

## 1.4. *Marsilea*

### Structure

- *Marsilea* is commonly known as “pepper wort” or “water fern” (although it is a fern but hardly resembles a true fern).
- It is represented by about 53 species which are cosmopolitan in distribution but abundantly found in tropical countries like Africa and Australia.
- Either the species are hydrophytic or amphibious i.e., they grow rooted in mud or marshes and shallow pools or are completely submerged or partially or entirely out of water in wet habitats.

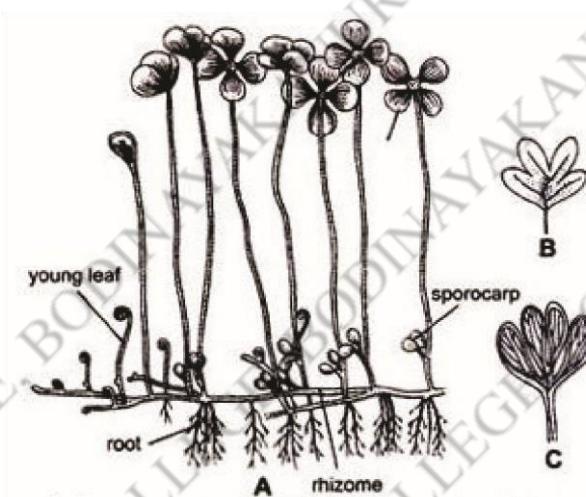


Fig.1.25. A-C). *Marsilea*. External features. A. External morphology; B. Leaf showing arrangement of segments as a result of three dichotomies, C. Pinna showing venation.

### External Features of *Marsilea*:

The mature sporophyte is an herbaceous plant. Its underground rhizome spreads in a diameter of 25 meter or more. The plant body is distinctly differentiated into rhizome, leaves and roots.

#### 1. Rhizome:

All the species possess a rhizome which creeps on or just beneath the soil surface. It is slender, dichotomously branched with distinct nodes and internodes and is capable of

indefinite growth in all directions as a result of which it occupies an area of 25 metre or more in diameter.

## 2. Leaves:

They are borne alternately on upper side of rhizome at nodes, in two rows. Young leaves show circinate vernation (like ferns). In some species young leaves are covered with multicellular hairs. The leaves are compound, with basal petiole and terminal lamina.

In submerged plants the petiole is a long and flexible structure and the lamina floats over the surface of water but in muddy or marshy plants the petiole of the leaf is short and rigid with short lamina spreading in the air.

The lamina consists of 4 leaflets (pinnae) which are present at the apex of petiole. The 4 leaflets arise as a result of 3 dichotomies of the lamina in close succession to each other i.e., 2 leaflets arise slightly higher than other two. At night the pinna are folded upwardly. This is known as sleeping movement of pinna. Near the base of petiole the stalked bean-shaped sporocarps are borne.

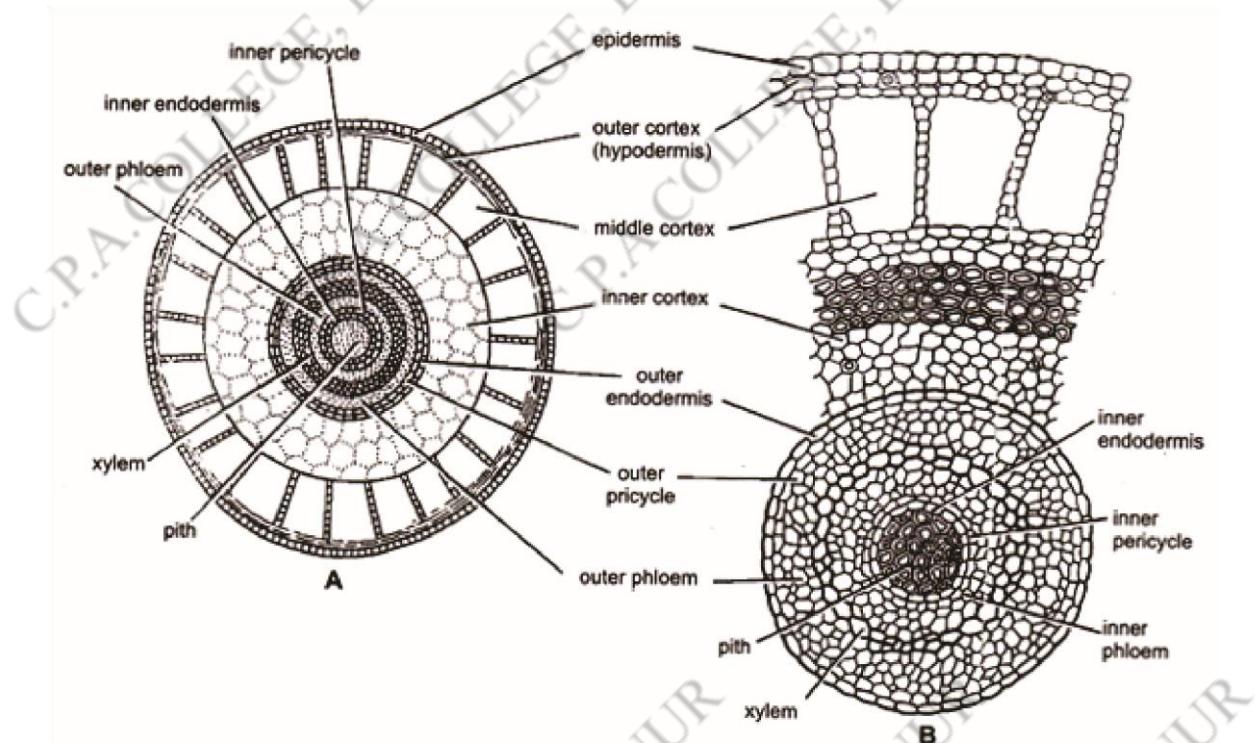


Fig. 1.26. A&B, *Marsilea* Internal structure on rhizome.

### **3. Roots:**

The roots are adventitious, arising from the underside of the node of rhizome, either singly or in groups. In certain cases the roots are given out even from the internodes (*M. aegyptiaca*).

### **Internal Structure of *Marsilea*: T.**

#### **S. Rhizome (stem):**

A T. S. of the young rhizome shows a protostelic structure i.e., pith is absent and xylem is completely surrounded by phloem but in the old stem pith is developed in the centre and the stele is amphiphloic siphonostelic type.

#### **Epidermis**

It is the outermost limiting layer of single celled thick parenchymatous cells. The stomata are absent.

#### **Cortex**

It is differentiated into three regions – the outer cortex, the middle cortex and the inner cortex.

#### **Outer cortex**

It is present just below the epidermis (also called hypodermis). It is parenchymatous and may be one to several cells thick. Some of its cells contain tannin.

#### **Middle cortex**

It is also called aerenchyma. It lies below the hypodermis. It consists of large air spaces (chambers) separated by one cell thick parenchymatous septa. In the xerophytic species e.g., *aegyptiaca* the air chambers are obliterated.

#### **Inner cortex**

It is a solid tissue of several cells thickness. The outer layers are thick walled (sclerenchymatous) while the inner layer of cells is thin walled (parenchymatous) and compactly arranged. Some of these cells are filled with starch or tannin.

#### **Stele**

Stele is amphiphloic siphonostele i.e., in the centre there is a pith which may be either parenchymatous (aquatic species) or sclerenchymatous (terrestrial muddy species).

Xylem is present in the form of a complete ring which is surrounded on both sides by a complete ring of inner and outer phloem, pericycle and endodermis.

In this way the continuation of different tissues in the form of complete ring in stele is as follows—outer endodermis, outer pericycle, outer phloem, xylem, inner phloem, inner pericycle and inner endodermis. The protoxylem may be well defined exarch (*M.vestita*) or mesarch (*M.aegyptiaca*) or ill defined (*M.quadrifolia*).

A T. S. of the nodal region shows an amphiphloic solenostelic condition and is provided with one leaf gap.

## 2. T. S. of Petiole:

A T. S. of the petiole is somewhat circular in outline and is differentiated into epidermis, cortex and stele.

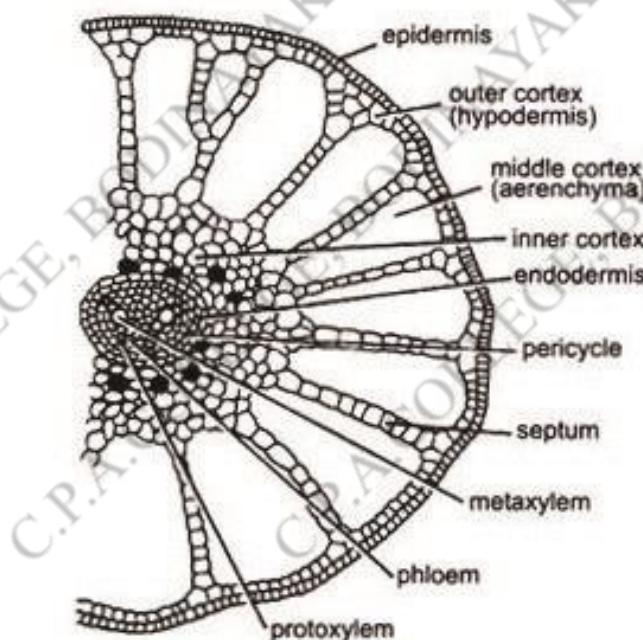


Fig. 1.27. *Marsilea quadrifolia*, T.S. of petiole

## Epidermis

It is the outermost layer of single cell thickness. The cells are parenchymatous and slightly elongated.

## **Cortex**

It is differentiated into three regions: The outer cortex, the middle cortex and the inner cortex.

### **Outer cortex**

It is present just below the epidermis, (also called hypodermis). It is made of thin walled cells (parenchymatous).

### **Middle cortex**

It lies below the hypodermis and called aerenchyma. It consists a ring of air chambers. The air chambers are separated by single layered partitions of thin-walled parenchymatous cells.

### **Inner cortex**

It is a solid tissue of several cells thickness. The cell layers are parenchymatous and contain starch and tannin filled cells. In *M. minuta* few sclerenchymatous layers are also present just inner to middle cortex.

## **Stele**

It is somewhat triangular in outline and is of protostelic type i.e. pith is absent. Xylem is "V" shaped with 2 distinct arms. Each arm is provided with metaxylem elements in the centre and protoxylem is situated at both the margins i. e., protoxylem is exarch.

The xylem is surrounded on all sides by phloem. Phloem is externally surrounded by a single layer of parenchymatous pericycle which, in turn, is bounded by a single layered endodermis.

## **Transverse Section of Leaflet:**

A. T. S. of the leaflet shows epidermis, mesophyll and vascular bundles.

## Epidermis

It is the outermost surrounding layer and is only one cell in thickness. It is differentiated into upper and lower epidermis. In floating leaflets the stomata are present on the upper epidermis but in case of plants growing in mud or moist soil where the leaves are aerial, the stomata are present both on upper as well as lower epidermis.

## Mesophyll

It occupies a wide space between upper and lower epidermis. It is usually differentiated into an upper palisade tissue and lower spongy parenchyma. The palisade tissue is made up of elongated cells provided with chloroplast. The spongy tissue is made up of loosely arranged parenchymatous cells with large air spaces separated by single layered septa. In submerged species, however, the mesophyll is not differentiated into palisade and spongy parenchyma.

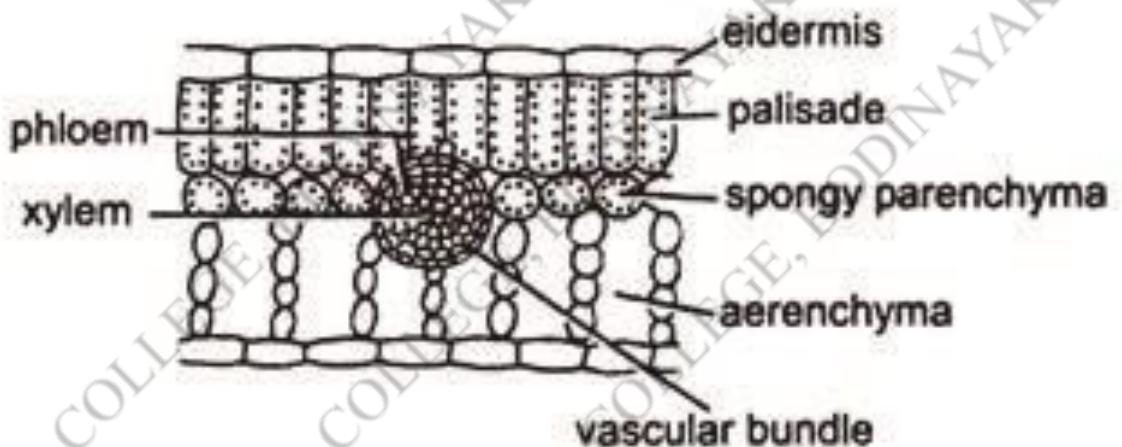


Fig. 1.28. *Marsilea quadrifolia*, V.S. of leaflet or pinna

## Vascular bundles

In between the mesophyll tissue are present several vascular bundles. Each vascular bundle is concentric and amphicribal type i. e., made up of a centrally situated xylem, surrounded on all sides by phloem. The phloem is enclosed by a single layered thick endodermis.

## T. S. Root

A T. S. of root is somewhat circular in outline and can be differentiated into epidermis or piliferous layer, cortex and stele.

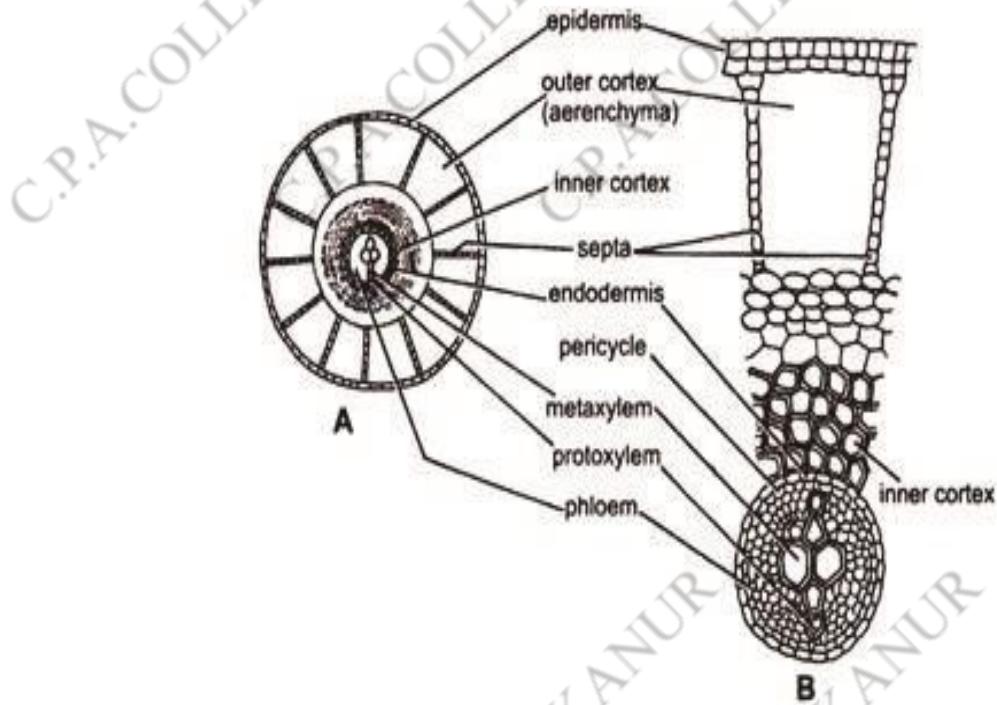


Fig. 1.29. A & B- *Marsilea quadrifolia*, internal structure of root.

### Epidermis

It is the outermost, parenchymatous, single layered covering.

### Cortex

It can be differentiated into two parts: outer cortex and inner cortex. The outer cortex consists of large air chambers arranged in the form of a ring (parenchymatous). These chambers are separated from each other by longitudinal septa. The inner cortex is differentiated into outer parenchymatous and inner sclerenchymatous regions. The inner cortex is delimited by single layered thick endodermis.

### Stele

It is of protostelic type and occupies the central position. It is devoid of pith. Xylem is situated in the centre which is diarch and exarch. It is surrounded by phloem. The phloem is bounded externally by a single layer of pericycle.

#### 1.4.1. Reproduction in *Marsilea*:

*Marsilea* reproduces by two methods:

(i) Vegetative reproduction (ii)

Sexual reproduction.

### **1. Vegetative reproduction:**

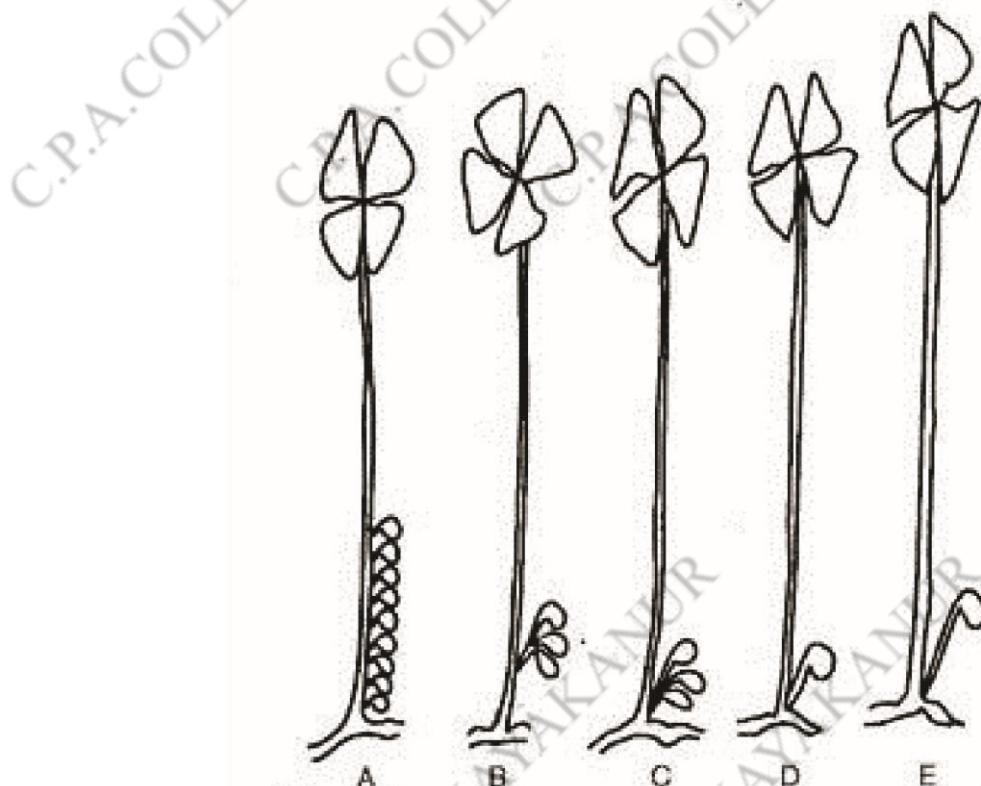
Under some unfavourable circumstances the subterranean branches of the rhizome form tubers.

These structures have reserve food in the form of oil globules which help them to overcome the unfavourable conditions.

On return of the favourable conditions, these tubers germinate and form new plant body (e.g., *M. hirsuta*, *M. minuta*, *M. erosa*).

### **2. Reproduction by Spores:**

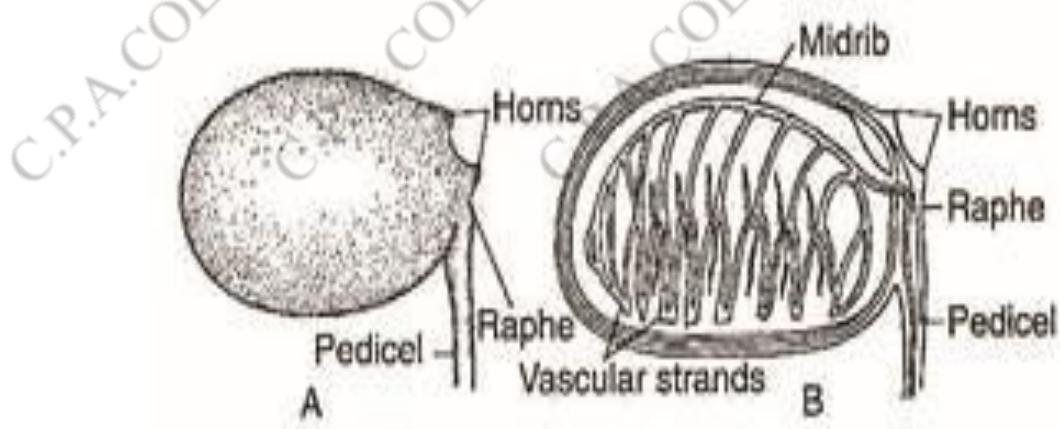
- *Marsilea* is a heterosporous fern. It produces two types of spores i.e., the microspores and the megaspores. The microspores and megaspores are produced in microsporangia and megasporangia, respectively, and the sporangia are enclosed in special bean-shaped structures called sporocarps.
- When young, the sporocarps are soft and green, but turns dark brown and hard at maturity. Sporocarps withstand desiccation and are reported to be viable even after twenty to twenty five years.
- The sporocarp develops at the short branch of petiole called pedicel or stalk. In most species they occur singly, but in some species the number varies from two to twenty.



**Fig.1.30 :** *Marsilea* : Attachment of sporocarps in various species. A. *M. polycarpa*, B. *M. quadrifolia*, C. *M. minuta*, D. *M. coromandelica*, E. *M. uncinata*

**The mode of attachment of sporocarps with the petiole is of following types:**

- Sporocarps are attached to one side of the petiole in a single row e.g., *M. polycarpa*, *M. caribaca*, *M. subangulata*, *M. detlexa*.
- Pedicels of the sporocarps are fused in groups and then attached to the petiole by a common stalk e.g., *M. quadrifolia*.
- Pedicels of all the sporocarps remain free and are attached to the petiole at a single point (e.g., *M. minuta*).
- A single sporocarp attached to the base of the petiole by the pedicel (e.g., *M. coromandelica*, *M. uncinata*).



**Fig 1.31. *Marsilea* : A. A sporocarp, B. L.S. of sporocarp showing vascular stands on one valve**

- The place of attachment of the pedicel with the body of the sporocarp is known as raphe. The distal end of the raphe in some species is marked by the presence of one or two protuberances or teeth-like projections known as horns or tubercles.
- The sporocarp wall is hard, thick, thus resistant against mechanical injury. Anatomically, the wall is differentiated into three layers. The outer layer is epidermis made up of single-layered cuboidal cell with sunken stomata.
- The middle layer is made up of radially elongated compactly arranged thick-walled palisade cells. This is followed by second palisade layer which is comprised of more elongated thin-walled palisade cells.
- A vertical longitudinal section (VLS) in the plane of the stalk shows that a single strong vascular strand enters the sporocarp near the lower horn and continues forward all along the dorsal or upper side of the sporocarp thus forming a midrib (dorsal bundle).
- From this midrib, the lateral side branches (lateral or commissural bundles) arise which eventually pass on to the two sides of the sporocarp. The sporocarp, therefore, has a bivalved structure.
- Another bundle called placental bundle develops from the point of forking of lateral bundle which enters into the receptacle bearing sporangia and dichotomises. Thus a closed network of vascular system is formed within the sporocarp.
- A vertical longitudinal section (VLS) of sporocarp away from the plane of the stalk reveals many sori arranged in vertical rows. In this plane of section either megasporangia or microsporangia are visible. Each sorus is surrounded by an indusium.

The development of sori is of gradate type. The gelatinous mucilage ring is more prominent in dorsal side.

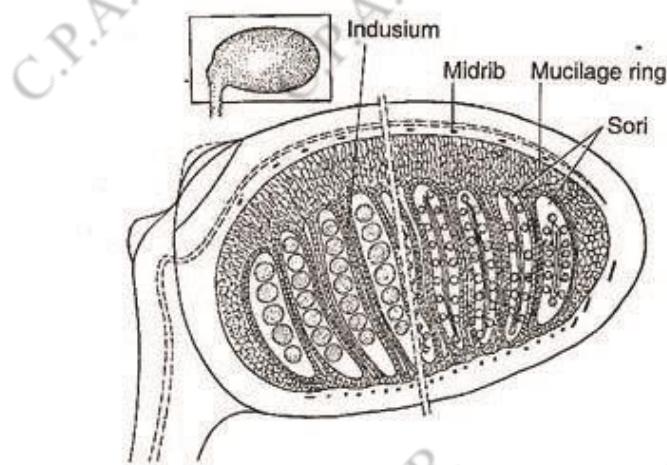
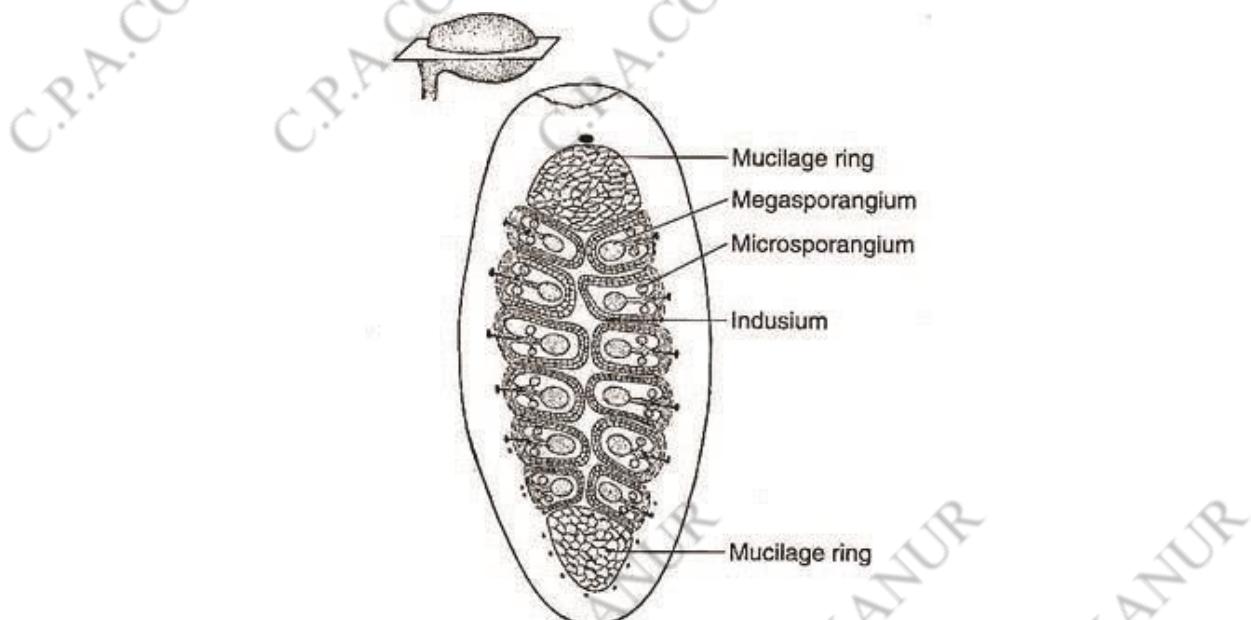


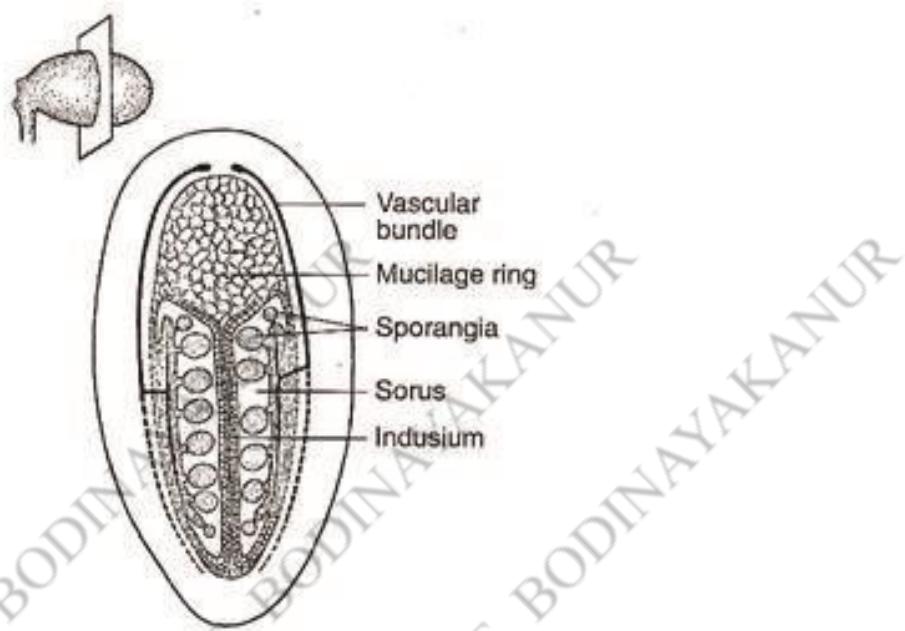
Fig 1.32. *Marsilea* (vertical) L.S. of sporocarp

- A horizontal longitudinal section (HLS) cuts each sorus transversely and it is seen that each sorus is an elongated structure, covered by a delicate indusium.
- The sori are gradate, basipetal in arrangement with a row of largest sporangia (megasporangia) at top and two rows of smaller sporangia (microsporangia) on two sides. The mucilage ring is present in the form of two masses, one in the dorsal and the other in the ventral sides.



**Fig 1.32. *Marsilea* (horizontal) L.S. of sporocarp**

- A vertical transverse section (VTS) of the sporocarp shows only the sori on two sides. Both the sori contain megasporangia if the section is taken through the megasporangia or the sori contain only microsporangia if it is taken through the microsporangia. The sporophore is seen in the form of two masses on either side. The mucilage ring is present only on the dorsal side.



**Fig 1.33. *Marsilea* (vertical) T.S. of sporocarp**

#### **Development of Sporangium:**

- Sporangial development is of the leptosporangiate type. The development of both the micro- and megasporangia is almost alike. The sporangial initials for megasporangium and microsporangium are formed at top and at the sides of the receptacle, respectively.
- The initial cell divides periclinally (transversely) into an outer and an inner cell. The inner cell does not take part in further development. The outer cell undergoes three successive diagonal divisions and forms a tetrahedral apical cell with three cutting faces.
- This apical cell cuts off two segments along each of its three cutting faces which forms the stalk of the sporangium. Now the apical cell divides with the help of an arched periclinal wall towards its outer face and forms an outer jacket initial and an inner

tetrahedral archesporial cell. The outer jacket initial divides anticlinally to form a single-layered jacket.

- The archesporial cell divides periclinally to form an outer tapetal initial and an inner primary sporogenous cell. Anticlinal and pericinal divisions of the tapetal initial forms a two-layered thick tapetum.
- The primary sporogenous cell divides to form a mass of either 8 or 16 spore mother cells. Spore mother cells ( $2n$ ) undergo meiotic division to form 32 or 64 haploid spores ( $n$ ). The developments of both the sporangia are similar up to this stage.
- In megasporangium, only one megaspore survives to become a large functional megaspore, while all the microspores are functional in microsporangium.

#### **Opening of the Sporocarp:**

- The sporangium wall of *Marsilea* shows no sign of cellular specialisation (e.g., formation of annulus) required for dehiscence of sporangium. A sporocarp is a hard structure and it does not open until two or three years after their formation. This delay is probably due to the imperviousness of the hard sporocarp wall.
- Hence the sporocarp may remain viable for even 50 years. The tissues slowly swell up by absorbing water in natural conditions. Thus the swelling puts pressure on the wall of the sporocarp and eventually it splits open along its ventral side into two halves.
- Splitting is followed by the emergence of a long, worm-like gelatinous structure to which the sori are attached. The mucilaginous cord may become ten or fifteen times larger than the sporocarp. Following the release of the sori from the sporocarp, the indusia and the sporangial wall disintegrate and the spores are liberated.

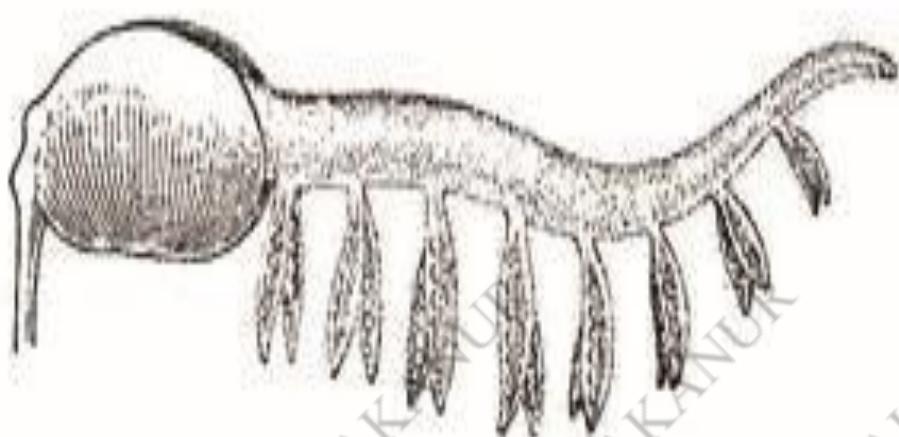


Fig 1.34. *Marsilea* Extrusion of the sori on the mucilaginous cord

**Gametophyte:**

*Marsilea* is heterosporous i.e., they produce microspores and megasporangia which eventually germinate to form the male and female gametophytes, respectively.

**Male Gametophyte:**

- The microspores are small, globose structures with a thick outer ornamented exine and inner thin intine. The outer exine is covered by a thin layer called perispore.
- The microspore contains a distinct haploid nucleus and its cytoplasm is rich in starch grains.

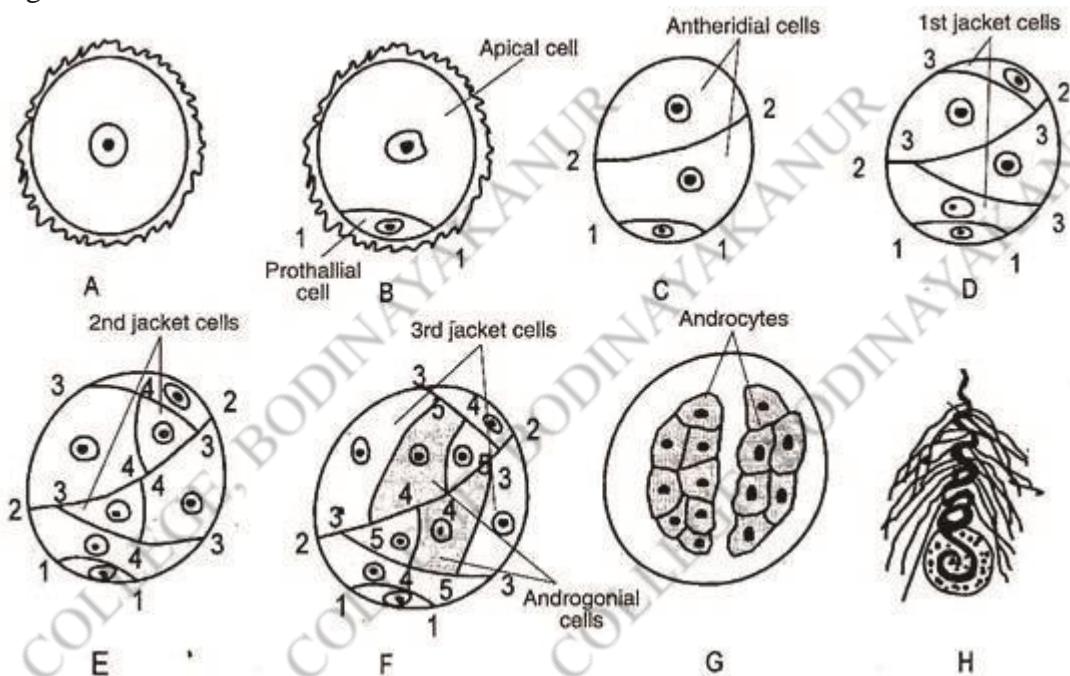


Fig 1.35. *Marsilea* : A-G Stages in the development of male gametophyte H. A sperm

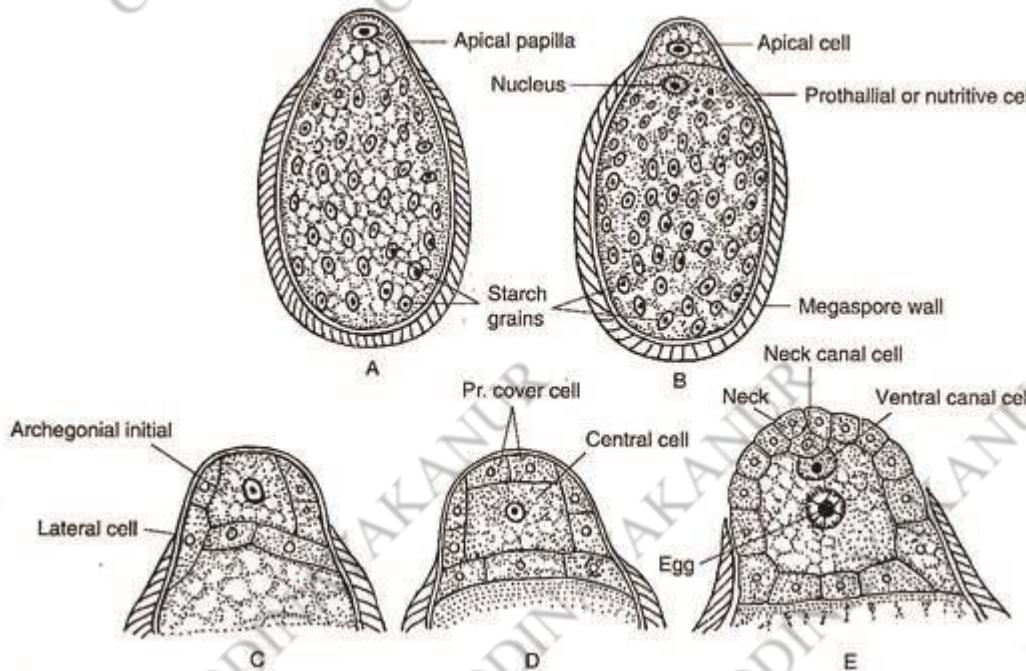
- The microspores germinate inside the spore wall (endosporic type) almost immediately after its release. It divides asymmetrically to form a small prothallial cell and a large apical cell (1-1). A division (2-2) of apical cell diagonal to prothallial cell forms two antheridial cells.
- Then both the antheridial cells divide diagonally (3-3) with curving wall forming the first jacket cell and large wedge-shaped cell. The jacket cells do not divide, but the wedge-shaped cell divides periclinally (4-4) to form smaller inner cell (2nd jacket) and a large outer cell. Further, the periclinal division (5-5) of outer cell forms 3rd jacket and primary androgonial cell.

- At this stage the male gametophyte consists of one prothallial cell, 6 jacket cells and 2 androgonial cells. After several divisions of the primary androgonial cells, sixteen androcytes are formed surrounded by jacket cells.
- Later the prothallial cell and the jacket cells disintegrate and the two groups of androcytes, representing the two antheridia, float freely in the cytoplasmic mass within the original spore wall. Each androcyte becomes a motile antherozoid by dissolution of the androcyte membrane.
- The antherozoids are corkscrew-shaped, multiflagellate structure characterised by the presence of a large posterior cytoplasmic vesicle.

### **Female Gametophyte:**

- The megasporangium is an oval or elliptic structure, the wall of which imbibes water and expands to form a gelatinous mass around the megasporangium.
- The spore wall expands to form a small papilla (protuberance) at the apical end where the nucleus is located in a dense part of cytoplasm. The remaining portion of the spore is filled with a frothy cytoplasm full of starch grains.
- The first division in the apical nucleus of the large megasporangium is transverse, forming a small nipple-shaped apical cell and a very large basal nutritive or prothallial cell. The prothallial cell provides the nutrition to the growing female gametophyte.
- The apical cell further divides by three intersecting vertical walls to establish an axial cell surrounded by three lateral cells. Now the axial cell functions as archegonial initial which divides periclinally to form an outer primary cover cell and an inner central cell.
- A small neck (2 tiers of 4 cells each) is derived from the primary cover cell. The central cell divides transversely to form an upper primary canal cell (behaves as neck canal cell) and lower primary venter cell.
- The primary ventral cell again divides transversely to form a ventral canal cell and an egg. The growth of the archegonial complex ruptures the megasporangium wall at the apical end and forms a conspicuous gelatinous mass with funnel-shaped papilla or protuberance. Now, the megasporangium splits through the triradiate fissure and archegonium becomes exposed.
- Fertilisation disintegration of the neck canal cell and ventral canal cell create a passage for the antherozoids to fertilise the egg. The antherozoids, after liberation from ruptured

male gametophyte, enter through the gelatinous protuberances and moves downwards to the archegonium. One of the antherozoids eventually fuses with the egg to form a diploid zygote.



**Fig 1.36. *Marsilea* megagametophyte : A. Megasporangium, B-D. The stages in the development of megagametophyte, E, A mature archegonium**

#### New Sporophyte (The Embryo):

- The zygote is the mother cell of the next sporophytic generation.
- The first division of the zygote is vertical (in relation to the neck of the archegonium) followed by a transverse division resulting in the quadrant stage (four-celled stage) of the embryo.
- Subsequent development of the upper two cells forms the root and leaf, whereas the lower two cells give rise to the foot and shoot apex.
- With the development of the embryo the vegetative cells of the surrounding gametophyte divides periclinally and form a two- to three-layered sheath (calyptra) around the embryo.
- The primary root grows vertically and establishes the sporeling in the soil. The young sporophyte has a well-developed primary root and leaf.

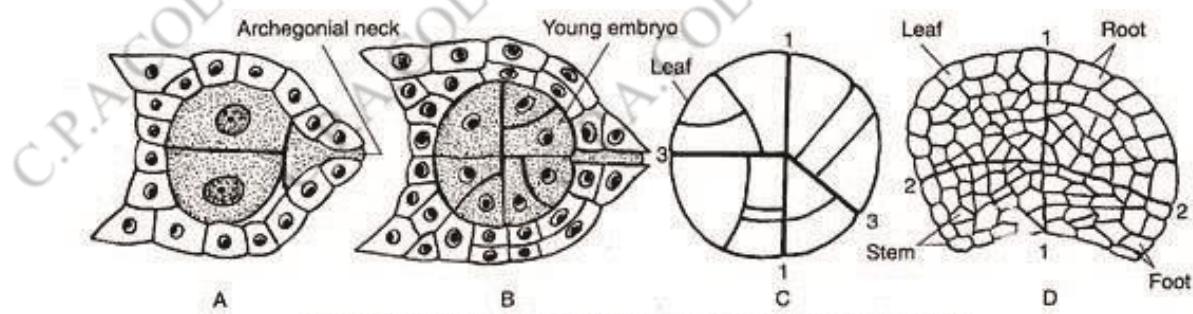


Fig 1.37. *Marsilea*: A-D the stages in the development of embryo 1.4.2.Life

cycle of *Marsilea*

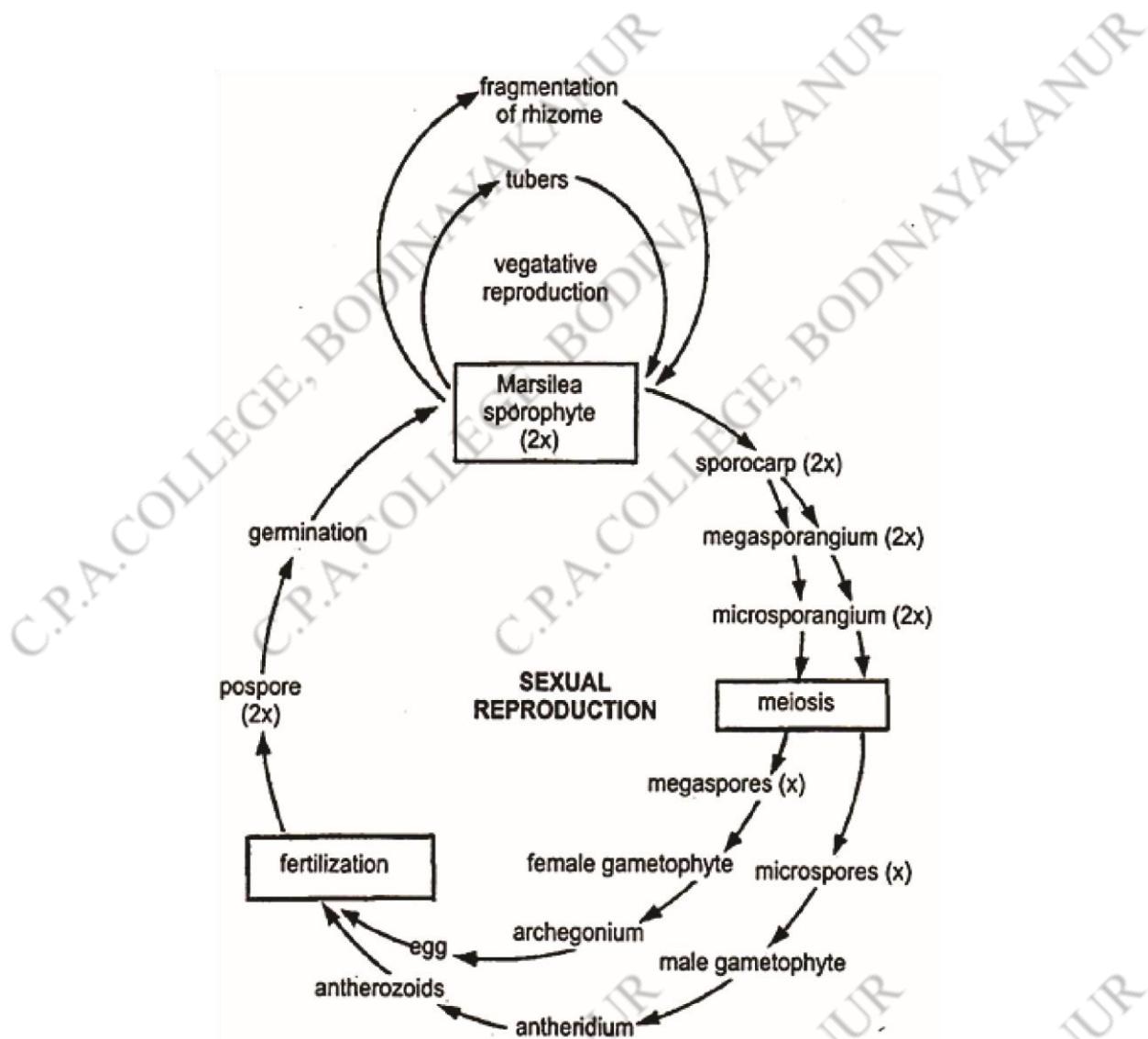


Fig 1.37. *Marsilea*: Schematic life cycle.