler interprets the three cells at the exine end of the pollen grain as the equivalents of antipodals and the other three as equivalents of the egg and synergids.

14.7 Summary

Microsporogenesis is the developmental process leading to the production of four haploid spores from a diploid sporocyte by meiosis and cytokinesis. Each microsporocyte produces four microspores arranged in various patterns, resulting in differently shaped tetrads: tetrahedral, tetragonal (or isobilateral), decussate, rhomboidal, T-shaped, and linear. The existing typological convention for microsporogenesis recognizes two patterns simultaneous type and successive type. Each microspore enters into microgametogenesis in which the nucleus of microspore divides mitotically to produce a bigger vegetative cell and a smaller generative cell. Generative cell divides mitotically and forms two male gametes and finally trinucleate microgametophyte is formed.

As far as allergy is concern grass-related allergenic activity is present throughout the year, demonstrating the existence of aeroallergens outside the pollen season. Symptoms in allergic patients may be related to airborne pollens concentrations. This fact should be taken into account in the management of allergic patients that is clinical a well as precautionary.

14.8 Glossary

- **Allergy:** An allergy is a condition which manifests as an exaggerated defense reaction of the body to allergens.

- **Allergen:** Any substance to which a person is allergic (for example, pollen, house dust mite droppings, animal dander, peanuts).
- **Asthma:** A disease in which the airways (the breathing tubes taking air in and out of the lungs) become inflamed and swollen, making breathing difficult. In many cases it is caused by an allergy.
- **Micro:** Suffix that originally denoted small but has, in a botanical sense, taken on the meaning "male".
- **Microgametophyte:** In seed plants this is the pollen grain.
- Microsporangiate cone = male cone: terminal clusters of microsporophylls such as the pollen cones of conifers.
- **Microsporangium:** Sporangium that bears microspores. In seed plants synonymous to a pollen sac.
- **Microspore Mother Cell:** Microsporocyte: Diploid cell destined to undergo meiosis to produce microspores in the seed plants.
- **Microspore:** Spore that develops into a microgametophyte.
- **Microsporophyll:** Modified leaf that bears microsporangia. Examples from the seed plants include the stamens of flowers and the subunits of the pollen cones of conifers.
- **Rhinitis:** It is an inflammation of the mucous membrane that lines the nose, often due to an allergy to pollen, dust or other airborne substances. Seasonal allergic rhinitis also is known as "hay fever," a disorder which causes sneezing, itching, a runny nose and nasal congestion.

14.9 Self-Learning Exercise

Section – A : (Very Short Answer Type)

1. What are allergens?
2. Define microgametogenesis.
3. Name various types of pollen tetrads.
4. How many nuclei in angiospermic microspore?
5. What is endomitosis?
6. What are restitution nuclei?

Section – B : (Short Answer type)

1. Write general characters of microsporogensis.

2. Give a note on pollen wall.
3. Describe tapetum briefly.
4. Briefly explain symptoms of pollen allergy.

Section – C : (Long Answer type)

1. Describe the structure of anther.
2. Give a detailed account on control measures against pollen allergy.
3. Describe microgametogenesis with suitable diagrams.
4. Give a detailed account on structure of microspore.

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Unit-15

Structure and Development of Female Gametophyte

Structure of the Unit:

15.0 Objectives

After studying this unit you will be understand about:

- the female reproductive parts
- Ovules: structure, development and types

- Process of megasporogenesis and megagametogenesis
- Female Gametophyte (Embryo sac): structure, development and types

15.1 Introduction

By studying this unit students will learn about the structure and development of ovules which are the site of megasporogenesis, embryo sac formation, fertilization and embryogenesis. Besides, they will also know about the various types of ovules diferring from each other in term of their location in the ovary, number and thickness of integuments, nucellus thickness, direction and degree of curvature etc.

Furthermore, students will come to know about the details of megasporogenesis and megagametogenesis process which result into formation of megaspore and female-gametophyte (embryo sac) respectively. Finally, they will also learn about the various modes of development of embryo sac and its structure.

15.2 Structure of Ovule

Ovules as developmental predecessors of seeds are important organs in flowering plants. Their evolution can be traced back to the most primitive seed plants. In angiosperms, ovules have diversified in relation to their location in the ovary, number and thickness of integuments, nucellus thickness, direction and degree of curvature and histological differences. Even though ovules have their specific developmental process, certain structural features of ovules, for instance symmetry and curving, are influenced by their position in the ovary.

Angiosperm ovules fundamentally comprise of a nucellus enclosed by two integuments (Fig. 15.1). They may be sessile or attached to the placenta by a stalk called the funiculus. Usually a vascular bundle runs from the placenta through the funiculus to the chalaza, i.e. the region right beneath the base of the nucellus where the integuments set out. The chalaza and the funiculus are intercalary structures and therefore less delineated than the integuments and nucellus. The nucellus is equivalent to the megasporangium, in which a megaspore mother cell go through meiosis producing four megaspores, characteristically only one of which grows into an embryo sac. Embryo sac represents the megagametophyte. It contains mostly four or eight nuclei, structured into four or seven cells, depending on number of mitotic divisions in the growing embryo sac. Embryo sac contains the egg cell, accompanying with two synergids, all three making the egg apparatus, a central cell with one or two

nuclei and three antipodal cells. The micropyle, a narrow passage through which a pollen tube reaches the nucleus, is formed by the inner or both integuments. Growing through micropyle, pollen tube enters into the nucellus and the embryo sac, and there into one of the synergids. The pollen tube carries two male gametes; one of them fertilizes the egg cell developing into the zygote and the other fuses with the nucleus of the central cell (double fertilization), which then grows into the endosperm. In classic embryo sacs having seven cells, the central cell incorporates two nuclei, which fuse into a diploid nucleus and the endosperm converts triploid. It is the most common type of embryo sac in angiosperms (Polygonum type, fig. 15.1).

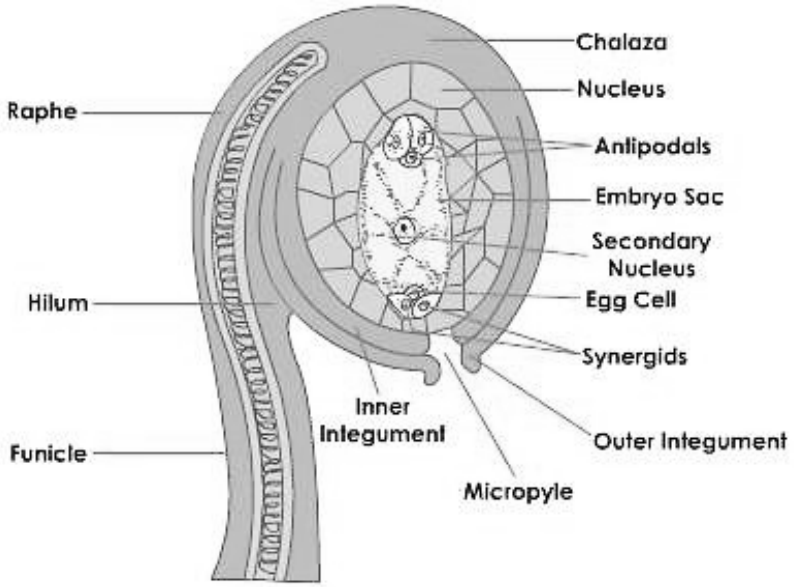

Fig. 15.1: Structure of an Ovule

15.3 Types of Ovules

Ovule can be classified in term of several aspects. The main features of ovule diversity are size, degree of curvature, nucellus thickness, integument number and thickness, development of the micropyle, funiculus length, extent of vascularization of the ovule and various histological variations (e.g. hypostase, postament and endothecium).

Generally in angiosperms, ovules are about 0·5 mm long during fertilization. But their length may be as short as approx. 0·15 mm or they may attain size of more than 2 mm.

Ovules vary in their degree of curvature (Fig. 15.2). Orthotropous and atropous (straight and uncurved) ovules are radially symmetric. Anatropous and

campylotropous ovules are curved but in the former nucellus is straight while in the latter one nucellus is also involved in the curvature. Hemitropous (hemianatropous) ovules are only slightly curved. The terms (in French) 'orthotrope', 'anatrope' and 'campulitrope' for the different degree of curvature were coined by Mirbel (1829). Bent ovules may be monosymmetric or, if twisted, may be asymmetric. In present angiosperms, anatropous ovules are most probably primitive. Some other forms, in addition to orthotropous, anatropous and campylotropous have been defined but their systematic importance is unknown.

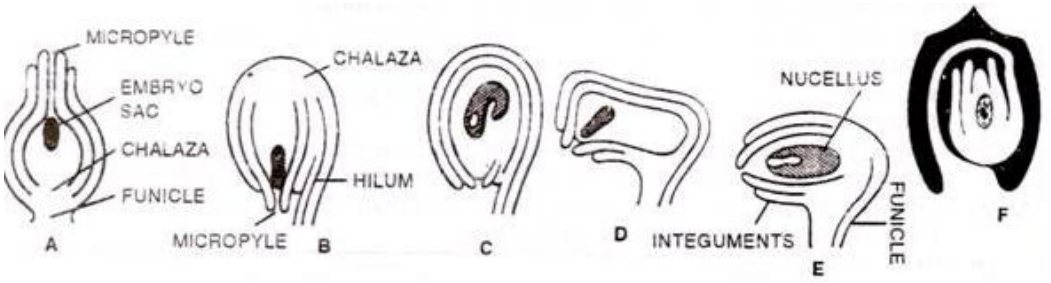

Fig. 15.2: Different types of Ovules on the basis of their degree of curvature. (A) Orthotropous, (B) Anatropous, (C) Amphitropous, (D) Campylotropuous, (E) Hemitropous, (F) Circinotropous

The nucelli vary in thickness (Fig. 15.3). Ovules having thick and thin nucelli are known as crassinucellar and tenuinucellar ovules respectively. Number and thickness of integuments also vary in ovules (Fig. 15.4). On the basis of integument numbers, Van Tieghem (1901) classified angiosperms as 'plantes bitegminées' and 'plantes unitegminées', plants with bi- and unitegmic ovules respectively.

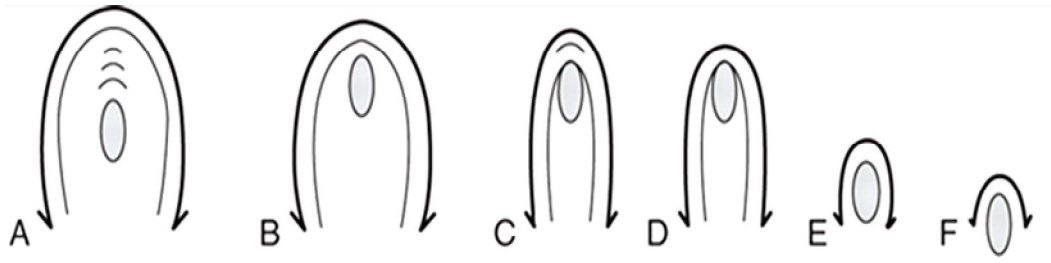

Fig. 15.3: Different types of Ovules on the basis of variation in nucellus thickness. (A) Crassinucellar (B) Weakly crassinucellar (C) Pseudocrassinucellar (D) Incompletely tenuinucellar (E) Tenuinucellar (F) Reduced tenuinucellar. (Thick lines, morphological surfaces; thin lines,

boundaries between cell layers; epidermal layer drawn in full, other layers only partially drawn). Megaspore mother cells are shaded light grey. (Source: Peter K 2011)

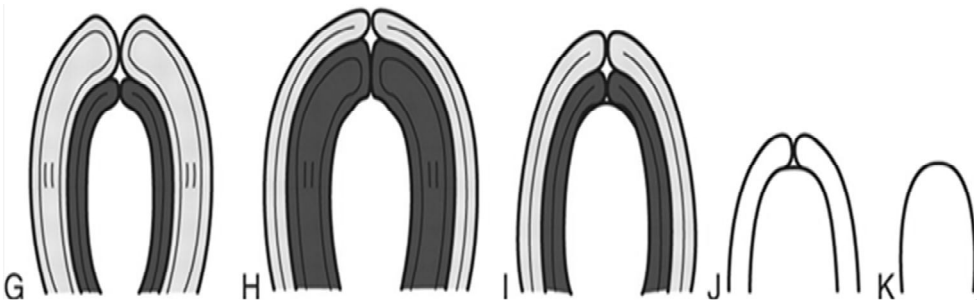

Fig. 15.4: Different types of Ovules on the basis of variation in integument number and thickness. In bitegmic ovules, the inner integument is shaded dark grey, the outer light gray. (G) The outer integument is thicker than the inner. (H) The inner integument is thicker than the outer. (I) both integuments are equally thick. (J) Unitegmic. (K) Ategmic. (Source: Peter K 2011)

One or both integuments may form the micropyle. In some circumstances micropyle is absent in mature ovule, and neighboring parts (obturator or funiculus) may be in contact with the integuments or the nucellus. Ovules develop on the placentae in the carpels. They may be stalked or sessile. Stalks (funiculi) also vary in their length.

A vascular bundle extends from the placenta through the funiculus and raphe to the chalaza in most ovules. In certain angiosperms clades having large seeds, vascular bundles also reach into one of the integuments. Contrary to this, some ovules contain only an undifferentiated procambial strand or no strand at all. Generally, these ovules are small or reduced.

15.4 Development of Ovule

As described above, ovules are derived from the placenta of the ovary wall. They are dedicated structures to produce the megasporocyte and are the site of megasporogenesis, embryo sac formation, fertilization and embryogenesis. Process of megasporogenesis and embryo sac formation is coupled with the development of ovule itself. Ovule development starts with formation of ovule primordium. Nucellus develops from its apical part and functions as the megasporangium. Soon after ovule origination, a single subdermal nucellar cell present below the apex of the nucellus expands and exhibits a

conspicuous large nucleus (Fig. 15.5). This cell functions as the archesporium which may act directly as the megasporocyte or it may go through one mitotic division to yield a megasporocyte and a somatic cell. The number of archesporial cells may be more than one. A multicellular archesporium is found in mustard and soybean, however only one of archesporial cells produces the megagametophyte. Factors that regulate archesporium development and decide the identity of the megasporocyte are yet to determined.

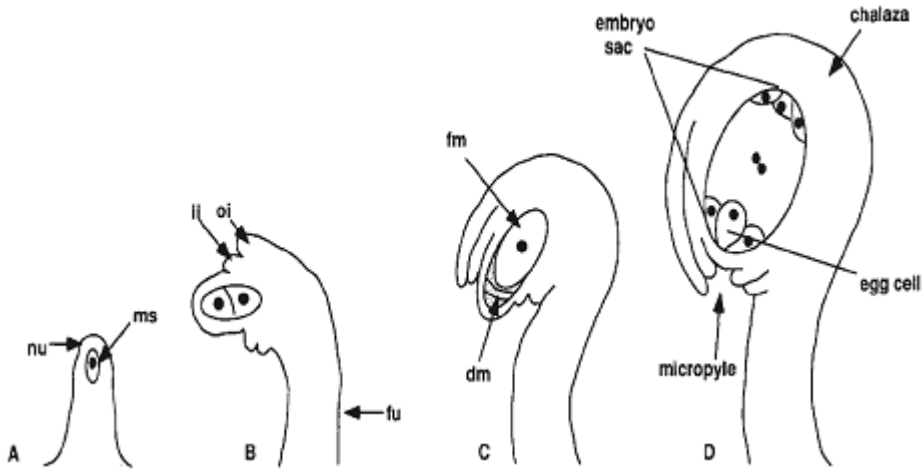

Fig. 15.5: Ovule Development

Stages are shown for anatropous ovule with *Polygonum*-type embryo sac development. A) Ovules shortly after initiation, showing a single megasporocyte (ms). nu, nucellus. (B) Ovule after both integuments have been initiated. Megasporocyte has undergone meiosis-I. The axis of the nucellus is transiently perpendicular to the axis of the funiculus (fu). ii, inner integument; oi, outer integument. (C) Ovule after meiosis. The functional megaspore (fm) at the chasal end has expanded and the nonfunctional megaspores are defenerated. The axis of nucellus is now parallel to the funiculus due to unequal growth, primarily of the integuments. dm, degenerate megaspores. (D) Ovule after megagametogenesis. The embryo-sac contains seven cells and eight nuclei. (Source: Leonore R and Robert LF, 1993)

15.5 Megasporogenesis and Development of Female Gametophyte

Development of embryo sac occurs in two stages: megasporogenesis and megagametogenesis (Fig. 15.6). During the first stage, the megasporocyte goes

through meiosis and four megaspores are formed. Later the functional megaspore undergoes mitotic divisions, nuclear migration and cytokinesis during megagametogenesis and produces embryo sac.

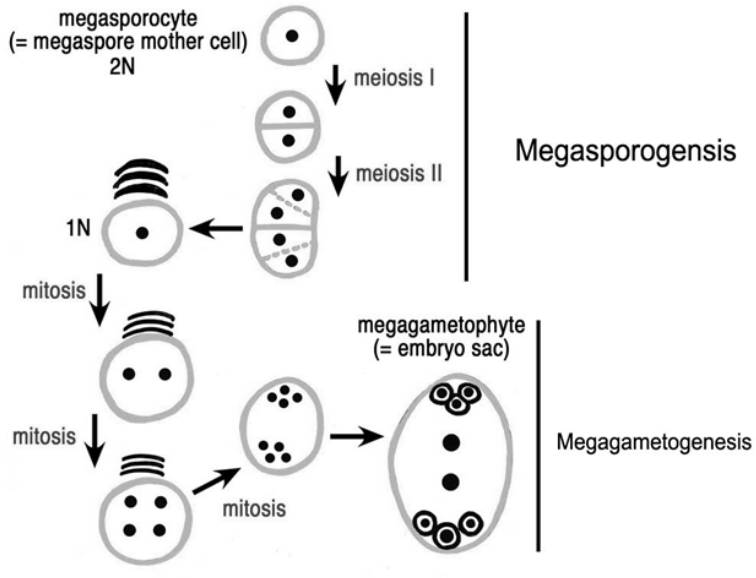

Fig. 15.6: Development of Embryo Sac: megasporogenesis and megagametogenesis

Pattern of embryo sac development differs significant among plant species. First, there are many differences in megasporogenesis itself. Frequently, all the four separate megaspore cells do not grow further.

In about 70% of the plant species, polygonum-type embryo sac development has been observed. In this pattern embryo sac develops from chalazally located functional megaspore. During meiosis-I of megasporogenesis, the orientation of spindle is parallel to the micropylar-chalazal axis of the nucellus. Cell wall is formed at right angles to this axis. After meiosis-II division, another transverse wall is formed; bring about linearly arranged four megaspores. The megaspore adjoining to the chalaza remains functional. The remaining three megaspores degenerate and are ultimately crushed by the growing functional megaspore. Callose is believed to play an important role in the selection of a functional megaspore. After megasporogenesis, callose is found in the walls of the nonfunctional megaspores while functional megaspore contains no callose or very less amount of callose in its wall. Selective deposition of callose perhaps makes sure that only the functional megaspore gets nutrients from the nucellus. The pattern of callose deposition reflects the pattern of megasporogenesis. For

instance, in Oenothera, callose is thinner at the micropylar end of the ovule, where the functional megaspore is found. In tetrasporic species, meiosis occurs without cytokinesis, and callose does not store in the walls of the single tetranucleate megaspore.

The female gametophyte is generated from the functional megaspore through the process of megagametogenesis. As described above, in Polygonum-type embryo sacs, the functional megaspore at the chalazal end enlarges before three rounds of free nuclear divisions. After the first mitosis, the two nuclei move to opposite ends and the smaller vacuoles merge to form a large central vacuole. Formation of this central vacuole is important for proper positioning of the nuclei before successive divisions. Each of the two nuclei then undergoes two more divisions, resulting in formation of four nuclei are at each pole. One nucleus from each pole migrates to the center of the embryo sac where they fuse to form a 2n secondary nucleus. Three nuclei at micropylar pole get surrounded by membranes. The central cell among them enlarges and turns into egg cell. The adjacent cells become synergids. These three cells form egg apparatus. Three antipodal cells are formed at chalazal end. Finally embryo sac is made encompassing 8-nucleli and later 7-cells during its development.

15.6 Types of Embryo Sac Development

The process of embryo sac development differs in plants (Fig. 15.6). The main differences are regarding number of spore nuclei involved in embryo sac formation, number of nuclear divisions during megasporogenesis and embryo sac development and arrangement of nuclei in the mature embryo sac. Basically, there are following three types of embryo sac development-

15.6.1 Monosporic type

Polygonum type: It is commonly found in plant. However, it was first clearly described in Polygonum. Therefore, it is also called as Polygonum type. In this type, the female gametophyte (embryo sac) develops from a single functional megaspore present towards chalazal end in the nucellus (Fig. 15.6).

Oenothera type: In Oenothera, the development is monosporic but the functional megaspore is located towards micropyle in nucellus and only 4 nuclei (the antipodals are eliminated) develop instead of 8.

15.6.2 Bisporic or Allium type

In Allium, the megaspore mother cell undergoes meiosis I giving rise to two haploid cells one of which degenerates while nucleus of the remaining one goes

through meiosis II resulting a cell with 2 nuclei (megapsore nuclei) which move towards opposite ends. These nuclei undergo two mitotic divisions forming 8 nuclei which get arranged as the polygonam type embryo sac. Thus a bisporic embryo sac contains 8 nuclei. This type of development is found in several monocot and dicot families (Fig. 15.6).

15.6.3 Tetrasporic type

In a number of cases, cytokinesis and cell wall formation are not accompanied by meiosis therefore the normal distinct megaspores are not produced. All four haploid megaspore nuclei are involved in the development of the embryo sac. The subsequent embryo sac may be 8 or 16 nucleate. Hence the embryo sac is tetrasporic as all the nuclei of the four megaspores are within the single cell. Many variations of the tetrasporic embryo sac are found as below (Fig. 15.6)-

Plumbago type (8-Nucleate): In this type, the 4 megaspore nuclei organize themselves in a cross-like fashion. One lies at the micropylar end and the other at the chalazal end. The other two occupy position at each side of the embryo sac. Each nucleus divides once. Thus four pairs of nuclei are produced. One nucleus from each pair moves to the center where they fuse to form 4n secondary nucleus. The micropylar nucleus forms the egg cell. The rest three nuclei degenerate. Therefore, no antipodals and synergids are present in the mature embryo sac.

Fritillaria type (8-Nucleate): Here, three out of four megaspore nuclei migrate to the chalazal end while one nucleus lies at the micropylar pole. The micropylar nucleus divides and two haploid nuclei are formed. The three chalazal nuclei fuse and the resulting nucleus divides once to form two triploid nuclei. At this time the embryo sac encompasses four nuclei, two haploid micropylar nuclei and two triploid chalazal nuclei. Later each nucleus divides. Thus four haploid nuclei at micropylar end and four triploid nuclei at chalazal end are formed. One nucleus from each pole migrates to the center. A tetraploid secondary nucleus is formed by their fusion. Egg apparatus is formed by the nuclei at micropylar end while those present at the chalazal end gives rise to antipodal cells.

Plumbagella type (4-Nucleate): This is a reduced form of Fritillaria type embryo sac. Four megaspore nuclei show 1 (micropylar) + 3 (chalazal) arrangement. Three chalazal nuclei fuse and form 3n nucleus. Now embryo-sac becomes 2-nucleate containing one triploid and one haploid nucleus. Both of them divide to form 4-nucleate stage. There is no further division. Haploid

micropylar nucleus organises into egg while one triploid nucleus form single antipodal cell. Remaining two nuclei (one haploid and one triploid) fuse to form tetraploid secondary nucleus.

Penaea type (16 Nucleate): In this case, 16 nuclei are organized in four groups and each group comprises of four nuclei. One group is present at each end of the embryo-sac and two groups are present at the sides. One nucleus from each quarter moves towards the center and act as polar nucleus. Three nuclei of each quarter become cells. Thus this type of embryo-sac contains four triads and four polar nuclei. One cell of the micropylar triad acts as egg. It is the only functional cell.

Drusa type (16 Nucleate): After formation of four megaspore nuclei, one nucleus migrates towards the micropylar end while the remaining three megaspore nuclei move towards chalazal end. Each nucleus divides twice. Thus four nuclei are produced at micropylar end and twelve at chalazal end. One nucleus from each migrates towards the center of the embryo sac. They fuse to form secondary nucleus. The three nuclei at micropylar end form egg apparatus. The eleven nuclei at chalazal end form antipodal cells.

Adoxa type (8-Nucleate): In Adoxa, 8 nuclei are formed and these nuclei get arranged as the normal (Polygonum) type though the development of megaspore mother cell is tetrasporic.

Peperomia type (16 Nucleate): In this case, each of four megaspores nuclei divides twice. 16 nuclei are formed. They become uniformly distributed at the periphery of the embryo sac. Two nuclei at micropylar end form an egg and a synergid. Eight nuclei fuse to form secondary nucleus. The remaining nuclei stay at the periphery of the embryo sac.

It is clear from the above mentioned details of developments of various embryo-sacs that there may be great differences in the final appearance of the female gametophyte, viz., the egg apparatus may contain only one synergid (Peperomia), the fusion nucleus may involve only one (Oenothera type) or more than two (Peperomia, Penaea, Plumbagella and Fritillaria types) polar nuclei, the number of antipodals may differ from none in Oenothera type to a numerous (Peperomia, Penaea and Drusa types; as many as 300 antipodal cells are reported in Sasa paniculata of Bambusae) and so on.

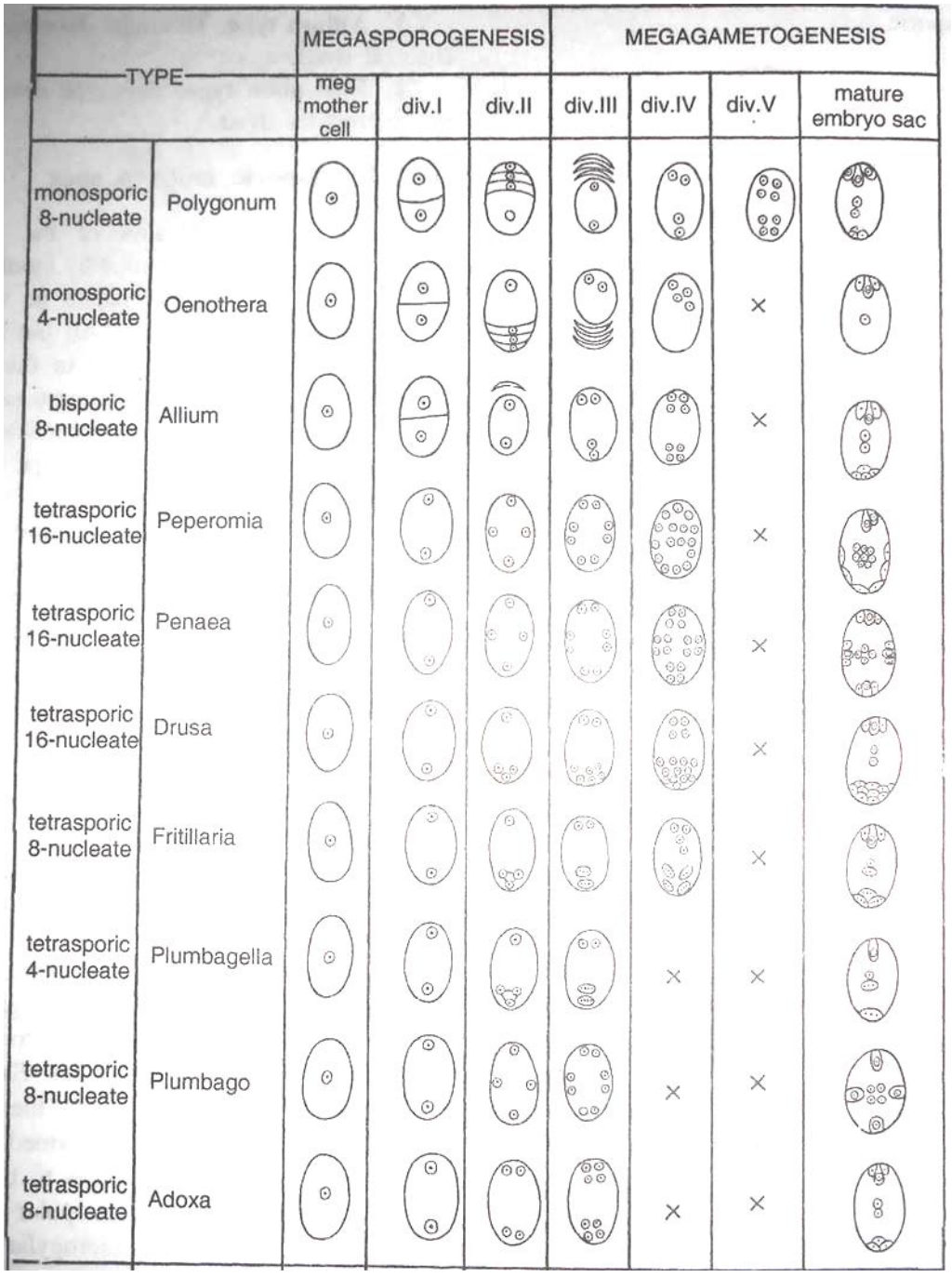

Fig. 15.6: Different types of Embryo Sac development among Angiosperms

15.7 Structure of the mature Embryo-Sac

As shown in figure, the *Polygonum*-type embryo sac contains one egg cell, two synergids, three antipodal cells and a central cell with two nuclei (Fig. 15.7). These cells make four groups that are involved in fertilization, embryogenesis, nourishment of the embryo sac and embryo.

15.7.1 Egg Cell

The egg cell is situated at the micropylar end and in the end fuses with a sperm nucleus to form a zygote. The egg cell lies between the two synergids. The plasmalemma alone or incomplete cell walls separate egg cell from synergids. The egg cell contains a large vacuole at the micropylar end. Due to this the cytoplasm within the egg cell is uneven distribution. Large vacuole confines most of the cytoplasm and the nucleus to the chalazal end.

15.7.2 Synergids

The synergids, present on both sides of the egg cell, are important for fertilization. The pollen tube releases its insides into one of the synergids before fusion of the sperm nuclei with the egg and central cells. Synergids contain filiform apparatus. This is a structure present at the micropylar end of the synergid cell wall that is thickened, forming finger-like projections into the synergid cell cytoplasm.

15.7.3 Central Cell

As the name suggests, this cell occupies the center of the embryo sac. It contains two polar nuclei, a large vacuole and several organelles. The polar nuclei are produced at both ends of the embryo sac during development and move to the center after cellularization. They may partly fuse before fertilization by a single sperm nucleus, producing a 3n primary endosperm nucleus.

15.7.4 Antipodal Cells

These cells are present at the chalazal end of the embryo sac. They do not perform specific function during reproduction but they are believed to be involved in the import of nutrients to the embryo sac.

Synergids, central cells and antipodals contain numerous ribosomes and mitochondria. It suggests they have high metabolic activities. By contrast, the egg cell has few ribosomes, plastids and other organelles and seems to be relatively quiescent.

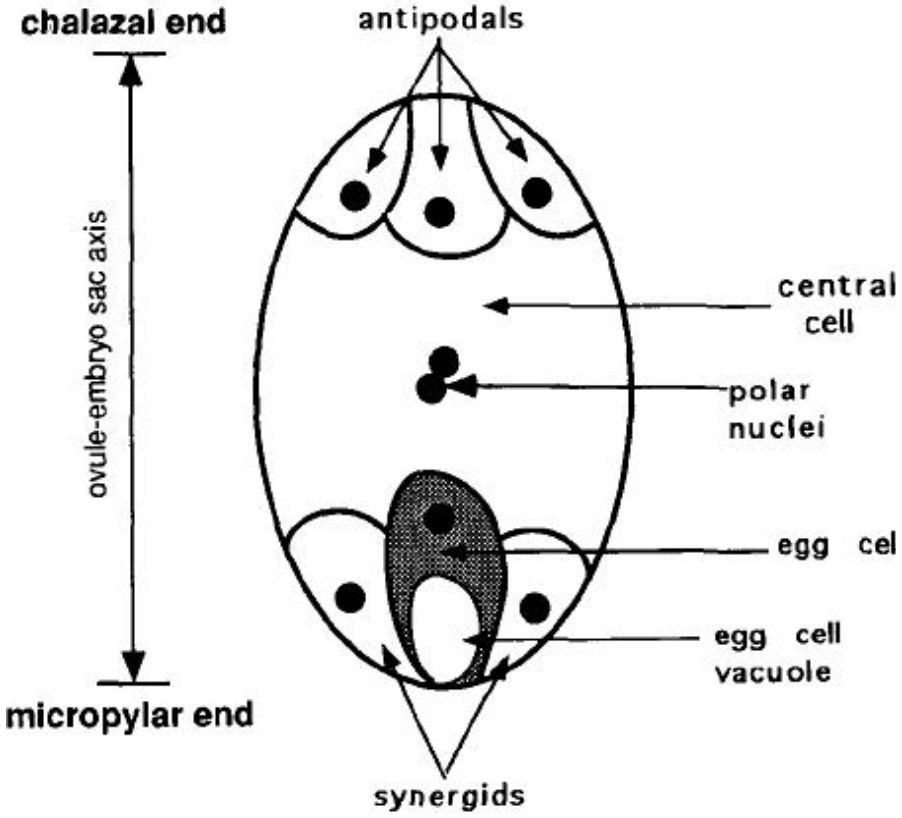

Fig. 15.8: Organization of the Embryo Sac

The orientation of the embryo sac with respect to the chalazal-micropylar axis of the ovule is indicated by the vertical arrow on the left. The egg apparatus, including the egg cell and synergids, is located at the micropylar end, where the pollen tube enters the embryo sac. The central cell contains two nuclei. Three antipodal cells are located at the chalazal end of the embryo sac. The egg cell is actually adjacent to, rather than between, the two synergids. Note the position of the vacuole in the egg cell. (Source: Leonore R and Robert LF, 1993)

15.8 Summary

The life cycle of land plants involves an alternation of generations between a haploid gametophyte and a diploid sporophyte. Whereas animal gametes are formed directly after meiosis, plant gametes are produced only after growth of the multicellular haploid gametophyte. The morphological complexity of the haploid generation ranges from the macroscopic moss gametophytes, which dwarf the sporophyte, to the three-celled male gametophyte (pollen) and seven-celled female gametophyte (embryo sac) that are characteristic of most flowering plants. The latter evolved through an extreme reduction from the female gametophytes of the gymnosperms, which frequently contain over a thousand cells, and is considered a key innovation in the evolution of flowering

plants. These 'stripped down to essentials' female gametophytes confer two major defining characteristics of the flowering plants. First, they are small enough to be packaged within an ovary. Second, they generate two gametes that undergo double-fertilization to produce the nutritive tissue called endosperm concordantly with the embryo, which allows more efficient resource allocation to fertilized seeds. The reduced female gametophyte of flowering plants enabled much more rapid seed setting (i.e. the production of seeds during reproductive growth) than is possible in gymnosperms, allowing for habitat adaptations that require short reproductive cycles and facilitating the expansion of flowering plants into diverse ecological niches.

15.9 Glossary

- **Double fertilization:** in flowering plants: the more or less simultaneous union of one sperm and one egg to form a diploid zygote and another sperm with two polar nuclei to form triploid endosperm in the ovule
- **Gametophyte:** the haploid phase of a life cycle on which gametes are produced
- **Integument:** the outermost layer(s) of an ovule which will develop into the seed coat; most seed plant ovules have one integument, angiosperm ovules have two integuments
- **Megagametophyte**: in heterosporous plants and in seed plants: the female gametophyte produced by a megaspore
- **Megasporangium**: a sporangium that produces megaspores
- **Megaspore**: a large, haploid spore of a heterosporous plant that produces a megagmetophtye (female gametophyte)
- **Micropyle**: a small opening in the integument at the apex of a seed through which either pollen (gymnosperms) or the pollen tube (angiosperms) enters
- **Nucellus**: ovule tissue within which an embryo develops (embryo sac); homologous with the megasporangium of a seed plant
- **Ovule**: unfertilized seed; the ovule contains the megasporangium with the megagametophyte, surrounded by one or two integuments integument

15.10 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. Name the part of the flower where female gametophyte development takes place.
2. Arrange the following terms in correct developmental sequence. Megasporocyte, Embryo sac, Megaspore, Archesporium,
3. Which part of the ovule functions as megasporangium?
4. Which cells form the embryo sac?
5. After double fertilization a zygote and…… are formed.
6. Which is the most common type of embryo sac development?
7. Name the structure which develops into seed after fertilization.

Section B : (Short Answer Type Questions)

1. What is meant by monosporic development of female gametophyte?
2. Write a short note on bisporic type of embryo sac development?
3. Write short note ovule development?
4. Mention the different types on ovules on the basis of nucellus thickness and number of integuments?
5. What is the mechanism of phototropism shown by seedlings?

Section C : (Long Answer Type Questions)

1. Mention about the structure of an ovule.
2. Describe the process of female gametophyte development.
3. Describe the types of development of embryo sac.
4. Write a note on the structure of a typical embryo sac.

Answer key of Section – A

1. Ovule
2. Archesporium, Megasporocyte, Megaspore, Embryo sac
3. Nucellus
4. Egg, Synergids, Central cell (Pollar nuclei) and Antipodals
5. Endosperm
6. *Polygonum* type
7. Ovule

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Unit-16

Pollination, Pollen- Pistil Interaction and Fertilization

Structure of the Unit:

16.0 Objectives

After studying this unit you will able to understand following concepts about pollination & fertilization:

- Introductory aspects about pollination present in Angiosperms
- Types of pollination and various pollination vectors
- Structure and Function of pistil
- Mechanism of pollen tube entry into ovule and embryo sac
- Double fertilization
- Self- incompatibility in angiosperms
- *in-vitro* fertilization

16.1 Introduction

Pollination is the process by which pollen is placed on the stigma. Pollen may be carried to the flower by wind or by animals, or it may originate within the individual flower itself. When pollen from a flower's anther pollinates the same flower's stigma, the process is called self-pollination. Cross pollination occurs when pollen is transferred to the stigma of a flower on another plant where pollen is often carried by insects and other animals, but sometimes by wind or water. Once the pollen grain reaches the stigma, it produces a pollen tube, which grows down through the style to the ovary. This enables male gametes to

fuse with the female gametes inside the ovule. This is the process known as fertilization. Both male gametes (sperms) fuse with nuclei in the embryo sac. One sperm fertilizes the egg to form the zygote. The other sperm combines with the two polar nuclei to form a triploid nucleus in the central cell. This large cell will give rise to the endosperm, a food-storing tissue of the seed. The union of two sperm cells with different nuclei of the embryo sac is termed **double fertilization**. Including fusion of polar nuclei to form secondary nucleus in embryo sac and two fertilizations the phenomenon is called **triple fusion**.

16.2 Pollination

In angiosperms pollen fall on the stigma as the ovules are place inside the ovary hence is called **indirect pollination**. In gymnosperms ovules are naked so pollen falls directly on them and hence called **direct pollination.** Pollination in angiosperms can be discussed under following sub topics.

16.2.1 Pollination in Early Seed Plants

Early seed plants were pollinated passively, by the action of the wind. As in present day conifers, great quantities of pollen were shed and blown about, occasionally reaching the vicinity of the ovules of the same species. Individual plants of any given species must grow relatively close to one another for such a system to operate efficiently. Otherwise, the chance that any pollen will arrive at the appropriate destination is very small. The vast majority of windblown pollen travels less than 100 meters. This short distance is significant compared with the long distances pollen is routinely carried by certain insects, birds, and other animals

16.2.2 Self-Pollination

All of the modes of pollination that we have considered thus far tend to lead to outcrossing, which is as highly advantageous for plants as it is for eukaryotic organisms generally. Nevertheless, self-pollination also occurs among angiosperms, particularly in temperate regions. Most of the self-pollinating plants have small, relatively inconspicuous flowers that shed pollen directly onto the stigma, sometimes even before the bud opens. You might logically ask why there are many self-pollinated plant species if outcrossing is just as important genetically for plants as it is for animals.

There are two basic reasons for the frequent occurrence of self-pollinated angiosperms:

1. Self-pollination obviously is ecologically advantageous under certain circumstances because self-pollinators do not need to be visited by animals to produce seed. As a result, self-pollinated plants expend less energy in the production of pollinator attractants and can grow in areas where the kinds of insects or other animals that might visit them are absent or very scarce as in the Arctic or at high elevations.
2. In genetic terms, self-pollination produces progenies that are more uniform than those that result from outcrossing. Remember that because meiosis is involved, there is still recombination and the offspring will not be identical to the parent. However, such progenies may contain high proportions of individuals well adapted to particular habitats. Self-pollination in normally out crossing species tends to produce large numbers of ill-adapted individuals because it brings together deleterious recessive genes; but some of these combinations may be highly advantageous in particular habitats. In such habitats, it may be advantageous for the plant to continue self-pollinating indefinitely. This is the main reason many self-pollinating plant species are weeds not only have humans made weed habitats uniform, but they have also spread the weeds all over the world.

Self-pollination is further classified into two types:

1. **Autogamy:** Transfer of pollen grains from anther to stigma of same flower is called autogamy. It is found in bisexual flowers Clistogamous flowers as well as in chasmogamous flowers.
2. **Geitonogamy:** Transfer of pollen grains from anther to stigma between two flowers present on the same plant is called Geitonogamy.

16.2.2.1 Contrivances for Self-Pollination

Although cross-pollination seems to be favoured by Nature, there are cases where self-pollination is ensured.

1. Cleistogamys

In these cases the flowers never open as opposed to most flowers which show chasmogamy (i.e., flowers open normally during anthesis). In cleistogamous flowers the pollens are shed within the closed flowers so that self-pollinaion is obligatory. Cleistogamy is seen in the underground flowers of *Commelina benghalensis* which are small and inconspicuous. This plant also bears normal chasmogamous blue flowers above. Such plants bearing normal as well as

cleistogamous flowers are called chasmocleistogamous. Many rice varieties also are cleistogamous in the sense that the anthers in them shed their pollens and pollination is complete before the flowers open. Cleistogamy or chasmocleistogamy is also seen in *Impatiens balsa- mina* (balsam), *Viola tricolor* (Pansy), *Oxalis, Portulaca,* etc. (Fig. 16.1)

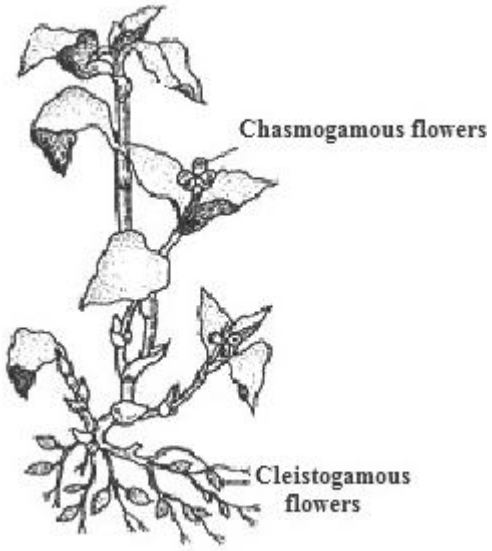


Fig. 16.1 : Commelina sps: Cleistogamous and Chasmogamous flowers

2. Homogamy

Homogamy, as opposed to dichogamy, simply means that the stamens and carpels of a flower mature at the same time. So, there is a greater chance of self- pollination although that is not obligatory. Some homogamous flowers, however, show special mechanisms for self-pollination. Thus, in *Mirabilis jalapa*, when the stamens mature the filaments recoil and bring the anthers near to the stigma so that when they burst self-pollination is achieved. Somewhat similar adaptations are seen in *Argemone mexicana. Grewia subtnaequaus,* etc. (Fig. 16.2)

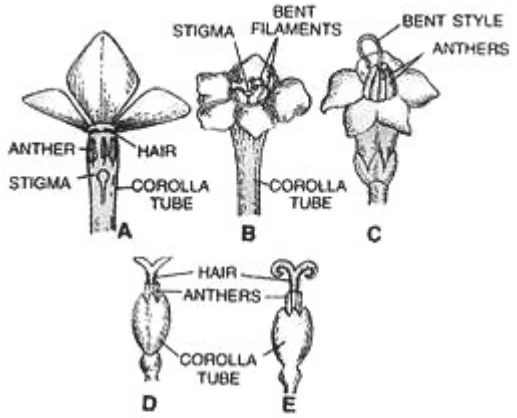


Fig 16.2 Self pollination through Homogamy and Mechanical Devices. A, Style growing to bring stigma in contact with ripe anthers in *Catharanthus* (= *Vinca*). B, Filaments curving over stigma in *Mirabilis jalapa*. C, Curved style bringing stigma in contact with ripe anthers in Potato. D, Normal position of stigma in Sunflower. E, Stigma curling to receive pollen grain present on brushing hair in Sunflower.

3. Dichogamous Flowers Showing Adaptations for Self-pollination:

(a) Flowers of *Ixora, Gardenia, Vinca*, etc., have their anthers placed at the mouth of the corolla tube. As the stigma elongates from below, it pushes out through these anthers which are already ripe and are pollinated in so doing.

(b) Compositae flowers are protandrous. In sunflowers the bilobed stigma, is still young and hidden within the syngenesious tube of anthers when the latter ripen and shed the pollens within the tube. At this stage self-pollination is not possible as the receptive surfaces of the stigmas are not only immature but also hidden. In the second stage the bifid stigma grows through the anther tube pushing out pollens and opens out above. Cross-pollination usually takes place at this stage through the agency of insects. But, if cross-pollination fails, it is seen that the stigma lobes curl back so that the receptive surfaces brush against any pollen still sticking on its surface and are thereby self-pollinated. It is seen in actual practice that a large number of agricultural crops are naturally self-pollinated. Among these are rice, wheat, barley, oats, potato, peas, beans, tobacco, linseed, tomato and *Corchorus capsularis*.

16.2.3 Cross-Pollination (Allogamy)

Transfer of pollen grains from the anthers of one flower to the stigma of another flower of the same species is called cross pollination or allogamy. It occurs in chasmogamous flowers. Followings are vectors helpful in cross pollination:-

16.2.3.1 Wind-Pollinated Angiosperms (Anemophily)

Many angiosperms, representing a number of different groups, are wind-pollinated a characteristic of early seed plants. Among them are such familiar plants as oaks, birches, cottonwoods, grasses, sedges, and nettles. The flowers of these plants are small, greenish, and odorless; their corollas are reduced or absent. Such flowers often are grouped together in fairly large numbers and may hang down in tassels that wave about in the wind and shed pollen freely. Many wind pollinated plants have stamen- and carpel-containing flowers separated among individuals or on a single individual. If the pollen-producing and ovule-bearing flowers are separated, it is certain that pollen released to the wind will reach a flower other than the one that sheds it, a strategy that greatly promotes outcrossing. Some wind-pollinated plants, especially trees and shrubs, flower in the spring, before the development of their leaves can interfere with the wind-borne pollen. Wind-pollinated species do not depend on the presence of a pollinator for species survival. (Fig. 16.3)

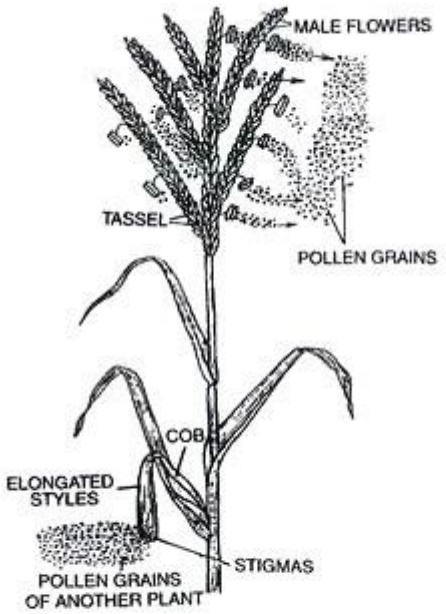


Fig. 16.3 : Anemophily in *Zea mays*

16.2.3.2 Pollination by Animals (Zoophily)

The spreading of pollen from plant to plant by pollinators visiting flowers of specific angiosperm species has played an important role in the evolutionary success of the group. It now seems clear that the earliest angiosperms, and perhaps their ancestors also, were insect-pollinated, and the coevolution of insects and plants has been important for both groups for over 100 million years. Such interactions have also been important in bringing about increased floral specialization. As flowers become increasingly specialized, so do their relationships with particular groups of insects and other animals.

Pollination by Insects (Entomophily): Bees (Melittophily)

Among insect-pollinated angiosperms, the most numerous groups are those pollinated by bees. Like most insects, bees initially locate sources of food by odor, and then orient themselves on the flower or group of flowers by its shape, color, and texture. Flowers that bees characteristically visit are often blue or yellow. Many have stripes or lines of dots that indicate the location of the nectaries, which often occur within the throats of specialized flowers. Some bees collect nectar, which is used as a source of food for adult bees and occasionally for larvae. Most of the approximately 20,000 species of bees visit flowers to obtain pollen. Pollen is used to provide food in cells where bee larvae complete their development. Only a few hundred species of bees are social or semi-social in their nesting habits. These bees live in colonies, as do the familiar honeybee, *Apis mellifera,* and the bumblebee, *Bombus*. Such bees produce several generations a year and must shift their attention to different kinds of flowers as the season progresses. To maintain large colonies, they also must use more than one kind of flower as a food source at any given time. Except for these social and semi-social bees and about 1000 species that are parasitic in the nests of other bees, the great majority of bees at least 18,000 species are solitary. Solitary bees in temperate regions characteristically have only a single generation in the course of a year. Often they are active as adults for as little as a few weeks a year. Solitary bees often use the flowers of a given group of plants almost exclusively as sources of their larval food. The highly constant relationships of such bees with those flowers may lead to modifications, over time, in both the flowers and the bees. For example, the time of day when the flowers open may correlate with the time when the bees appear; the mouthparts of the bees may become elongated in relation to tubular

flowers; or the bees' pollen-collecting apparatuses may be adapted to the pollen of the plants that they normally visit. When such relationships are established, they provide both an efficient mechanism of pollination for the flowers and a constant source of food for the bees that "specialize" on them. (Fig. 16.4)

Pollination by Insects (Entomophily): Insects other than Bees

Among flower-visiting insects other than bees, a few groups are especially prominent. Flowers such as phlox, which are visited regularly by butterflies, often have flat "landing platforms" on which butterflies perch. They also tend to have long, slender floral tubes filled with nectar that is accessible to the long, coiled proboscis characteristic of Lepidoptera, the order of insects that includes butterflies and moths. Flowers like jimsonweed, evening primrose, and others visited regularly by moths are often white, yellow, or some other pale color; they also tend to be heavily scented, thus serving to make the flowers easy to locate at night.

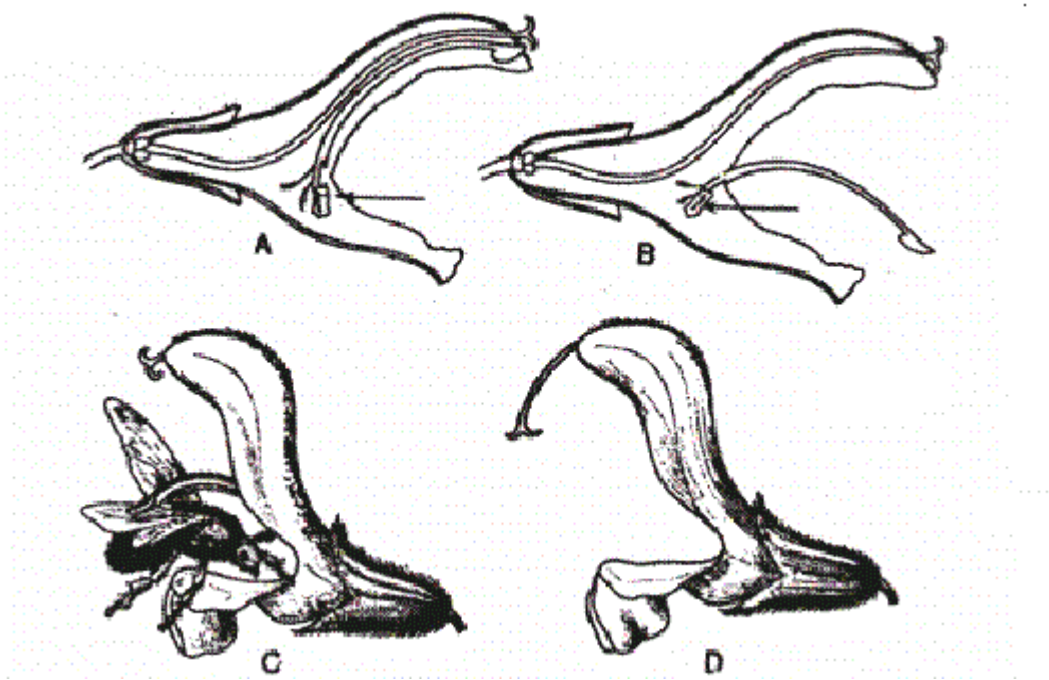


Fig. 16.4 : Pollination in *Salvia* A & B L.S. of flower showing immature pistil and movement of stamen, C A Bee becoming dusted with pollen D A flower with mature stigma

Pollination by Birds (Ornithophily)

Several interesting groups of plants are regularly visited and pollinated by birds, especially the hummingbirds of North and South America and the sunbirds of Africa. Such plants must produce large amounts of nectar because if the birds do not find enough food to maintain themselves, they will not continue to visit flowers of that plant. Flowers producing large amounts of nectar have no advantage in being visited by insects because an insect could obtain its energy requirements at a single flower and would not cross-pollinate the flower. How are these different selective forces balanced in flowers that are "specialized" for hummingbirds and sunbirds? Ultraviolet light is highly visible to insects. Carotenoids, yellow or orange pigments frequently found in plants, are responsible for the colors of many flowers, such as sunflowers and mustard. Carotenoids reflect both in the yellow range and in the ultraviolet range, the mixture resulting in a distinctive color called "bee's purple." Such yellow flowers may also be marked in distinctive ways normally invisible to us, but highly visible to bees and other insects. These markings can be in the form of a bull's-eye or a landing strip. Red does not stand out as a distinct color to most insects, but it is a very conspicuous color to birds. To most insects, the red upper leaves of poinsettias look just like the other leaves of the plant. Consequently, even though the flowers produce abundant supplies of nectar and attract hummingbirds, insects tend to bypass them. Thus, the red color both signals to birds the presence of abundant nectar and makes that nectar as inconspicuous as possible to insects. Red is also seen again in fruits that are dispersed by birds. (Fig: 16.5)

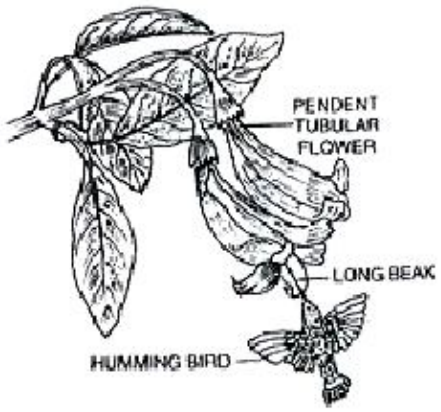


Fig. 16.5: Humming bird collecting nector from *Bigonia capteolata* flowers thereby pollinating them

Other Animals

Other animals including bats **(Cheiropteriphily),** snails **(Malacophily)** and small rodents may aid in pollination. The signals here also are species specific. These animals also assist in dispersing the seeds and fruits that result from pollination. Monkeys are attracted to orange and yellow and will be effective in dispersing those fruits.

16.2.2.3 Contrivances for Cross-Pollination

It seems that nature favours cross-pollination as opposed to self-pollination. It is a study of this partiality on the part of Nature that so greatly impressed Darwin. All unisexual flowers and a large majority of bisexual flowers are naturally cross-pollinated. Special contrivances ensuring cross-pollination as noted below are very conspicuous:

1. Dicliny:

Cross-pollination is the rule among diclinous plants, i.e., those bearing unisexual flowers. In dioecious plants nothing else can take place. In monoecious plants the only alternative is geitonogamy which, however, has the same effect as self-pollination.

2. Self-sterility:

This is the condition when a flower cannot be fertilized by the pollen of the same flower or, sometimes, from a flower of the same strain of plants. In some orchids, flowers wither away if pollinated by its own pollen. Many species of Solanum (potato, tobacco, etc.) and the tea plant are self-sterile because of genetic reasons. Cross-pollination is obligatory in such plants.

3. Dichogamy:

When stamens and carpels of a bisexual flower mature at different times, pollination between them becomes ineffective. Sometimes, however, it is found that self-pollination may take place at a later stage if cross-pollination fails. Dichogamy may be of two types:

(a) Protandry:

The anthers ripen first as in most Compositae, many Umbelliferae, Malvaceae, etc. As a result, when the anther bursts, it pollinates stigmas of other flowers but not its own stigma which is not yet ripe.

(b) Protogyny:

The carpel matures first as in many members of Annonaceae (e.g., *Annona*, *Polyalthia*) and Magnoliaceae (e.g., *Magnolia, Michelia*) as well as in Arum maculatum. When the stigma is receptive, its own pollen is riot ripe so that it has to depend on foreign pollens.

4. Herkogamy:

In some flowers there may be some physical barrier between the anther and the style so that pollination between them is rendered difficult or even impossible. In many Cruciferae and Caryophyllaceae, the stigma extends far beyond the stamens so that pollens from the latter are not likely to reach the former. The extrorse anthers of *Gloriosa* dehisce the anthers out of reach of its own stigmas. In *Calotropis* and orchids, where the pollens are aggregated in pollinia, the pollination (described later) is entirely at the mercy of insects.

5. Heteromorphism:

In certain plants there are flowers of two (dimorphic) or three (trimorphic) different forms with anthers and stigmas at different levels. This dimorphism or trimorphism usually involves beterostyly (styles of different lengths) and heteroanthy (i.e., different types of anthers). The primrose ` of Primulaceae) shows an interesting case of dimorphism. In the first form, the anthers are placed deep down in the corolla lube and the stigma lies at the entrance. The pollens of this type are smaller and the stigma papillae larger. In the second type the anthers arc placed at the entrance while the stigma is deep down. Moreover, in this case the pollens are larger and the stigma papillae smaller. Different types of insects moving about these flowers will naturally touch floral organs at the same level because of the difference in the lengths of their organs (proboscis, legs, etc.), so that the short style will be cross-pollinated by pollens from low anthers and vice versa. This will involve pollens and stigmas of similar luxuriance and growth. Such dimorphism is also shown by jasmine and other sweet scented flowers of Oleaceae, by *Linum,* by *Fagopyrum* (buckwheat) of Polygonaceae, etc. Some species of *Oxalis, Linum* and *Lythrum* (Lythraceae) show trimorphism. The three types of flowers show three positions of anthers and stigmas so that there is cross-pollination involving three heights. According to Tischler, dimorphic or trimorphic conditions may be altered by changed nutrition.

16.2.3.4 Pollination by Human (Artificial pollination)

Mechanical pollination or Human pollination (Antrophily) or artificial pollination is a technique used when natural, or open pollination is insufficient or undesirable. For example in Cucurbitaceae poor pollination and poor maturation is exhibited. Anthrophily is better option in these cases used by small market gardeners. In this simple action method pollens are taken on an artist's brush, cotton swab or on forceps from male flower and transferred on stigma of female flowers of same species. Sometimes the corolla is removed from male flowers and the flower itself is brushed against the stigmas of female flowers.

16.3 Structure and Function of Pistil

16.3.1 Pollen Pistil Interactions

A developmental analysis of pollination responses implicates pollen as well as stigma maturation factors in the acquisition of reproductive function. In the pistil, competence to support pollen germination and tube growth extended over a broad developmental window, and abundant as well as efficient pollen tube development has been observed on pistils at anthesis. In contrast, pollen tube growth on immature pistils found to proceed at low efficiency, at reduced growth rates, and with lack of directionality. Based on the pattern of pollen tube growth at different stages of pistil maturation, temporally regulated signals emanating from specialized cells of the pistil are inferred to be operative in each of the four identified phases of pollen tube growth. In the stigma and the stylar transmitting tissue, these signals directed the path of intra-specific pollen tubes as well as pollen tubes. By contrast, in the ovary, signaling by the ovule was effective only on intra-specific pollen tubes and was thus identified as the basis of inter-specific incompatibility. Furthermore, the acquisition of reproductive function found to involve, in addition to the induction of a variety of stimulatory signals, a heretofore unrecognized developmental restriction in the capacity of epidermal surfaces of the flower to support pollen tube growth.

16.3.2 Pollen Germination

Exact information on the time taken by pollen to germinate on the stigma is available for only a few plants, but the following examples will illustrate the range that has been observed: 2 days in *Garrya elliptica* (Hallock, 1930), 3 hours in *Reseda spp*. (Eigsti, 1937) ; 2 hours in *Beta vulgaris* (Artschwager and Starrett, 1933); and 5 minutes in *Taraxacum* (Poddubnaja- Arnoldi and

Dianowa, 1934), *Zea mays* (Randolph, 1936), and Hordeum distichon (Pope, 1937). In *Saccharum oflicinarum (Artschwager et al, 1929) and Sorghum vulgare* (Artschwager and McGuire, 1949) germination takes place almost immediately.

The first step in germination is the expansion of the pollen grain by the absorption of liquid from the moist surface of the stigma and the protrusion of the intine through a germ pore. The small tubular structure which arises in this way then continues to elongate, making its way down the tissues of the stigma and style. Only the distal part of the tube has living cytoplasm, and as the nuclei pass forward callose plugs are left in the empty portions behind them. Most pollen grains are monosiphonous, i.e., only a single pollen tube emerges from each pollen grain; others, like those of the Malvaceae, Cucurbitaceae and Campanulaceae, are polysiphonous. In *Althaea rosea* 10 tubes, and in *Malva neglecta* even 14 tubes, areknown to come out from the same pollen grain (Stenar, 1925). Eventually, however, only one of them makes further progress. Sometimes the same pollen tube may divide into one or more branches. Such a condition seems to be frequent in the Amentiferae, where the branching tubes give the appearance of a ramifying fungous mycelium (see Finn, 19286). In plants whose pollen grains are united into tetrads or into pollinia, several pollen tubes are produced at the same time. The stigma is believed to play an important part in the germination of pollen, but in many plants germination can also be induced in a sugar solution of appropriate strength. Martin (1913) germinated the pollen of *Trifolium pratense* on hog's bladder moistened with distilled water and suggested that the only use of the stigma lies in controlling the water supply. Katz (1926) agreed with this view and said that the chief function of the stigmatic secretion is to protect the pollen as well as the stigma from desiccation. In her experiments the pollen germinated even on the cut surface of the style, provided the stigmatic secretion was applied to the stump and the latter was kept moist for some time.

Pollen grains may also germinate on other parts of the flower besides the stigma. In cleistogamous flowers (Frisendahl, 1927; Madge, 1929; West, 1930; Maheshwari and Singh, 1934) germination takes place within the anther loculi, and in *Aeginetia indica* (Juliano, 1935) even on the moist surface of the corolla tube. Frequently pollen grains germinate on a foreign stigma, i.e., stigma of a different species (see Eigsti, 1937; Sanz, 1945). If fertilization takes place, it results in the formation of interspecific and intergeneric hybrids.

16.3.3 Course of Pollen Tube

After the tube has emerged from the pollen grain, it makes its way between the stigmatic papillae into the tissues of the style. The latter is extremely variable in length. In some plants it is so short that the stigma is described as sessile, while in *Zea mays* the so-called "silk" may attain a length of 50 cm. Depending on the presence or absence of the transmitting tissue and on the extent of its development, styles have been classified into three main types called open, half-closed, and closed (Hanf, 1935). In the first type there is a wide stylar canal and the inner epidermis itself assumes the function of the nutrition and conduction of the pollen tube, as in the Papaveraceae, Aristolochiaceae, Ericaceae, and many monocotyledons. In the second type the canal is surrounded by a rudimentary transmitting tissue of two or three layers of glandular cells, as in several members of the Cactaceae. In the third or closed type, illustrated by *Datura* and *Gossypium*, there is no open channel but instead a solid core of elongated and richly protoplasmic cells through which the pollen tube grows downward in order to reach the ovary. Finally, there are other plants like Salix, Acacia, and many grasses in which the styles are solid but are not provided with any specialized transmitting tissue. In open styles the pollen tube grows on the surface of the cells lining the stylar canal (often in the mucilage secreted by them); and in solid styles through the intercellular spaces between the cells of the transmitting tissue, enlarging the spaces by the hydrostatic pressure of its contents and secreting some enzymatic substances which bring about a dissolution of the middle lamellae. Only rarely does the pollen tube pass through the cells themselves.

16.3.4 Entry of Pollen Tube into Megasporangium (Ovule)

After arriving at the top of the ovary, the tube may enter the ovule either through the micropyle or by some other route. The former is the usual condition and is known as **Porogamy,** but evenin plants ordinarily classed as porogamous there are several modifications. To mention a few examples, in *Acacia* (Newman, 1934) the integuments are still below the apex of the nucellus at the time of fertilization so that a micropyle does not exist at this stage and in *Philadelphus, Utricularia, Vandellia*, and *Torenia* the embryo sacprotrudes out of the micropyle so that the pollen tube comes in direct contact with it. In several members of the Loranthaceae there is no integument and therefore nothing that can be called a micropyle. Here the embryo sacs undergo a remarkable elongation and meet the pollen tubes at some point in the stylar region.

In some plants the pollen tube enters the ovule through the chalaza. This condition, known as **Chalazogamy,** was first reported in *Casuarina* (Treub, 1891) and soon afterwards in several members of the Amentiferae. Nevertheless, it is not confined to them, being also known in *Rhus* (Grimm, 1912), *Circaeaster* (Junell, 1931), and a few other genera. Recent studies have, however, shown that even in such cases, where entry into the ovule is affected through the chalaza, the tube usually continues its growth over the surface of the embryo sac and penetrates it only after arriving near the egg apparatus. As examples *Ostrya carpinifolia* (Finn, 1936), *Juglans regia* (Nast, 1941), and *Casuarina equisetifolia* (Swamy, 1948) may be mentioned.

In *Alchemilla* (Murbeck, 1901), *Cucurbita* (Longo, 1901; Kirkwood, 1906), and *Circaeaster* (Junell, 1931) the pollen tube enters through the funiculus or the integument. This is known as **Mesogamy.**

Formerly considerable phylogenetic significance was attached to the route taken by the pollen tube during its entry into the ovule, but now this point is considered to be of physiological rather than phylogenetic importance, for we sometimes find considerable variation in this respect even in one and the same species. In *Brassica oleracea* (Thompson, 1933) the tube normally enters through the micropyle, but sometimes it may do so by way of the chalaza. In *Ulmus,* Shattuck (1905) speaks of its branching and apparently aimless wandering through the funiculus, the integuments, and occasionally the nucellus. In *Boerhaavia* (Maheshwari, 1929), although the tube actually enters through the micropyle, it first makes a horizontal crossing through the funiculus. In *Gossypium* (Gore, 1932) it often passes from the funiculus to the base of the ovule and then travels up along the wall of the latter to enter the micropyle.

An organ of special significance in facilitating the entry of the pollen tube into the ovule is the so called **Obturator**, to which reference had already been made by Hofmeister in the year 1849. Usually it is a swelling of the placenta which grows towards the micropyle and fits like a hood or canopy over the nucellus, serving as a sort of bridge for the pollen tube. Often the cells of the obturator may be greatly elongated or may have a glandular appearance. Some other structures having a different origin but serving the same function may also be included under the general term obturator. In the Thymelaeaceae (Fuchs, 1938) the cells belonging to the base of the stylar canal elongate and grow down as hairyprocesses approaching the nucellus (Fig. 111). In Pilea (Fagerlind, 1944) a tuft of papillate cells extends from the base of the style to the apex of the ovule,

coming in intimate contact with the latter. In *Myriocarpa* and *Leucosyke* (Fagerlind, 1944), on the other hand, it is the cells of the inner integument which elongate upward and penetrate into the stylar canal (Fig. 110A, B), forming what may be called an integumentary obturator. Usually there are no special modifications in the cells lining the micropylar canal, but sometimes, as in *Berkheya* (Gelin, 1936), *Grevillea* (Brough, 1933), and *Cynomorium* (Steindl, 1945), they become mucilaginous or glandular and seem to contribute to the nutrition of the pollen tube. In Cardiospermum (Kadry, 1946) not only the cells belonging to the inner integument but also those forming the apical portion of the nucellus give rise to a mucilaginous mass which facilitates the entry of the pollen tube. In plants with a many-layered nucellar tissue, like *Beta* (Artschwager and Starrett, 1933), those of its cells which are in continuity with the micropyle become elongated and richly protoplasmic and give an impression as though they were designed to lead the pollen tube through the path of least resistance. It is of interest to note that even during its passage through the nucellus the pollen tube usually makes its way between the cells and not through them. Normally it causes but little disturbance in their position and they soon return to their original shape, but in a few families like the Lythraceae, Sonneratiaceae, Onagraceae, and Cucurbitaceae the tubes are so broad that they destroy the cells which lie in their way and cause a permanent break in the tissues.

16.3.5 Entry of Pollen Tube into Embryo Sac

After penetrating the wall of the embryo sac, the pollen tube may either pass between the egg and one synergid as in *Fagopyrum* (Mahony, 1935), or between the embryo sac wall and a synergid as in *Cardiospermum* (Kadry, 1946), or directly into a synergid as in *Oxalis* (Krupko, 1944), *Elodea* (Ernst-Schwarzenbach, 1945), and *Daphne* (Venkateswarlu, 1947). In *Viola* it not only enters a synergid but is said to force its way through the base of the latter (Madge, 1929). As a rule only one synergid is destroyed by the impact of the pollen tube and the other remains intact until some time afterward, but in *Mimusops*, *Achras*, and *Bassia* (Murthy, 1941) both are destroyed and in *Phryma* (Cooper, 1941) and *Tropaeolum* (Walker, 1947) neither of them seems to be affected. In some genera, such as *Tacca*, *Wormia* (*Paetow*, 1931), and *Nelumbo* (Ohga, 1937), the synergids degenerate even before the entry of the pollen tube, and in others like *Plumbago*, *Vogelia*, and *Plumbagella* (see Maheshwari, 1948) they are not formed at all. This seems to indicate that they are not essential for fertilization, and the view that they secrete substances

which exercise a chemotactic influence over the pollen tube, or that they act as shock absorbers against its impact, does not rest on a sound basis (see also Dahlgren, 1938). In *Zauschneria latifolia* (Johansen, 19316) pollen tubes were found to enter even those ovules whose embryo sacs had degenerated and virtually disappeared. Detailed information regarding the exact manner of discharge of the male gametes is lacking. In *Crepis capillaris* (Gerassimova, 1933) and *Taraxacum koksaghys* (Warmke, 1943) the tip of the pollen tube becomes "wedged in" between the egg and the polar fusion nucleus, so that both the male gametes are discharged in close proximity to their mates. Fagerlind (1939) noted some embryo sacs of *Peperomia* in which the tip of the tube had divided into two short branches, one of which was directed toward the egg. Cooper (1940, 1941, 1946) refers to a similar bifurcation of the tip of the pollen tube in *Portulaca, Phryma*, and *Petunia* (Fig. 112), one branch becoming closely appressed to the egg and the other extending in the direction of the polar nuclei, and suggests that the two male gametes reach their destinations by way of these separatebranches. In *Coffea arabica* (Mendes, 1941) the pollen tube does not bifurcate but shows two subterminal openings through which the two male gametes are discharged into the cavity of the embryo sac.

16.4 Double Fertilization

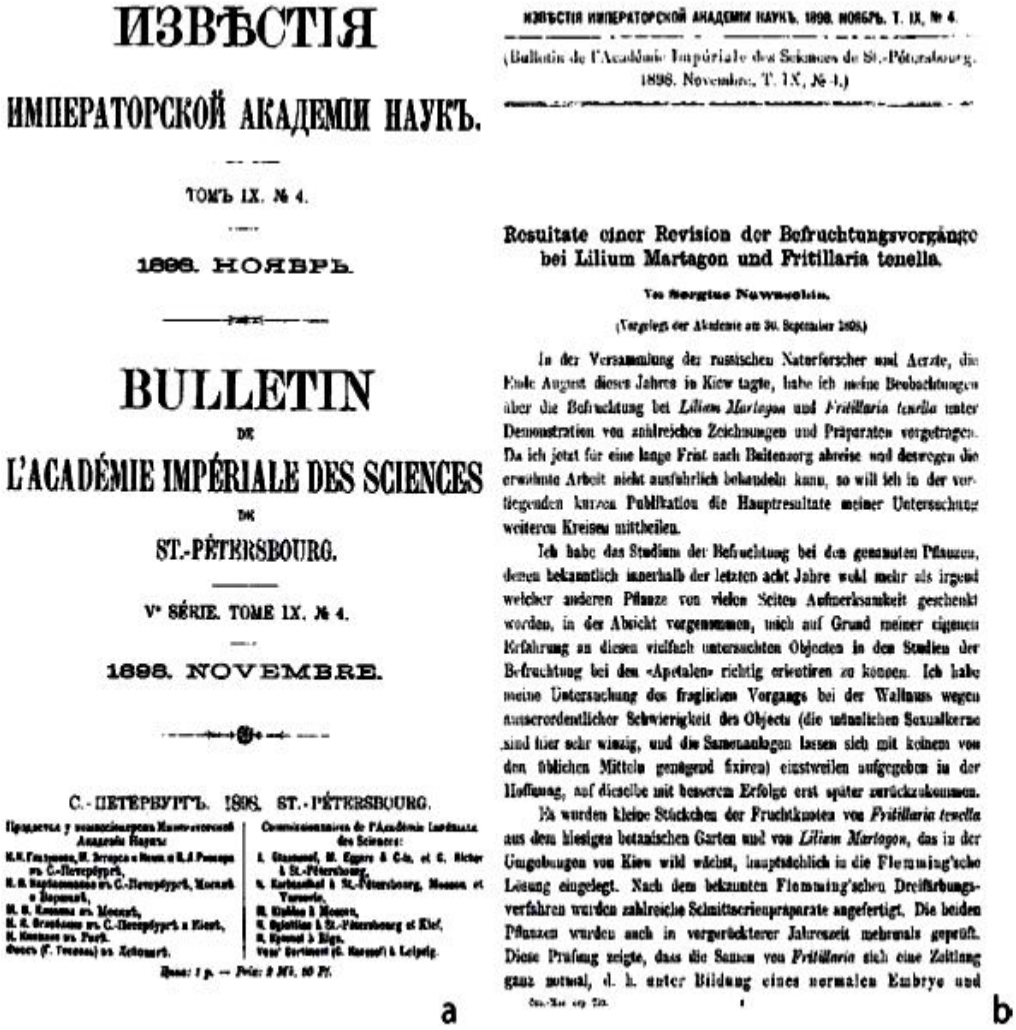
16.4.1 Discovery of Double Fertilization

Unambiguous proof of the actual fusion of the male and female gametes embodied in fertilization in flowering plants can be traced to a monographic publication of Strasburger (1884). This work was devoted mostly to the nuclear cytology of pollen grains and pollen tubes of plants belonging to a wide range of families, and to the fate of male gametes delivered by pollen tubes in the embryo sacs of *Gloxinia hybrida* (Gesneriaceae), *Himantoglossum hircinum*, *Orchis latifolia* (Orchidaceae), and *Monotropa hypopitys* (Pyrolaceae). The most complete, illustrated details were provided on *M. hypopitys*, in which it was shown that one of the two male gametes conveyed by the pollen tube fused with the nucleus of the egg. At that time the male gametes were known as generative nuclei and it was uncertain whether these gametes were true cells or naked nuclei. However, the observation that a male gamete fused with the egg in the act of fertilization was contrary to a previous puzzling finding that this event was orchestrated by the diffusion of the cytoplasmic contents of the pollen tube (see Maheshwari 1950). Although Strasburger's work identified the embryo as the resulting product of fertilization, understanding of the fate of the

second male gamete discharged by the pollen tube, and the source of origin of the endosperm (albumen), remained major hurdles in gaining a complete insight into the dynamics of fertilization in angiosperms.

The breakthrough in the discovery of double fertilization occurred when **S. Nawaschin** in Russia showed that, in ovules of *Lilium martagon* and *Fritillaria tenella* (Liliaceae), both male gametes from the pollen tube penetrated the embryo sac; whereas one of them fused with the nucleus of the egg cell, the other fused with the polar fusion nucleus (at that time known as the definitive nucleus) floating in the central cell, initiating a second fertilization event (Nawaschin 1898, 1899). The results of this work were presented orally on 24 August 1898 to the botanical section of the "Naturforscherversammlung" (Fig. 16.6 a & b) held in Kiew, Russia (20–30 August 1898) and published as an abstract in the following year (Nawaschin 1899); the full paper appeared a few months after the meeting (Nawaschin 1898). Thus, reverent credit is due to Nawaschin for this legendary discovery of the two fusion events during fertilization in flowering plants. The phenomenon observed by Nawaschin was also independently confirmed in *L. martagon* and *Lilium pyrenaicum* by L. Guignard (1899) in France. The account of this investigation was communicated to the Academy of Sciences in Paris on 4 April 1899 and was published soon afterwards in its report. Exactly the same paper, with a footnoted reference to the earlier paper with volume number and a middle page number, was also published in another journal in the same year (Guignard 1899b). The work described in these two papers, which included a reference to Nawaschin's 1899 abstract, was accompanied by a series of illustrations in the form of line drawings showing the two fusion events. Guignard's description and figures portrayed a precise two-step sequence of events involving the fusion of the second sperm with the upper polar nucleus, followed by integration of this fusion product into the lower polar nucleus. Within a few months of the publication of Guignard's papers, full confirmation of the startling discovery of fusion of the second sperm with the polar fusion nucleus came from a reexamination of previously prepared slides of fertilized ovules of L. martagon by E. Sargant in England (Sargant 1899). The coincident choice of ovules of species of *Lilium* and *Fritillaria* by investigators working in three European countries as the classic experimental system in these pioneering studies is not surprising because of the relatively large size of the embryo sac and its equally conspicuous nuclei as seen in microscopic preparations of ovules of these two genera. Indeed, because of this and other advantages, slides

demonstrating embryo sac development in various species of *Lilium* and *Fritillaria* have been popular in the teaching of general plant biology; species of these genera have also been favored systems of subsequent investigators because embryo sac development in them appeared to be a simplified version of a complex series of nuclear fusions and divisions that did not have parallels in other plants studied (Maheshwari 1950). To designate the two fertilization events those occur at the inception of the sporophytic phase in flowering plants, Guignard (1899a, 1899b), in a seemingly visionary act, used the term 'double copulation' in the title of the first two papers and 'Double Fecondation' in later publications. Strasburger (1900)



ИЗВѢСТІЯ

ИМПЕРАТОРСКОЙ АКАДЕМІИ НАУКЪ.

ТОМЪ IX. № 4.

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ИЗВѢСТІЯ ИМПЕРАТОРСКОЙ АКАДЕМІИ НАУКЪ. 1898. НОЯБРЬ. Т. IX, № 4.

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Resultate einer Revision der Befruchtungsvorgänge bei Lilium Martagon und Fritillaria tenella.

Von Sergius Nawaschin.

(Vorgelegt der Akademie am 30. September 1898.)

In der Versammlung der russischen Naturforscher und Aerzte, die Ende August dieses Jahres in Kiew tagte, habe ich meine Beobachtungen über die Befruchtung bei *Lilium Martagon* und *Fritillaria tenella* unter Demonstration von zahlreichen Zeichnungen und Präparaten vorgetragen. Da ich jetzt für eine lange Frist nach Baitenzorg abreise und deswegen die erwähnte Arbeit nicht ausführlich behandeln kann, so will ich in der vorliegenden kurzen Publikation die Hauptresultate meiner Untersuchung weiteren Kreisen mittheilen.

Ich habe das Studium der Befruchtung bei den genannten Pflanzen, denen bekanntlich innerhalb der letzten acht Jahre wohl mehr als irgend welcher anderen Pflanze von vielen Seiten Aufmerksamkeit geschenkt worden, in der Absicht vorgenommen, mich auf Grund meiner eigenen Erfahrung an diesen vielfach untersuchten Objecten in den Stadien der Befruchtung bei den «Apetalen» richtig orientiren zu können. Ich habe meine Untersuchung des fraglichen Vorgangs bei der Wallnuss wegen ausserordentlicher Schwierigkeit des Objects (die winzlichen Sexualkerne sind hier sehr winzig, und die Samenanlagen lassen sich mit keinem von den üblichen Mitteln genügend fixiren) einstweilen aufgegeben in der Hoffnung, auf dieselbe mit besserem Erfolge erst später zurückzukommen.

Es wurden kleine Stückchen der Fruchtknoten von *Fritillaria tenella* aus dem hiesigen botanischen Garten und von *Lilium Martagon*, das in der Umgebungen von Kiew wild wächst, hauptsächlich in die Flemming'sche Lösung eingelegt. Nach dem bekannten Flemming'schen Dreifärbungsverfahren wurden zahlreiche Schnittserienpräparate angefertigt. Die beiden Pflanzen wurden auch in vorgerückterer Jahreszeit mehrmals geprüft. Diese Prüfung zeigte, dass die Samen von *Fritillaria* sich eine Zeitlang ganz normal, d. h. unter Bildung eines normalen Embryo und

b

Fig: 16.6 a & b Discovery of double fertilization. a Cover page of the journal in which Nawaschin's discovery of double fertili-zation was first published. b First page of the article describing double fertilization

referred to the two fertilization events as 'doppelten Befruchtung' in the title of a paper, and nearly the same term 'die doppelte Befruchtung' appeared in the text of two papers by Nawaschin (1900). The term '**Double fertilization**' now

in universal use was first employed in the title of a paper by Thomas (1900) and in the text of a paper by Sargant (1900). Putting to rest the prevalent assumption that the endosperm was generated by fusion of the two polar nuclei, the above mentioned investigators also concluded correctly that the product of fusion of the second sperm with the polar fusion nucleus gives rise to the endosperm, typically constituted of cells with chromosomes of biparental origin from the coalescence of three nuclei. The discovery of double fertilization in the liliaceous species, and the confirmation of its occurrence in many other angiosperms, including both monocotyledons and dicotyledons, within a period of just over a year for example, additional species within the **Liliaceae** such as *Fritillaria meleagris, Scilla bifolia, Lilium candidum, Tulipa celsiana, Tulipa gesneriana,* and *Tulipa sylvestris* ; *Himantoglossum hircinum, Orchis latifolia, Orchis maculata,* and *Orchis mascula* of the **Orchidaceae;** *Erigeron philadelphicus, E. strigosa, Guizotia oleiflora, Helianthus annuus, Heliopsis patula, Rudbeckia grandiflora, Rudbeckia laciniata, Rudbeckia speciosa, Silphium integrifolium, Silphium laciniatum, S. terebinthinaceum,* and *Spilanthes oleracea* of the **Asteraceae** ; *Hibiscus trionum* of the **Malvaceae**; *Anemone nemorosa, Caltha palustris, Clematis viticella, Delphinium elatum, Helleborus foetidus, Nigella sativa,* and *Ranunculus flammula* of the **Ranunculaceae;** *Reseda lutea* of the **Resedaceae**; *Juglans sp*. of the **Juglandaceae**, and *Monotropa hypopitys* of the **Pyrolaceae.**

16.4.2 Universality of Double Fertilization in Flowering Plants

The momentum created in the waning years of the nineteenth century to establish double fertilization as a ubiquitous feature in the reproductive biology of flowering plants was followed by a sustained effort in the twentieth century leading to the discovery of this phenomenon in additional members of the Ranunculaceae, Liliaceae, Juglandaceae, and Pyrolaceae (Shibata 1902), as well as in plants belonging to Poaceae, Najadaceae, Solanaceae, Gentianaceae (Guignard 1901), Asclepiadaceae (Frye 1902), Brassicaceae (Guignard 1902), and Ceratophyllaceae (Strasburger 1902). Guérin (1904), in a monograph devoted entirely to the topic of fertilization in seed-bearing plants, and Coulter and Chamberlain (1912) in their classic book on the "Morphology of Angiosperms", refer to 16 families of angiosperms, encompassing about 40 genera and over 60 species definitely known to have a second fertilization event; these two publications surveyed the literature up to the end of 1902. From that time onwards, along with the presence of a reduced female gametophyte and embryo-nourishing endosperm, the occurrence of double

fertilization was accepted as a general feature of the reproductive biology of angiosperms. Indeed, under this assumption, there were only occasional references to double fertilization in the numerous publications dating from the early 1900s to the present dealing with the variability and diversity of reproductive processes in flowering plants with special reference to their embryogenesis and endosperm development. However, this period was notable for providing the first glimpses of electron microscopic details of double fertilization in several plants, including cotton (*Gossypium hirsutum*; Malvaceae; Jensen and Fisher 1967), maize (*Zea mays;* Poaceae; Diboll 1968; van Lammeren 1986), barley (*Hordeum vulgare*; Poaceae; Cass and Jensen 1970; Mogensen 1982, 1988), spinach (*Spinacia oleracea*; Chenopodiaceae; Wilms 1981) and *Plumbago zeylanica* (Plumbaginaceae; Russell 1982, 1983).

16.4.3 Gametic Fusion

After the pollen tube has discharged its contents into the embryo sac, one male gamete fuses with the egg (syngamy) and the other with the two polar nuclei (triple fusion). Because of the technical difficulties encountered in studying the process, very few detailed accounts of it have appeared up to this time. The time between the beginning and the end of the gametic fusions is so short that one rarely succeeds in "catching" the material at the right stage. There is also an element of chance in obtaining proper median sections; for the embryo sac is usually large enough at this stage to run into several sections, and thick sections do not stain satisfactorily. Besides, even if the material has been properly selected and the desired stages are actually at hand, detailed observations may still prove difficult for the following reasons:

(1) The pollen tube discharges a deeply staining material into the embryo sac which surrounds the egg and decreases visibility;

(2) One or both of the synergids disorganize at this time and their contents become converted into a tenacious mucus-like material which stains densely; and

(3) The vegetative nucleus (or its fragments) and the nuclei of the synergids "wander" into the upper part of the embryo sac and are liable to be confused with the male gametes.

In view of these difficulties it is not surprising that our knowledge of the events concerned with fertilization has not advanced very far beyond where it stood during the early part of this century. Several workers have confessed with a feeling of disappointment that, in spite of repeated efforts and the study of

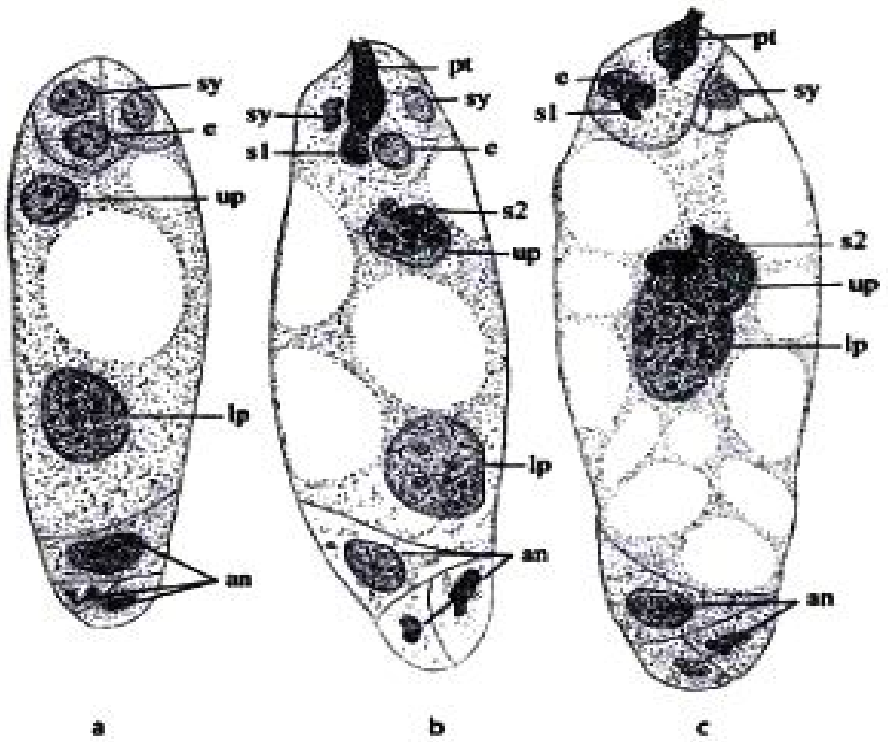

Fig: 16.7 a–c Double fertilization in *Lilium martagon*. **a** Mature embryo sac showing the egg apparatus, consisting of the egg and synergids, antipodals, upper polar nucleus, and lower polar nucleus. **b** Mature embryo sac after discharge of male gametes from the pollen tube. The nucleus of one sperm has entered the egg and that of the second sperm is in contact with the upper polar nucleus. The nucleus of one of the synergids is disintegrating. **c** Union of one sperm with the egg nucleus and of the second sperm with the two polar nuclei. *an* Antipodals, *e* egg cell, *lp* lower polar nucleus, *pt* pollen tube, *s1* sperm that fuses with the egg, *s2* sperm that fuses with the polar nucleus, *sy* synergid, *up* upper polar

hundreds of ovules, they failed to find many of the critical stages in the process. The present status of the subject may be dealt with in two parts:

1. Form and structure of the male gametes
2. Details of the two fusions—one between the egg and the first male gamete, and the other between the polar nuclei and the second male gamete.

In form, the male gametes may be spherical as in *Erigeron* (Land, 1900), ellipsoidal as in *Levisticum* (Hakansson, 1923), rod-shaped as in *Urtica* (Strasburger, 1910), or vermiform as in *Lilium* (Guignard, 1899 **Fig. 16.7**) and Fritillaria (Sax, 1916). Frequently they may change their shape after their discharge into the embryo sac.

Nawaschin (1898) reported that in *Fritillaria* the sperms lose their worm-like form just before fertilization. In *Silphium* (Land, 1900), *Monotropa* (Shibata, 1902), *Taraxacum* (Poddubnaja-Arnoldi and Dianowa, 1934*), Lacluca* (Jones, 1927), and *Nicotiana* (Goodspeed, 1947) they are at first elongated or oval, but gradually become shorter and more spherical as they approach the female nuclei. In Juglans (Nawaschin, 1900), on the other hand, they are spherical in the beginning but become curved afterward. The male cells of Vallisneria (Wylie, 1923, 1941) are originally only slightly longer than broa but become considerably elongated as the pollen tube enters the ovarian cavity. Finally they once again present a contracted appearance at the time of their discharge into the embryo sac. Gerassimova (1933) found that the changes undergone by the male gametes of *Crepis* are so rapid that it is difficult to follow them satisfactorily. Eventually the sperms become more or less band-shaped and appear to consist of two definite halves, folded along their entire length. Sometimes they roll up into a ball-shaped body, but the two longitudinal halves still remain distinguishable. There are a few reports of differences in the size and shape of the two male nuclei discharged by a pollen tube. Guignard (1899) stated that in *Scilla nonscripta* the male nucleus destined to fertilize the egg is smaller than the one fusing with the polar nuclei, and Hoare (1934) has confirmed this although admitting that the size difference is not a constant feature. In *Lilium auratum*, *Iris versicolor, Fritillaria pudica (*Sax*, 1916, 1918), Trillium grandiflorum (*Nothnagel*, 1918), Acacia baileyana (*Newman*, 1934)*, and *Camassia leichtlinii* also, the male gamete entering the egg is said to be somewhat smaller than the one fusing with the polar nuclei and is sometimes also not so vermiform. In some other plants the reverse condition has been reported. Persidsky (1926) states that in *Orobanche cumana* the sperm nucleus fertilizing the egg is shaped like a hemisphere and is larger than the other, which has an oval outline. In *O. ramosa* (Finn and Rudenko, 1930) also, the sperm nucleus fusing with the egg is looser and morefaintly staining than the other.

In *Vallisneria* (Wylie, 1923) both nuclei are oval at the time of their discharge from the pollen tube but the second soon becomes spherical. Very recently Kadry (1946) has reported that in *Cardiospermum* the male nucleus fertilizing the egg is swollen and rounded in front but tapering behind, and that it is about four times as long as the other male nucleus, which is more or less spherical.

In *Vinca minor* also, the sperm cells are unequal, one with a larger and the other with a shorter plasma tail, but it could not be determined which fuses with the

egg and which with the polar nuclei. Although slight differences in the size and shape of the two male gametes are possible, the few examples cited above do not seem adequate to justify any generalization. In the first place considerable care has to be taken to make sure that the observed differences are not due to the plane of sectioning. Stages in fertilization are in frequent, and in the same section one male gamete may be cut across its longer diameter and the other in a plane at right angles to it, so that the two appear to be of different sizes, although in fact they are quite similar. Further, the male gametes often change their form and, as Gerassimova (1933) has suggested, the reported size differences may well be due to a disparity in the rates of their transformation.

Long ago, Strasburger (1884) put forth the view that the male gametes are carried passively by the streaming movements of the cytoplasm inside the pollen tube. On the other hand, Nawaschin (1898) and some other workers believed that the vermiform appearance of the male gametes, observed in several members of the Liliaceae and Compositae, is indicative of an independent power of movement. Studies on living pollen tubes (Wulff, 1933) seem to support Nawaschin's view. The cytoplasm in the tube shows several plasma strands running in opposite directions, while the male gametes move in the forward direction only. This would hardly be possible unless the male gametes have the power to move independently of the strands of cytoplasm. Concerning the actual course of fusion of the gametic nuclei, only a few observers have described it in sufficient detail.

In conclusion, it may be well to emphasize that until a few years ago the male gametes of angiosperms were usually considered to be naked nuclei. Recent studies on the subject leave no doubt, however, that the cytoplasmic sheath remains intact at least for the period during which the male gametes are in the pollen tube. Finn (1941) has suggested that in order to decide the point with certainty the whole series of events should be studied in living material, but this seems to be impracticable with most plants, as the embryo sac is enclosed in several opaque layers which interfere with a direct and detailed observation of its contents. The only alternative is to look for some suitable material in which fertilization stages may be found abundantly, process does not take place too rapidly and the gametic cells not only are fairly large but also respond more favorably to our staining methods.

16.4.4 Multiple Fusions and Polyspermy

Compton (1912) recorded an ovule of *Lychnis* with two embryo sacs, each of which had been penetrated by a pollen tube. Another ovule with a single embryo sac had also received two pollen tubes, but only one of them had entered the sac, the other remaining behind in the nucellus. He concluded that there is a quantitative relation between embryo sacs and pollen tubes, two embryo sacs secreting enough chemotactic material to attract two pollen tubes, while one can attract only one tube. Pope (1946) also recorded an ovule of *Hordeum* with one pollen tube inside the micropyle and four at its mouth, but how the embryo sac admitted the first and excluded the others could not be determined. Although one pollen tube to an embryo sac may thus be considered as the usual condition, the entry of accessory tubes is not unknown. The entry of additional pollen tubes naturally results in the release of supernumerary male gametes inside the embryo sac. Rarely, one and the same pollen tube may also carry more than two sperms. This abnormality may originate either in the pollen grain or in the pollen tube. To mention a few examples, three sperms were sometime seen in the pollen grains of *Cuscuta epithymum* and four in *Helosis cayennensis* (Umiker, 1920), *Vinca herbacea*, *Parthenium argentatum* and *P. incanum* (Dianowa, Sosnovetz, and Steschina, 1935). Four sperms have also been seen in the pollen tubes of *Allium rotundum*, *A. zebdanense* , *Crepis capillaris* (Gerassimova, 1933) and *Polygonatum canaliculatum* (Eigsti, 1941). The presence of extra sperms inside the embryo sac, whether they are derived from one or more than one pollen tube, may result in two kinds of abnormalities. Either some of the supernumerary sperms enter the egg, resulting in a polyploid offspring, or more than one cell of the egg apparatus may be fertilized, resulting in multiple embryos.

Fertilization of more than one cell of the egg apparatus has been reported in several plants of which *Sagittaria graminea* (Johri, 19366) and *Crepis capillaris* (Gerassimova, 1933) may be cited as examples. In *Sagittaria* fertilization usually occurs normally, on female gamete fusing with the egg and the other with the two polar nuclei. But the synergids often assume an egg-like appearance and sometimes a second pollen tube enters the embryo sac, releasing two additional sperms. Although an actual fertilization of the synergids was not seen, the presence of two pollen tubes and three proembryos in the upper part of the embryo sac leave no doubt that this may happen. In *Crepis capillaris*, Gerassimova (1933) observed some embryo sacs with two to five eggs in addition to the two synergids. Usually only one of the eggs gives

rise to an embryo and the others eventually degenerate and disappear, but if a pollen tube carrying more than two sperms enters the embryo sac there is a possibility of the production of additional embryos.

16.4.5 Single Fertilization

Although double fertilization is the rule in angiosperms, the question arises whether development can proceed with only a single fertilization, i.e., if there is syngamy without triple fusion or triple fusion without syngamy. Cooke and Shively stated that in *Epiphegus virginiana* endosperm formation begins before fertilization, and Anderson (1922) reported the same in *Martynia louisiana*. In *Ramondia nathaliae* and *R. serbica* (Glisic, 1924) syngamy occurs regularly, but triple fusion is said to be "facultative" and is frequently omitted. Wiger (1935) stated that in some members of the Buxaceae and Meliaceae, endosperm formation is entirely independent of fertilization. All these reports are, however, of a doubtful nature. Without going into details it may be said that some of the above workers seem to have overlooked the pollen tube, and others mistook the unfused polar nuclei for the first pair of endosperm nuclei. It is only rarely that development can take place without triple fusion. Guignard (1921) reported a case in *Vincetoxicum nigrum* in which the zygote had divided several times while the polar nuclei were still lying unfused and the second male gamete had not yet been discharged from the pollen tube. Dahlgren (1930, 1939) has figured embryo sacs of *Mitella pentandra* and *Zostera marina* in which a several-celled embryo is associated with an undivided secondary nucleus, and Johansen (1931) has reported similar occurrences in *Taraxia ovata* and *Zauschneria latifolia*. However, such embryos are likely to stop growth so that no viable seeds are produced. The second of the two alternatives i.e., the occurrence of triple fusion without an accompaniment of syngamy has been reported in several plants, but the ovules soon begin to degenerate. If seeds are formed, they are without embryos and therefore nonviable. Rarely, however, the unfertilized egg may develop into a haploid embryo. Such cases will be discussed in connection with apomixes.

16.5 Self Incompatibility (SI)

Self incompatibility is a genetically controlled cell–cell recognition system that acts as a barrier to self-pollination in a wide range of flowering plant species. In plants with self-incompatibility, when a pollen grain produced in a plant reaches a stigma of the same plant or another plant with a similar genotype, the process of pollen germination, pollen-tube growth, ovule fertilization and

embryo development is halted at one of its stages and consequently no seeds are produced. Various self incompatibility mechanisms have been identified.

The best studied mechanisms of SI act by inhibiting the germination of pollen on stigmas, or the elongation of the pollen tube in the styles. These mechanisms are based on protein-protein interactions, and the best-understood mechanisms are controlled by a single locus termed S, which has many different alleles in the species population. Despite their similar morphological and genetic manifestations, these mechanisms have evolved independently, and are based on different cellular components; therefore, each mechanism has its own, unique S-genes. The S-locus contains two basic protein coding regions - one expressed in the pistil, and the other in the anther and/or pollen (referred to as the female and male determinants, respectively). Because of their physical proximity, these are genetically linked, and are inherited as a unit. The units are called S-haplotypes. The translation products of the two regions of the S-locus are two proteins which, by interacting with one another, lead to the arrest of pollen germination and/or pollen tube elongation, and thereby generate an SI response, preventing fertilization. However, when a female determinant interacts with a male determinant of a different haplotype, no SI is created, and fertilization ensues. This is a simplistic description of the general mechanism of SI, which is more complicated, and in some species the S-haplotype contains more than two protein coding regions. Following are the types of self compatibility:

16.5.1 Gametophytic self-incompatibility (GSI)

In this type of self-incompatibility the phenotype of the pollen is determined by its own gametophytic haploid genotype. This is the more common type of self-incompatibility, existing generally in Solanaceae, Rosaceae, and Papaveraceae. Following are two mechanisms of GSI:

The RNase Mechanism

In this mechanism, pollen tube elongation is halted when it precedes approximately one third of the way through the style. The female component ribonuclease, termed S-RNase probably causes degradation of the ribosomal RNA (r RNA) inside the pollen tube, in the case of identical male and female S alleles, and consequently pollen tube elongation is arrested, and the pollen grain dies.

The S-Glycoprotein Mechanism

It was described in detail in *Papaver rhoeas*. In this mechanism, pollen growth is inhibited within minutes of its placement on the stigma. The female determinant is a small, extracellular molecule, expressed in the stigma; the identity of the male determinant remains elusive, but it is probably some cell membrane receptor. The interaction between male and female determinants transmits a cellular signal into the pollen tube, resulting in strong influx of calcium ions; this interferes with the intracellular concentration gradient of calcium ions which exists inside the pollen tube, essential for its elongation. The influx of calcium ions arrests tube elongation within 1–2 minutes. At this stage, pollen inhibition is still reversible, and elongation can be resumed by applying certain manipulations, resulting in ovule fertilization. Subsequently, the cytosolic protein p26, a pyrophosphatase, is inhibited by phosphorylation, possibly resulting in arrest of synthesis of molecular building blocks, required for tube elongation. There is depolymerization and reorganization of actin filaments, within the pollen cytoskeleton. Within 10 minutes from the placement on the stigma, the pollen is committed to a process which ends in its death.

16.5.2 Sporophytic self-incompatibility (SSI)

In this self-incompatibility, the SI phenotype of the pollen is determined by the diploid genotype of the anther (the sporophyte) in which it was created. SSI is determined by a diploid genotype, the pollen and pistil each express the translation products of two different alleles, i.e. two male and two female determinants. Dominance relationships often exist between pairs of alleles, resulting in complicated patterns of compatibility/self-incompatibility. These dominance relationships also allow the generation of individuals homozygous for a recessive S allele. It is generally found in Brassicaceae, Asteraceae, Convolvulaceae, Betulaceae, Caryophyllaceae, Sterculiaceae and Polemoniaceae. Mechanism of SSI has been described in detail at the molecular level, in *Brassica* (Brassicaceae).

The SI Mechanism in *Brassica*

In *Brassica*, the pollen coat, derived from the anther's tapetum tissue, carries the translation products of the two S alleles. These are small, cysteine-rich proteins. The male determinant is termed SCR or SP11, and is expressed in the anther tapetum as well as in the microspore and pollen. The female determinant of the SI response in *Brassica*, is a transmembrane protein termed SRK, which has an

intracellular kinase domain, and a variable extracellular domain. SRK is expressed in the stigma, and probably functions as a receptor for the SCR/SP11 protein in the pollen coat. Another stigmatic protein, termed SLG, is highly similar in sequence to the SRK protein, and seems to function as a co-receptor for the male determinant, amplifying the SI response. The interaction between the SRK and SCR/SP11 proteins results in autophosphorylation of the intracellular kinase domain of SRK, and a signal is transmitted into the papilla cell of the stigma. Another protein essential for the SI response is MLPK, a serine-threonine kinase, which is anchored to the plasma membrane from its intracellular side.

16.5.3 2-loci gametophytic self-incompatibility

Family Poaceae shows gametophytic self-incompatibility system that involves two unlinked loci referred to as S and Z. If the alleles expressed at these two loci in the pollen grain both match the corresponding alleles in the pistil, the pollen grains are recognized as incompatible.

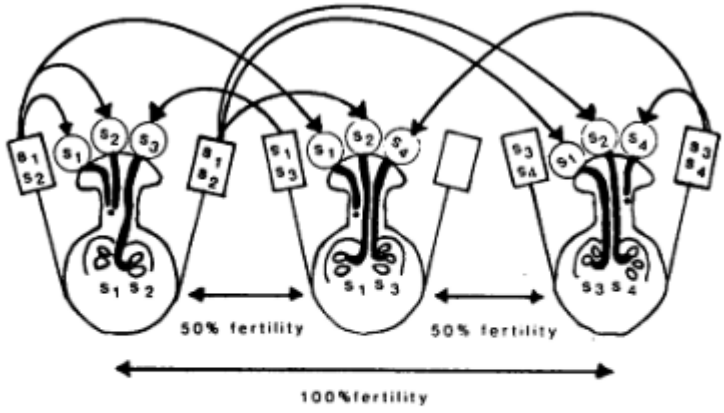

Fig. 16.8 Homomorphic gametophytic SI. In the classic mechanism genetic control is by means of a single locus S with a series of alleles in the population. Each allele, s_1, s_2, s_3,... acts independently and (self) incompatibility occurs when products of the s allele in the pollen tube encounter products of the same s allele in the stylar transmitting tissue. In some taxa there is polyfactorial control with two (Gramineae) or rarely more (*Ranunculus acris, Beta vulgaris*) multiallelic SI loci.

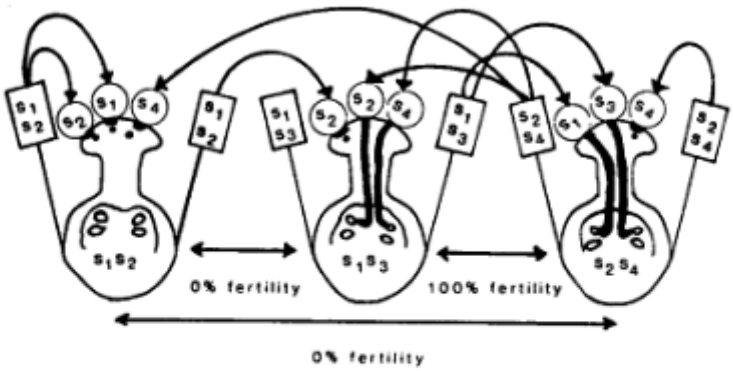

Fig. 16.9 Homomorphic, sporophytic SI. Genetic control is also determined by a single locus S with multiple alleles in the population. Each haploid pollen grain, although clearly bearing only one s allele in its nucleus, carries the incompatibility reaction of *both* s alleles present in the pollen parent. As a consequence note that pollen grains with the s_2 allele derived from a s_1 s_2 plant are incompatible on a s_1 s_3 stigma whereas s_3 -bearing pollen grains from a s_2 s_4 pollen parent are compatible on s_1 s_3 stigmas. Hierarchical dominance-recessive reactions may exist between s alleles in the pollen or stigma which, for simplicity, are omitted in Fig. 2.

16.5.4 Heteromorphic self-incompatibility

This SI mechanism exists in heterostylous flowers and termed heteromorphic self-incompatibility. The loci responsible for SI in heterostylous flowers are strongly linked to the loci responsible for flower polymorphism, and these traits are inherited together. **Distyly** is determined by a single locus, which has two alleles; **tristyly** is determined by two loci, each with two alleles. Heteromorphic SI is sporophytic, i.e. both alleles in the male plant, determine the SI response in the pollen. SI loci always contain only two alleles in the population, one of which is dominant over the other, in both pollen and pistil. Variance in SI alleles parallels the variance in flower morphs, thus pollen from one morph can fertilize only pistils from the other morph. In tristylous flowers, each flower contains two types of stamens; each stamen produces pollen capable of fertilizing only one flower morph, out of the three existing morphs.

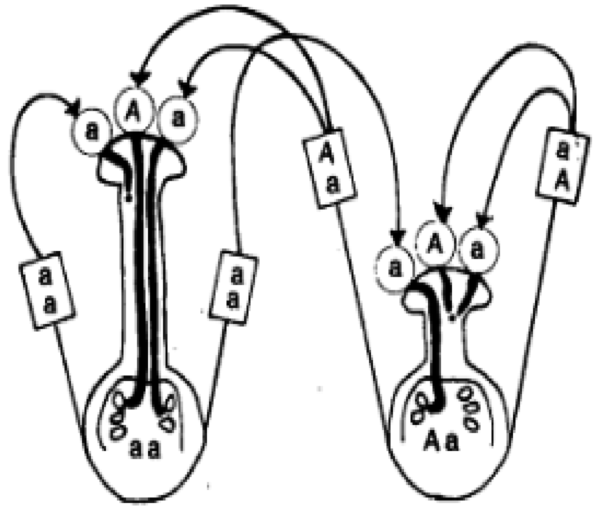

Fig.16.10 Heteromorphic SI. Simplistically, genetic control is determined by one locus (A) with two alleles, one dominant, A and one recessive, a. The mechnism is associated with floral dimorphy, commonly but by no means always, including heterostyly. In *Primula* spp. long-styled morphs are homozygous recessives, aa, and short-style morphs are heterozygotes, Aa. The system functions by a sporophytic mechanism in the sense that all the pollen grains and also the stigma/stylar tissues of the heterozygotes function phenotypically with the incompatibility reaction of the dominant allele. Consequently, compatible pollinations are effectively: a x A and A x a. In fact, rather than a single locus, genetic control is heteroallelic with a 'supergene' or tightly clustered series of loci with dominant or recessive alleles which determine not only the SI reaction but also the facets of the floral architecture -style length, anther level, pollen size, etc.

16.5.5 Cryptic self-incompatibility (CSI)

This self-incompatibility exists in a limited number of taxa for example *Silene vulgaris,* and Caryophyllaceae. In this mechanism, the simultaneous presence of cross and self pollen on the same stigma, results in higher seed set from cross pollen, relative to self-pollen. However, as opposed to complete or absolute SI, in CSI, self-pollination without the presence of competing cross pollen, results in successive fertilization and seed set; in this way, reproduction is assured, even in the absence of cross-pollination. CSI acts, at least in some species, at the stage of pollen tube elongation, and leads to faster elongation of cross pollen tubes, relative to self-pollen tubes.

16.5.6 Late-acting self-incompatibility (LSI)

It is also termed as ovarian self-incompatibility (OSI). In this mechanism, self-pollen germinates and reaches the ovules, but no fruit is set. LSI can be pre-zygotic or post-zygotic. In pre zygotic LSI deterioration of the embryo sac prior to pollen tube entry is seen e.g. *Narcissus triandrus*. In post zygotic

malformation of the zygote or embryo is seen e.g. *Asclepias, Spathodea campanulata* etc.

16.6 *in-vitro* Fertilization

The technique of in vitro fertilization (IVF), in which isolated sperm and egg cells are induced to fuse under controlled conditions, removes much of the interfering presence of somatic tissues, as well as interference of immediately surrounding maternal gametophytic cells. The use of IVF in higher plants is therefore an important contemporary research area in plant developmental and reproductive biology with potentially significant scientific applications.

The first isolations of living male gamete were reported by Cass (1973), in which he described cellular characteristics of male gametes of barley (*Hordeum vulgare)* isolated by bursting anthesis pollen grains in a 20% sucrose solution. Following the first mass isolation of sperm cells in 1986, success in male gamete isolation was repeated with modification in numerous angiosperms. Isolation of living egg and central cells was first reported in tobacco by Hu et al. (1985) and in *Plumbago* by Huang and Russell (1989).

In angiosperms little is known about the underlying mechanisms of these processes and, in spite of its fundamental importance, regulation of early embryonic development is only poorly understood. New data suggest that the asymmetric division of the zygote separates determinants of apical and basal cell fates and that programmes of transcription are initiated in the domains of single cells of the early embryo.

An in vitro fertilization (IVF) system was developed whereby maize zygotes produced in vitro by electrical fusion of an isolated egg cell with an isolated sperm cell are able to develop into an asymmetrical two-celled embryo, proembryo and transition-phase embryo via zygotic embryogenesis in a similar manner to that in planta. A major benefit of the in vitro gamete fusion and subsequent culture of zygotes is that the first unequal division of zygotes can be observed directly, and the zygote and two celled embryo can be used as materials for further analyses Recently, a procedure for isolating the apical and basal cells from two-celled maize embryos was established, and these isolated cells were used as starting points for detecting genes that are up- or down-regulated in the apical or basal cell. In this review, single cell manipulation and IVF techniques are outlined, genes expressed in apical and basal cells or zygotes of maize and *Arabidopsis* are described, and finally prospects for further investigations of early higher plant embryogenesis are outlined. Clearly,

difficulties in isolating gametes of higher plants have impeded our understanding of gamete physiology, activation of development and early embryogenesis in flowering plants. Increasing number of tools, however, are now available to manipulate male and female gametes of higher plants, providing numerous opportunities for scientific and biotechnological progress. Isolated gametes can be analyzed directly during IVF with modern cellular and physiological probes, while means of regulating sexual reproductive development are being refined. Research on reproductive biology of angiosperms has therefore entered a new phase where methods of molecular biology will permit many questions to be answered about fundamental mechanisms of fertilization and early developmental activation. Molecular characterization of male and female gametes and zygotes is expected to become a new focus of plant molecular understanding and bioengineering, which will combine cell hybridization techniques with transformation and regeneration of transgenic plants. Interestingly, parallel techniques are also being applied to gymnosperms (Fernando et al. 2005) that are uncovering the potential advantages of using gametic cells with IVF as fertilization models, and as founder populations for transgenic plants.

Presently, the most significant obstacle to using IVF has been the isolation of male and female gametes, especially the isolation of egg cells, for manipulation of living gametic cells. Although initial successes have been limited to only two species, both are critical crop plants. Findings from these plants may also provide a model for obtaining insights on fertilization and activation of development that apply to numerous other angiosperms, including other molecular and crop models. Developing successful experimental systems for IVF in other plants will be needed to confirm results obtained from maize and wheat using IVF. Both of our current best developed models are grasses, which are highly specialized angiosperms. Perfecting IVF in other plants, particularly dicotyledonous models and crops may provide critical data allowing better understanding of critical molecular events during fertilization and improving methods for the production of fertile sporophyte plants and new, stable genetic combinations of high efficiency.

16.7 Summary

Pollination and fertilization are essential activities for life cycle of angiosperms. Self-pollination and cross pollination are two major types of pollination. Anemophily, hydrophily, entomophily, zoophily and human itself are the

vectors they play their roles for pollination. After pollination gametes enter into pistil and finally reach at female gametes. Embryo sacs are the place where double fertilization takes place. Double fertilization only presents in angiosperms and alternates with pollination micsporogenesis, megasporigenesis and pollination (Fig.16.11). Self incompatibility is the method that is used by plant to confirm cross pollination and intraspecific crossing. In-vitro fertilization is artificial measure that is used by plant breeder to improvise species or to develop new varieties.

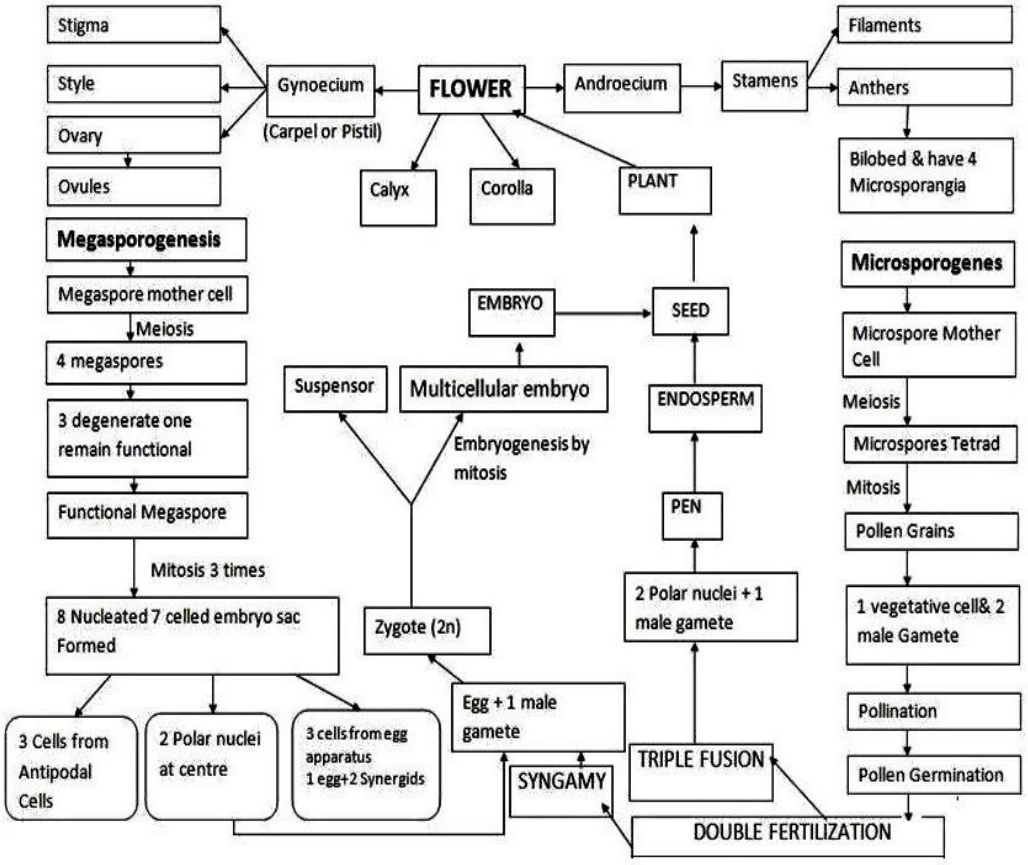

Fig. 16.11 : Life cycle of an angisperm showing pollination, double fertilization and related phenomenons

16.8 Glossary

- **Anemophily:**Pollination that occurs through wind is called anemophily.
- **Chasmogamous flowers:** Flowers which open are called chasmogamous flowers.
- **Cheiropterophily:**Pollination that occurs through water is called bats.

- **Cleistogamous flowers:** Flowers do not open are called cleistogamous flowers.
- **Dichogamy:**When stamens and carpels of a bisexual flower mature at different times.
- **Embryo sac:**The mature female gametophyte of angiosperms containing the egg cell.
- **Embryo:**The non-self-supporting immature organism formed from the zygote by cell- division and differentiation; the rudimentary plant within the seed.
- **Entomophily:**Pollination that occurs through insects is called Entomophily.
- **Fertilisation:**The union of the nucleus and other cellular constituents of a male gamete (sperm) with those of a female gamete (egg) to form a zygote.
- **Herkogamy:**When there are some physical barrier between the anther and the style to ban self- pollination.
- **Heterostyly:**Presence of styles in different lengths in the flower.
- **Hydrophily:**Pollination that occurs through water is called Hydrophily .
- **Integuments:** Sterile tissues surrounding the nucellus. These tissues mature into a seed coat.
- **Malacophily:** Pollination by snails or slugs.
- **Megasporangium:** nucellus in the seed plants: It is surrounded by sterile tissues called integuments. The integuments retain both the nucellus and the megagametophyte keeping this generation bound to the sporophyte.
- **Melittophily:** Pollination by bees. When Pollinated is occurred by small bees it is called Micromelittophily.
- **Micro:** Suffix that originally denoted small but has, in a botanical sense, taken on the meaning "male".
- **Microgametophyte:** In seed plants this is the pollen grain.
- **Micropyle:**Minute opening in the integuments of an ovule through which the pollen grain or pollen tube passes to reach the embryo sac.
- **Microsporangiate cone:** male cone: terminal clusters of microsporophylls such as the pollen cones of conifers.
- **Microsporangium:** Sporangium that bears microspores. In seed plants synonymous to a pollen sac.

- **Microspore:** Spore that develops into a microgametophyte.
- **Ornithophily:** Pollination that occurs through birds is called Ornithophily.
- **Ovule:** The integuments together with the nucellus form the ovule. Later stages include the megagametophyte and embryo.
- **Ovule:** A structure in seed plants consisting of the nucellus which contains the female gametophyte, one or two integuments and the funiculus.
- **Pollen tube:** Tubular protuberance of maturing male gametophytes in seed plants, occurring during fertilisation.
- **Pollination:** Transfer of pollen from anthers to stigma is called pollination
- **Protandry:**When anthers are matured/ripen before gynoecium phenomenon is called Protandry.
- **Protogyny:**When gynoecium are matured/ripen before anthers phenomenon is called Protandry
- **Zygote:** The fertilised egg is zygote.

16.9 Self-Learning Excercise

Section – A : (Very Short Answer Type)

1. What are Cleistogamous flowers?
2. Define dichogamy with its types.
3. Define double fertilization.
4. What is Melittophily?
5. What is Cheiropterophily?

Section – B : (Short Answer type)

1. Describe various ways of entry of pollen tube into ovule.
2. Give the ways to entry of pollen tube into embryo sac.
3. What is difference between autogamy and allogamy?
4. Describe entomophily in brief.
5. Briefly explain zoophily.

Section – C : (Long Answer type)

1. Describe various adaptations which have been developed by plants to confirm autogamy.
2. Give a detailed account on double fertilization.
3. Describe various contrivances in cross pollinated plants.
4. Give a detailed account on self incompatibility.

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Unit-17

Seed Development and Fruit Growth

Structure of the Unit:

17.0 Objectives

After studying this unit you will be understand the fundamentals of :

- Endosperm development and its types
- Embryogenesis in dicots and monocots
- Fruit development

17.1 Introduction

By studying this unit students will learn about the specific features of development of endosperm, embryo and fruit. All these developmental processes are triggered by fertilization. Embryogenesis establishes basic body plan of plant in form of miniature embryo. Endosperm formation establishes nutrient reservoir for germinating embryo until it become self-dependent. Fruit is formed by gynoecium to assist seed dispersal. All these processes are tightly coupled with each other. Various plant hormones play important roles during embryo formation, mobilization of food reserve from endosperm and fruit development.

17.2 Development of Endosperm and its Types

Endosperm is a food storage tissue of angiosperm seed which is formed after a second fertilization of the Secondary medei of control cell with male nucleus within embryo sac. Usually, endosperm cells are triploid and are closely

organized. Carbohydrates, protein and lipids are found in these cells as reserved foods, though proportions of these reserves differ depending on the species.

Secondary nuclei + male gamete = primary Endosperm nucleous:-- Endosperm

(2n) (n) (3n)

Initially endosperm was identified as an independent structure resulting from a second fertilization incident that is characteristic of angiosperms. After the finding of double fertilization, two theories, which still continue today, were suggested for the evolutionary origin of the angiosperm endosperm. In one, the endosperm was assumed to originate as an altruistic twin embryo, which changed into a nurturing endosperm for the surviving embryo, whereas in the second, the endosperm was regarded as a continual development of the megagametophyte activated by fertilization. The endosperm develops after the fertilization of Secondary nuclei with second male gamete nucleus. Three types of endosperm development are known: nuclear, cellular and helobial (Fig. 17.1).

17.2.1 Nuclear

The first one, free nuclear type, is the most common mode of endosperm development. In this, the triploid nucleus divides repeatedly without cytokinesis and cell wall formation. Simultaneously, the central vacuole appears in the primary endosperm cell and it undergoes expansion resulting into peripheral arrangement of nuclei and enlargement of the cell (Fig. 17.1). Number of peripheral nuclei increase by free nuclear divisions, leading to the endosperm coenocyte. The vacuole then shrinks and finally disappears as the nuclei multiply and cytoplasm increases. Finally, the coenocytic cytoplasm undergoes cellularization to produce a multicellular tissue. Examples are many Archichlamydeae plants and Monocots.

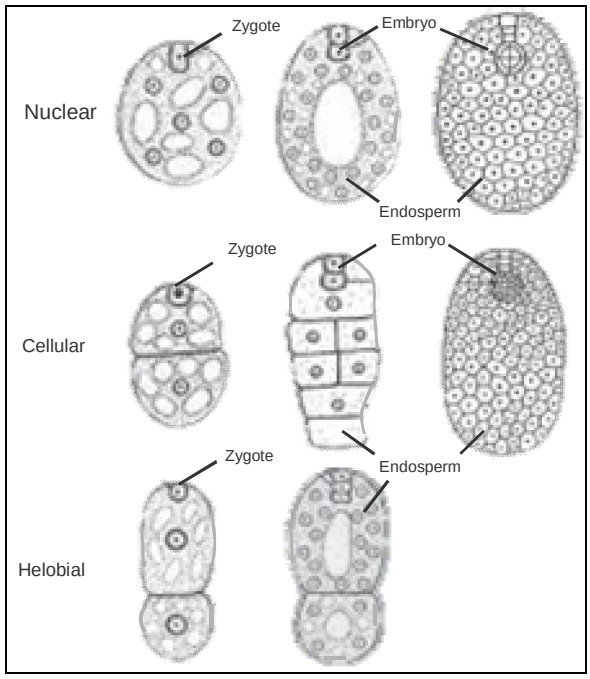

Fig. 17.1: Types of Endosperm Development

17.2.2 Cellular

In this type, every nuclear division is accompanied by cell wall development, so that the endosperm becomes cellular from the starting (Fig. 17.1). Examples- *Petunia, Datura* and most gamopetalae plants.

17.2.3 Helobial

This type of endosperm development is infrequent. It is a transitional form between the nuclear and cellular types (Fig. 17.1). In this type, a horizontal cell wall is formed between the two nuclei after first division. Later the cell at the chalazal end develops endosperm along the cellular pattern while the other one located at the micropylar end of the embryo sac develops along the nuclear pattern. Generally, chalazal cell divide only once or twice. Example- Helobiales.

The developing endosperm crushes the nucellus. The endosperm may persist in the seed (endospermic or albuminous seed e.g., castor) or may get completely absorbed by the growing embryo and the food reserve gets stored in the cotyledons (non-endospermic or exalbuminous seed e.g., Cucurbita).

17.3 Mechanism of Cellularization during Endosperm Development

In nuclear endosperm development, cellularization of the endosperm coenocyte is initiated by the formation of an array of microtubules radially emanating from the membrane of each endosperm nucleus (Fig. 17.2 c). This array is known as radial microtubular system (RMS). RMS in developing nuclear endosperm are well documented and have been observed in the endosperm coenocyte in a number of species including rice, wheat, barley, *Arabidopsis* etc. Initially, microtubules originating from neighboring nuclei may overlap (Fig. 17.2 b), but soon they get separated from each other (Fig. 17.2 c), marking out cytoplasmic interzones between individual RMS. Formation of phragmoplasts and subsequently formation of anticlinal walls take place in these RMS interzones.

Soon walls are deposited around each nucleus. As shown in the figure 17.2 these walls surround each nucleus and form a tube-like structure known as alveolus. The alveolus walls extend centripetally (toward the central vacuole). Mitotic divisions of nuclei within alveoli, followed by periclinal cell wall formation, lead to one peripheral layer of cells and a new layer of alveoli (Fig. 17.2 e). Repeated rounds of the same cycle of events lead to cell files that eventually completely invade the central vacuole (Fig. 17.2 f). In this way, cellularization process completes during the first few days after fertilization.

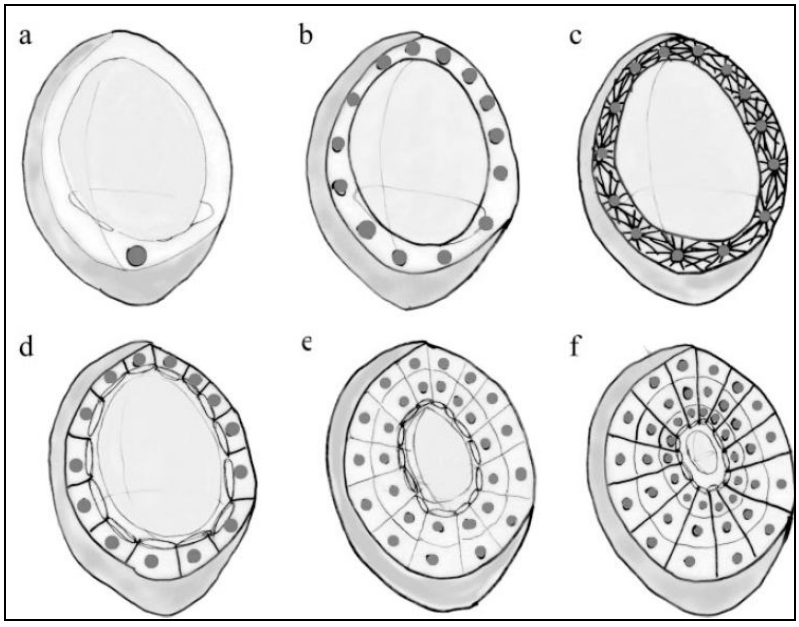

Fig. 17.2: Diagrammatic overview of the endosperm cellularization process

(*a*) In the central cell, the fertilized triploid nucleus is situated in the proximal end of a cytoplasm which surrounds a large central vacuole. (*b*) Mitotic divisions occur without cell wall formation and leads to a multinucleate cell with a large central vacuole; the endosperm coenocyte. (*c*) Endosperm cellularization is initiated by the formation of RMS (radial microtubular systems) at the surface of the endosperm nuclei. (*d*) Cell wall formation is facilitated by interaction between RMSs from each nucleus forming the cytoplasmic phragmoplast, and each nucleus becomes surrounded by a tube-like wall structure (alveolus) with its open end towards the central vacuole. (*e*) Continued growth of alveoli towards the central vacuole, and periclinal cell divisions with wall formation between the daughter nuclei within the alveoli, leads to two cell layers, one complete layer in the periphery, and a new layer of alveoli internally. (*f*) After one repetition of the alveolation process and a mitotic division, two layers of peripheral cells are formed, the new layer of alveoli extending almost to the center of the central vacuole. After further centripetal growth of the cell files, the central vacuole is completely closed. Please note that the figure has not been drawn to scale, and only one layer of endosperm cells is shown (After Olsen, 2001).

17.4 Types of cells in Endosperm and their Functions

The completely developed endosperm contains four main cell types: the embryo surrounding region, the transfer cells, the aleurone layer and the starchy endosperm (Fig. 17.3).

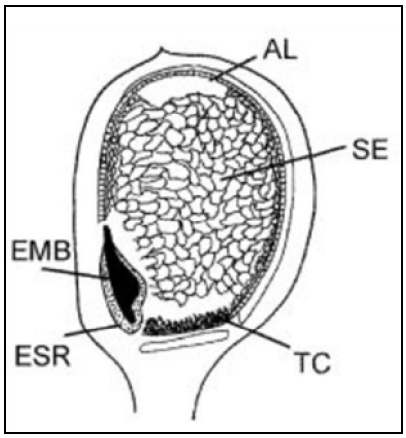

Fig. 17.3: Diagram of longitudinal section of 15 DAP maize grain consisting of the aleurone layer (AL), the starchy endosperm (SE), the transfer cell region (TC), and the embryo surrounding region (ESR) around the basal part of the embryo (EMB) (After Olsen, 2001).

17.4.1 The Embryo Surrounding Region

The cells of this region make the lining of the cavity of the endosperm in which the embryo develops (Fig. 17.3). Generally, they have dense cytoplasmic contents. Their function is still unknown; probably they have a role in embryo nutrition or in creating a physical barrier between the endosperm and the embryo during seed development.

17.4.2 Transfer Cells

These cells are present over the vascular tissue of the maternal plant (Fig. 17.3), where they carry out transport of photosynthate (e.g. sucrose) into the endosperm. Presence of prominent secondary wall ingrowths is characteristic feature of endosperm transfer cells in cereals. Latter on during early stage of the grain, these cells contain a widespread and complex endomembrane system.

17.4.3 Aleurone Cells

The aleurone layer makes the covering of the entire periphery of the endosperm excluding the transfer cell region (Fig. 17.3). The aleurome layer may be one (wheat and maize), three (barley) or several (rice) cells thick. The aleurone cells contain several aleurone grains and small vacuoles with inclusion bodies. Therefore, their cytoplasm is dense and granular. Aleurone cells may contain two main kinds of inclusion bodies — protein-carbohydrate bodies and the globoid bodies filled with a matrix of lipid, phytin and protein. The aleurone grains are surrounded by lipid droplets. The aleurone contains well developed endoplasmic reticulum and numerous mitochondria. Mature aleurone cells contain anthocyanins pigments which are accountable for the colorful grains of corn. The aleurone cells may be highly polyploidy as observed in barley.

17.4.4 Starchy Endosperm Cells

The starchy endosperm represents the major part of the endosperm. These cells stores bulk amount of starch synthesized within amyloplasts by the four enzymes, ADP-glucose pyrophosphorylase, starch synthases, branching enzymes, and debranching enzymes. Prolamin storage proteins are the second major cell-specific component of starchy endosperm cells. These cells undergo endo-reduplication following the phase of cellularization and cell fate specification. With the termination of the grain-filling stage, the starchy endosperm cells die, involving a procedure that is similar to programmed cell death in animals.

17.5 Embryogenesis

In plants, fertilization triggers four events: embryo, endosperm, seed and fruit developments. Embryogenesis includes transformation of single celled zygote into multicellular, miniature embryonic plant. The processes of embryogenesis occur in an organized manner. Cell division, differentiation and growth are tightly regulated. This fact indicates that transformation of zygote into embryo is genetically programmed. During embryogenesis, the basic body plan of the sporophyte is established. The main challenges of embryogenesis are:

1. To establish the basic body plan. Radial patterning produces three tissue systems, and axial patterning establishes the apical-basal (shoot-root) axis.
2. To set aside meristematic tissue for postembryonic elaboration of the body structure (leaves, roots, flowers, etc.).
3. To establish an accessible food reserve for the germinating embryo until it becomes autotrophic.

During embryogenesis, the fundamental process of establishment of the basic body plan is similar in all angiosperms (Fig. 17.4). Differences occur in pattern elaboration. The precision of cell division patterns, the extent of development of endosperm, cotyledon and shoot meristem differ in different species.

Axial patterning: Arrangement of tissues and organs occurs in an accurate order along the linear apical-basal (shoot-root) axis in all plants. Shoot apical meristem and root apical meristem are found at opposite ends of the axis. In angiosperms, one or two cotyledons attached to the axis are found just below the shoot apical meristem. Following this attachment hypocotyl, root, root apical meristem and root cap are found in sequence. This axial pattern is established during embryogenesis.

Radial patterning: Different tissues within a plant organ are organized in a particular manner. For example in roots and stems epidermis, cortex, endodermis, pericycle, phloem and xylem are radially arranged from the periphery to the center. Three types of meristems- protoderm, ground meristem and procambium lay foundation of radial patterning.

Four different stages of embryos development

The process of embryogenesis has been studied comprehensively in Arabidopsis (a dicot). The most significant phases of embryogenesis in angiosperms including Arabidopsis are:

1. **Globular stage:** Apical and basal cells are formed after the first division in zygote. The apical cell goes through a series of precise cell divisions, producing an octant (eight-cell) globular embryo. Further, well-ordered divisions increase the number of cells of embryo.
2. **Heart stage:** Rapid cell divisions occur in two areas on both sides of the future shoot apex. Further division and growth in these two regions give rise to the cotyledons later. At this stage, embryo becomes bilaterally symmetric.
3. **Torpedo stage:** This stage is formed as a consequence of cell elongation throughout axis of the embryo and additional growth of the cotyledons.
4. **Maturation stage:** At the termination of embryogenesis process, the embryo and seed undergoes desiccation and become metabolically inactive before entering into dormancy phase.

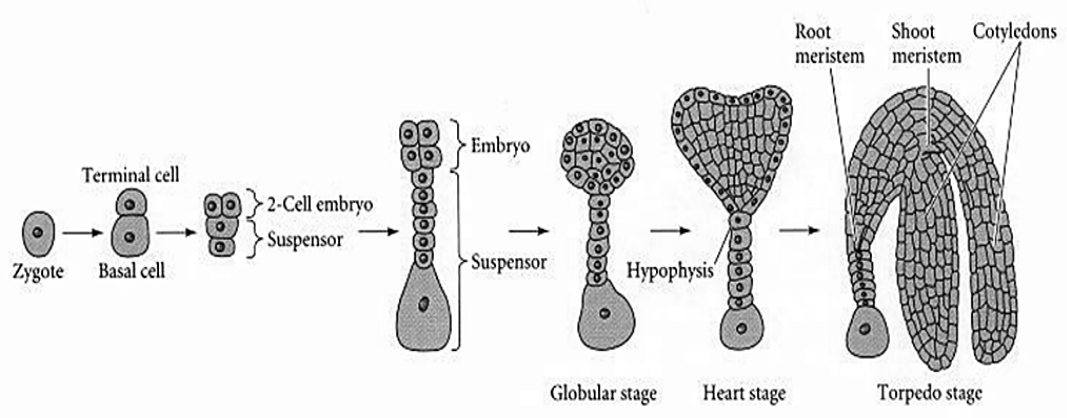

Fig. 17.4: Dicot embryogenesis

A representative dicot is shown; a monocot would develop only a single cotyledon. While there are basic patterns of embryogenesis in angiosperms, there is tremendous morphological variation among species (Source- Gilbert, Developmental Biology, 2000).

17.6 Embryogenesis in Dicots and Structure of Dicot Embryo

Polarity is established in the first cell division following fertilization. Embryogenesis initiates with an asymmetrical (transverse) cell division occurring after elongation of zygote, resulting into two unequal cells- a terminal cell and a large basal cell (Fig. 17.4). The terminal cell forms the embryo proper. The basal cell (located near the micropyle) undergoes few transverse

divisions and produces a filamentous suspensor of 6-10 cells. The suspensor pushes the embryo in the endosperm. The suspensor is present at the absorptive surface of the embryo toward its food source and assists as a nutrient channel for the growing embryo. The suspensor cell located near the micropylar end becomes swollen and develops wall ingrowths and acts as a haustorium. The suspensor cell present adjacent to the embryo at the other end of suspensor is known as hypophysis. It later produces the radicle and root cap. Filamentous suspensor degenerates later in embryogenesis. Experiments conducted on embryo culture (with and without the suspensor) in scarlet runner beans demonstrated the requirement for a suspensor through the heart stage in dicots (Fig. 17.5).

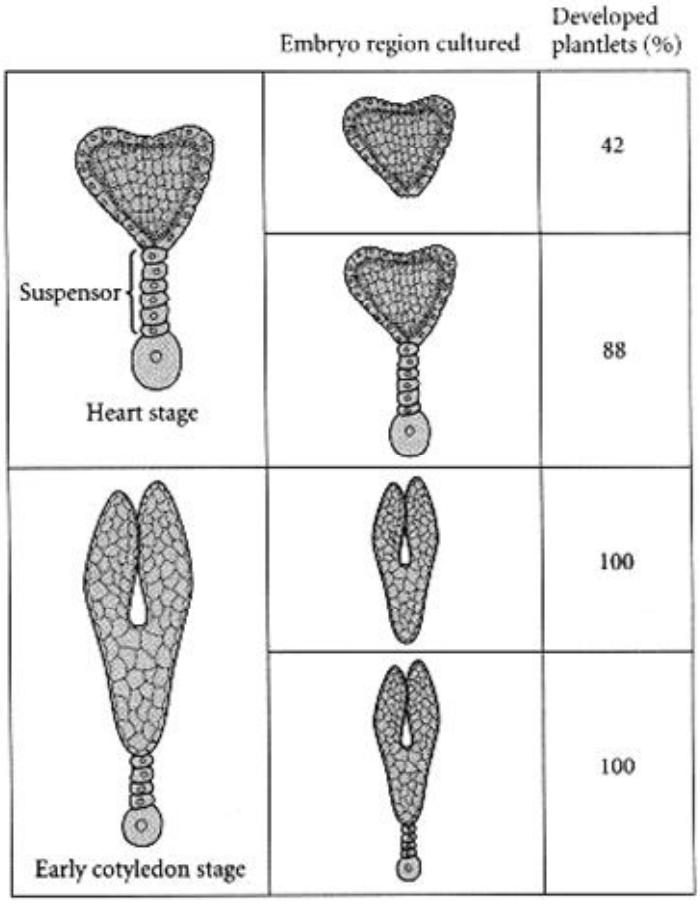

Fig. 17.5: Role of the suspensor in dicot embryogenesis

Culturing scarlet runner bean embryos with and without their suspensors has demonstrated that the suspensor is essential at the heart-shaped stage, but not later. (After Yeung and Sussex 1979.)

Radial and axial patterns are established as cell division and differentiation continue (Fig. 17.6). The cells of the embryo proper undergoes two vertical divisions (quadrant stage) and one transverse division give rise to a globular

(octant) stage embryo with eight cells arranged in two layers; terminal layer- epibasal and second layer near the suspensor- hypobasal. The two cotyledons and the plumule are derived from the epibasal cells. The hypobasal cells give rise to the hypocotyl later during the course of embryogenesis.

The emerging shape of the embryo is determined by orientations of the planes of cell division and cell expansion. Three tissues- dermal, ground, and vascular which are responsible for radial patterning, emerge at the globular stage. The eight cells of octants go through periclinal division. Cells of outer layer give rise to protoderm while the inner cells differentiate further into procambium and ground meristem. Epidermis is formed from the protoderm which acts as outmost protective layers of the plant. Ground meristem present beneath the protoderm, gives rise to cortex and pith in mature plant. Present at the core of the embryo- the procambium is responsible for development of the steal or vascular tissue in mature plant. The differentiation of each tissue system is partly independent. The early stage of embryo having radial symmetry is also known proembryo.

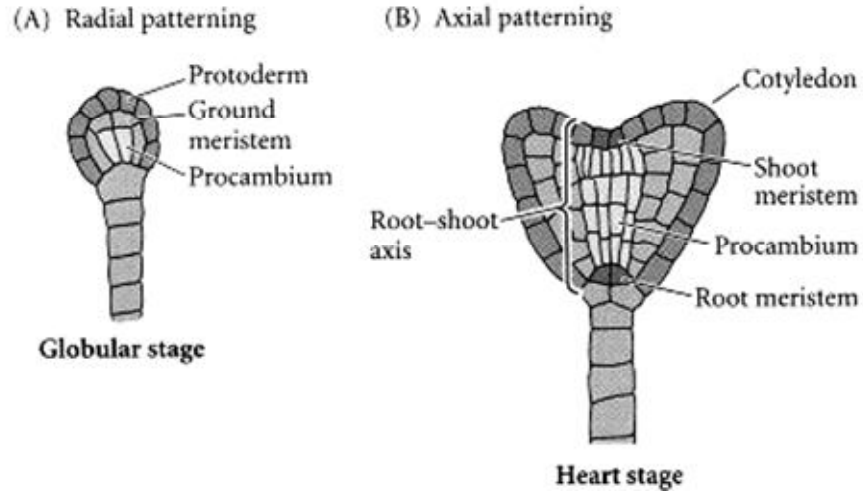

Fig. 17.6: Radial and axial patterning

(A) Radial patterning in angiosperms begins in the globular stage and results in the establishment of three tissue systems. (B) The axial pattern (shoot-root axis) is established by the heart stage (Source- Gilbert, Developmental Biology, 2000).

The globular stage of the embryogenesis terminates with the emergence of cotyledons (Fig. 17.4). Formation of two cotyledons in dicots gives the embryo a heart-shaped form. They elongate enormously with high growth rate while plumule also develops as small mound of undifferentiated tissue in apex region of embryonic axis. The axial patterning becomes evident with the development

of heart stage. The transition from radial to bilateral symmetry is mediated by plant hormones specially, auxin. Single cotyledon emerges in monocots.

In several plants, the cotyledons support in nourishing the plant by becoming photosynthetic after germination. Though in some species, they do not emerge from the ground. Yet in some plants such as peas, the food stored in the endosperm gets consumed before germination and the cotyledons provide the nutrients to the germinating seedling. Even in the presence of a persistent endosperm (maize), the cotyledons accumulate starch, lipids, and proteins. In many monocots, the single cotyledon helps in nutrient transfer to the seedling. Vertical cotyledons give the embryo a torpedo shape. In some plants, the cotyledons grow sufficiently and bend to fit within the confines of the seed coat. By this point, the suspensor is degenerating.

Two clusters of stem cells representing shoot apical meristem and root apical meristem are produced during embryogenesis (heart stage; Fig. 17.6 B). They persist in the postembryonic plant and built most of the sporophyte body. The root meristem is partially derived from the hypophysis in some species. Genetic evidence shows that the foundation of the shoot and root meristems is controlled independently.

The shoot apical meristem not only gives rise to leaves after germination and but eventually forms reproductive structures. In *Arabidopsis,* the cotyledons are formed from general embryonic tissue, not from the shoot meristem. Similarly in cotton, the cotyledons and the first two true leaves are derived from embryonic tissue rather than an organized meristem. Most embryonic cells are pluripotent and meristems possess this capacity in the postembryonic plant body.

A typical dicot embryo is made of of an embryonic axis and two cotyledons. The part of embryonic axis above and below the attachment of cotyledons is known as epicotyl and hypocotyl respectively. Plumule and radicle are found at opposite ends of the embryonic axis. A root cap (calyptra) covers radicle end.

With the development of embryo, the ovule transforms into the seed. Its integuments eventually form hard protective coverings. At this time the embryo enters into resting state in seed.

17.7 Embryogenesis in Monocots and Structure of Monocot Embryo

The zygote elongates and then divides transversely to produce basal and terminal cells. Like the process of embryogenesis in dicot, the basal cell forms suspensor cell. It may acts as haustorium. The terminal cell undergoes another transverse division to form two cells.

After going through sequential divisions, the top cell forms plumule and a single cotyledon while the other cell produces hypocotyl and radicle. The single cotyledon is known as scutellum. It shows rapid growth which pushes the terminal plumule to one side. Scutellum is situated towards lateral side of embryonic axis. Epicotyl is found above the level of attachment of scutellum. Shoot apex and few leaf primordia are found enclosed in the coleoptile. Epiblast denotes rudiments of second cotyledon.

17.8 Fruit

True fruits are found only in the angiosperms or flowering plants; the name angiosperm means hidden seed. Fruits are often defined as structures derived from a mature ovary containing seeds, but many structures we might call as fruit are in fact composed of a variety of tissue types. True fruits are derived from the flower gynoecium. Fruits have evolved to assist seed protection and dispersal. Diverse range of fruits within angiosperm species exists and these variations are exemplified between fleshy fruits that have evolved with an enlargement of the tissue surrounding the seed to create attractive flesh for seed dispersing animals and "dry" fruit that split open (dehisce) to release the seed via abiotic dispersal mechanisms. The myriad of fruit types recognized in angiosperms can be categorized on basis of different parameters.

17.9 Fruit Types

17.9.1 On basis of origin

Fruits are of three types based upon floral origin- simple, aggregate and multiple fruits. In some cases one flower develops into just one fruit. This is because that one flower contained just one pistil that matured into a simple fruit (Cherries and Tomatoes). The pistil might be simple (monocarpous) or compound (multicarpous but syncarpous).

In other species, one flower develops into several fruits in a cluster. This happens because that one flower contained several pistils (unfused simple

pistils- multicarpous but apocarpous condition) that each became a fruit. It is known as aggregate fruit, examples are Raspberries and strawberries.

In yet other species, a whole cluster of flowers--an inflorescence--gets pollinated and fruits develop. However as these several fruits grow, they merge and fuse into a single fruit body. Because many flowers produce fruits that fuse into just one single body, it is known as a multiple fruit. Examples are Mulberries and Pineapple etc.

17.9.2 On basis of composition

Another way to categorize fruits is to classify them on the basis of what parts of the flower contribute to the overall fruit. Here there are just two categories.

A true fruit is one that consists of nothing but ovary wall. There are no other contributions to the fruit body. A good example is cherry and peach. Here, after pollination, all the other parts of the flower fall off of the pedicel. Even the style and stigma abscise, leaving just an ovary. The ovary wall thickens. The outer layers of ovary become fleshy and red; the inner layers become hard and stony and they surround a single seed.

An accessory fruit is one that has more than ovary wall as part of the fruit body. Other parts of the flower swell along with the expanding ovary wall. Frequently the receptacle participates. Ovaries that are inferior or that are in perigynous flowers often have accessory tissues surrounding or subtending the true fruit. A good example of an accessory fruit is a watermelon. The true fruit is red at maturity, the accessory is white. Another example is apple. The cartilaginous core is the true fruit; the white tissue you like to eat is the accessory.

17.9.3 On basis of fruit repining

This is yet another way of fruit categorization based on ripening physiology. Fruit that have a strong requirement for ethylene to ripen such as tomatoes, Peaches, Bananas, Apples, and Melon have been known as climacteric fruits (Fig. 17.7.). In fruit that have a lower requirement of ethylene to ripen are referred as non-climacteric fruit such as grape and citrus. Climacteric fruits show increased respiration rate during ripening stage while non-climacteric fruits don’t (Fig. 17.7). Non- climacteric fruits can ripe only on tree while climacteric fruits can also ripe after detachment from tree.

17.10 Dynamics of Fruit Growth

In all types, fruits undergo a progression of specific steps including: fruit set, fruit growth, maturation, and ripening/senescence. Fruit shows sigmoidal growth curve (Fig. 17.7).

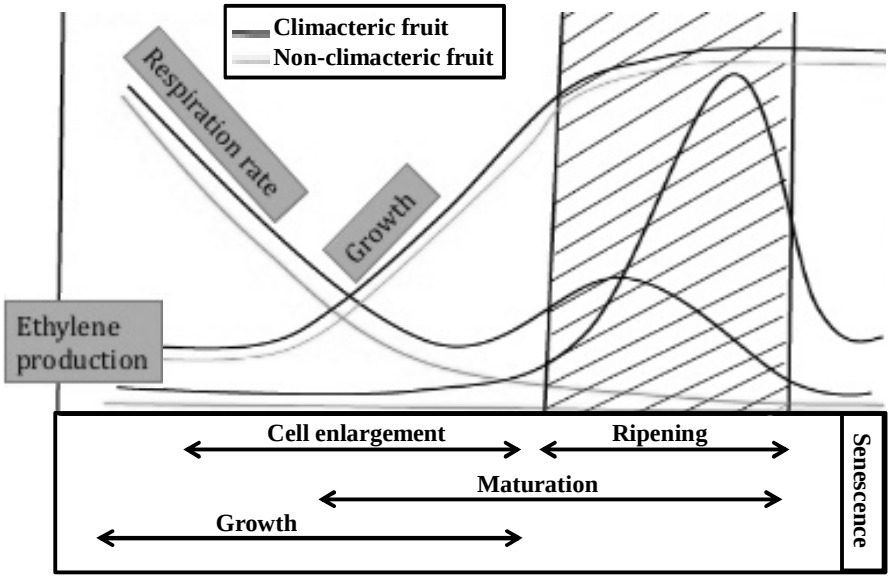

Fig. 17.7: Dynamics of Fruit Growth

Ripening graph is showing growth curve, respiration rate and level of ethylene hormone during different stages of fruit development in climacteric and non-climacteric fruits.

17.10.1 Fruit set

Fruit set is the first step in fruit development; it is established during and soon after fertilization. Fruit develops from gynoecium. The gynoecium is derived from the fusion of carpels and forms in the center of the flower. Many regulators of carpel development identified in *Arabidopsis* also have roles during leaf development, thereby confirming the evolutionary origin of carpels as modified leaves. The developmental switch that turns a gynoecium into a growing fruit depends on the fertilization of ovules that form along the placenta. In most angiosperms, the gynoecium senesces and dies if not fertilized.

The fruit initiation process has traditionally been thought to involve phytohormone activities. Upon fertilization, a seed-originating auxin signal is generated that is thought to upregulate biosynthesis of another hormone, gibberellin. This leads to activation of gibberellin signaling in the ovules and ovary, thereby stimulating fruit growth. Fruit set has traditionally been

attributed to the action of three hormones, auxin, and/or gibberellin, and/or cytokinin. Interplay between these hormones is necessary for fruit set and fruit growth. Developing seed influences fruit development. The "seed control" hypothesis suggests that the seeds communicate through hormones to the surrounding tissue(s) to promote fruit growth through firstly cell division and later on cell expansion (Fig. 17.8 and 17.9). At the molecular level, the main advances have been on how gibberellin and auxin pathways interact to promote fruit set. Elevated levels of gibberellins and auxin are present in fruits from plants that exhibit parthenocarpy and auxin levels increase during seed development while gibberellin levels increase in the ovaries following fertilization.

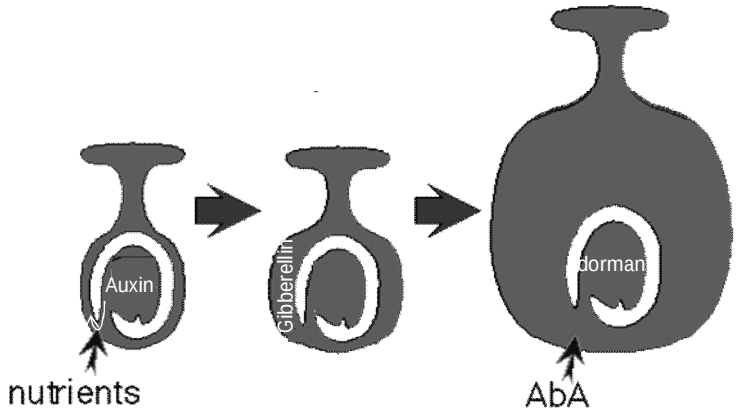

Fig. 17.8: Fruit development is linked with seed growth through hormones

The seeds developing inside the ovary produce hormones (Fig. 17.8). At first they produce auxins (probably cytokinins too) which are hormones that are exported from ovules after fertilization and cause cell division in the ovary wall. Ovary wall forms the pericarp of fruit. This adds some thickness to the wall of the growing fruit. Auxin also induces ovary to produce gibberellic acid which causes rapid expansion of each of the cells in the wall of the ovary (Fig. 17.8). The combination of more cells and expanding cells leads to tremendous increase in the size of the ovary. Meanwhile, the mother plant produces another hormone, abscisic acid, which causes the embryo in the developing seeds to become dormant (Fig. 17.8). This is adaptive because it prevents the seed from sprouting inside the warm, moist fruit. This sequence of events is diagrammed above.

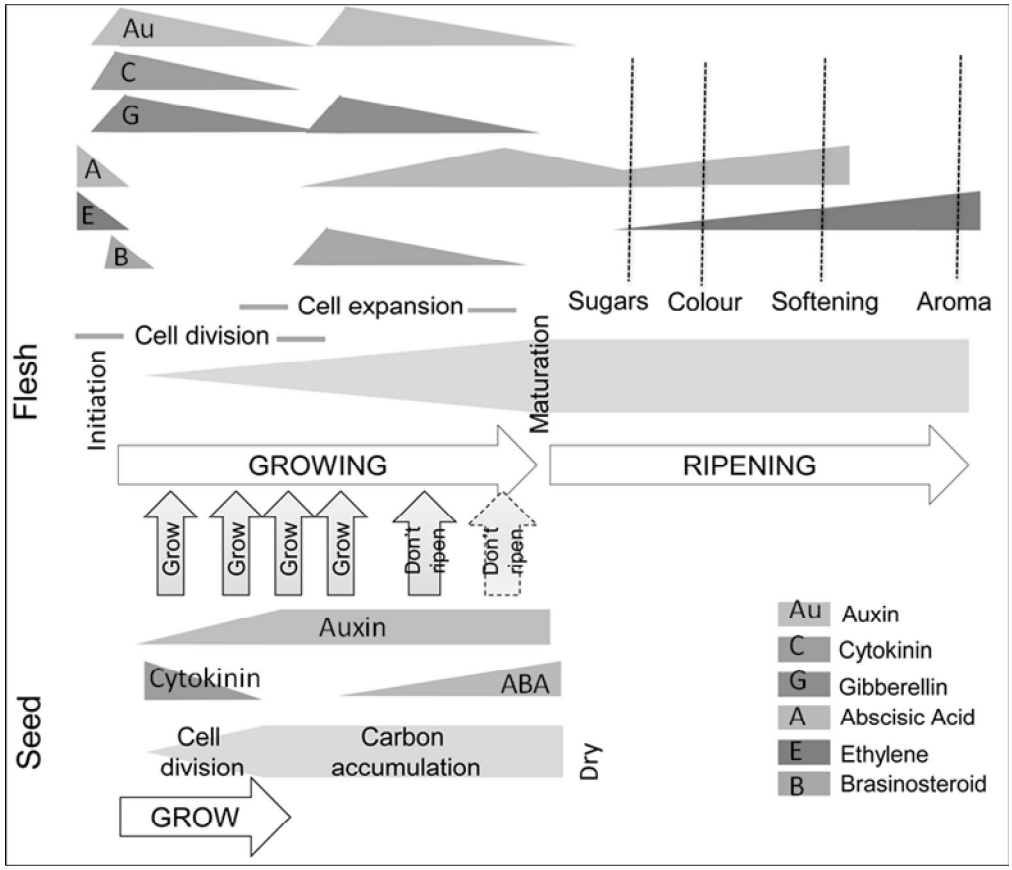

Fig. 17.9: Hormonal changes that occur in a generic fruit during development and ripening.

Differential hormone concentrations occur in the seed and the surrounding tissue with the developing seed inthiening its environment. Multiple studies have shown that increases in auxin, cytokinin, gibberellin, and brassinosteroid at fruit set, and an involvement of auxin, gibberellin, and brassinosteroid at fruit growth. For fruit maturation there is an inhibition of auxin transport from the seed and increase in ABA. This triggers the ripening/senescence program which leads to an increase in ABA and/or ethylene biosynthesis and response in the surrounding tissue (After Peter et al, 2013).

17.10.2 Fruit growth

The developing seed continually sends signals to the surrounding tissue to expand and there is usually a positive correlation between seed number and fruit size (Fig. 17.9). The developing fruit must also signals back to the rest of the plant so that it is provided with enough nutrients and does not abort. The extent of growth of the fruit from anthesis to maturity is extremely variable; in some species the fruit enlarge relatively little while in others they may increase in volume many thousand times. Unique to fleshy fruit, concomitant with cell

expansion, there is an accumulation of storage products and an increase in sugar accumulation. Fruit expansion is a key event during its growth.

Cell enlargement depends on both cell wall loosening and increases in turgor pressure. While auxin mostly controls cell division during fruit set, it is thought to play an important role during the growth phase by influencing cell enlargement together with gibberellins (Fig. 17.9). A range of cell wall-related proteins are up-regulated during the expansion stage of the fruit, as well as sugar transport proteins and various glycolytic enzymes. Some genes belonging to the expansins, endo-xyloglucan transferase and pectate lyases families have been shown to be regulated by auxin, gibberellin, or both in tomato.

17.10.3 Fruit maturation

Fruit maturity is a developmental point where the fruit has reached the competence to ripen, but has yet to start the ripening process. Auxin and may be cytokinin appear to be key regulators of fruit maturation (Fig. 17.9). During fruit growth, auxin levels in the seed are higher than in the surrounding fruit tissue and this suggests as the seeds become dormant, auxin biosynthesis or transport to the rest of the fruit is inhibited, allowing the mature fruit to ripen. This appears to be supported across fruit species as addition of auxin to mature fruit invariably delays ripening.

17.10.4 Fruit ripening/senescence

The progression of fruit ripening or senescence is a complex process involving changes to the metabolic and physiological traits of a fruit. In all fruits, in the tissue surrounding the seed, there is a color change and a change in cell wall composition causing either a dehiscence or a softening. Unique to fleshy fruit there is often a breakdown of stored carbohydrates to sugars and a decrease in acidity along with an increase in flavor and aroma volatiles. The control of ripening appears to be achieved predominantly through the ripening hormones ABA and ethylene, ethylene being the most studied (Fig. 17.9). Climacteric fruits require ethylene for ripening while in case of non-climacteric fruits ABA appears to have a stronger role in ripening. Tomato is a good model for understanding ripening in fleshy fruits (Fig. 17.10). Developments taking place during entire course have been shown in the diagram below.

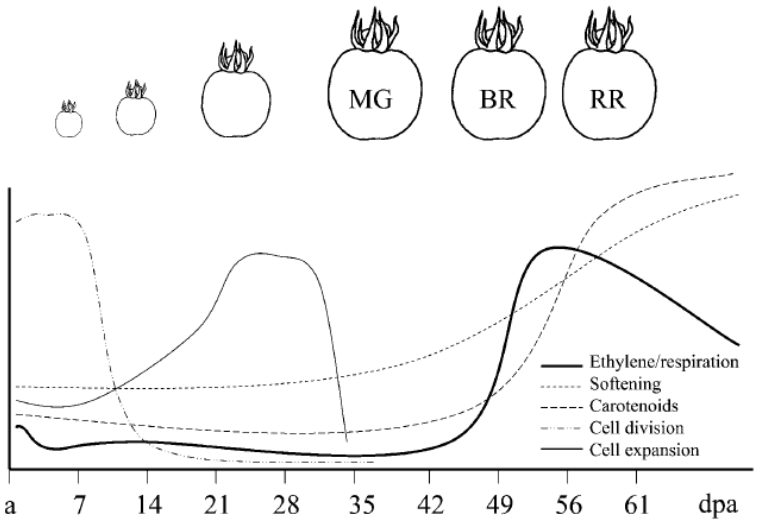

Fig. 17.10: Major Developmental Changes during Tomato fruit development and ripening. Relative changes in cell division, cell expansion, respiration, ethylene synthesis, fruit softening, and carotenoid accumulation are shown over the course of fruit development. The time from anthesis (a) to **m**ature **g**reen (MG; fully expanded unripe fruit with mature seed), **br**eaker (BR; first visible carotenoid accumulation), and **r**ed **r**ipe (RR) can vary substantially among cultivars. dpa, days after anthesis (After Giovannoni, 2004).

17.11 Biochemical changes during the ripening process

17.11.1 Chloroplast to chromoplast conversion and color change

Fruit color changes during fruit ripening. This is achieved by a combination of chlorophyll loss (degreening) and production of secondary color metabolites such as carotenoids and anthocyanins. The production of secondary color metabolites is strongly ethylene regulated in tomato, though some intermediates can be produced in the absence of ethylene.

One of the most critical developmental acts associated with ripening and palatability is the conversion of chloroplasts to chromoplasts. There are a number of important structural changes that accompany chromoplast conversion. These changes have a major influence on the nutrient and flavor composition of the fruit. The photosynthetic capacity of the chloroplast is lost as the thylakoid structures begin to disassemble. Within the chromoplast, plastoglobules accumulate. These are the sites for accumulation of large quantities of carotenoids, principally lycopene and □-carotene, in the form of crystal structures. Accumulation of these carotenoids provides a visual indication that the fruit is mature and suitable for consumption. These carotenoids are also important human nutrients, providing the precursor of

Vitamin A as well as abundant antioxidants to the diet. The chromoplast and plastoglobule proteomes have been examined in multiple species. The tomato chromoplast is highly metabolically active, and enzymes associated with synthesis and metabolism of carotenoids, amino acids, and fatty acids are found in it. The enzymes responsible for carotenoid synthesis are found within these structures. The conversion of chloroplasts to chromoplasts and associated degradation of chlorophyll and membrane structure/photosynthetic activity with concomitant carotenoid accumulation are the main hallmarks of the ripening transition of tomato and many fruits and are irreversible. The primary enzymatic regulator of flux into the carotenoid pathway, phytoene synthase (PSY), is under strong positive ethylene control during ripening, indicating a link between chromoplast development and the primary ripening hormone. Also under ethylene control is repression of the gene encoding the lycopene □-cyclase (LYC) enzyme that would otherwise convert red lycopene to orange □-carotene.

17.11.2 Flavour and aroma biochemistry

Most fruits, such as tomato, have evolved to attract animal seed dispersers. When tomato seeds are fully mature, chemical changes occur in the fruit to make it attractive to animals. Cell walls undergo textural changes that result in softening. As chloroplasts are converted to chromoplasts, large amounts of carotenoids, principally lycopene and □-carotene, are synthesized, providing visual cues that the fruit is ripe. Ethylene also induces changes in glycoalkaloid content. High levels of □-tomatine are present in immature fruits, peaking at mature green. Upon ripening, levels are significantly reduced in an ethylene-dependent manner. Given that □-tomatine is linked to bitter flavor, the effect is to increase palatability upon ripening. Finally, certain chemicals that contribute to taste begin to accumulate. In tomato, there are three major classes of chemicals responsible for flavor: sugars, acids, and volatiles. Although sugars and acids are absolutely essential for good taste, it is the volatiles that really determine the unique flavor of a tomato. There have been a number of studies that have documented the metabolic changes that occur during maturation and ripening.

17.11.2.1 Sugars and Acids

At the onset of ripening, starch that has accumulated throughout development is metabolized to glucose and fructose, the two main sugars in a ripe fruit. These two sugars can constitute 2%–4% of the fresh weight of a fruit. Organic acids,

principally citric and malic acids are also abundant. The pH of a ripe fruit is typically in the range of 4. Both the sugars and acids are critical to good flavor, although different individuals have different preferences for sweetness and acid balance. Interestingly, recent work has shown that malic acid influences starch content via redox control of the enzyme responsible for starch synthesis, ADP glucose pyrophosphorylase. Given that starch synthesis is positively correlated with reducing sugar content in ripe fruits, malic acid content of the immature fruit is predicted to be inversely correlated with reducing sugar content of the ripe fruit.

17.11.2.2 Volatiles

A set of 20–30 volatile chemicals positively contribute to tomato flavor; that is, they are present in sufficient quantities to noticeably stimulate the olfactory system. They are a diverse set of chemicals derived from amino acids, fatty acids, and carotenoids. Despite their chemical diversity, they share one important trait; each is derived from some chemical that is essential for the human diet. Thus, volatiles are derived from essential amino acids (phenylalanine, leucine, and isoleucine) and essential fatty acids (principally linolenic acid). □-carotene, the precursor of retinal (Vitamin A) is the immediate precursor of one of the most important flavor volatiles, □-ionone. Plants make the compounds that are attractive to animals and animals, in turn, have evolved to recognize and seek out the compounds that indicate the presence of important nutrients. The content of most of the flavor volatiles in a tomato increases at the onset of ripening and peak either at or shortly before full ripening. This timing suggests that synthesis of flavor volatiles is highly regulated. At least some part of the regulation occurs at the level of transcription. For example, the rate-limiting step for synthesis of several Phe-derived volatiles, including 2-phenylethanol, phenylacetaldehyde, and 1-nitro-2-phenethane, is performed by a small family of aromatic amino acid decarboxylases (AADCs). Expression of several of the genes encoding these enzymes is upregulated during ripening and increased expression of AADC enzymes results in increased metabolic flux into this volatile synthesis pathway. Synthesis of the volatiles that are upregulated during ripening is dependent upon ethylene.

Depending on the fruit type these can manifest as a formation of a dehiscence zone, or through the softening of the fleshy tissue. In each case there is a suite of cell wall-related genes that are up-regulated, and in many instances each is differentially regulated.

17.12 Regulation of ethylene synthesis during fruit development and ethylene signaling

Ethylene is the simplest of plant hormones, consisting of just two carbons and four hydrogens. It is a readily diffusible gas. Ethylene mediates fruit development and ripening as well as organ abscission. Ethylene synthesis is highly regulated. The biosynthetic pathway is simple, consisting of only two enzymes (Fig. 17.11). S-adenosylmethionine (SAM) is converted to 1-aminocyclopropane-1- carboxylate (ACC) by ACC synthase (ACS). ACC is subsequently converted to ethylene by ACC oxidase (ACO) (Fig. 17.11). Formation of ACC is the rate limiting step. ACO is not normally limiting for ethylene synthesis and although induced during ripening, substantial ACO activity is present prior to ripening initiation. A major point of regulation for ethylene synthesis occurs at the level of ACS transcription. On the onset of fruit ripening, multiple ACS genes are activated resulting into increased production of ethylene. Characterization of ripening mutants (*ripening-inhibitor- rin, nonripening- nor and Colorless nonripening- Cnr*) have revealed a network of transcription factors that regulate ethylene synthesis and additional ripening processes. Some of these belong to MADS-box family of transcription factors and they are widely conserved among fruit species.

After production ethylene binds to its ER membrane localized receptors which function as dimers (Fig. 17.11). Ethylene binding requires a copper ion cofactor. Ethylene receptors act as negative regulators of ethylene signaling and upon ethylene binding these proteins get inactivated and thereby permit signaling to proceed. Ethylene binding also causes the inactivation of the associated negative regulators CTR 1 which, in turn, leads toward the activation of a MAP kinase pathway which ultimately results in altered expression of the ethylene regulated ripening genes (Fig. 17.11).

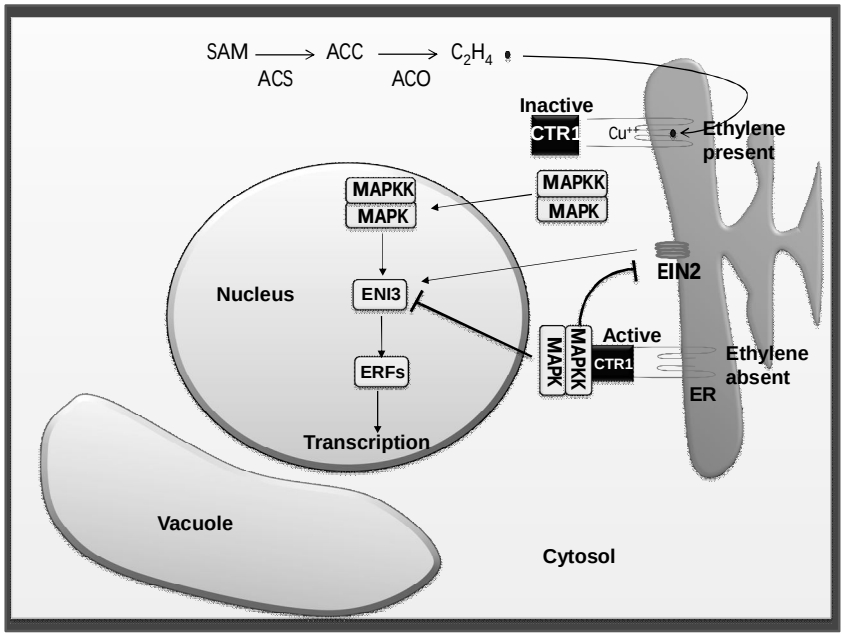

Fig. 17.11: Ethylene signaling.

Ethylene is perceived by receptors localized ER or Golgi (not shown in diagram). These receptors suppress the ethylene responses in the absence of the hormone. Binding of ethylene results in the inactivation of the receptors as well as the associated negative regulatory protein, CTR1.This activates a MAP kinase pathway which facilitates the ethylene response factors (ERFs) through the eventual activation of EIN3. CTR1 (constitutive triple response 1) is a MAPKKK serine/ threonine protein kinase (mitogen-activated protein kinase kinase kinase) and EIN2 (ethylene-insensitive 2) is a channel-like transmembrane protein. EIN3 and ERFs are transcription factors.

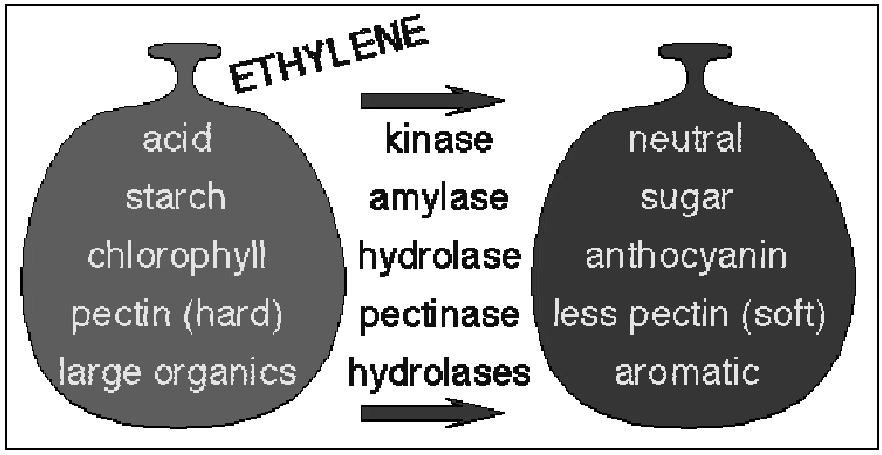

Fig. 17.12: Biochemical changes during fruit ripening

An unripe fruit is hard, green, sour, has no smell, is mealy (starch present Fig. 17.12). The way fruits ripen is that there is commonly a ripening signal-a burst of ethylene production (climacteric fruit). Ethylene is a simple hydrocarbon gas

(H2C=CH2) that ripening fruits make and shed into the atmosphere. This ethylene signal causes developmental changes that result in fruit ripening. New enzymes are made because of the ethylene signal (Fig. 17.12). These include hydrolases to help break down chemicals inside the fruits, amylases to accelerate hydrolysis of starch into sugar, pectinases to catalyze degradation of pectin. Ethylene apparently "turns on" the genes that are then transcribed and translated to make these enzymes. The enzymes then catalyze reactions to alter the characteristics of the fruit. The action of the enzymes causes the ripening responses. Chlorophyll is broken down and sometimes new pigments are made so that the fruit skin changes color from green to red, yellow, or blue. Acids are broken down so that the fruit changes from sour to neutral. The degradation of starch by amylase produces sugar. This reduces the mealy quality and increases juiciness. The breakdown of pectin between the fruit cells separates them. That result in a softer fruit. Also enzymes break down large organic molecules into smaller ones that can be volatile (evaporate into the air) and we can detect as an aroma. Thus ethylene signal causes the fruit to change from green to yellow, from hard to soft, from mealy to juicy, from tart to sweet, from odorless to fragrant. Above mechanism of ethylene mediated fruit ripening has been illustrated in the figure below.

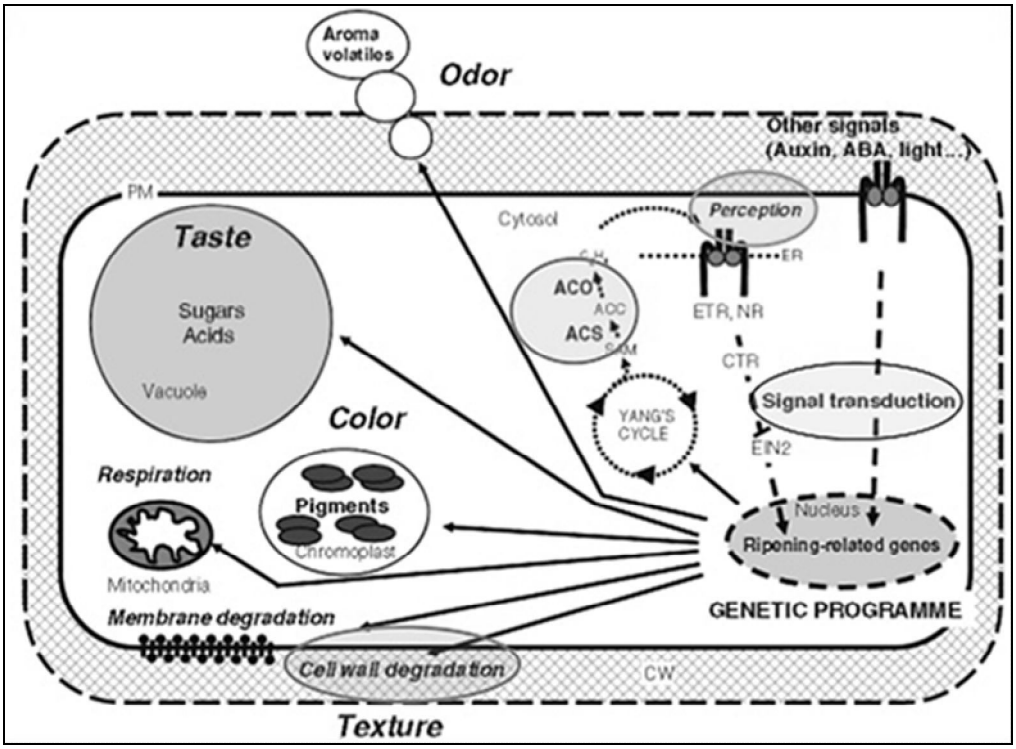

Fig. 17.13: Molecular Mechanisms controlling the ripening of fruit (Source-Open archive TOULOUSE archive ouverte)

The fruit-ripening process described above, also affects a layer of cells in the pedicel near the point of attachment to the stem of the plant. This layer of cells in the pedicel is often called the abscission zone because this layer eventually separates and the fruit drops from the plant. Just as the cells inside the fruit, the cells in this cross sectional layer in the pedicel get the ethylene signal from the ripening fruit. Reception of the signal causes new enzymes to be made. Pectinases enzymes unglue the cells of the abscission zone. The connections between cells become weak-enough that weight of the fruit cause it to fall from the plant. So an animal can pick it up and carry it off to disperse seeds.

17.13 Summary

Endosperm is a nutritive tissue formed after double fertilization. The first step in endosperm development is coenocyte formation, which results from free nuclear divisions. Later, cellularization of the coenocyte is initiated by RMS formation and cell wall deposition and alveoli formation. Following the formation of the first layer of alveoli, mitosis resumes and periclinal cell wall is deposited. Cellularization is completed Apart from free nuclear type of development, two more modes- cellular and helobial are also found in angiosperms. Four types of cells are found in endosperm- Embryo surrounding cells, Transfer cells, Aleurone cells and Starchy endosperm cells.

Embryogenesis can be seen as a hierarchy of events that culminates in the production of a morphologically complex structure. The polarity of the embryo, which may reflect the asymmetric organization of the egg cell, establishes an axis upon which the plant body is elaborated. An early compartmentation of the embryo sets off domains that appear to be involved in establishing the organization of the plant body. Other events organize the embryonic tissue and organ systems and partition progenitors of the shoot and root apices early in embryogenesis.. During embryogenesis, embryo obtains different shapes in different stages. The fundamental processes such as embryonic polarity establishment, pattern formation and establishment of shoot and root apices are similar in embryogenesis in dicots and monocots.

There is a diversity of fruit types among the flowering plants. The fleshy fruit attends its full size but it is green and immature. Ripening starts and makes the fruit attractive and rewarding for an animal to carry it off dispersing the seeds.

17.14 Glossary

- **Coleoptile:** Protective sheath around epicotyl in grasses. A protective sheath for the young shoot in the embryo, usually limited to the poaceae.
- **Embryo:** An embryo is a developing plant still inside the seed. The embryo has cotyledons (embryonic leaves), a root cap, a food source and a plumule (shoot), all located inside the protective seed coat.
- **Endosperm:** The nutritive substance within the embryo sac of the ovule, a food supply in which the embryo is embedded. Food reserve tissue in seed derived from fertilized polar nuclei; or food reserve derived from megametophyte in gymnosperms.
- **Epicotyl:** The epicotyl is the part of the stem that is above the first leaves. The embryonic axis part above the cotyledons. The shoot apical meristem is located here.
- **Fusiform:** Spindle shaped; widest in the middle and tapered to each end.
- **Gynoecium:** The female reproductive structure, formed by fusion of carpels.
- **Pericarp:** The structure developing from the wall of the ovary that protects or encloses the seed or seeds in an angiosperm.
- **Periclinal:** Parallel to a surface.
- **Radicle:** The original root that develops from the germinating seed.
- **Scutellum:** The flattened cotyledon of a monocotyledonous plant embryo

17.15 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. Mention the events which are triggered by double fertilization in angiosperms.
2. Which one is the most common type of endosperm development?
3. …….and …… patterns are established during embryogenesis.
4. ……..cells of endosperm contain anthocyanins pigments and are responsible for the color of the grains.
5. Axial pattern is established during …… stage of embryogenesis.

6. The plant hormone responsible for enlargement of cells ovary wall is……..
7. Name the plant hormone which is necessary for fruit ripening in climacteric fruits.

Section B : (Short Answer Type Questions)

1. Write short note on different types of cells present in endosperm.
2. Describe the structure of monocot embryo.
3. What are four stages of embryogenesis?
4. Differentiate between climacteric and non-climacteric fruits.
5. Write the role of ethylene in fruit ripening.

Section C : (Long Answer Type Questions)

1. Mention about the process of endosperm development and also describe different modes of endosperm development.
2. Write notes on cross-talk among different plant hormones regarding seed germination.
3. Describe the process of embryogenesis in dicot.
4. Describe the biochemistry of fruit ripening and also mention the molecular mechanism of fruit ripening mediated by ethylene hormone.

Answer key of Section – A

1. Embryo, endosperm, seed and fruit developments.
2. Free nuclear endosperm development
3. Radial and axial
4. Aleurone
5. Heart stage
6. Gibberellin
7. Ehylene

17.16 References

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Unit 18

Apomixis, Polyembryony and Parthenocarpy

Structure of the Unit:

18.0 Objectives

. These are the main objectives of the study of this unit:

- Study of different abnormal reproductive process in angiosperms like Apomixis, Polyembryony, Parthenocarpy.
- To know about the many asexual reproductive processes they are substitutes of sexual reproduction.

- To give the knowledge of different conditions they are responsible for abnormality of reproduction.
- To provide the information and importance of these asexual processes of reproduction.

18.1 Introduction

The haploid and diploid phases regularly alternate each other and this is known as alternation of generations. Meosis and syngamy are the two characteristics of sexual reproduction. Meosis and syngamy are supplement to each other and essential in sexual reproduction. In many plants, however, this normal process of sexual reproduction is substituted by an asexual, process. Alternative processes are also utmost significance in embryological studies in angiosperm. There are so many reasons for abnormal reproductive process in angiosperms. Abortion of embryos before attaining maturity of fruits, fruit development due to stimulus received by pollination followed by unsuccessful fertilization, immaturity of gametes and abnormal structure of reproductive parts.

Apomixis, Polyembrony, and Parthenocary have been observed in a large number of taxa in angiosperm. In some plants sexual and asexual processes occur simultaneously. Hence, the asexual processes have great significance in development and evolution of angiosperms. Study of these alternative processes helped in plant breeding and horticulture. These have also thrown light on how the plants grown more and more for better environment.

This unit deals with introduction and knowledge of many reproductive asexual processes in angiosperms like Apomixis, Polyembrony and Parthenocary.

18.2 Apomixis

The normal sexual cycle involves two important processes; first is meiosis and second one is fertilization. In meiosis diploid sporophytic cells transforms in to four haploid gametophytic cells. In fertilization two haploid gametes fuse and establishing the diploid sporophytic generation. Meiosis and syngamy are the two main characteristics of sexual reproduction (or amphimixis). By meiosis the diploid cells of the sporophyte give rise to the haploid gametophytes which bear male and female gametes. Syngamy is the fusion of haploid gametes, results in the restoration of the diploid sporophytic generation. The haploid and diploid phases regularly alternate each other and this is known as alternation of generations. In many plants, however, this normal process of sexual

reproduction is substituted by an asexual, process. This phenomenon of substitution of sexual process by asexual methods is known as apomixis and the plants which show it are called apomictic plants.

According to Winkler (1908), the term apomixis (= away from mixing) refers to the substitution of sexual reproduction. Apomixis may be defined as the substitution for the usual sexual reproduction of a form of reproduction which does not involve meiosis and syngamy. The amphimictic species are not different from apomictic species, the former under certain circumstances may be responsible for apomixis.

18.2.1 Types of Apomixis

Maheshwari (1950) recognised the following three types of apomixes:

(1) Non-recurrent apomixis

(2) Recurrent apomixis

(3) Adventive apomixis

(1) Non-recurrent apomixis

In this type of apomixis, the megaspore mother cell undergoes, normal meiotic division and one of the four megaspores thus develops into haploid female gametophyte (i.e., embryo sac). However, there is no fertilization and the embryo develops either from the unfertilized egg (haploid parthenogenesis) or from some other cell of the embryo sac (haploid apogamy). The embryo, thus formed, is naturally haploid.

1. Haploid parthenogenesis. It is the development of embryo from an unfertilized egg. Jorgensen (1928) has shown that haploid parthenogenesis can be induced in some species of *Solanum* by stimulating but not fertilizing the egg with the pollen of another species. He observed that when the stigma at *Solanum nigrum* was pollinated with the pollen of *S. luteum,* the male nucleus penetrated the egg but there was no effective nuclear fusion and the male nucleus soon began to disintegrate. The egg, however, was activated as if by normal fertilization and an embryo was formed.

In plants like *Orchis maculata, Epipactus latifolia* and *Platenthera chlorantha* pollen tubes enter, the ovule and the egg develops into diploid embryo after fertilization, but occasionally pollen tube does not enter the ovule or enters at a time when it is too late for fertilization (fig18.1 A-b).

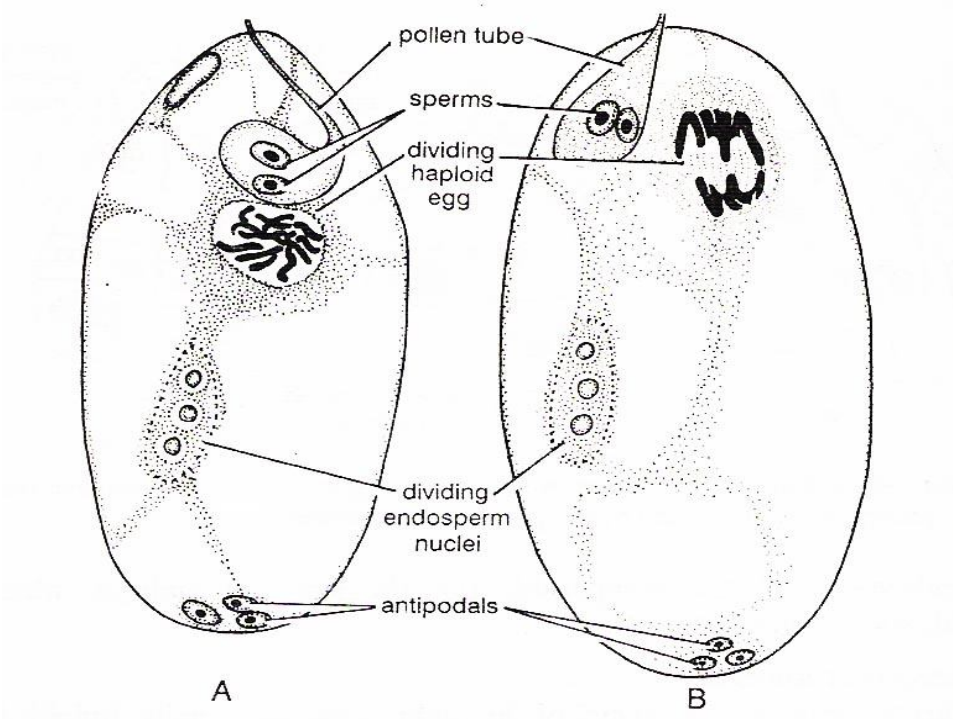

Fig. : A-B. 18.1Haploid parthenogenesis in *Epipactus latifolia*:

A. Division in egg before the release of male gametes from the pollen tube, B. Anaphase of dividing egg before the release of male gametes.

In their event of pollen tube not penetrating the embryo sac, the egg divides to form haploid embryo without fertilization. According to Maheshwari (1950), egg may fail to fertilize due to the following causes:

(i) Non functional pollen tube.

(ii) Inability of the tube to discharge its contents.

(iii) An insufficient attraction between the male and female nuclei.

(iv) An early degeneration of sperms.

(v) Maturation of egg and the entrance of male gametes may not be synchronized.

The occurrence of haploid parthenogenesis may be considerable value in plant breeding and genetics .In this process fertilization is enables to obtain true breeding only obtains homozygous forms.

2. Haploid apogamy. It is development of embryo from any cell of the embryo sac apart from the egg. Twin proembryos have been observed in *Lilium martagon, Bergenia delavayi and Erythraea centaurium.* One of these embryos develops from the egg by normal fertilization and the other from a stimulated

haploid synergid cell (fig18.2A-C). The initial development of the two embryos is closely comparable but the synergid embryo, which is haploid, soon degenerates.

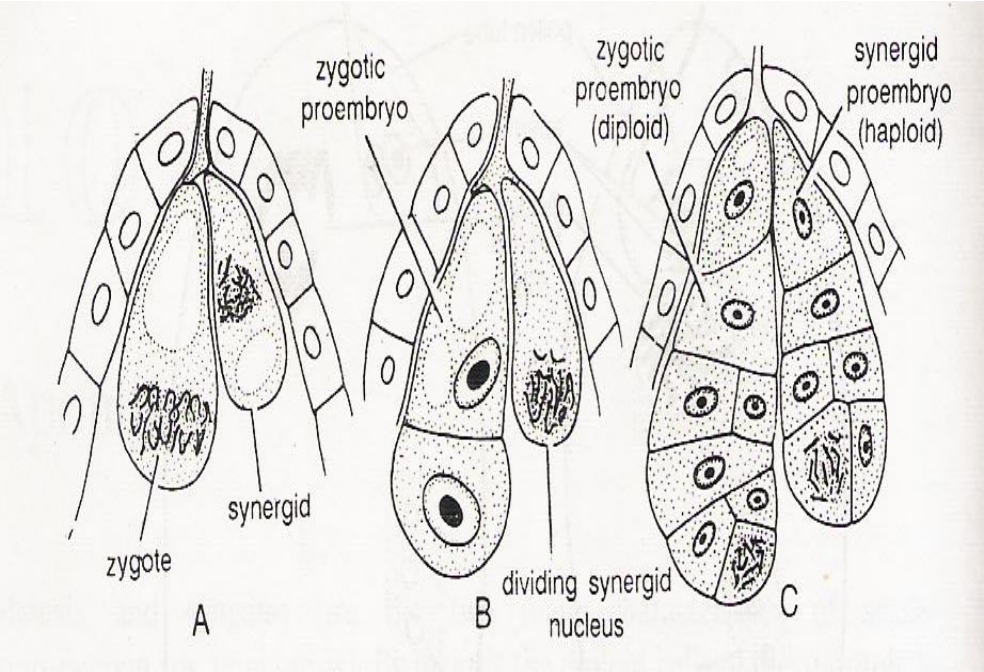

Fig. : 18.2 Haploid apogamy in *Lilium*:
A-B. Dividing egg and synergid; C. Haploid (synergid proembryo) and diploid (zygotic proembryo) proembryos.

(2) Recurrent apomixis

In recurrent apomixis the nuclei of the embryo sac are usually diploid. Such embryo sacs may arise either from a cell of the archesporium (generative apospory) or from some other cell of the nucellus (somatic apospory).

1. Generative apospory

It is the development of diploid embryos from diploid cells of the archesporium. *Parthenium argentatum*, which has two varieties, is the common example of generative apospory. In one of the varieties (2n = 36) the megaspore mother cell undergoes normal meiotic division and eventually forms the female gametophyte. After syngamy and double fertilization there is coordinated development of the embryo and endosperm.

On the other hand, in the other variety (2n = 72), the nucleus of the megaspore mother cell undergoes meiosis but does not give rise to dyad or spore tetrad,

and instead it enlarges and itself functions as an embryo sac. In such embryo sacs the egg develops into embryo without fertilization. In this variety there is only very little or no correlation between the development of embryo and endosperm. Although the development of embryo proceeds normally in the absence of pollination, for the proper development of endosperm pollination is necessary. In the absence of proper development of endosperm, the embryo does not reach maturity.

2. Somatic apospory

Apospory in angiosperms was the first time reported by Rosenberg (1907) in *Hieracium* species. Here the MMC undergoes the usual meiotic divisions and forms tetrad. At this stage a nucellar cell becomes activated and starts developing an embryo sac. The megaspores gradually degenerate and the aposporic embryo sac matures.

The development of diploid embryo sacs from the cell of the nucellus or integument is known as somatic apospory. *Hieracium* is the most common example of somatic apospory. Here the megaspore mother cell goes through usual meiotic division and gives rise to a megaspore tetrad. Soon after the meiotic division is over, a somatic cell situated at the chalazal end of the ovule becomes enlarged and vacuolated. It encroaches upon the megaspore and eventually forms an aposporic embryo sac (Fig. 18.3A,B). As the development of aposporic embryo sac does not involve meiosis, it is diploid. The unfertilized eggs of such embryo sacs give rise to diploid embryos.

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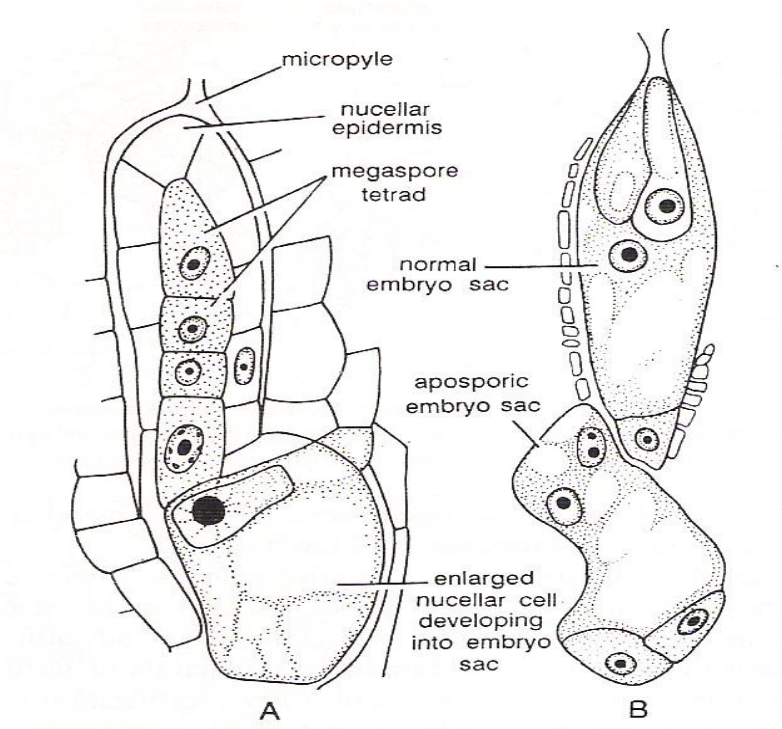

Fig. : A-B 18.3 : Development of aposporic embryo sac in *Hieracium*

Apospory has been reported in many plant species like *Parthenium* and *Rubus*. In the grasses, on the other hand, more then one aposporic embro sac may develop in the same nucellus. The organization of the mature sac is 4-nucleated. Aposporus embryo sacs may co-exist with the sexual embryo sac formed by a haploid megaspore. Aposporic embryo sac may develop from the cells of the nucellar epidermis or integuments. Besides *Hieracium*, such embryo sacs have also been observed in *Malus, Crepis, Ranunculus*, etc.

(3) Adventive embryony

The development of embryo from any diploid cell of the ovule lying outside the embryo sacs is referred to as adventive embryony. It is also called sporophytic budding. In this process there is no alternation of generation.

The cells of the nucellus or integuments, forming adventive embryo, become densely protoplasmic and divide actively to form a small mass of meristematic cells. This tissue mass grows actively, pushes it's to the embryo sac and eventually gives rise to what appears to be an embryo (Fig.18. 4A-C). The zygotic as well as the adventive embryo may grow simultaneously in the same embryo sac. The normal (zygotic) embryo has a suspensor, whereas in the nucellar adventive embrye suspensor is, absent (Fig.18.4 4C).

Citrus is the most common example of adventive embryony. As many as ten viable embryos may be found in a single seed of *Citrus*. Adventive embryony is also of common occurrence in the members of the Euphorbiaceae, Cactaceae, Buxaceae, and Orchidaceae.

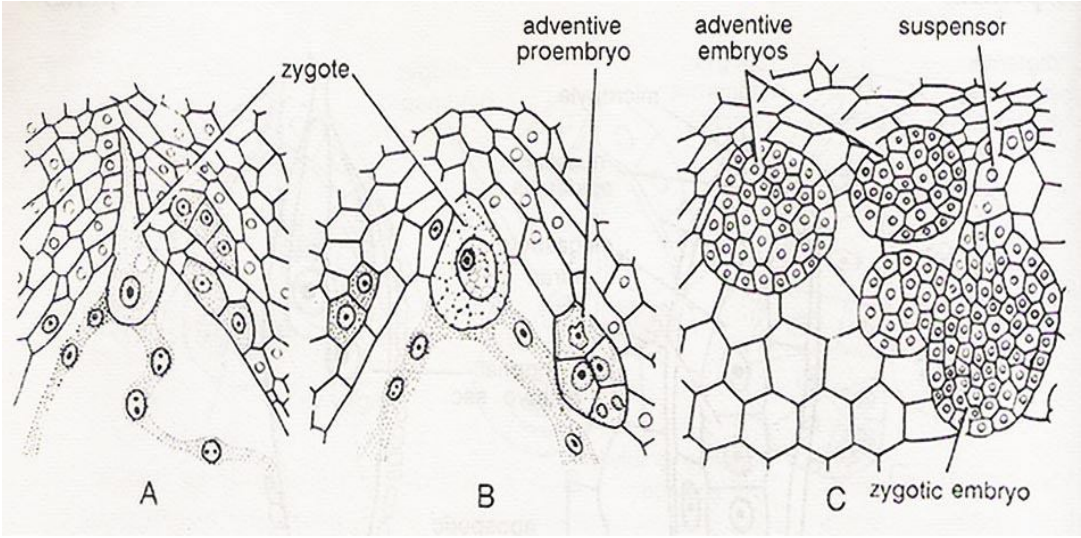

Fig. : A-C 18.4 : Development of adventive embryos in *Citrus:*

A. Differentiation of some embryonic cells in necellus, B. Development of adventive proembryos, C. Zygotic embryo (with suspensor) and adventive embryos (without suspensor)

According to Maheshwari (1950), adventive embryos originate from the cells of the nucellus if it is intact. But if the nucellus is disorganised, they may originate in the integument. The development of adventive embryos may take place with or without the stimulus of pollination or fertilization. *In Mangifera indica* and most of the species of *Citrus*, fertilization is necessary the maturation of adventive embryos. But in *Eugenia jambos* adventive embryos originate without pollination, however, their full development is attained only if the egg is fertilized.

18.2.2 Genetics of Apomixis

Parthenium argentatum, which has two varieties, is the common example of generative apospory. In one of these varieties (2n= 36) the megaspore mother cell undergoes normal meiotic division and eventually forms the female gametophyte. After syngamy and double fertilization there is coordinated development of the embryo and endosperm. On the other hand, in the other variety (2n = 72), the nucleus of the megaspore mother cell undergoes meiosis but does not give rise to dyad or spore tetrad, and instead it enlarges and itself functions as an embryo sac.

Most of the polyploid forms of *Parthenium argenatum* are obligate apomics, and the diploid individuals are sexually reproducing. Experimental studies have indicated that three pairs of genes determine the breeding behavior of individual plants of the species. In homozygous condition, gene 'a' leads to the formation unreduced egg, gene 'b' prevents fertization and gene 'c' causes the egg to develop without fertilization. Plants with genetic constitution aaBBCC from unreduced eggs but these cannot develop into embryos without fertilization. Plants with the genotype AAbbCC produce reduced eggs but embryos are not formed because fertilization is prevented. Those with AABBcc genotype have a normal sexual behavior. The gene c has no effect in the presence of A and B since the eggs are reduced and fertilization takes place. Only those plants with as aabbcc genetic condition would be apomictic. In facultative apomicts, environmental conditions seem to play a key role in the shift from sexual mode reproduction to apomixes.

18.2.3 Significance of Apomixis

1. The plants where the usually sexual reproduction has been completely replaced by a type of asexual reproduction are called apomitic, and the phenomenon is known as apomixes. As apomixis does not involve meiosis, there is no segregation and recombination of chromosomes. Apomixes is controlled by recessive genes.
2. Apomixis offers the possibility indefinite multiplication of especially favorable biotypes without any variation due to segregation or recombination.
3. Thus it is useful in preserving desirable characters for indefinite periods. But- the importance of meiosis in evolution and variation cannot be ignored.
4. In obligate apomictic species though desirable characters are preserved for quite a long time, they are deprived of evolution.
5. On the contrary, in facultative apomictic species, sexual and asexual processes occur simultaneously and hence there is great significance of apomixis.
6. However, in facultative apomicts or group of plants where sexual and apomictic members co-exist, the phenomenon is of special significance.

18.3 Polyembryony

Polyembrony has been defined as the occurrence of more than one embryo in a seed. Polyembrony includes there is a clear indication of the actual occurrence

of two or more pro-embryos or embryos in a developing ovule. Few taxa (*Citrus* and *Mangifera*), polyembryony occurs only as an abnormal feature.

After fertilization, ovules mature into seeds. Normally, a single embryo is present in each seed. But sometimes more than one embryo is present in a seed. This condition is known as polyembryony. In nature, there are many plants whose seeds have more than one embryo. But in such plants normally only one embryo matures and the rest degenerate during the course of seed development. Thus the mature seed has only one embryo. Strictly speaking, polyembryony includes instances where there is actual occurrence of two or more proembryos or embryos in a developing ovule, irrespective of the fact that only one succeeds to mature and the rest degenerate. Polyembryony was first time reported by Antony van Leeuwenhoek in 1719 in the seeds of orange. Since then it has been observed in a large number of taxa of angiosperms.

Origin of Polyembryony

On the basis of its origin, the following four types of polyembryony have been recognized in angiosperms:

1. Cleavage polyembryony, which develops due to splitting or cleavage of the pro-embryo.
2. Formation of embryos by cells of the embryo sac other than the egg.
3. Development of more than one embryo sac within the same ovule.
4. Development of embryo sac from any sporophytic cell of the ovule.

18.3.1 Cleavage Polyembryony

It is the common and simplest method of the development of more then one embryo in a seed. Two or more embryos are formed in a seed by the cleavage of the zygote or proembryo. Cleavage polyembryony is of widespread occurrence in the gymnosperms, but it is rare in the angiosperms. It was first time observed by Jeffery (1895) in *Erythronium americanum* (Liliaceae), and subsequently reported in several other taxa. Cleavage polyembryony develops due to splitting and proliferation of any sporophytic cell of the ovule.

Cleavage polyembryony may arise in different ways. In *Erythronium americanum*, the first division of the zygote is normal and as a result a basal and an apical cell are formed. Repeated divisions of the basal cell give rise to an irregular mass of cells which is known as embryonic mass (fig. 18.5 A). Several cells at the distal end of the embryonic mass develop into separate embryos (fig.18.5B, C). This type of cleavage polyembryony has also been

reported in *Limnocharis emarginata, Tulipa gesneriana.* In *Isotoma Longiflora* (Campanulaceae) and *Exocarpus* (Santalaceae), additional embryos are developed from the suspensor cells of the polymbryony. The formation of plural embryos during seed germination is known only in vanda.

Cleavage polyembryony is also fairly common in orchids. In *Eulophia epidendraea*, reported three different modes of cleavage polyembryony

1. A mass of cells formed by irregular divisions of the zygote and several cells of this mass present at the chalazal end develop into embryos (fig. 18.5A)
2. The pro-embryo gives rise to small bud like outgrowths which develop into supernumerary embryo (Fig. 18.5B).
3. The filamentous pro-embryo becomes branched, and an embryo develops at the tip of each branch.

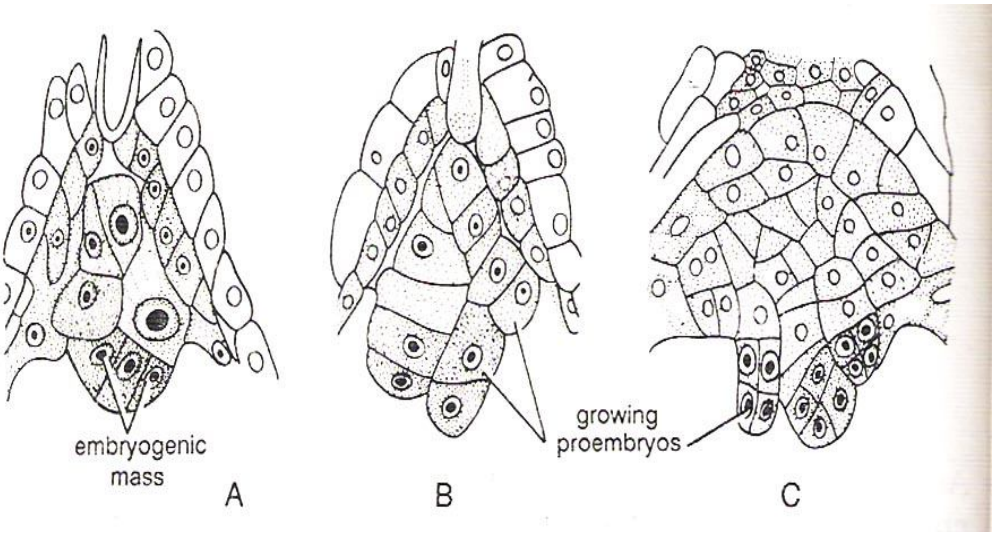

Fig. 18.5 : Cleavage polyembryony :

A Embryonic mass formed by the basal cell of the zygote in *Erythronium americamum,*

B-C. Differentiation of embryos from the cell of the embryonic mass.

[II] Origin of embryo from cells of the embryo sac than the egg cell

Here additional embryos originate from cells of the embryo sac other than the egg cell. Commonly, the additional embryos develop from synergids. In *Argemone mexicana* and *Phaseolus vulgaris,* these embryos develop from unfertilized synergids and hence they are haploid in nature. In *Sagittaria*

graminea, *Aristolochia bracteata* and *Crepis capillaris*, one or both the synergids may get fertilized by the entry of more than one pollen tube into the embryo sac or by the presence of one or more male gametes in the same pollen tube.

Suspensor polyembyony is a common feature in the genus *Exocarpous*, a member of santalaceae. As many as six embryos may develop simultaneously in an ovule by the proliferation of suspensor cells. However, only one of them takes the lead and reaches maturity.

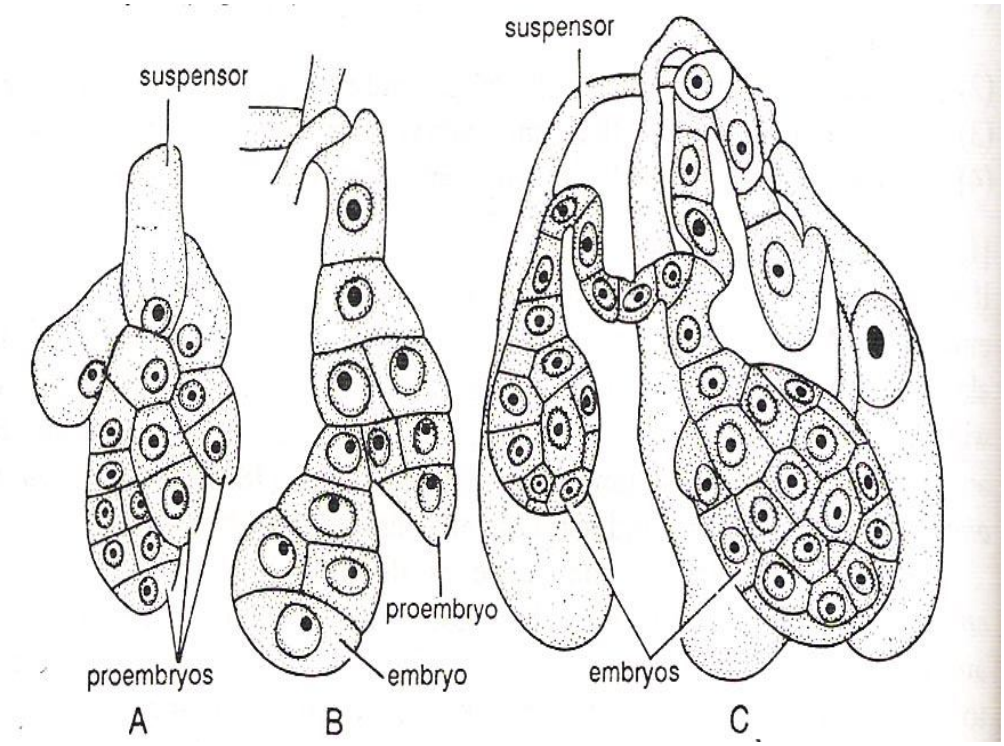

Fig. 18.6 : A-C. Cleavage polyembryony :

A Embryonic mass formed by the division of the zygote in *Erythronium edipendraea*, B. A proembryo formed in addition to the normal embryo, C. Two embryos formed by the splitting of a single embryo.

Through the formation of embryos from antipodals is of rare occurrence, such embryos have been reported from *Allium odorum, Paspalum scrobiculatum, Sedum fabaria and Ulmus americana,* etc. However, it is not certain whether such embryos are viable or not viable (fig.18.7 A).

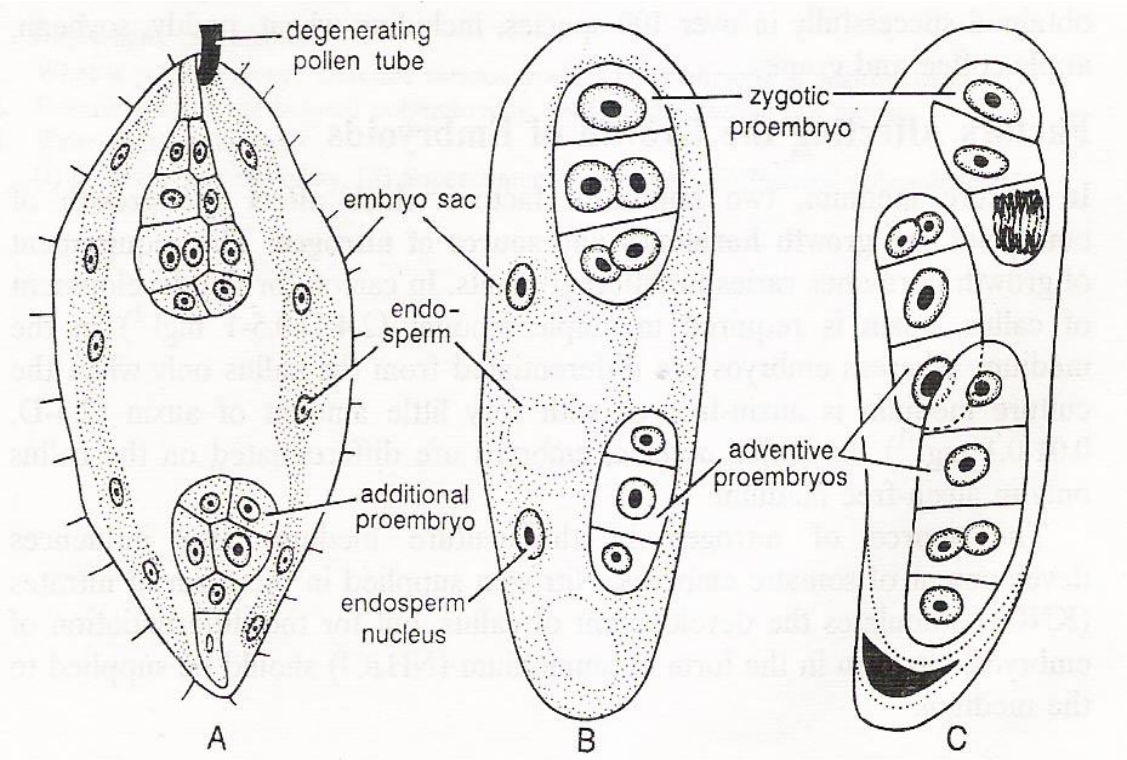

Fig. 18.7: Polyembryony : A Development of embryo from antipodal cells. B-C. Adventive pro-embryos developed from the cells of the nucellus (they grow along with the zygotic embryos).

[III] Embryos developing from additional embryo sacs in an ovule

Sometimes additional embryo sacs are present in the ovule, and fertilization of their eggs results in the formation of extra embryos. Multiple embryo sacs in an ovule may arise:

1. By differentiation of more than one megaspore mother cells in the ovule as in *Hydrilla verticelata, Solanum melongena, Jussiaea repens* and *Casuarina montana.* Formation of twin embryo sac within an ovule is found in *Casurina* and *Citrus.*
2. By activation of two or more spores out of the four megaspores formed by the reduction division of the megaspore mother cell, as in Rosa .
3. From the cell of the nucellus (e.g., *Citrus, Mangifera, Opuntia*) or integument (e.g., *Limnanthes*).

[IV] Activation of some sporophytic cells of the ovule

The embryos arising from the maternal sporophytic tissue are called adventive embryos. The embryos developed from nucellus or integument is known as adventive embryos. Since the development of such embryos is not synchronous, embryos in various stages of development can be seen in the same ovule.

Studies on *Citrus* clearly indicate that the origin of adventive embryos is not affected by pollination, fertilization or the development of zygotic embryo, but the development of endosperm plays an important role. In the absence of endosperm, nutrients become scarce and proper development of adventive embryos does not take place. Some species of *citrus* are monoembryonate while others are polyembryonate. In polyembyonate species the adventive embryos arise from micropylar half of the nucellus.

Nucellar polyembrony occur in *Opuntia*. In the *Opuntia* the egg apparatus, antipodals and polars invariably degenerate, several adventive embryos develop from the nucellar cell. However, usually only one of them attains maturity. Nucellar embryos can be distinguished from the zygotic embryos by their lateral position in the embryo sac, irregular shape and lack of suspensor.

18.3.2 Classification of Polyembryony

Mostly polyembryony is of two types -

1. Spontaneous: Includes instances of naturally occurring polyembrony.
2. Includes instances of experimentry polyemer.

Spontaneous polyembry has been subdivided into two categories:

A-True polyembryony : Two or more embryos arising in the same embryo sac. The zygote or embryo develops from synergides, from antipodal cells or from nucellus or integuments.

B-False polyembryony : Development of embryos in more than one embryo sac in the same ovule or placenta.

Spontaneous polyembrony divide on the basis of genetics

A-Gametophytic : Arising from any haploid cell of the embryo sac after or without fertilization.

B-Sporophytic : Arising from the zygote, proembryo or the initial sporophytic celled of the ovule (nucellus or tntegument).

II Induced Polyembryony

Induced polyembryony is includes instances of experimentally polyembrony. In nature polyembryony arises spontaneously and additional embryos are developed only from the ovular tissue like embryo sac, nucellus and integuments. Earlier, it was considered that a specific physical and chemical environment is required for the development of the embryo which is available only in the embryo sac. But the recent studies clearly indicate that not only the

ovular tissue but all the cells of a plant are capable to develop into embryos, and embryo development can also be carried out in culture medium. The embryos developed in culture medium are known as adventitious embryos, somatic embryos, supernumerary embryos or embryoids.

Besides zygote and ovular tissue, embryoids can also be obtained from the cells of the root, leaf, fruit, pollen grain, etc. Somatic embryos have been obtained successfully in over 100 species, including wheat, paddy, soybean, apple, coffee and grapes.

Factors Affecting the Growth of Embryoids

In culture medium, two important factors which affect the growth of embryoids are growth hormones and source of nitrogen. The requirement of growth hormones varies in different plants. In carrot, for the development of callus, auxin is required in ample amount (2,4-D, 0.5-1 mgl^{-1}) in the medium. Whereas embryos are differentiated from the callus only when the culture medium is auxin-free or with very little amount of auxin (2,4-D, 0.01-0.1 mgl^{-1}). A *Coffea arabica*, embryos are differentiated on the callus only in auxin free medium. Some studies on carrot clearly indicate that a minimum level of endogenous and exogenous auxin is necessary for in vitro embryogenesis.

The source of nitrogen in the culture medium also influences development of somatic embryos. Nitrogen supplied in the form of nitrates (KNO_3) stimulates the development of callus, but for them differentiation of embryos, nitrogen in the form of ammonium (NH_4Cl) should be supplied to the medium.

Causes of Polyembryony

Several theories have been to explain the occurrence of polyembryony. Two important views are given below:

[I] Necrohormone theory

This theory, proposed by Haberlandt (1921, 1922) and states that degenerating cells of the nucellus act as source of stimulus to the adjacent cells to divide and form adventive embryos. But no clear evidence is available to show that adventive embryony is induced by the secretary substances of the dying cells.

[II] Recessive gene theory

In Linum, Kappert (1933) has shown that polyembryony is a recessive genetic character, controlled by multiple genes. Due to hybridization in different strains, genes recombine in an individual plant and this exhibits the

phenomenon of polyembryony. Leroy (1947) also considered that in *Mangifera indica* polyembryony is caused by one or more recessive genes.

18.3.3 Importance of Polyembryony

Role of Polyembryony in Plant Breeding and Horticulture

Adventive polyembryony is of great significance in horticulture and plant breeding. It provides uniform seedlings of the parental type, as obtained through vegetative. Moreover, the nucellar embryos are free from diseases.. Adventive embryos are also of utmost significance in morphogenetic studies. Haploid are of great economic importance in genetics and plant breeding studies because of the ease with which homozygous diploid can be obtained from them by colchicines treatment.

Nucellar seedlings of plants furnish better clones of orchard stock than cuttings. This is because of the following reasons:

1. The nucellar seedlings have a tap root and therefore, develop a better root system than do the cuttings. The latter have only small lateral root system.
2. The nucellar embryos are free from disease. So far nucellar polyembryony is the only practical approach to raising virus free clones of polyembryonate verities in nature. Nucellar polyembryony is thus the only practical approach to raise virus-free clones of polyembryonate Citrus varieties in nature. For monoembryonate cultivars of citrus there is no *in-vivo* method for raising virus free clones. However, it can be achieved by culturing their nucellus and including embryoid formation.

18.3 Parthenocarpy

Parthenocarpy means the development of fruits without pollination or any other stimulus. According to Noll (1902), who introduce the term parthenocarpy. Parthenocarpy is of widespread occurrence, especially among species which have a large numbers of ovules per ovary, such as banana, pine apple and others. The definition of parthenocarpy has undergone slight modification, and according to the present concept it refers to “the formation of fruits without fertilization” (Nitch, 1965).

It means formation of fruits with seed (seedless fruit). Such fruits have no advantage to plants but may be of great interest and commercial use for human beings. “Seedless fruits” should not be considered synonymous to

"parthenocarpic fruits" because in a seedless fruit the ovule have been fertilized and later degenerated, as in some strains of *Vitis vinifera* var. They are may be parthenocapic fruits with seed them.

It is also a natural process as in the case of bananas. Bananas are sterile and develop no viable ovaries.They do not produce seeds, which mean they must propagate vegetatively. Pineapples and figs are also examples of parthenocarpy which occur naturally.

This situation of parthenocarpy in plants can occur in two types, vegetative and stimulative parthenocarpy. Some parthenocarpic cultivars are of ancient origin. The oldest known cultivated plant is a parthenocarpic fig first grown at least 11,200 years ago.

Parthenocarpy in plants is a relatively unusual condition but it does occur in some of our most common fruit. This circumstance occurs when the ovary of a flower develops into a fruit without fertilization. The result is a seedless fruit.

18.4.1 Main causes of Parthenocarpy

1. Parthenocarpy (or stenospermocarpy) occasionally occurs as a mutation in nature; if it affects every flower the plant can no longer sexually reproduce but might be able to propagate by apomixis or by vegetative means.
2. Fruit development without fertilization occurs in banana.
3. Abortion of embryos before attaining maturity of fruits.
4. Fruit development due to stimulus received by pollination followed by unsuccessful fertilization.
5. Induction of parthenocarpy has been found by the use of hormones such as auxins, gibberellins and cytokinins.

18.4.2 Types of Parthenocarpy

Nitch ,1963 has recognized three types of pathenocarpy namely

1. Genetical parthenocarpy

Many of the plants cultivated for their fruits show seeded as well as parthenocarpic varities. This type of parthenocarpy is known to arise due to mutation or hybridization. The famous navel orange arose from a normal seeded variety through mutation in an axillary bud which grew out into a branch bearing seedless oranges. Some parthenocarpic varities have been developed as genetically modified organisams.

2. Envorionmental parthenocarpy

Variation in environmental conditions such as frost, fog, and low temperature affect the normal functioning of sexual organs and bring parthenocarpy. Botanist observed that a heavy fog in the month of June caused the formation of seedless olives. Osborne and Went (1953) were able to induce parthenocarpy in tomatoes with low temperature and high light intensity.

3. Chemically induced parthenocarpy

Auxins and gibberellins at low concentration have been successfully used to induce parthenocarpy in a number of plants which normally bear seeded fruits. These substances are applied to flowers in the form of a lanoline paste or its sprays. This process is more convenient for commercial purpose. Growth regulating hormones, when used on crops, also halt the fertilization process. When sprayed on flowers any plant hormones gibberellin, auxin and cytokinin could stimulate the development of parthenocarpic fruit. This is termed **artificial parthenocarpy or** chemically induced parthenocarpy. Auxin treatments are known to produce seeded parthenocarpic fruits in citrus and grapes. The seeds in such fruit are really pseudo-seeded because lacking a sexual embryo. Gibberelic acid has been reported to induce parthenocarpy in a number of rosaceous fruit trees, grapes and tomatoes.

Plant hormones are seldom used commercially to produce parthenocarpic fruit. In cultivated plants, parthenocarpy is introduced with plant hormones such as gibberellic acid. It causes ovaries to mature without fertilization and produces bigger fruits. This process is being introduced to all kinds of crops.

Stimulative parthenocarpy

In some plants, pollination or other stimulation is required for parthenocarpy. This is termed **stimulative parthenocarpy**. Stimulative parthenocarpy is a process where pollination is required but no fertilization takes place. It occurs when a wasp inserts its ovipositor into the ovary of a flower. It can also be simulated by blowing air or growth hormones into the unisexual flowers found inside something called a syconium. The syconium is basically the flask-shaped structure lined with the unisexual flowers.

Vegetative parthenocarpy

Plants that do not require pollination or other stimulation to produce parthenocarpic fruit have **vegetative parthenocarpy**. Seedless cucumbers are

an example of vegetative parthenocarpy, seedless watermelon is an example of stenospermocarpy. One can imagine annoyance caused by seeds while eating a watermelon. Parthenocarpy may also increase proportion of edible part of the fruits.

18.4.3 Importance of Pathenocarpy

It's important in economically because seedless fruits are consider as good quality and farmers get high value. However, parthenocarpy of some fruits of a plant may be great value. Seedless ness is a very desirable trait in edible fruit with hard seeds such as pineapple, banana, orange and grapefruit.

1. Parthenocarpic seeds have importance in horticulture because seedless fruit are ideal for jam and juice industries.
2. Parthenocarpy may also increase proportion of edible part of the fruits.
3. Parthenocarpy allows the grower to keep insect pests from his crop without chemicals. This is because no pollinating insect is required for fruit formation so the plants can be covered to prevent the bad insects from attacking the crop.
4. In the world of organic production, this is a significant improvement from the use of even organic pesticides and improves crop yield and health. Fruits and vegetables are bigger, the growth hormones introduced are natural and the results are easier to achieve and more healthful.
5. In dioecious species parthenocarpy increases fruit production because unisexual trees do not need to be planted to provide pollen.
6. Parthenocarpy of some fruits on a plant may be of value. Up to 20% of the fruits of wild are parthenocarpic.
7. The ability to produce seedless fruit when pollination is unsuccessful may be an advantage to a plant because it provides food for the plants.
8. Parthenocarpy is also desirable in fruit crops that may be difficult to pollinate or fertilize, such as tomato and summer squash.
9. Plants moved from one area of the world to another may not always be accompanied by their pollinating partner and the lack of pollinators has spurred human cultivation of parthenocarpic varieties. Some parthenocarpic varieties have been developed as genetically modified organisms

Fig. 18.8 : A seedless watermelon as parthenocaepic fruit

18.5 Summary

The plants where the usual sexual reproduction has been completely replaced by a type of asexual reproduction are called apomitic, and the phenomenon is known as Apomixis. Apomixis may be found in the hybrid origin of the species and polyploid plants. Apomixis may occur due to the disturbance and the failure of synopsis and normal contraction of the chromosomes.

Apospory is the part of the apomixis. In this situation a somatic cell in the nucellus directly forms an unreduced emryosac, and the diploid egg partheno-genetically develops in to embryo. The apomitic embyosac may develop in addition to the haploid embryo sac from a functional MMC.

Types of apomixes are: Recurrent- Its include vegetative propagation and agamospermy and Non-recurrent –It's include where egg or some other cells of the haploid embryos sac develops in to embryo without fertilization.

As apomixis does not involve meiosis, there is no segregation and recombination of chromosomes. Apomixes is controlled by recessive genes. Apomixis offers the possibility indefinite multiplication of especially favorable biotypes without any variation due to segregation or recombination.

In nature polyembryony arises spontaneously and additional embryos are developed only from the ovular tissue like embryo sac, nucellus and integuments. Earlier, it was considered that a specific physical and chemical environment is required for the development of the embryo which is available only in the embryo sac. But the recent studies clearly indicate that not only the ovular tissue but all the cells of a plant are capable to develop into embryos, and embryo development can also be carried out in culture medium. The embryos developed in culture medium are known as adventitious embryos, somatic embryos, supernumerary embryos or embryoids.

Besides zygote and ovular tissue, embryoids can also be obtained from the cells of the root, leaf, fruit, pollen grain, etc. Somatic embryos have been obtained successfully in over 100 species, including wheat, paddy, soybean, apple, coffee and grapes.

Parthenocarpy in plants is a relatively unusual condition but it does occur in some of our most common fruit. This circumstance occurs when the ovary of a flower develops into a fruit without fertilization. The result is a seedless fruit.

This situation of parthenocarpy in plants can occur in two types, vegetative and stimulative parthenocarpy. In botany and horticulture, **parthenocarpy** (literally meaning virgin fruit) is the natural or artificially induced production of fruit without fertilization of ovules. The fruit is therefore seedless. Stenospermocarpy may also produce apparently seedless fruit, but the seeds are actually aborted while still

18.6 Glossary

- **Apomixis** - The plants where the usual sexual reproduction has been completely replaced by a type of asexual reproduction are called apomitic, and the phenomenon is known as Apomixis.
- **Fertilization** - In fertilization two haploid gametes fuse and establishing the diploid saprophytic generation.
- **Amphimixis** - Sexual reproduction is known as amphimixis.
- **Haploid parthenogenesis** - It is the development of embryo from an unfertilized egg.
- **Haploid apogamy**- It is development of embryo from any cell of the embryo sac apart from the egg.
- **Adventive embryony** - The development of embryo from any diploid cell of the ovule lying outside the embryo sacs is referred to as adventive embryony.
- **Somatic apospory** - The development of diploid embryo sacs from the cell of the nucellus or integument is known as somatic apospory.
- **Polyembryony**- Sometimes more than one embryos are present in a seed. This condition is known as polyembryony.

- **Adventive embryos** - The embryos arising from the maternal sporophytic tissue are called adventive embryos.
- **True polyembryony** - Two or more embryos arising in the same embryo sac.
- **False polyembryony** - Development of embryos in more than one embryo sac in the same ovule or placenta.
- **Induced polyembryony** - Includes instances of experimentally polyembrony.
- **Parthenocarpy** -The formation of fruits without fertilization
- **Stimulative parthenocarpy -**Pollination or other stimulation is required for parthenocarpy.
- **Genetical parthenocarpy**- This type of parthenocarpy is known to arise due to mutation or hybridization
- **Induced parthenocaepy** - Growth harmones could stimulate the development of parthenocarpic.
- **Vegetative parthenocarpy-** Plants that do not require pollination or other stimulation to produce parthenocarpic fruits.

18.7 Self -Learning Exercise

Section A : (Very Short Answer Type Questions)

1. Define the apomixes.
2. What is polyembryony ?
3. What is Parthenocarpy ?
4. Give the example of three parthenocarpic fruit.
5. Who Introduce the term parthenocapy?
6. Define vegetative parthenocarpy.
7. What is Apospory ?

Section B : (Short Answer Type Questions)

1. Write the four significances of apomixes.
2. Explain the role of polyembryony in Plant breeding and horticulture.
3. What are the main causes of polyembryony?
4. Explain the cleavage polyembryony.
5. What do you understand by Induced polyembryony?

6. Describe the different types of pathenocarpy.

Section C : (Long Answer Type Questions)

1. What is Apomixis ? How is different from the normal sexual reproduction ?
2. Desccribe various apomitic methods found in angiosperm.
3. Write short noted on:
   A. Recurrent apomixes
   B. Non recurrent apomixes
   C. Definition and origin of polyembryony
   D. Induced pathenocarpy
4. Describe various type of polyembryony in angiosperm.
5. Describe briefly the induced polyembrony and factor affecting this process.
6. Expalin the different type of parthenocarpy.

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Unit-19

Latent Life-Dormancy

Structure of the Unit:

19.0 Objectives

A dormant seed is one that is unable to germinate in a specified period of time under a combination of environmental factors that are normally suitable for the germination. Dormancy is a mechanism to prevent germination during unsuitable ecological conditions, when the probability of seedling survival is low. Following are the main objectives of the study of seed dormancy:

- Study of the seed structure.
- To correlate the environmental factors with seed germination.
- To know Different types of seed dormancy

- To provides knowledge about different methods to overcome seed dormancy.

19.1 Introduction

If the three normal conditions necessary for germination (i.e., necessary oxygen, water and temperature) are available, the seeds of most plants germinate. But in some plants (e.g. *Xanthium, Ginkgo* and *Cucurbita*) the seed do not germinate in spite of the availability of these three necessary conditions. Such seeds are called dormant seeds and this phenomenon is called seed dormancy.

Seed germination can be defined as the resumption of growth of the embryo of the mature seed. It depends on the same environmental conditions as vegetative growth does. Water and oxygen must be available, the temperature must be suitable, and there must be no inhibitory substances present.

In many cases a viable (living) seed will not germinate even though all the necessary environmental conditions for growth are satisfied. This phenomenon is termed seed dormancy. In a large number of seeds geminate cannot take place immediately after harvesting on account of the innate inhibition. This innate inhibition of germination of a viable seed is termed as seed dormancy. Dormancy is also called rest by certain botanists. Another term used in connection with seed is quiescence. During seed maturation, the embryo enters a quiescent phase in response to desiccation. Quiescence is the inability of a viable seed to germinate because important environmental conditions required.

19.2 Types of Seed Dormancy

Dormancy is best defined as temporary suspension of active growth. The cause of this temporary inactivation may be unfavorable environmental conditions or some internal exigency. The former type is known as quiescence while the later is known as true or innate dormancy.

Often seed dormancy is divided into two major categories based on what part of the seed produces dormancy: exogenous and endogenous.

19.2.1 Exogenous Dormancy

Exogenous dormancy is caused by conditions outside the embryo and is often broken down into three subgroups:

Physical Dormancy

Dormancy that is caused by an impermeable seed coat is known as physical dormancy. Physical dormancy is the result of impermeable layers that develops during maturation and drying of the seed or fruit. This impermeable layer prevents the seed from taking up water or gases. As a result, the seed is prevented from germinating until dormancy is broken. In natural systems, physical dormancy is broken by several factors including high temperatures, fluctuating temperatures, fire, freezing/thawing, drying or passage through the digestive tracts of animals. Physical dormancy has not been recorded in any gymnosperms.

Generally, physical dormancy is the result of one or more palisade layers in the fruit or seed coat. These layers are lignified with malphigian cells tightly packed together and impregnated with water-repellent. In the Anacardiaceae and Nelumbonaceae families the seed coat is not well developed. Therefore, palisade layers in the fruit perform the functional role of preventing water uptake. While physical dormancy is a common feature, several species in these families do not have physical dormancy or produce non-dormant seeds.

In nature, the seed coats of physically dormant seeds are thought to become water permeable over time through repeated heating and cooling over many months-years in the soil.

Mechanical Dormancy

Mechanical dormancy occurs when seed coats or other coverings are too hard to allow the embryo to expand during germination. This mechanism of dormancy was ascribed to a number of species that have been found to have endogenous factors for their dormancy instead. These endogenous facts include physiologically dormancy cased by low embryo growth potential

Chemical Dormancy

Includes growth regulators and chemicals are present in the coverings around the embryo. They may be leached out of the tissues by washing or soaking the seed, or deactivated by other means. Other chemicals that prevent germination are washed out of the seeds by rainwater or snow melt.

19.2.2 Endogenous Dormancy

Endogenous dormancy is caused by conditions within the embryo itself. This type of seed dormancy is embryo dormancy, which refers to a dormancy that is inherent in the embryo and is not due to any influence of the seed coat or other

surrounding tissues. In some cases, embryo dormancy can be relieved by amputation of the cotyledons. A fascinating demonstration of the cotyledon's ability to inhibit growth is found in species (e.g., peach). In some species the isolated dormant embryos germinate but grow extremely slowly to form a dwarf plant. If the cotyledons are removed at an early stage of development, however, the plant abruptly shifts to normal growth.

Endogenous dormancy broken down into three subgroups: physiological dormancy, morphological dormancy and combined dormancy, each of these groups may also have subgroups.

Physiological Dormancy

Physiological dormancy prevents embryo growth and seed germination until chemical changes occur. These chemicals include inhibitors that often retard embryo growth to the point where it is not strong enough to break through the seed coat or other tissues. Physiological dormancy is indicated when an increase in germination rate occurs after an application of gibberellic acid (GA3). It is also indicated when dormant seed embryos are excised and produce healthy seedlings when up to 3 months of cold (0–10°C) or warm (=15°C) stratification increases germination. In some seeds physiological dormancy is indicated when scarification increases germination.

Embryo dormancy is thought to be due to the presence of inhibitors, especially ABA, as well as the absence of growth promoters, such as GA (gibberellic acid). The loss of embryo dormancy is often associated with a sharp drop in the ratio of ABA to GA.

Conditions that affect physiological dormancy of seeds are:

Drying; some plants including a number of grasses and those from seasonally arid regions need a period of drying before they will germinate. The seeds are released but need to have lower moisture content before germination. If the seeds remain moist after dispersal, germination can be delayed for many months or even years. Many herbaceous plants from temperate climate zones have physiological dormancy that disappears with drying of the seeds. Other species will germinate after dispersal only under very narrow temperature ranges, but as the seeds dry they are able to germinate over a wider temperature range.

Photo dormancy or light sensitivity affects germination of some seeds. These pho toblastic seeds need a period of darkness or light to germinate. In species with thin seed coats, light may be able to penetrate into the dormant embryo.

The presence of light or the absence of light may trigger the germination process. Inhibiting germination in some seeds buried too deeply or in others not buried in the soil.

Thermo-dormancy is seed sensitivity to heat or cold. Some seeds including Amaranths germinate only at high temperatures (30C or 86F). Many plants that have seeds to germinate in early to mid summer have thermo-dormancy and germinate only when the soil temperature is warm. Other seeds need cool soils to germinate, while others are inhibited when soil temperatures are too warm. Often thermo-dormancy requirements disappear as the seed ages or dries.

Morphological Dormancy

Some seeds have fully differentiated embryos that need to grow more before seed germination. The embryos are not differentiated into different tissues at the time of fruit ripening.

Immature embryos – Some plants release their seeds before the tissues of the embryos have fully differentiated. The seeds ripen after they up take water while on the ground. Germination can be delayed from a few weeks to a few months.

Combined Dormancy

Seeds have both morphological and physiological dormancy.

Morpho-physiological or physio- morphological dormancy occurs when seeds with underdeveloped embryos and have physiological components to dormancy. These seeds therefore required dormancy-breaking treatments as well as a period of time to develop fully grown embryos.

Coat-imposed dormancy is dormancy imposed on the embryo by the seed coat and other enclosing tissues, such as endosperm, pericarp, or extra floral organs. The embryos of such seeds will germinate readily in the presence of water and oxygen once the seed coat and other surrounding tissues are either removed or damaged. There are five basic mechanisms of coat-imposed dormancy:

Prevention of water uptake - Prevention of water uptake by the seed coat is a common cause of seed dormancy in families found in arid and semiarid regions, especially among legumes, such as clover (*Trifolium* spp.) and alfalfa *(Medicago* spp.). Waxy cuticles, suberized layers, and lignified sclereids all combine to restrict the penetration of water into the seed. Seed coat has impermeable to water due to hardness. The seed coats of several plants

belonging to the families Leguminoceae, Malvaceae, Chenopodiaceae and Solanaceae are impermeable to water.

Mechanical constraint- The first visible sign of germination is typically the radical breaking through the seed coat. In some cases, the seed coat may be too rigid for the radical to penetrate. Nuts with hard, lignified shells are examples of dormancy caused by mechanical constraint. Such shells must be broken by biotic or environmental forces for the seed to germinate. Even non-lignified tissues, such as the endosperm of lettuce seeds, can suppress expansion of the embryo.

Interference with exchange of gases - Some seed coats are considerably less permeable to oxygen than an equivalent thickness of water-e.g., (*Xanthium*). This lowered permeability to oxygen suggests that the seed coat inhibits germination by limiting the oxygen supply to the embryo. In support of this idea, investigators can break the dormancy of such seeds either by making a small hole in the coat with a pin (without weakening the coat mechanically), or by treating the coat with concentrated oxygen. However, other studies suggest that the oxygen consumption of the embryos from such seeds is considerably less than the amount of oxygen able to penetrate the seed coats under normal aerobic conditions. Thus the role of oxygen impermeability in seed coat dormancy remains unresolved.

Inhibitor production - Seed coats and pericarps may contain relatively high concentrations of growth inhibitors that can suppress germination of the embryo. ABA is a common germination inhibitor present in these maternal tissues. In certain cases where repeated washing (leaching) removes seed dormancy.

Seed Dormancy May be Primary or Secondary

Different types of seed dormancy also can be distinguished on the basis of the timing of dormancy rather than the cause of the dormancy.

The primary dormancy is that which is produced in the organs originally by innate or imposed conditions. Seeds that are released from the plant in a dormant state are said to exhibit primary dormancy.

Seeds that are released from the plant in a non dormant state but which become dormant if the conditions for germination are unfavorable exhibit secondary dormancy. For example, seeds of *Avena sativa* (oat) can become dormant in the presence of temperatures higher than the maximum for germination. Seeds of *Phacelia dubia* (small-flower scorpion weed) become dormant at

temperatures below the minimum for germination. Various factors induce secondary dormancy. The mechanisms of secondary dormancy are poorly understood.

19.3. Factors Responsible for Seed Dormancy

19.3.1 Environmental Factors

Various external factors release the seed from dormancy, and dormant seeds typically respond to more than one condition. Many seeds lose their dormancy when their moisture content is reduced to a certain level by drying. This method of breaking seed dormancy is called after-ripening, and is usually performed in a special drying oven. On the other hand, if the seed becomes too dry (5% water content or less), the effectiveness of after-ripening is diminished.

Seeds requiring, chilling treatment is another factor that can release seeds from dormancy is low temperature, or chilling. Many seeds require a period of cold (0 to 10°C) while in a fully hydrated (imbibed) state in order to germinate. Chilling treatment of seeds to break their dormancy is a time-honored practice in horticulture and forestry. Traditionally it has been referred to as stratification. This term is derived from the old agricultural practice of allowing seeds with a chilling requirement to over winter outdoors in layered mounds of moist soil. Today the seeds are simply stored in a refrigerator.

The seeds require a low temperature treatment before they can become capable of germination. The chilling requirement is essential under natural conditions in the winter. Examples are found in many plants of the temperate areas (e.g., Peach, Plum and Cherry). The temperature most suitable for chilling treatment is about-5°C. All seeds requiring a chilling treatment have been found to contain growth inhibitors. Chilling treatment increases the synthesis of gibberellins.

Light Sensitive Seeds. Some seeds require a specific light treatment before being able to germinate, e.g., Lettuce, *Viscum album*. Light sensitive seeds are thought to have either an impermeable seed coat or possess inhibitors which are broken down by light.

The second external factor that plays an important role in breaking seed dormancy is light. Many seeds have a light requirement for germination, which may involve only a brief exposure as in the case of lettuce and an intermittent treatment (e.g., succulents of the genus *Kalanchoe*). The Specific duration of photoperiod involving short or long days also break the dormancy. Phyto-

chrome is the main sensor for light-regulated seed germination. Interestingly, all light-requiring seeds exhibit seed coat dormancy and removal of the outer tissues of the seed allows the embryo to germinate in the absence of light. The effect that light has on the embryo is thus to enable the radical to penetrate the seed coat. This penetration often involves some enzymatic weakening of the enclosing tissues.

Seed germinate after-ripening. These seeds gain power to germinate after a period of dormancy when kept under dry environment at normal temperature, e.g., Oat, Wheat and Barley. The exact cause of dormancy is not known. Some of these can germinate within a narrow range of temperature soon after shedding. Others can become non dormant when kept at high temperature.

19.3.2 Reasons for Seed Dormancy

Immaturity of the Embryo: The embryo is immature at the time of seed shedding. Therefore, germination will not occur till the complete development of the embryo has taken place, e.g. *Anemone nemorosa, Caltha palustris, Ranuculus ficaria* (Ditots).

Seeds coats Impermeable to Water (Hard Seed). The seed coats of several plants belonging to the families Leguminosae, Malvaceae, Solanaceae, Convolvulaceae and Chenopodiaceae are impermeable to water. These remain dormant till their outer coverings are broken down by mechanical injury and microbial action.

Inhibiting Pericarps and Glumes- The dormancy of a number of cereals has been known to be due to the glumes. The hull of *Avena fatua* does not allow the leaching of inhibitors present in the caryopsis. Certain achenes are dormant because of the presence of an outer pericarp. A number of fruits make their seeds dormant because of their pulp.

High Osmotic Concentration- Atriplex seeds have a very high osmotic concentration. This prevents their germination. The seeds germinate only when sufficient solutes have come out on account of good rainfall.

Chemical Inhibitors- Besides the above types, seed dormancy can also be due to the presence of growth inhibitors. The latter are of diverse types, e.g., phenolic inhibitors, abscisic acid, alkaloids, cyanogenic chemicals, etc. The inhibitors can be found in the seed coats, endosperm or embryo, e.g., Apple, Peach, Ash, etc. The inhibitors are destroyed in nature by the production of anti-inhibitors, growth hormones or their oxidation.

19.4 Importance of Seed Dormancy

One important function of dormancy is most seeds are delayed germination, which allows time for dispersal and prevents germination of all the seeds at the same time. True dormancy or innate dormancy is caused by conditions within the seed that prevent germination under normally ideal conditions. Seed dormancy introduces a temporal delay in the germination process that provides additional time for seed dispersal over greater geographical distances. It also maximizes seedling survival by preventing germination under unfavorable conditions. These are the main Importance of seed dormancy

1. Dormancy allows seeds, buds and other organs to pass through unfavorable conditions like drought and cold without any injury.
2. It is an important component of seed dispersal of many plants. For example, a number of seeds adapted to dispersal by birds require passage through their digestive system in order to weaken their seed coats for future germination. The best example is kalkaria tree's seed germinated after run through the digestive tract in Dodo bird. The extinction of Dodo bird the Kalkaria trees come under extinct state in Mauritius.
3. The seeds of some plants of arid areas have impermeable testa, e.g., *Convolvulus* sps. Permeability is achieved after an interval of few years. This protects the species from extinction in the year of drought. Some weeds of this type are very difficult to eradicate because new plants will continue to grow over the years. Even when all plants coming out in a single year are completely destroyed.
4. Dormancy caused by inhibitors found in the seed coats of desert plants allows them to remain inactive during the dry periods. They germinate only when the inhibitors get leached by a good rainfall. It ensures rapid growth under favorable conditions.
5. The knowledge of dormancy, both induction and its breaking is of great importance to human welfare. Delayed germination of tubers, corms, rhizomes, bulbs, grains and seeds can help us in their storage and for their proper distribution both in time and space. Similarly quick growth is important to enhance productivity of the plants. Various synthetic chemicals have been developed which will cause prolongation of the

period of rest or dormancy, e.g., IBA, NAA, 2: 4: 5-T, maleic hydrazide, etc.

19.5 Methods to Overcome Dormancy

Many seeds fail to germinate after processing and placement in favorable growing conditions—such seeds are said to be dormant. In some dormant seeds morphological changes must take place before germination can start. For other parts of the seed must undergo physiological changes before germination can occur. Under natural conditions necessary changes take place gradually under varying combinations of aeration, moisture, temperature, and light. By duplicating key conditions of the natural environment in the laboratory or nursery dormant seeds can be induced to germinate with a reasonable length of time.

Seed dormancy is nature's way of setting a time clock that allows seeds to initiate germination when conditions are normally favorable for germination and survival of the seedlings. For example, dogwoods produce mature seeds in the fall but conditions are not suitable for seedling survival at that time. Thus, dogwoods have developed a mechanism that keeps the seeds dormant until spring when conditions are favorable for germination as well as seedling growth and survival. Viable seeds that do not germinate are said to be dormant.

Dormancy can be regulated by the environment or by the seed itself. If a seed is not exposed to sufficient moisture, proper temperature, oxygen and light, the seed will not germinate. In this case the seed's dormancy is due to unfavorable environmental conditions. On the other hand some seeds may not germinate because of some inhibitory factor of the seed itself. Several treatments have been developed which can break the dormancy of seeds. Type of the treatment depends upon the type of seed dormancy. Dormancy caused by the immaturity of the embryo cannot however be overcome.

Methods of breaking seed coat dormancy include scarification, hot water, dry heat, fire, charate, acid and other chemicals, mulch, water, cold and warm stratification, and light etc.

1. Scarification

Seed coat (external dormancy) results from a seed's hard seed coat that is impermeable to water and gases. The seed will not germinate until the seed coat is altered physically. Any process of breaking, scratching or mechanically altering the seed coat to make it permeable to water and gases are known as

scarification. For mechanical scarification seed coats can also be filed with a metal file, rubbed with sandpaper, nicked with a knife, or cracked gently with a hammer to weaken the seed coat. Scarification can also occur as seeds pass through the digestive tract of various animals.

Following scarification, the seeds should be dull in appearance but not deeply pitted or cracked as to damage the embryo. Scarified seeds do not store well and should be planted as soon as possible after treatment.

2. Microorganisms Reaction

In nature, this often occurs by fall seeding. Freezing temperatures or microbial activities modify the seed coat during the winter.

3. Sulphuric Acid Treatment

Commercial growers scarify seeds by soaking them in concentrated sulfuric acid. Seeds are placed in a glass container and covered with sulfuric acid. The seeds are gently stirred and allowed to soak for 10 minutes to several hours it's depending on the species. When the seed coat has been modified (thinned) and washed and sown. Sulfuric acid can however be very dangerous for an inexperienced individual and should be used with extreme caution.

Vinegar is safer (but less effective treatment) and can be used for some species that do not have an extremely hard seed coat this technique is the same as with sulfuric acid. After treatment and a thorough washing, the seeds may be sown or dried and stored for several months. Since sulfuric acid is caustic and dangerous to handle its use is recommended only for those familiar with the use of caustic chemicals. All workers should wear suitable safety clothing, gloves and goggles or other eye protection.

4. Hot Water Treatment

Another method is hot water scarification. Bring water to a boil (212°F), remove the pot from the stove and place the seeds into the water. Allow the seeds to soak until the water cools to room temperature. Remove the seeds from the water and sow.

5. Role of Chemicals and Growth Promoting Substances in Breaking Seed Dormancy

About 50 years ago researchers in various agencies and private industry began experimenting with chemicals to neutralize dormancy conditions present in seeds. Results have shown that inhibiting chemicals can be present in one or more parts of the seed. Dormancy-causing factors (i.e., immature embryos or

impermeable seed coats) may also be present in a given seed. These chemicals are very helpful in breaking certain types of dormancy.

1. Gibberellic acid (GA3) 2. Potassium nitrate 3. Thiourea.

   The aqueous solutions of these chemicals should be used at room temperature. The concentration and length of treatment depends on the species to be treated.

Seeds soaked in GA3, or thiourea should be stirred occasionally and not rinsed in water but sown immediately. After this soaking they can also be air-dried and stored for short periods and then sown or given a subsequent treatment. The non-rinse-afterwards also applies to the use of potassium nitrate and hydrogen peroxide, other chemicals occasionally recommended as aids to germination. Great care should be taken in working with these chemicals as some are poisonous. Due to their toxic or poisonous nature, some are difficult to obtain. The main advantages of these chemicals are speed, ease of use, and unaltered physical condition of the seeds following treatment.

6. Mulch

The mulch treatment is the microbial breakdown or softening of the seed coats. It is a slow method but is often occurs in the wild. For this treatment, fill a six- to eight-inch deep container half full with seedbed medium. Then the sown seeds should be covered with a mulch of wood shavings (not redwood or cedar). A one-inch thick layer of old composted shavings is best but if not available, a three-inch layer of fresh shavings is satisfactory. If fresh shavings are to be used, they should be soaked a few hours in a bucket of water first and mixed with a compost starter of microbial inoculants. Neither the seeds nor the medium should be treated with a fungicide. If this treatment is initiated in early spring or early summer and if the shavings are kept moist all summer, germination will require three to four months or longer, depending on the species. This mulching technique also works well in a ground bed. However, transplanting may be a bit more difficult (Emery 1987).

7. Cold Stratification

The more common method for breaking internal dormancy is cold stratification. In some cases, the use of chemicals can be substituted for part or the entire stratification requirement (Emery 1987). In one general type there is seed coat dormancy plus internal dormancy. Seeds with this dormancy combination must be treated for the impermeable seed coat first, then for internal dormancy. In

another type there are two or more distinct internal dormancy factors, which unlock sequentially at different temperatures.

Cold stratification or pre-chilling used for seeds with internal dormancy stimulates cold winter conditions. For small quantities of seeds, mix a ratio of 1:3 or more with moist peat moss place in a tightly sealed polyethylene bag or glass jar and store in the refrigerator at a temperature of 35º- 41º F.

With a few species, freezing the seeds at 28º-32º F is required. For bulk seeds, soak in water for a few hours first, then place wet in a sealed container. Containers can be boxes, tanks, trays, cans, or barrels, as long as they have perforated bottoms to allow drainage of excess water and to facilitate gas exchange between the seeds and the storage room. Of course, polyethylene bags can be used as well. In any case, the seeds must be kept moist during the entire length of the treatment. This will require periodic checking and the addition of water if necessary.

In contrast, to cut the stratification period short by even a few days could be harmful if no radicals are visible. By prematurely discontinuing stratification, primary dormancy may not be broken. Consequently, a secondary dormancy may be induced which is more difficult to break than the original dormancy. The cold stratification period necessary to break dormancy may last from a few days to several months, depending upon the species, with one to three months being the most common. Some species even require up to 3 years of stratification. After stratification, the seeds should be sown promptly before they have a chance to dry out.

The following sequence of operations is normally used in cold stratification with a moisture-holding medium:

1. **Moisten the medium uniformly**: Peat moss should be just moist enough so that a little free water can be easily squeezed out by hand. Excessive moisture can be harmful to some species. Mixing cracked ice with medium and seed to promote quick and uniform chilling should also help distribute moisture.
2. **Mix seeds with the medium:** The most common practice is to place lots of seeds in loosely woven bags, which are flattened into disks no more than 3 inches thick.

   The bags of seeds are then alternated with layers of the moist media in the container. Putting the same dry weight of seeds in each sack permits easy allocation of seeds in subsequent planting operations despite gains

in weight during stratification. Mixing can also be accomplished by placing seeds in thin layers alternating with layers of medium; layers may be separated by cheese cloth.

3. A third step is mixing the seed directly into the moist medium. The volume of the medium should be three times that of the seeds. This method is very effective, but since it creates a cleaning problem when treatment is finished, may have abandoned the practice.

However, another type of internal dormancy requires special treatments to overcome. Seeds having this type of dormancy will not germinate until subjected to a particular duration of moist-pre chilling and moist warm periods. Cold stratification (moist-pre chilling) involves mixing seeds with an equal volume of a moist medium (sand or peat, for example) in a closed container and storing them in a refrigerator.

19.6 Bud Dormancy

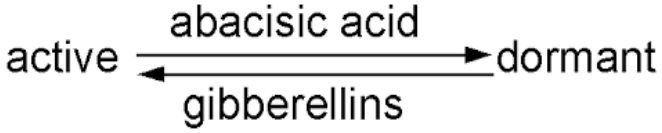
Bud dormancy is a suspension of most physiological activity and growth that can be reactivated. It may be a response to environmental conditions such as seasonality or extreme heat, drought, or cold. The exit from bud dormancy is marked by the resumed growth of the bud. Buds of any types of woody plants undergo dormancy and they fail to grow and develop even under favorable environmental conditions. The dormancy can be overcome, and in some cases, by almost same treatments which are employed for breaking bud dormancy.

Some examples are:

Chilling for several weeks at 0-5°C (stratification) is effective in breaking dormancy of buds, rhizomes, corms and many types of seeds.

Many substances will break the dormancy of several kinds of organs. Thiourea and GA remove the bud dormancy rhizomes, corms and all types of dormancy.

Some tree plants bud dormancy may be induced to grow by exposure to long days, where as short days is ineffective.



Knowledge of bud dormancy comes from temperate deciduous trees, especially fruit crops such as apples and stone fruit. Trees detect environmental signals, mainly shortening day length. Cold and winter trigger reductions in growth rate or bud dormancy and development of bud scales and leaf fall. As buds enter dormancy, warm temperatures (>15°C) no longer promote growth. Several weeks or months of chilling (0–12°C) are required to overcome dormancy. In some tropical species such as coffee, water stress is an alter-native clue for breaking flower bud dormancy. Buds then exist in an ecodormant state ready to respond by rapid floral growth as soon as the first rains fall at the end of the dry season.

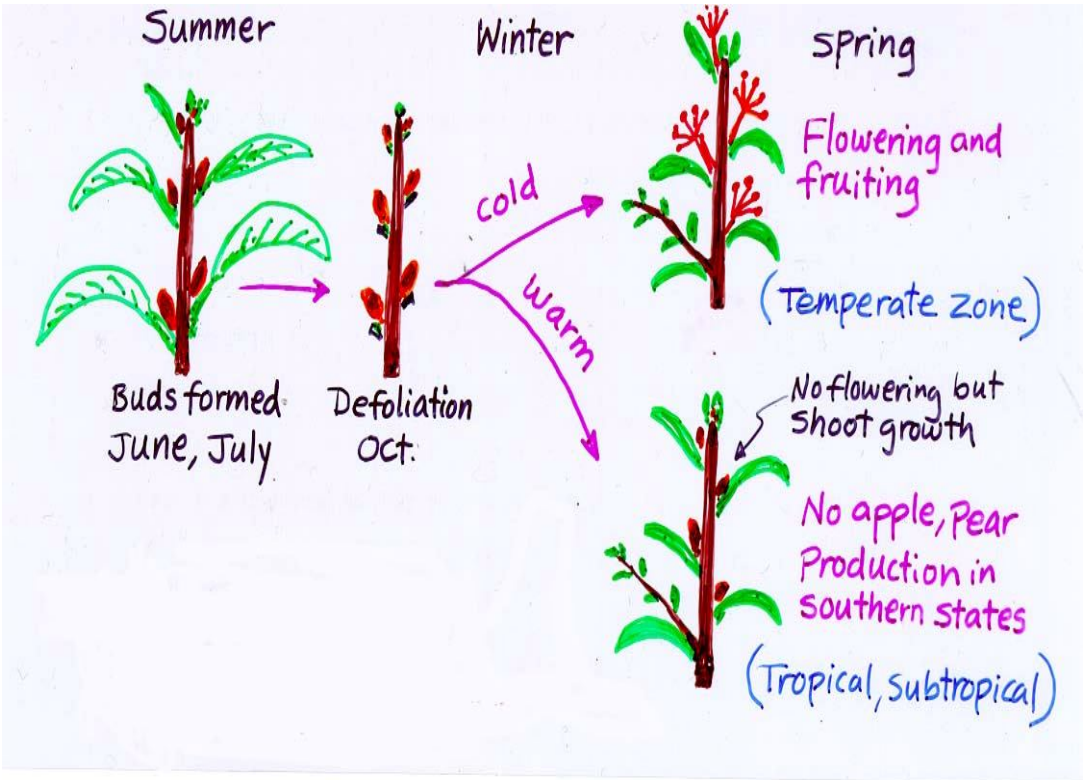


Fig. 19.1 : Bud Dormancy

Several models have been proposed to describe dormancy and to attempt to predict responses to different growing conditions. One problem is a lack of measurable indicators of endo-dormancy other than an inability to grow. Researchers typically quantify 'depth' of dormancy by the duration of chilling required to break bud dormancy. The ability of warm temperatures to 'force' bud growth on cut shoots. Entry into and exit from bud dormancy are often gradual transitions rather than abrupt events.

19.7 Summary

If the three normal conditions necessary for germination (i.e., necessary oxygen, water and temperature) are available, the seeds of most plants germinate. But in some plants (e.g. *Xanthium, Ginkgo* and *Cucurbita*) the seed do not germinate in spite of the availability of these three necessary conditions. Such seeds are called dormant seeds and this phenomenon is called seed dormancy.

Dormancy is also called rest by certain workers. Another term used in connection with seed is quiescence (imposed dormancy). Quiescence is the inability of a viable seed to germinate because the environmental conditions required for it (e.g. favourable moisture, temperature, light, etc.) are not available. Dormancy is best defined as temporary suspension of active growth. Internal dormancy is a general term encompassing a number of physiological conditions that delay germination.

In nature, dormancy period coincides with the unfavorable period for the seedlings of the species. This period, which varies from species to species it is called dormancy period. It is assumed that these seeds undergo physiological maturation during the dormancy period. The physiological changes may include the disappearance of growth inhibitors, appearance of growth promoters. Seed dormancy occurs due to the many internal or external reasons.

Dormancy may be divided in to two types, primary and secondary. The primary is that which is produced in the organs originally by innate or imposed conditions. Secondary dormancy is always induced by a change in the environment before the organs have completed their post dormancy or after ripening. Seed dormancy is, therefore, the inability on the part of the viable seeds to germinate under suitable conditions.

Causes of seed dormancy: Some of the major causes of the seed dormancy are following:

(i) Mechanically hard seed coat.

(ii) Seed coat impermeable to water and air.

Several treatments have been developed which can break the dormancy of seeds. Type of the treatment depends upon the type of seed dormancy. Methods of breaking seed coat dormancy include scarification, hot water, dry heat, fire, charate, acid and other chemicals, mulch, water, cold and warm stratification, and light. The fire or mulch treatments can be used with thinner-coated seeds.

Hot water or scarification is also satisfactory. Dormancy caused by the immaturity of the embryo cannot, however, be overcome by

Scarification : It is a treatment which ruptures or weakens the seed coat. Scarification is meant for breaking all types of dormancies which are imposed by the seed coats, viz., impermeability of the seed coats to water and oxygen, presence of growth inhibitors or mechanical resistance. The treatment involves the rupturing or weakening of the seed, coats by two methods - mechanical and chemical.

Pressure: This treatment involves subjecting of seeds to a high hydraulic pressure (about 2000 atm) at a temperature of 18°C - 20°C for a short duration of 5-20 minutes. It weakens seed coats and increases their permeability. Germination increases by 50- 200% in Sweet clover (*Melilotus alba*) and Alfalfa (*Medicago sativa*) by this method.

Chilling Treatment: The seeds, which require chilling winter for natural breaking of dormancy, can be made to germinate by artificially providing low temperature in a most medium. The seeds are kept at 0°- 5°C for a period ranking from a couple of weeks to a few months e.g., Peach, Plum, Cherry, Apricot, etc.

High concentration of oxygen : It induces germination in those Seeds which have seed coat impermeable to oxygen. Higher concentration of oxygen can also cause destruction of growth inhibitors.

Light : Application of high temperature reduces the requirement of red light. Certain light requiring seeds are actually influenced by photoperiod which may be short (e.g. *Veronica persica*) or long (e.g.- *Begonia*).

Growth Regulators : The seeds with a requirement of chilling after-ripening or light treatment can be made to germinate on the application of growth promoters, e.g., gibberellins, 2-chloroethanol or ethylene chlorohydrin ($ClCH_2CH_2OH$), thiourea (NH_2CSNH_2) etc.

The length of time it takes to break dormancy varies with particular species. Warm stratification is similar except temperatures are maintained at 68°F to 86°F depending on the species. Seeds of some species exhibit what is known as double dormancy. Some chemicals are effective in breaking dormancy, e.g., 2-chloroethanol (=ethylene chlorohydrin CICH2CH2OH), thiocyanate, thiourea (NH2 CS. NH2), etc.

Occasionally the dormancy is caused by an inhibiting chemical in the epidermis or adjacent interior membranes. Under natural conditions these seeds remain on or in the ground without germinating until they have weathered sufficiently. Allow penetration of water, exchange of gases, or neutralization of inhibiting chemicals. The length of time involves and it can be several years or more and may be depends upon the species and the environmental conditions. If seeds of some plants are harvested when slightly green or immature and sown immediately before they dry out, germination problems may be reduced.

Study of seed dormancy is to provide knowledge about different methods to overcome dormancy and it is essential for dormant seeds to properly germination. Dormancy caused by inhibitors found in the seed coats of desert plants allows them to remain inactive during the dry periods. They germinate only when the inhibitors get leached by a good rainfall. It ensures rapid growth under favorable conditions. Knowledge about seed dormancy is helped in the storage of seeds. It has also thrown light on how some of the complex structure seeds prolonged the dormancy.

19.8 Glossary

- **Dormancy :** Resting stage of seeds.
- **Quiescence-** Inability of a viable seed to germinate.
- **Cold stratification:**Pre-chilling
- **Scarification:**Any process of breaking or scratching of seed coat
- **Exogenous dormancy:** Exogenous dormancy is caused by conditions outside the embryo
- **Endogenous dormancy:** Endogenous dormancy is caused by conditions within the embryo itself.

19.9 Self -Learning Exercise

Section A : (Very Short Answer Type Questions)

1. Which Chemicals are helpful in breaking certain types of dormancy?
2. What is scarification
3. What is seed dormancy?
4. What do you know about quiescence?

Section B : (Short Answer Type Questions)

1. Which growth regulators are can be employed to overcome seed dormancy?
2. How the application of growth regulators can be effect the germination?
3. What do you know chilling treatment?

Section C : (Long Answer Type Questions)

1. What is dormancy Describe various types of dormancy?
2. Give an account of different types of seed dormancy.
3. Describe the important methods to break dormancy.
4. Give an account of the environmental factors affecting seed dormancy.
5. Discuss various causes of seed dormancy.
6. Briefly explains the importance of dormancy.
7. Write short notes on
    1. Stratification
    2. Scarification
    3. Chemical Inhibitors
8. What are the various physiological changes that take place at the time of seed germination?
9. How will you explain the mechanism of dormancy? Of what practical importance is the knowledge of dormancy to man?

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Unit-20

Plant Senescence and Programmed Cell Death

Structure of the Unit:

20.0 Objectives

After studying this unit you will be able to understand about :

- Senescence
- Programmed Cell Death
- Abcission
- Metabolic changes associated with Senescence
- Hormonal regulation of Senescence

20.1 Introduction

Senescence is an integral component of a plant's lifecycle, which refers to changes that take place as the plant matures. Senescence is the terminal phase of development in the life of a plant. Like human beings, plants also grow old and undergo ageing and then die.

The process of growing old is called ageing and death is the termination of functional life. The stages of developmental processes that ultimately lead to death of an organ or organism are called senescence. Ageing and senescence are thus, two different phenomena. Ageing is the sum total of changes occurring in the whole plant or some of its constituent organs. It includes all chemical and structural changes in cells, tissues, organs and the whole plant during their life cycle. Senescence, on the other hand, is a consequence of ageing. Metabolic failure and cellular breakdown increase while the functional activities decrease during the senescence.

It occurs due to some highly ordered degenerative processes and lead ultimately to death. Senescence, therefore, is a highly ordered degenerative process and finally terminates the functional life of an organ or organism. Senescence is a phase of the aging process.

Senescence is not confined only to whole plant. It may be limited to a particular plant organ such as leaf and flowers or cells or cell, organelles.

Senescence is a metabolic process; therefore, it requires energy. It is not simply the ending of growth. One of the first materials to degrade is the energy-converting pigment chlorophyll. As the bright green color of chlorophyll fades, the yellow-orange colors of the carotenoids become prominent and combine with the red-blue anthocyanins to produce the vivid colors of autumn in the trees and shrubs

The major characteristic of senescence is that the metabolic processes are catabolic and eventually become irreversible and terminate to death.

Senescence is closely associated with the phenomenon of aging. Aging leads to senescence. Senescence is the growth phase in a plant or plant part (as a leaf) from full maturity to death.

According to Curtis "Senescence is a genetically rather than a physiological wearing out." He proposed that it is caused by somatic mutation.

Senescence is also considered as the result of an altered hormone balance that prevents the genes from continuing to code for the same enzymes as and when the cells were younger. Considered in this way, senescence may be thought of as a final stage of differentiation which itself is continuous from juvenility to death. It is the culmination of the morphogenetic information programme.

20.2 Senescence

20.2.1 Types of senescence

Leopold (1961) has proposed types of senescence patterns in plants which are as follows.

(a) Overall Senescence

This type of senescence occurs in annuals where whole plant is affected. It is also called whole plant senescence. The process of senescence begins with the reproductive maturity and the whole plant dies after seed production. E.g. Paddy, wheat, soybean

(b) Top Senescence

In top senescence, the parts remaining above the ground or (shoot system) may die, but the root system and underground system remain viable. It is also called shoot senescence. E.g. perennials like Zingier, Musa, Dock and Chrysanthemum

(c) Deciduous Senescence

In deciduous woody plants, all the leaves die but the bulk of the stem and root system remains viable. It is called deciduous senescence or simultaneous or synchronous senescence. It is a type of Organ senescence.

E.g. Leaf falls in deciduous trees

(d) Progressive Senescence

It is a gradual death of old leaves from the base to the top of the plants. It may occur at any time. It is also called sequential senescence. . It is also a type of Organ senescence. E.g. Green tree

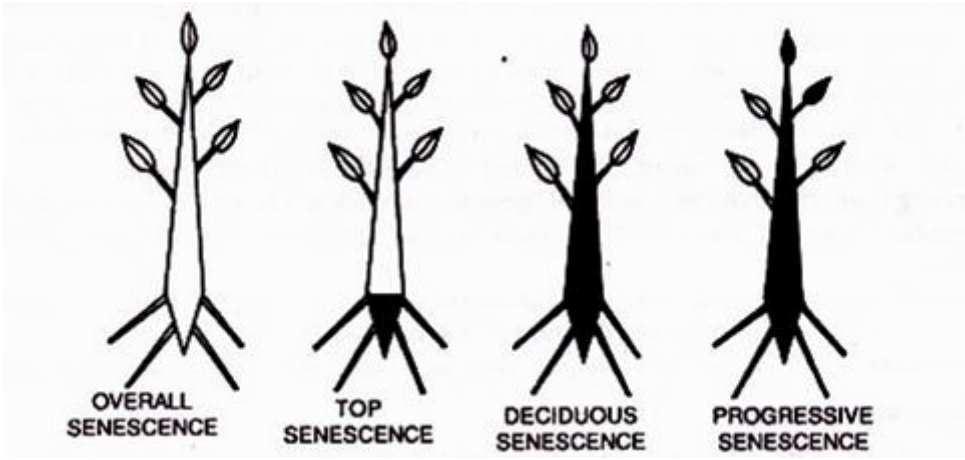

Fig. 20.1: Different Senescence pattern in Plants

20.2.2 Mechanism of Senescence

Three different theories have been proposed to explain the phenomenon of Senescence.

1. Nutritional theory

2. Harmonal theory

3. Suicidal theory

1. **Nutritional theory**

This theory was proposed by Molish in 1928. This theory assumes that senescence is caused due to nutritional deficiency. There are competitions for nutrients between different parts of plants. This theory is not universal.

2. **Hormonal theory**

This theory assumes that senescence is caused by Hormones like as Ethylene and Abscisic acid. Abscisic acid is produced in developing fruits and seeds. L.D.Nooden and A.C.Leopoid (1978) coined the term 'death hormone' for causal agent of senescence. According to them .each hormone is chemical produced in developing seeds from where it is translocated through the xylem to vegetative parts of monocarpic species. This hormone is different is from ABA or ethylene. In fact this hormone is

not detected in plants as yet .However three chemicals were isolated recently.

4-chlorindole acetic acid, 4-chlorindole acetyl aspartate monoethyl ester and jasmonic acid may act as death hormone. Deficiency of auxin and cytokinin may also induce senescence. Cytokinine divert nutrients to the leaves and prevent senescence of leaves.

Senescence is faster in detached leaf than attached leaf perhaps due to supply of cytokinine by root in attached leaves. Detached leaf lacks root, hence lacks supply. This senescence is the result of an altered hormone balance that prevents the genes from continuing to code for the same enzyme as when the cells are younger.

3. Suicidal theory (Hydrolytic enzymes exudation theory by vacuoles)

In senescence leaves there is progressive loss of membrane bilayer which results into leakage of solute and loss of membrane functions. During the process per oxidation of membrane lipid part may occurs by free radicals produced at senescence time. Tonoplast breaks, releasing hydrolytic enzymes of the vacuoles which digest the useless cellular materials. According to some physiologists vacuoles are allied to lysosomes of the animals.

20.2.3 Causes of Senescence

- Leaf senescence is accompanied by early loss in chlorophyll, RNA and enzymes.
- Cellular constituents are decreased due to slower synthesis or faster break down.
- Competition between vegetative and reproductive organs for nutrients.
- A senescence factor (a hormone) is produced in soybean fruits that move to leaves where it causes senescence.
- Short-day and long-night conditions induce flowering and leaf senescence.
- Degradation of food reserves and loss of integrity in food storage cells of seeds.
- Senescence is also hormonally controlled.

20.2.4 Physiology of Senescence

The following physiological changes occur during senescence.

- Photosynthesis stops.
- Chlorophyll degradation: The colour of leaf changes from green to yellow.
- Anthocyanin pigments accumulation in the leaves causing reddening in leaves.
- The vacuoles function as lysosomes and digest the cellular materials.
- The starch content decreased.
- RNA and proteins are decreased.
- DNA molecules are degraded by the enzyme DNase.
- Growth promoting hormones such as cytokinine decrease.
- The deteriorative hormones such as ethylene and abscisic acid (ABA) content are increased.

20.2.5 Hormonal Regulation of Senescence

The major plant growth regulators have been implicated in Senescence. Hormonal regulation of Senescence is achieved through interactive effects of the various growth regulators. Senescence is promoted by hormones such as abscisic acid and ethylene. The senescence accelerating ability of abscisic acid is well documented. The function of ABA as a promoter of flower tissue senescence including initiation of colour fading or blueing has been established. The ABA content of aging leaves increases markedly as senescence is initiated. Ethylene plays a very important role in the senescence of certain plant parts, particularly fruit and petals and in the abscission process. It is an inducer in the senescence of flower tissue.

Senescence Retardants: The primary plant hormones involved are auxin, gibberellins and cytokinine.

20.2.6 Significance of Senescence

- The whole plant senescence occurs in monocarpic plants coinciding the seed setting and seed dispersal.

- Due to the formation of abscission layer, the older leaves tend to fall down so that the nutrients will be diverted to the next young leaf.
- The senescence process helps the mobilization of nutrients and of the vegetative parts of the plant into the fruits.
- Plants escape the influence of seasonal adversity by undergoing senescence of its organs. Leaf fall in deciduous trees reduces the rate of transpiration to survive under adverse conditions.
- Falling of leaves due to senescence adds to the humus content of the surface layer of the soil and thereby makes to soil reach in nutrients for germination and growth of new seedlings.

20.2.7 Factors Influencing Senescence

1. **Hormones and growth regulators**

   As indicated earlier, cytokinine and auxins in some cases delay senescence .Abicisic acid accelerates senescence. Ethyelene also accelerates senescence mostly in fruits .Polyamines retard the process in some plants. Natural senescence is related to the lower levels of polyamines: spermidine and spermine in some cases.

2. **Light & dark**

   Senescence is rapid in dark as compared to light. In *Avina* more leaves in 72 h at 25ºC ,about 70% of the chlorophyll is lost in dark, while there is only about 20-30% loss in light .The effect of light on senescence ,seems to be though stomatal opening. Artificial closing of stomata accelerates senescence .This causes the accumulation of abiscisic acid.

3. **Water stress**

   Water stress also accelerates senescence, although the effects are less dramatic than that of darkness or heat. Water stress causes the accumulation of ABA in leaves, which is a senescent hormone. The stress also reduces the transport of cytokinine from roots to leaves, which is an anti senescent hormone.

4. **Temperature**

   Elevated temperature or heat can accelerate the process of senescence .The leaves of heat stressed tobacco plants turns yellow and their protein contents also decreases. If the plants are kept at low

temperatures, the greening and the protein content can be retained for longer times, and i.e. senescence is delayed.

5. Nutrients

Nutritional deficiency especially of nitrogen enhances leaf senescence. Supply of nitrogen to mature plants can delay senescence for sometimes. Calcium also delays senescence, its acts synergistically with cytokinine. In maize leaf discs, calcium delays the loss of chlorophyll and protein contents.

20.3 Abscission

Shedding of leaves, flowers and fruits is called abscission. Abscission is distinct in deciduous trees and shrubs. In autumn, all the leaves of deciduous plants fall, at about the same time giving the plants a naked appearance. In evergreen plants there is gradual abscission of leaves. The older leaves fall while new leaves are developed continuously throughout the year. In most of the herbaceous species, abscission is a complex physiological process. During abscission, the color of the leaves, flowers and fruits changes due to degradation of chlorophyll and the synthesis of anthocyanin pigment. Leaf abscission takes place at the base of the petiole. The site of abscission is internally marked by a distinct zone called abscission zone. The abscission zone is pale or brown in colour.

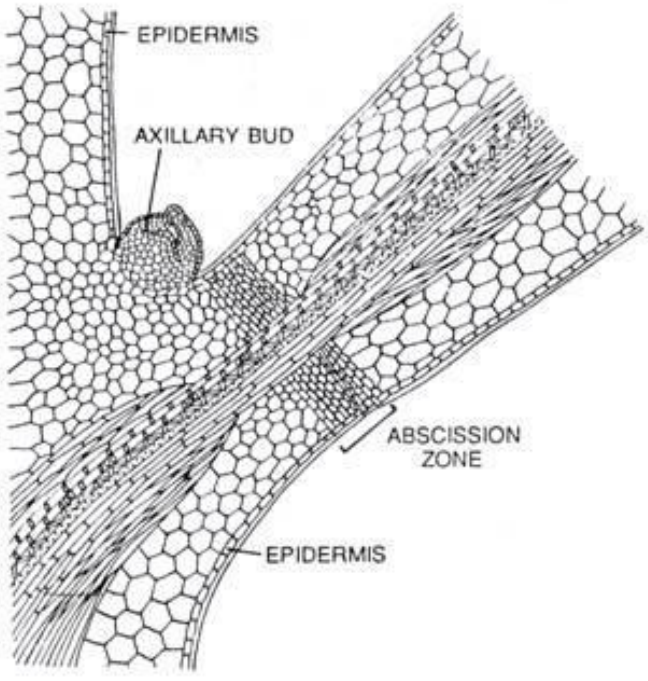

Fig. 20.3 : Leaf Abscission-Formation of the Abscission Zone

20.3.1 Morphological and Anatomical Changes

At the junction of multicellular organs (like leaf, flowers, fruit) and parent plant body an abscission zone (separation zone) is formed .In leaves it is at the base of petiole, where leaf joins the stem (like i.e. node) In compound leaves the abscission zone lies at the junction of pulvinous and the supporting tissues. Abscission zone may be swollen or constricted in respect to the other part of the petiole. The abscission zone is very prominent in some plants of Palmae family like coconut, date palm, borassus, cycas etc. Prior to abscission leaves are either yellow or dried. Abscission zone formed may be 1-2 row of cell or sometimes 15 or more cells tiers thickness .Cells are parenchymatous and devoid of lignin and suberin like thickening material. Such cells have dense protoplasm, large amount of starch, smaller intercellular spaces and highly branched plasmodesmata. Middle lamella of cells in abscission zone gets dissolved resulting into loosening of cells. After formation of abscission layers leaves are connected to parent plant by weak vascular tissues .Later vascular connection between leaves and stem gets broken, and leaves fruits and flowers get separated from parent plant.

After the abscission, outer layer of cells of stem or branch form a protective covering on exposed part (like i.e. abscission zone) by the development of periderm. These cells are suberized, tyloses are formed in vessels, and tyloses are bladder like protrusios from xylem parenhymma cells, which block xylem vessels.

20.3.2 Biochemical Changes

Biochemical Changes during abscission include production of hydrolytic enzymes such cellulose, peptic enzyme and lignase- which cause dissolution of middle lamella and primary walls of the cells in the abscission zone. The calcium pectate in the middle lamella is hydrolysed to pectic acid and water soluble pectin. Cellulose causes cell expansion in abscission zone. Lignases hydrolyse the lignin in xylem. Peroxidase activity is found to be also increased which might cause auxin destruction

20.3.3 Role of Hormones in Abscission

Hormones also play a significant role in abscission. Auxin and cytokinins retard abscission while abscisic acid and ethylene accelerate it. During the process endogenous auxin and cytokinins content decrease sharply ABA and ethylene increase sharply. Exogenous application of auxin or cytokinine delays abscission.

Endogenous gibberellin is also low at abscission zone but it is high in young developing leaves. Application of exogenous ABA at proximal end or distal end accelerates abscission normally. Exogenous application of ethylene promotes abscission of leaves, lowers petals and fruits. It is supposed that ethylene accelerates abscission by increasing respiration as well as permeability of the membrane.

20.3.4 Significance of Abscission

- It helps in diverting water and nutrients to the young leaves.
- It is a self pruning process through which fruits and injured organs are shed from the parent plant.
- It helps in disseminating fruits and vegetative propagates.
- Abscission serves as function in removing plant parts containing waste materials.
- Abscission separates senescent and dead parts of plants. Abscission of fruits helps in their dispersal and to repeat their life cycles.
- Abscission of leaves in deciduous plants helps in its water conservation during summer, when there is shortage of water. Leaf transpiration nil in such plants.
- In lower plants separation of vegetative parts like gemmae or plantules helps in vegetative reproduction.

20.3.5 Factors influencing Abscission

Many environmental factors influence abscission such as light, temperature, soil water status, minerals stress, oxygen and air pollutants.

1. Light

Low light intensity promotes abscission of organs like leaves, buds and fruits. Leaves of sciophytes (shade trees) are shed off prematurely

heliophytes (full light intensity plants) do not shad of prematurely. Longer photoperiod's retards abscission while shorter photoperiod accelerates abscission of leaves.Continuous red light represses abscission than far – red light. The inhibition of abscission by red light is reversible with far red light.

2. Soil water status

Abscission is enhanced under low water status to conserve water.

3. Minerals stress

Minerals stress also enhances abscission .N_2 reduces endogenous auxin production.

4. Oxygen content

If oxygen content is decreased beyond certain limit, abscission is also retarded. Oxygen has role in energy production through respiration and secretion of hydrolytic enzymes.

5. Air pollutants

Air pollutants generally accelerate abscission. Ozone induces abscission in leaves and a fruit, hydrogen fluoride accelerates leaf abscission in many species (e.g. *Bougainvillea*), Nitrogen dioxide induces abscission in *Citrus* spp.

20.4 Programmed Cell Death (PCD) or Apoptosis

Cell death occurs in almost all plant cells and tissues. Programmed cell death (PCD) is a fundamental process of life. During the evolution of multicellular organisms, the actively controlled demise of cells has been recruited to fulfil a multitude of functions in development, differentiation, tissue homeostasis, and immune systems. PCD occur as integral parts of plant development in a remarkable variety of cell types, tissues, and organs. PCD is involved in numerous processes.

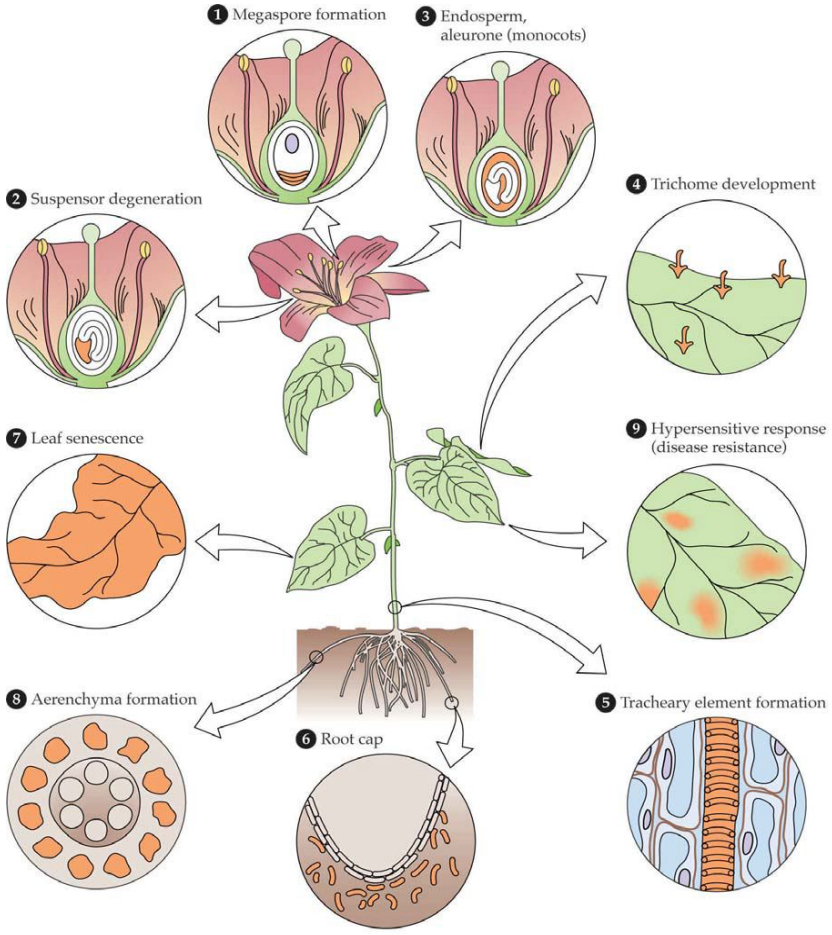

Fig. 20.2: PCD during Plant Development

The cells of a multicellular organism are members of a highly organized community. The number of cells in this community is tightly regulated—not simply by controlling the rate of cell division, but also by controlling the rate of cell death. If cells are no longer needed, they commit suicide by activating an intracellular death program. This process is therefore called **programmed cell death**, although it is more commonly called apoptosis (from a Greek word meaning "falling off," as leaves from a tree).

PCD in plants is a crucial component of development and defence mechanisms. There is no doubt as to the importance of PCD to plant growth and development and finding ways to manipulate plant PCD pathways may have agronomic benefits. Moving the field of plant PCD forward involves giving serious thought as to what form of cell death is actually being studied in a chosen system while remaining aware that these pathways may overlap. The elucidation of molecular pathways of caspase-dependent apoptotic cell death and the domination of this research

in studying eukaryotic PCD has led to the term apoptosis being used interchangeably with PCD. However, in plants, where true apoptosis doesn't appear to exist, this has led to use PCD as the 'blanket' term for different modes of cell death.

The field of plant 'PCD' it is apparent that at least three forms of cell death are described:

1. Autophagy
2. AL-PCD
3. Necrosis

The most common form of PCD induced in cell culture systems is AL-PCD, with necrosis occurring at higher levels of insult. Autophagy appears to be associated with the starvation response in cultured cells while AL-PCD has been shown to occur in both developmental death and hypersensitive response-induced death. Autophagy is the more common form of death during development. PCD is essential for normal vegetative and reproductive development. One example is the development of xylem tracery elements. In order to function efficiently as a conduit for water transport, the protoplast of the developing tracheary element must die and be removed at maturity.PCD also operates in the formation of parenchyma, a loose parenchyma tissue with large air spaces. Aerenchyma normally forms in the stems and roots of water lilies and other aquatic plants. These air spaces, created by a cell death program, provide channels for oxygen transport to the submerged portion of the plant. Even corn (Zea mays) and other terrestrial plants can be induced to form aerenchyma when subject to flooding. In the development of unisexual flowers, primordial for both the male and female flowers are present in the early stages. One or the other then aborts via a cell death program, leaving only one type of organ to complete development.

PCD is also an important factor in plant responses to invading pathogens and abiotic stress. When a plant recognizes a pathogen, for example, host cells in the immediate area of the infection undergo PCD. This deprives the invading pathogen of living tissue and either slows or prevents it spread.

20.5 Summary

The process of growing old is called ageing and death is the termination of functional life. The stages of developmental processes that ultimately lead to death of an organ or organism are called senescence.

Abscission is a complex physiological process that causes shedding of leaves, flowers and fruits.

If cells are no longer needed, they commit suicide by activating an intracellular death program. This process is therefore called programmed cell death, although it is more commonly called apoptosis.

20.6 Glossary

- **Senescence:** "to grow old," or biological aging (also spelled biological ageing)
- **Abscission:** is shedding of various parts of an organism, such as a plant dropping a leaf, fruit, flower or seed
- **Apoptosis:** In multicellular organisms, cells are a threat to the organism are destroyed by a tightly regulated cell suicide process
- **Autophagy:** an intracellular degradation system that delivers cytoplasmic constituents to the lysosome
- **Necrosis:** "death, the stage of dying, the act of killing" , is a form of cell injury which results in the premature death
- **Starvation:** a severe deficiency in caloric energy intake needed to maintain life.

20.7 Self-Learning Exercise

Section A : (Very Short Answer Type Questions)

1. Who proposed various types of senescence?
2. Write down full form of PCD.
3. Who proposed nutritional theory of senescence?
4. What do you mean by death hormone?
5. What is abscission?

Section B : (Short Answer Type Questions)

1. Define senescence

2. Write importance of plant senescence.
3. Write short note on pattern senescence in plant.
4. Write short note on PCD.
5. Write a note on hormonal regulation of senescence

Section C : (Long Answer Type Questions)

1. Describe physiological and biochemical changes during leaf.
2. Describe various type of senescence.
3. What are main causes of plant senescence?
4. Give a detailed description on plant senescence.
5. Explain the factors that influence senescence.

20.8 References

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