Course: B.Sc Botany
SEMESTER II
PAPER CODE: BOT CC 204
PAPER: Archegoniate
TOPIC: *Gnetum*
FACULTY: Isha Gaurav
Department of Botany
Email: ishagaurav86@gmail.com

Genus: *Gnetum*

1. Distribution of Gnetum:

Gnetum, represented by about 40 species is confined to the tropical and humid regions of the world. Nearly all species, except *G. microcarpum,* occur below an altitude of 1500 metres. Five species (*Gnetum contractum, G. gnemon, G. montanum, G. ula and G. latifolium*) have been reported from India . *Gnetum ula* is the most commonly occurring species of India.

2. Habit of *Gnetum*: Majority of the *Gnetum* species are climbers except a few shrubs and trees. *G. trinerve* is apparently parasitic. Two types of branches are present on the main stem of the plant, i.e. branches of limited growth and branches of unlimited growth. Each branch contains nodes and intenodes. Some scaly leaves are also present.

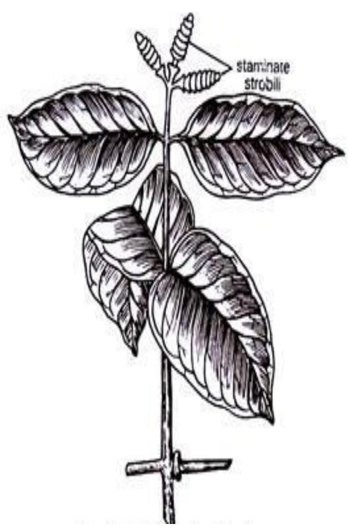


Fig: A branch of *Gnetum*

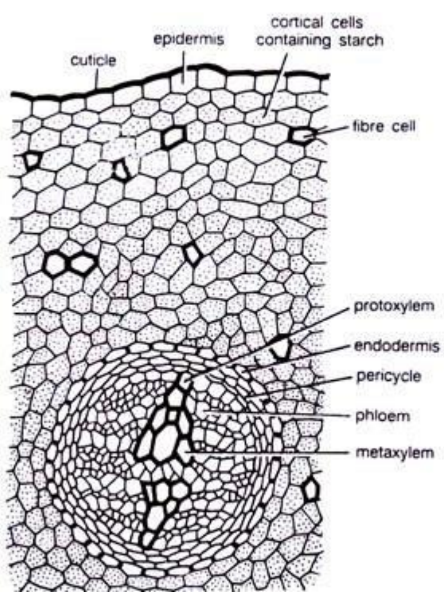
3. Anatomy of *Gnetum*:

(i) Root:

Young root has several layers of starch-filled parenchymatous cortex, the cells of which are large and polygonal in outline. An endodermal layer is distinguishable. Casparian strips are seen in the cells of the endodermis. The endodermis follows 4-6 layered pericycle. Roots are diarch and exarch. Small amount of primary xylem, visible in young roots, becomes indistinguishable after secondary growth.

The secondary growth is of normal type. A continuous zone of wood is present in the old roots . It consists of tracheids, vessels and xylem parenchyma. The tracheids have uniseriate bordered pits.

Vessels have simple or small multiseriate bordered pits. Some of the xylem elements have starch grains. Phloem consists of sieve cells and phloem parenchyma



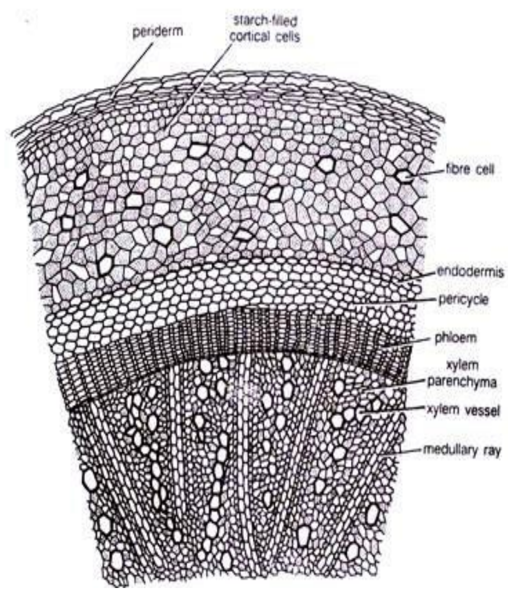


Fig: *Gnetum* T.S of young root **Fig: Gnetum T.S of old root showing secondary growth**

(ii) Young Stem:

The young stem in transverse section is roughly circular in outline, and resembles with a typical dicotyledonous stem. It remains surrounded by a single-layered epidermis, which is thickly circularized and consists of rectangular cells. Some of the epidermal cells show papillate outgrowths. Sunken stomata are present. The cortex consists of outer 5-7 cells thick chlorenchymatous region, middle few-cells thick parenchymatous region and inner 2-4 cells thick sclerenchymatous region. Endodermis and pericycle regions are not very clearly distinguishable. Several conjoint, collateral, open and endarch vascular bundles are arranged in a ring in the young stem. Xylem consists of tracheid and vessels. Presence of vessels is an angiospermic character. Protoxylem elements are spiral or annular while the metaxylem shows bordered pits which are circular in outline. The phloem consists of sieve cells and phloem parenchyma. An extensive pith, consisting of polygonal, parenchymatous cells, is present in the centre of the young stem.

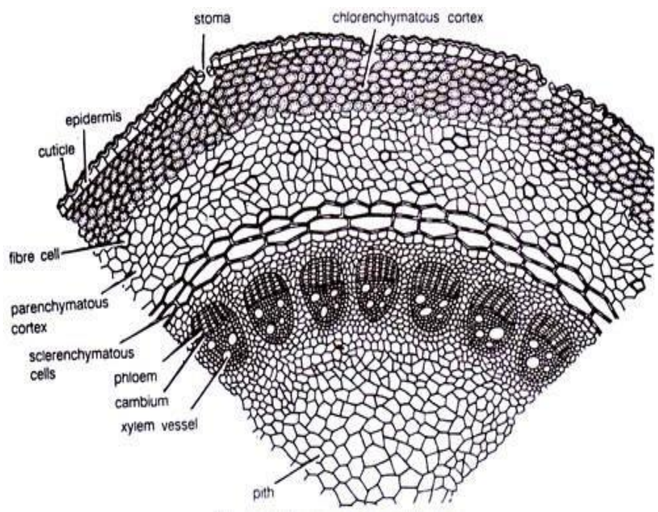


Fig: Gnetum T.S of young stem

(iii) Old Stem:

Old stems in *Gnetum* show secondary growth. In *G. gnemon* the secondary growth is normal, as seen also in the dicotyledons. But in majority of the species (e.g., *G. ula, G. africanum,* etc.) the anamolous secondary growth is present.

The primary cambium is ephemeral, i.e., short-lived. The secondary cambium in different parts of cortex develops in the form of successive rings, one after the other . The first cambium cuts off secondary xylem towards inside and secondary phloem towards outside. This cambium ceases to function after some time.

Another cambium gets differentiated along the outermost secondary phloem region, and the same process is repeated. In the later stages, more secondary xylem is produced on one side and less on the other side, and thus the eccentric rings of xylem and phloem are formed in the wood.

This type of eccentric wood is the characteristic feature of angiospermic lianes. The periderm is thin and develops from the outer cortex. It also possesses lenticels. The cortex also contains chlorenchvmatous and parenchymatous tissues along with many sclereids. In old stems the secondary wood consists of tracheids and vessels. Tracheids contain bordered pits on their radial walls while vessels contain simple pits. Transitional stages , containing one too many perforations in the terminal part of the vessels, are also seen commonly.

In tangential longitudinal section (T.L.S) of the stem, the wood xylem and medullary rays are visible. Bordered pits on both the radial and tangential walls are present. Medullary rays are either uniseriate or multiseriate and consist of polygonal parenchymatous cells. They are boat-shaped and their breadth varies from 2 to many cells. Sieve cells of the phloem contain oblique and perforated sieve plates.

(iv) Leaf:

Internally, *Gnetum* leaves also resemble with a dicot leaf. It is bounded by a layer of thickly circularized epidermis on both the surfaces. Stomata are distributed all over the lower surface except on the veins. The mesophyll is differentiated generally into a single-layered palisade and a well-developed spongy parenchyma.

The latter consists of many loosely-packed cells. Many stellately branched sclereids are present near the lower epidermis in the spongy parenchyma. Many stone cells and latex tubes are present in the midrib region of the leaf.

Several vascular bundles in the form of an arch or curve are present in the prominent midrib region . A ring of thick-walled stone cells is present just outside the phloem. Each vascular bundle is conjoint and collateral.

The xylem of each vascular bundle faces towards the upper surface while the phloem faces towards the lower surface. The xylem consists of tracheids, vessels and xylem parenchyma while the phloem consists of sieve cells and phloem parenchyma.

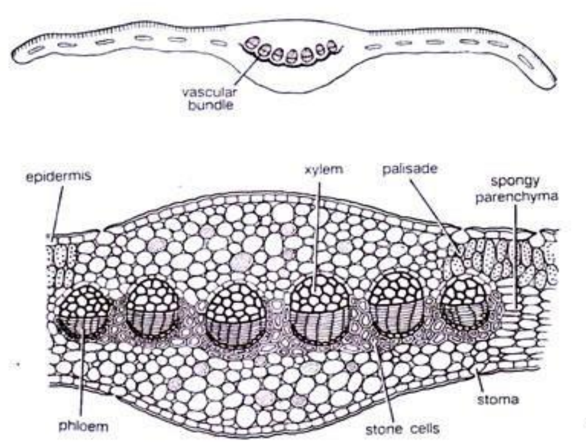


Fig: *Gnetum* T.S of leaf diagrammatic and cellular

4. Reproduction of Gnetum:

Gnetum is dioecious. The reproductive organs are organised into well-developed cones or strobili. These cones are organised into inflorescences, generally of panicle type. Sometimes the cones are terminal in position.

A cone consists of a cone axis, at the base of which are present two opposite and connate bracts. Nodes and internodes are present in the cone axis. Whorls of circular bracts are present on the nodes. These are arranged one above the other to form cupulas or collars . Flowers are present in these collars. Upper few collars may be reduced and are sterile in nature in *G. gnemon*.

Male Cone and Male Flower:

The male flowers are arranged in definite rings above each collar on the nodes of the axis of male cone. The number of rings varies between 3-6. The male flowers in the rings are arranged alternately. There is a ring of abortive ovules or imperfect female flowers above the rings of male flowers.

Each male flower contains two coherent bracts which form the perianth . Two unilocular anthers remain attached on a short stalk enclosed within the perianth. At maturity, when the anthers are ready for dehiscence, the stalk elongates and the anthers come out of the perianth sheath. In Gnetum gnemon a few (2-3) flowers are sometimes seen fusing each other.

Development of Male Flower

In very young cones, certain cells below each collar become meristematic. They divide repeatedly and form a small hump-like outgrowth. Certain cells on the upper side of this annular outgrowth start to differentiate into the initials of the ovules. They develop into abortive ovules which form the uppermost ring. The cells of the lower side of this annular outgrowth form the primordium of male flower.

A central cushion of cells develops by the repeated divisions in the male flower primordium. This cushion gets surrounded by a circular sheath called perianth. The sheath-like perianth encloses the central cushion-like mass only partially. With the development of a depression or notch in the central mass two lobes differentiate and later on develop into two anther lobes.

With the help of many divisions the basal portion of this central mass of cells starts to differentiate into a stalk. This stalk elongates and pushes the anther lobes towards the outer side. Each anther lobe remains surrounded by an epidermal layer and a few wall layers which enclose a microsporangium.The innermost wall layer enclosing the sporogenous tissue is known as tapetum.

The sporogenous cells become loose, contract, round up and change into the spore mother cells. In the process of microspore formation the tapetum and two wall layers are used for the developing microspores. The spore mother cells undergo meiosis and ultimately the spore tetrads are formed.

The characteristic radial thickenings develop in the epidermal cells. They help in the dehiscence of microsporangium. The microspores are ornamented.

Female Cone:

The female cones resemble with the male cones except in some definite aspects. A single ring of 4-10 female flowers or ovules is present just above each collar . Only a few of the ovules develop into mature seeds .

In the young condition, there is hardly any external difference between female and male cones. All the ovules are of the same size when young but later on a few of them enlarge and develop into mature seeds. All the ovules never mature into seeds.

Ovule or Female Flower:

Each ovule consists of a nucellus surrounded of three envelopes. The nucellus consists of central mass of cells. The inner envelope elongates beyond the middle envelope to form the micropylar tube or style. The nucellus contains the female gametophyte. There is no nucellar beak in the ovule of *Gnetum*.

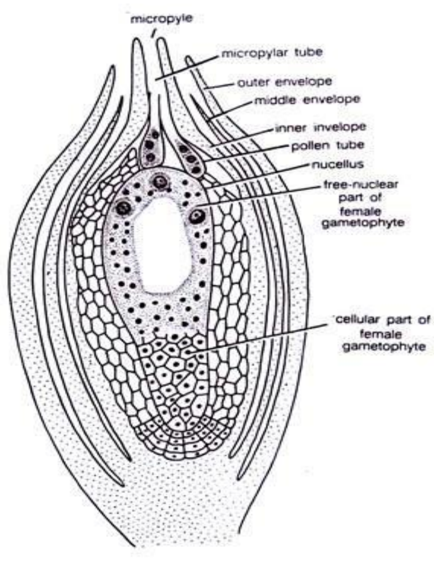


Fig: L.S of Ovule

Abnormal Cones:

More than one rings of ovules in the male cones in *Gnetum gnemon* have been reported by Thompson (1960) and Madhulata (1960). Collars, arranged spirally in the female cones of *G. gnemon* and *G. ula* have been observed by several workers including Maheshwari (1953). Pearson (1912) reported some cones bearing only two collars in *G. buchholzianum*. Rarely, the lower collars in the male cones bear one or two fertile ovules whereas normal male flowers are present in the upper collars of the same cone.

Mega-Sporangium, Mega-Sporogenesis and Female Gametophyte:

Four to ten ovular primordia differentiate on the annular meristematic ring. This ring develops below each collar of the female cone in the same manner as that of the male cone. The ovular primordium divides and re-divides several times to form a mass of cells.

All the three envelopes of the female flower develop around this mass of cells The innermost third envelope remains fused with the nucellus at the base while its upper portion remains free and form the long micropylar tube or **'style'.**

In the young conditions, an outer epidermal layer is distinguishable in the nucellus. Two to four archesporial cells develop below the epidermis at a later stage. The archesporial cells divide periclinally to form outer primary' parietal cells and inner sporogenous cells. The primary parietal cells and the epidermal layer divide periclinally and anticlinally several times resulting into a massive nucellus.

The sporogenous cells divide and re-divide to form megaspore mother cells which remain arranged in linear rows. All the megaspore mother cells may divide reductionally and form tetrasporic embryo-sacs but ultimately all, except one, degenerate.

As many as 256 (*Gnetum gnemon*) to 1500 (*G. ula*) free-nuclei are formed in the female gametophyte leaving a vacuole in the centre . The female gametophyte is tetrasporic in development. It is broader towards the micropylar end and it tapers towards the chalazal end.

The nuclei near the chalazal end get surrounded by cell walls while those towards micropylar end remain free. Gametophyte is thus partly cellular and partly-nuclear. The archegonia are absent in *Gnetum*.

Certain nuclei near the micropylar end start to function as egg nuclei. According to Swamy (1973) the only nucleus in a uninucleate cell or one of the nuclei in a multinucleate cell enlarges and functions as the egg in *G. ula*. The nucellar beak is absent in Gnetum.

The megaspore mother cell divides reductionally and forms four free haploid nuclei in the mother cell. Megaspore tetrads are never formed in Gnetum.

Microsporangium and Micro-Sporogenesis:

Development of the microsporangium can be studied only in young anthers. Two archesporial cells are distinguished below the epidermal layer . Archesporial cells divide and re-divide to form many-celled archesporium . The outermost layer of the archesporium divide periclinally to form an outer layer of parietal cells and inner layers of sporogenous cells .

The parietal cells form the wall layers and tapetal layer by periclinal divisions The sporogenous cells develop into microspore mother cells by some irregular divisions. Tapetal cells later on become bi-nucleate . Microspore mother cells divide reductionally to form haploid microspores. The microspores may be arranged in isobilateral, decussate or tetrahedral manner in their earlier stages. Side by side the wall cells and the tapetal cells degenerate and ultimately dis-organise. The epidermal cells become thick, cutinized and radially elongated. Many fibrous thickenings also develop in these cells. Small globular structures are present on the inner surface of the epidermis in Gnetum ula and *G. gnemon*. Anthers dehisce along a double row of small cells which extends from the tip towards the base.

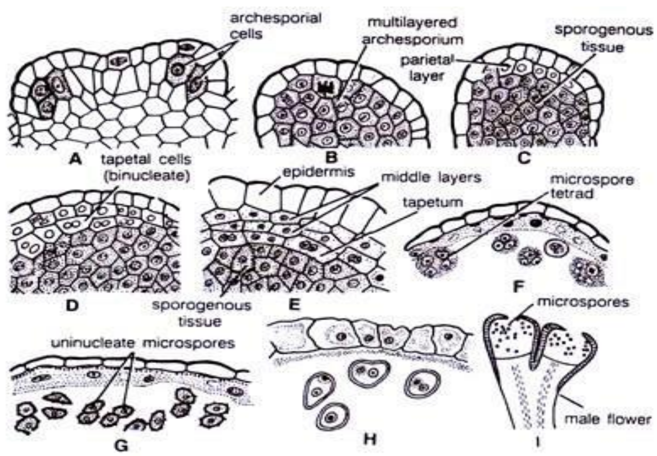


Fig: Development of microsporangium in *Gnetum*

Male Gametophyte:

Pollen grains or microspores are roughly spherical in outline. They are uninucleate and remain surrounded by a thick and spiny exine and thin intine. Mature pollen grains are shed at three-nucleate stage. These include prothallial nucleus, tube nucleus and generative nucleus .

This three-nucleate stage is reached by first dividing the microspore nucleus mitotically into two and then one of them again gets divided. Further development is affected only in the pollen chamber. The intine comes out by rupturing the exine and forms a pollen tube.

The tube nucleus migrates into the pollen tube. The generative nucleus also adopts the same course and divides into two unequal male gametes in the tube. Prothallial nucleus does not enter the pollen tube.

Thompson (1916) opined that the prothallial cell does not form at all in the male gametophyte . The microspore nucleus divides into a tube nucleus and a generative cell. The latter divides into a stalk cell and body cell. The tube nucleus and body cell enter in the pollen tube where the body cell divides into two equal male gametes.

According to Negi and Madhulata (1957) the microspore nucleus in Gnetum gnemon and G. ula divides into a small lenticular cell and a large cell (Fig. 13.20, Lower). The lenticular cell does not take part in the further development and ultimately disappears.

The other large nucleus divides into a tube nucleus and a generative cell, both of which pass into the tube. The generative cell divides into two equal male gametes in the tube. A stalk cell is never formed in these species.

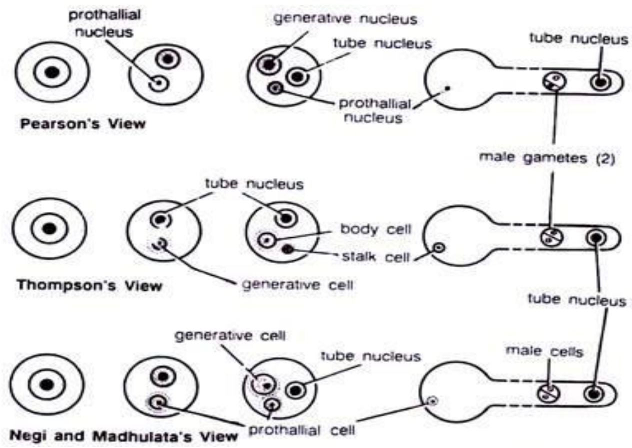


Fig: showing development of male gametophyte

Pollination:

Wind helps in carrying the pollen grains up to the micropylar tube of the ovule. The micropylar tube secretes a drop of fluid in which certain pollen grains get entangled and reach up to the pollen chamber. The nucellus cells below the pollen chamber are full of starch.

Fertilization:

The fertilization in *Gnetum* has been studied only by a few workers. Vasil (1959) studied this phenomenon in *G. ula*. At the time of fertilization, the pollen tube pierces through the membrane of the female gametophyte just near to a group of densely cytoplasmic cells. The tip of pollen tube bursts and the male cells are released. One of the male cells enters the egg cell.

The male and female nuclei, after lying side by side for some time, fuse with each other and form the zygote. According to Swamy (1973), the only identifying features of the zygote are its spherical shape and dense cytoplasm. Both the male cells of a pollen tube may remain functional if two eggs are present close to the pollen tube.

Endosperm:

In all gymnosperms, except Gnetum, a cellular endosperm develops before fertilization. In *Gnetum*, the cell formation, although starts before fertilization, a part of the gametophyte remains free-nuclear at the time of fertilization.

After fertilization the wall formation in the female gametophyte starts in such a way that the cytoplasm gets divided into many compartments. Each of these compartments contains many nuclei.

All the nuclei of one compartment fuse and form a single nucleus. The wall formation starts from the base and proceeds upwards. The wall formation varies greatly in *Gnetum*. Only the lower portion of the gametophyte may become cellular leaving the remaining upper portion free-nuclear. Sometimes the entire gametophyte may become cellular.

The Embryo:

The embryo development in several species of *Gnetum* has been studied by many different workers including Lotsy (1899), Coulter (1908) and Thompson (1916), but the details put forward by these wokers are highly variable.

Maheshwari and Vasil (1961) have stated that in all the angiosperms the first division of the zygote is accompanied by a wall formation but in all gymnosperms, except Sequoia sempervirens, these are free-nuclear divisions in the zygote. Gnetum in this respect forms a link in between gymnosperms and angiosperms by showing both free-nuclear divisions as well as cell divisions.

Thompson (1916) opined that a two-celled pro-embryo is formed . From each of these two cells develops a tube called suspensor . Now the nucleus divides and one of the two nuclei undergoes free-nuclear divisions forming four nuclei. The embryo gets organised by these four nuclei There is no division in the other larger nucleus..

Madhulata (1960) has worked on the zygote development in Gnetum gnemon. According to her 2-4 or sometimes up to 12 zygotes may develop in a gametophyte, of which normally one remains functional. From the zygote develops generally one or sometimes 2-3 small tubular outgrowths.

Only one of these tubes receives the nucleus and survives while the remaining tubes disintegrate and soon die. The surviving outgrowth elongates, becomes branched and grows into different directions through the intercellular spaces of the endosperm. All the primary suspensor tubes usually remain coiled round each other.

A small cell is cut off at the tip of the primary suspensor tube in *Gnetum gnemon*. It soon divides first transversely and then longitudinally resulting into four cells. Now irregular divisions take place forming a group of cells. Some of these cells divide and elongate to form secondary suspensor . The remaining cells at the tip form the embryonal mass.

In *Gnetum ula* a small cell is cut off at the tip of the tube called peculiar cell. This peculiar cell soon divides and forms a group of cells. The secondary suspensor and embryonal mass are differentiated from this group of cells. By this time, the wall of the tube starts to become thick.

What so ever may be the pattern of formation of the embryonal mass and secondary suspensor, the cells of the former are small, compact, dense in cytoplasm and develop into embryo-proper while that of the latter (i. e. secondary suspensor) are thin-walled, uninucleate and highly vacuolated.

The primary and secondary suspensors help in pushing the embryo into the endosperm. Soon a stem tip with two lateral cotyledons form in the tip region of the embryonal mass. On the opposite side develop the root tip with a root cap.

A feeder develops after the formation of stem and root tips . The feeder is a protuberance-like structure present in between root and stem tips. Thus, the stem tip, two cotyledons, feeder, root tip and root cap are the parts of a mature embryo.

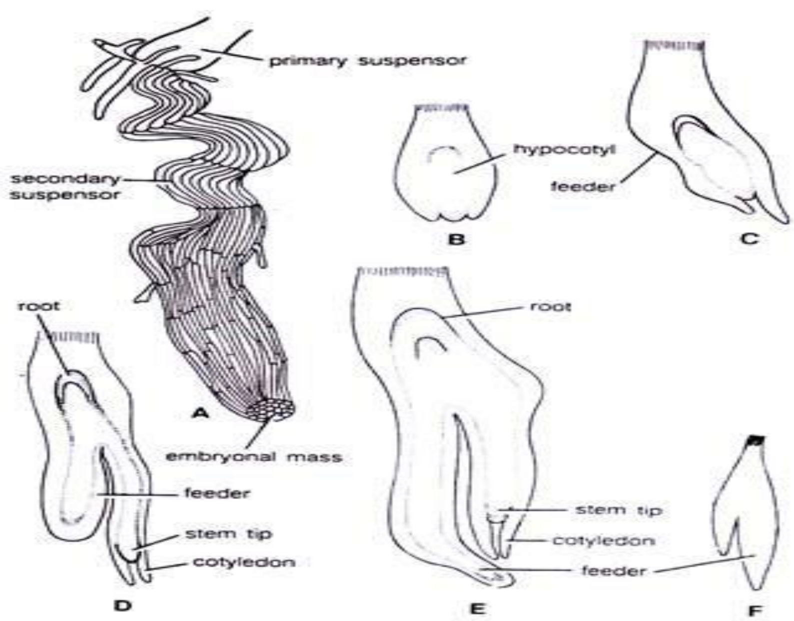


Fig: Development of embryo in *Gnetum ula*

Seed:

Gnetum seeds are oval to elongated in shape and green to red in colour. It remains surrounded by a three-layered envelope which encloses the embryo and the endosperm. Outer envelope is fleshy, and consists of parenchymatous cells. It imparts colour to the seed.

The middle envelope is hard, protective and made up to three layers, i.e., outer layer of parenchymatous cells, middle of palisade cells and innermost fibrous region. The inner envelope is parenchymatous. Branched vascular bundles traverse through all the three envelopes.

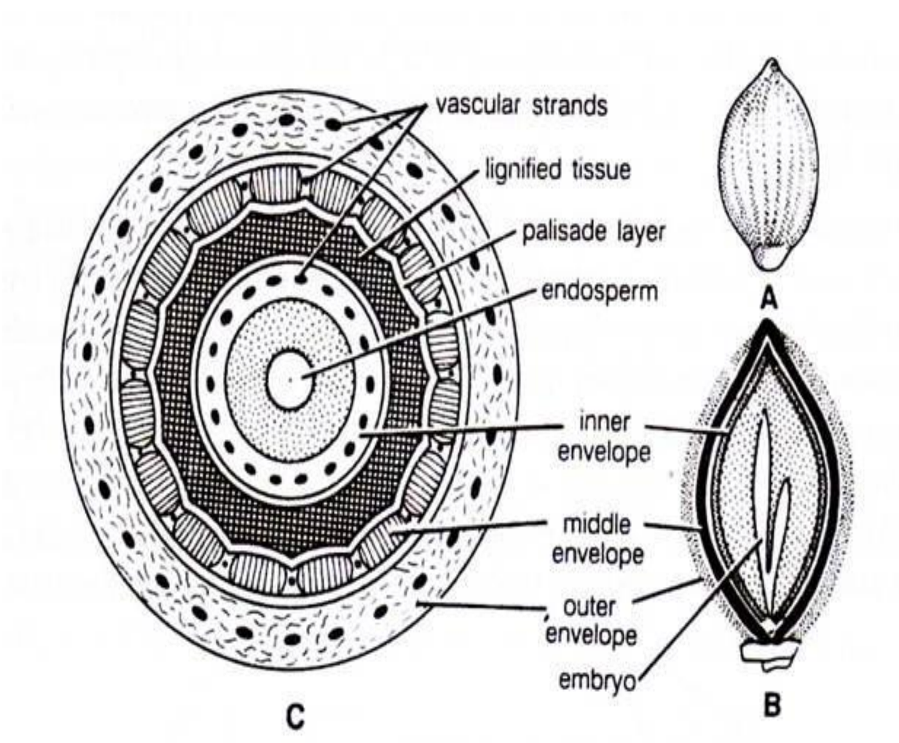


Fig: *Gnetum spp* A) entire seed B) L.S of seed C) T.S of seed

Germination of Seed:

Germination is of epigeal type . The cotyledons are pushed out of the seed. The hypocotyl elongates, and this brings the cotyledons out of the soil. The first green leaves of the plant are formed by the cotyledons. The first pair of foliage leaves is produced by the development of plumule. A persistent feeder is present up to a very late stage in the seed.

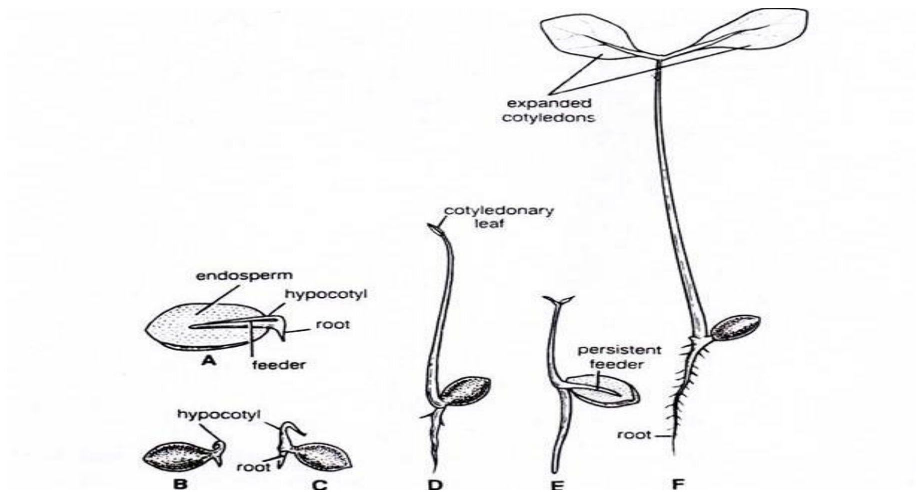


Fig: Germination of seed in *Gnetum gnemon*