MARSILEACEAE

The family includes three genera, (Fig. 5.29 A-C) *Marsilea, Pilularia* and *Regnellidium*.

MARSILEA

The genus *Marsilea* with about 60-65 species is distributed in temperate as well as tropical regions. Most of the species occur in Australia and about ten species have been recorded from India.

Marsilea has a slender creeping, dichotomously branched rhizome of indefinite growth, which by forming a mat can extend up to 25 metres in diameter. At each node are borne one or two adventitious roots and an upright leaf (Fig. 5.29 A) of flexible petiole and lamina divided into four leaflets. The leaves give the appearance of four-leaf clover and the plant has been occasionally used as a true substitute for clover. The leaf is circinate when young, and the leaflets are folded together upwards until nearly mature. At night the leaflets are folded upwards assuming a "sleeping" position. Presence of latex in petiole and sleep movement of leaflets are features comparable with angiosperm leaf (Loyal, 1990).

The reproductive structures, bean-like sporocarps are borne on short or long stalks, inserted a short distance above the base of petiole.

The plants are adapted to grow in shallow water or wet places. A few species also grow on soil. *M. hirsuta* is an extreme xerophytic form from

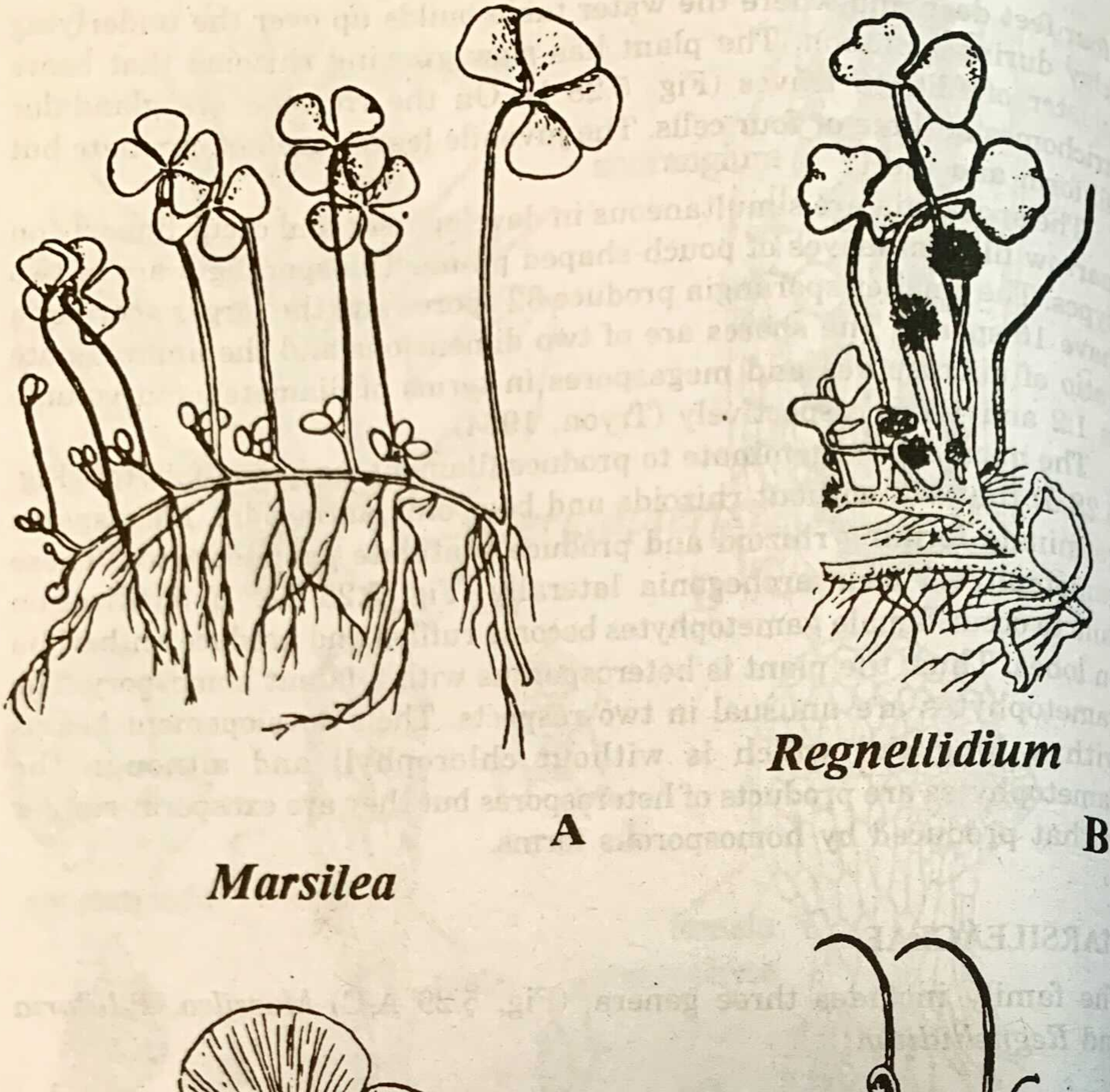

Fig. 5.29 Marsiliaceae, morphology.
A. *Marsilea quadrifolia* and *M. polycarpa.* B,C, *Regnellidium* and *Pilularia* respectively.
(Figures after: A, Eames, Bierhorst; B, Lindman; C, Meunier)

Australia; capable of surviving long periods of drought and is the only species which forms underground tubers on rhizomes. *M. minuta* and *M. aegyptiaca* are also xerophytic forms; the former can grow and form sporocarps in water but the latter grows vegetatively and fails to form sporocarps.

Occasionally, the same species may be found to grow in water as well as on land and consequently land form and water form are known to occur in the genus. Land and water forms can be easily distinguished on morphological grounds. Land forms have short internodes, branched roots, a few air spaces and more sclerenchyma in vegetative organs. The leaves have long petioles and stomata are distributed on both the surfaces of leaflets. Water forms, in contrast, have long internodes, unbranched roots and flexible petiole. There is practically no sclerenchyma in vegetative organs and there are extensive air spaces. Stomata are restricted largely to upper surface of leaflets.

Rhizome in *Marsilea* grows by means of an apical cell, with three cutting faces, that regularly forms segments (merophytes) which are the building units of stem. There is evidence for a slow rate of division in older rhizomes (Kuligowski-Andres, 1978). The vascular cylinder is an amphiphloic siphonostele (solenostele). Associated with the departure of a simple leaf trace there is a gap in the stele. The xylem cylinder is surrounded externally as well as internally by endodermis, pericycle, and phloem (Fig. 5.30 A). The maturation pattern of xylem is mesarch. Some species, however, have conspicuous protoxylem masses that are exarch (Fig. 5.30 B). In land forms the inner cortex and pith are sclerotic whereas in water forms these zones are uniformly parenchymatous. Middle cortex is aerenchymatous with a single ring of air chambers. The air chambers are separated from one another by one layered septum. Aerenchyma is more extensive in water forms and less extensive in land forms. The outer cortex is parenchymatous, one to several cells in thickness. Lying scattered in the outer and inner cortical zones are some tannin cells.

The stele of petiole (Fig. 5.30 C) is nearly triangular and is bound by single endodermis. There are two arms of xylem which are curved away, from each other, on adaxial side and are relatively close on the abaxial side. Each xylem arm comprises one or two metaxylem elements in centre and a few protoxylem elements at both ends. The cortex is similar to that of stem and is variable in land and water forms.

Root meristem consists of an apical cell with 3 cutting faces. These forms segments or merophytes which are the building units. About the regularity of merophyte formation there are contradictory evidences. The root apical meristem is reported to be active in early stages and growth ceases due to endopolyploidy (Avanzi & D'Amato, 1970). Contrary to it, root apical meristem is reported to remain active (Vallade & Bungon, 1979; Kurth,, 1981) for a very long period. The cortex of the root also has aerenchyma. It is extensive in water forms. The roots are diarch and

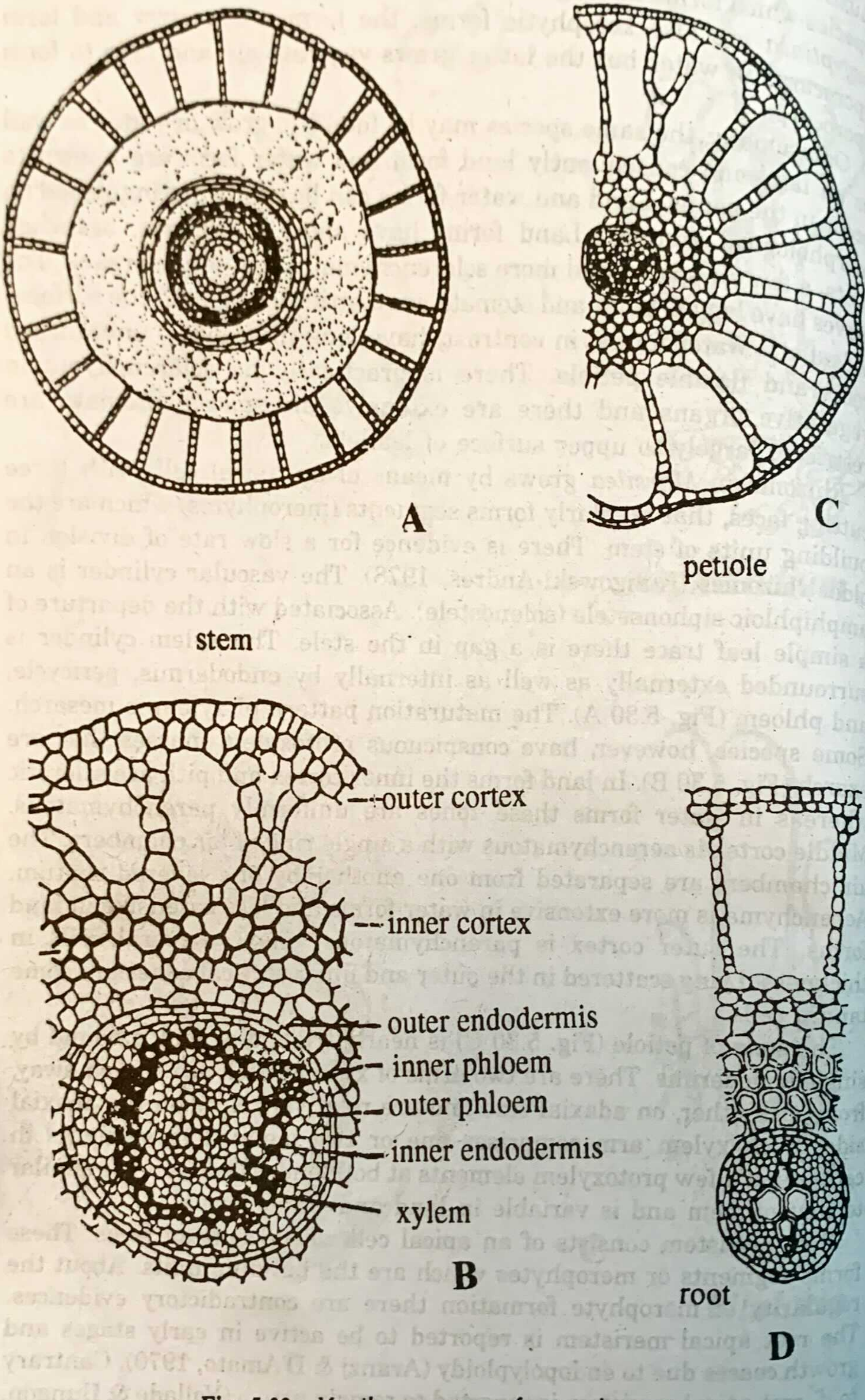

Fig. 5.30 *Marsilea*, structure of vegetative parts.
A, cross section of rhizome of *Marsilea* (diagrammatic). B, a portion enlarged to show cellular details. C, a portion of petiole (enlarged). D, a portion of root (enlarged).

exarch (Fig. 5.30 D). In the centre are two metaxylem elements and there are two groups of protoxylem elements on the periphery. There are reports of vessels in several species of *Marsilea*. In two species, *M. drummondii* and *M. alata* vessels occur in root, petiole as well as rhizome (White, 1961, Bharadwaja & Baijal, 1977).

Reproduction

The reproductive structures, the sporocarps, are flattened bean-shaped, spherical to ovoid, epipetiolar, stalked structures, inserted a short distance above the base of petiole. The length of stalk and its point of attachment is variable in different species. Normally, there is one sporocarp per petiole but in forms like *M. polycarpa* there are large number of sporocarps attached on one side of petiole (Fig. 5.29 D) and in *M. quadrifolia* the stalk is occasionally branched forming 2-3 sporocarps per petiole (Fig. 5.29 A). In some species the sporocarps have distinct external ridge, the raphe; and two bumps. The raphe represents the end of attachment of the stalk. One of the bumps is stouter and is present at the end of the raphe.

The sporocarps have a thick wall resistant to injury and dessication and are reported to remain viable up to 35-40 years. Inside the sporocarp, around the cavity, extends a ring of gelatinous tissue (Figs. 5.31 A). Also, there are rows of elongate sori, on either side, extending transversely to the long axis of the *sporocarp*. Each sorus is borne on an elongate receptacle, which is attached to the gelatinous ring, at its two ends as well as to the wall of sporocarp on one side, along its length; the other side faces the lumen of sporocarp. Number of sori in each sporocarp may vary from two in *M. aegyptiaca* to 20 in *M. quadrifolia*. Each sorus is enclosed by a delicate diaphragm, the indusium. On the crest of the receptacle are borne a number of larger sporangia, megasporangia (Fig. 5.32 B) each having a single large megaspoie (Fig. 5.32 C) and along the sides are borne smaller sporangia; microsporangia each with numerous microspores (Fig. 5.32 D).

The vascular supply to the sporocarp branches from the margin of petiolar trace. From the main trace arise pinnate series of traces, on each side (Fig. 5.31 D). These lateral bundles alternate with one another. Each of the lateral bundle bifurcates and the vascular supply to the individual sorus known as placental bundle arises at the point of forking. The placental bundle in turn bifurcates to form dorsal and ventral traces. Towards the lower margin of sporocarp, there is a network of veins due to anastomoses of the pinnate series of bundles.

The position and arrangement of sporangia in *Marsilea* sporocarp can be seen after sectioning it in following different planes (Fig. 5.31 A-C).

In a vertical longisection of sporocarp, can be seen either megasporangia or microsporangia. The megasporangia are seen if it is a median section and microsporangia in a tangential section. The two sections are shown (Fig. 5.31 A). The first half (on the left) is median

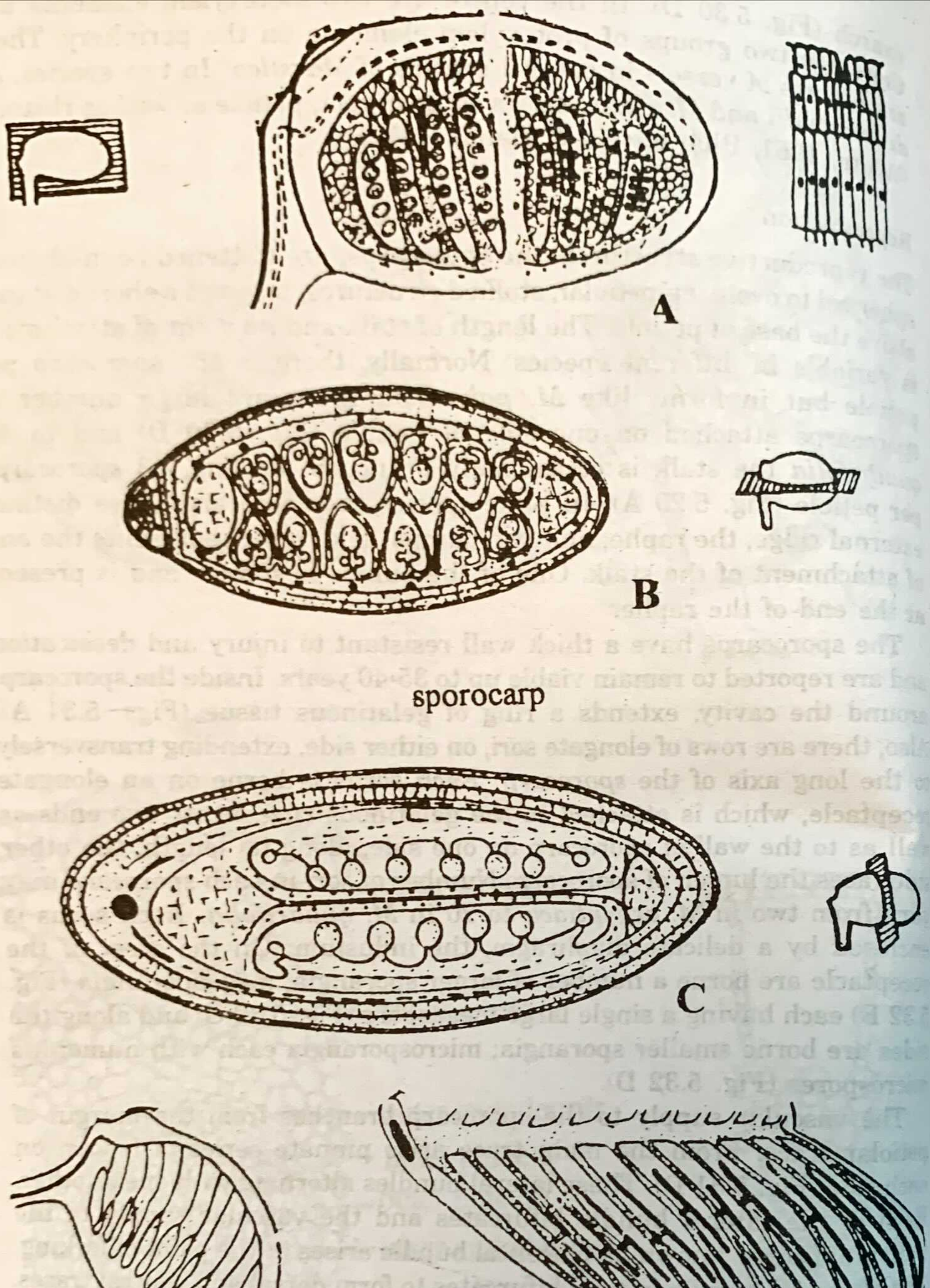

Fig. 5.31 *Marsilea*, schematic representation of sporocarp structure. A, vertical longisection. B, longisection. C, transection. D, a sporocarp cleared to show vasculature in surface view and E, veination pattern of sporocarp.

section showing megasporangia and the other half (on the right) is tangential section showing microsporangia. Around the sori is the gelatinous ring.

In a longisection of sporocarp (Fig. 5.31 B) are seen two alternating rows of sori. Each sorus has its vascular trace, cut transversely, and is enclosed by an indusium. On the crest of receptacle is borne a single megasporangium and flanking it on either side are microsporangia. The wall of the sporocarp consists of three layers (Fig. 5.31 A). The outermost layer is epidermis consisting of broad columnar cells and bears stomata. It is followed by first palisade layer of thick-walled elongate cells. The second palisade layer has more elongate cells which are thin-walled. In this section can also be seen cut portions of the gelatinous ring at the two ends. Also, at one end is peduncle with its vascular bundle cut in a transverse plane.

In a transection of sporocarp (Fig. 5.31 C) are two rows of sori; each one is attached to the receptacle, and is covered with indusium. The sorus shows only one kind of sporangia. In the section can be seen main bundle, lateral bundles and placental bundles. The cut portions of gelatinous ring are at the two ends. The gelatinous mass at the dorsal end is more prominent.

The sporocarps open only in water, splitting bivalve-like (Fig. 5.32 A). The process is initiated by the absorption of water by gelatinous ring which expands and frees the sori from the wall of sporocarp by an abscission along the receptacle. This is followed by the breaking of ring at one end. The broken gelatinous ring results into an elongate worm-like structure dragging the sori along with it. In laboratory, this natural course of germination can be simulated after making a slit at the apical end of sporocarp, by a sharp scalpel, and boiling the sporocarp in concentrated alkaline solution.

The dry sporocarps are like nuts, capable of surviving long period of drought and can remain viable up to 35-40 years. Steps in gametogenesis and embryogeny of *Marsilea* are, however, rapid. If mature sporocarps are split open and placed in water, within a week young sporophytes can be obtained. The plants are adapted to wet conditions but can survive long periods of drought due to thick-walled sporocarps and are capable of rejuvenating in a very short time. In nature, following 'germination' the spores are released by tissue decomposition.

The microspores are small globular cells with a dense wall and a centrally placed prominent nucleus. Lying close to the wall are some starch grains. The microspores germinate (Fig. 32 E) immediately after being shed. The nucleus migrates to the periphery and first division is unequal, forming smaller lens-shaped cell with scanty contents. The larger cell in turn divides equally and from these cells, through a series of divisions, differentiate two spermatogenous cells. Each cell by successive divisions produces 16 sperms which are characteristic of genus, multiflagellate cork-screw-shaped (Fig. 5.32 F) with a prominent vesicle.

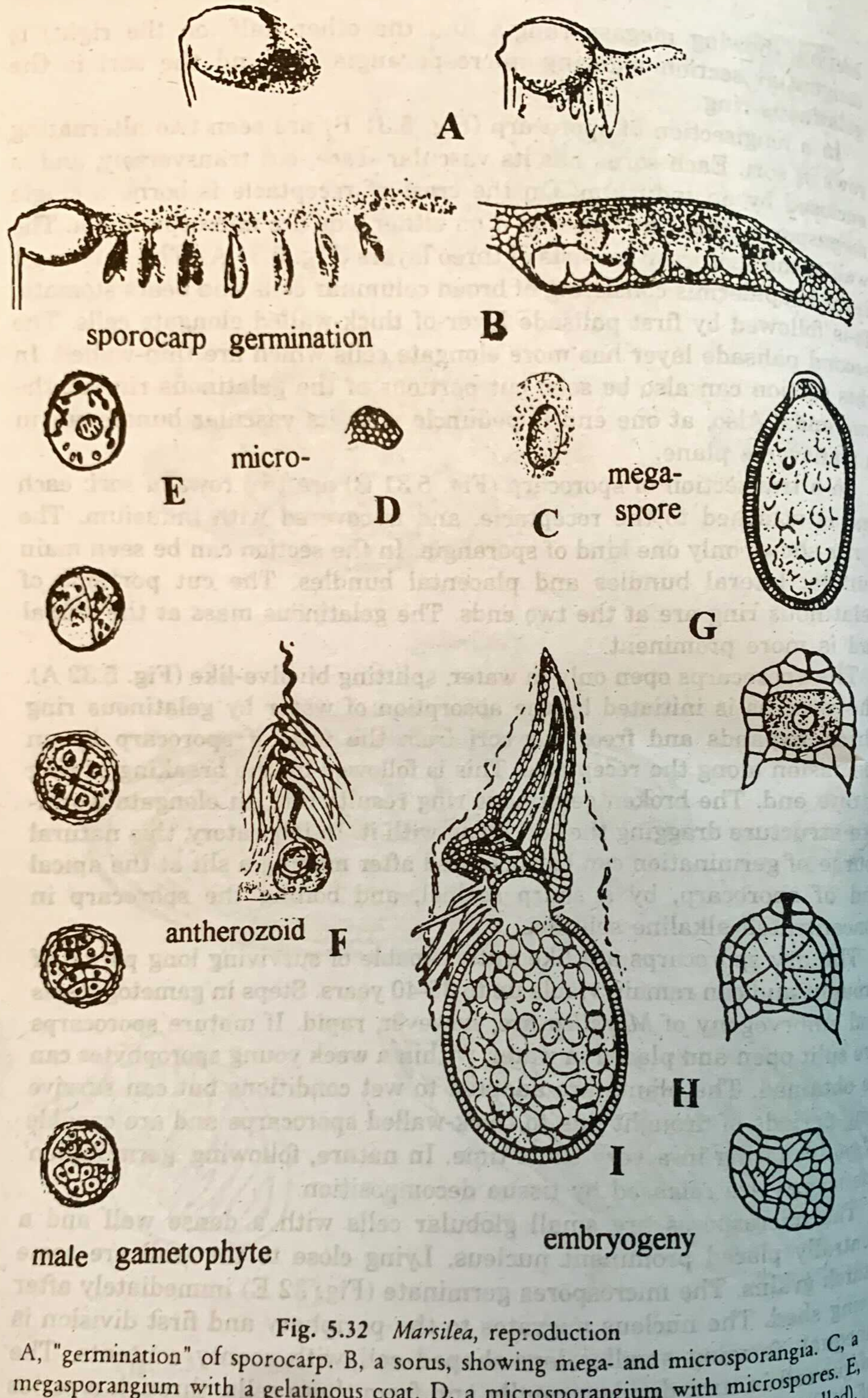

Fig. 5.32 *Marsilea*, reproduction

A, "germination" of sporocarp. B, a sorus, showing mega- and microsporangia. C, a megasporangium with a gelatinous coat. D, a microsporangium with microspores. E, development of male gametophyte. F, a sperm. G, female gametophyte (two-celled). H,I, stages of embryogeny.

(Figures after: A,E, Sharp; B,C,D, Eames; G,H, Campbell; I, Sachs)

The loss of cytoplasmic vesicle may make a sperm non-motile (Myles, 1975).

The megaspores are large ivory white papillate structures (Fig. 5.32 F) enclosed by a gelatinous layer, and can be seen with unaided eye. Towards the apical region the gelatinous mass is in the form of longitudinal folds -the papillar envelop- which is like a "bell" and inside there is liquid, the "sperm lake". At the base, sheath has variable folds. The megaspore nucleus is located near papilla. The first division produces a small cell, which lies in papilla (Fig. 5.32 F). The larger basal cell occupies the rest of spore, storing abundant food material. The gametophyte which characteristically produces only one archegonium is derived from apical cell. In a mature archegonium, the neck is only two cells long with a single neck canal cell. The gelatinous sheath around megaspore has an opening at the apex. When sperms are in the vicinity of megaspore they are trapped in the gelatinous sheath, pass downwards to the archegonium through the opening in the sheath, and can be seen swimming in "sperm lake".

The embryo (Fig. 5.32 G) grows entirely in the papillar region. First division is longitudinal and the organs formed; root, stem, leaf and foot are traceable to the quadrant stage. Two outer segments form leaf and root and two inner ones form stem and foot (Fig. 5.32 H). The cells of archegonial venter also divide and form a sheath around the developing embryo (Fig. 5.32 I).

Of the two other closely related genera, *Pilularia* (Fig. 5.29 C) with six species is equally widespread as *Marsilea*. Monotypic *Regnellidium diphyllum* occurs in Brazil. In habit and habitat, the two genera are similar to *Marsilea* but leaves of *Pilularia* are filiform (Fig. 5.29 C) without any lamina and those of *Regnellidium* bear two opposite pinnae (Fig. 5.29 B). *Regnellidium* is another exceptional non-flowering plant which produces latex. Unbranched and occasionally septate latex tubes occur throughout the plant body. Vessels have been recorded in roots of *Regnellidium* (Tiwari, 1975).

The sporocarps tend to be spherical in these two genera. The structure of *Regnellidium* sporocarp is similar to *Marsilea* but *Pilularia* sporocarp has four sori. In *Marsilea* and *Pilularia*, the sporangia are attached to the wall of sporocarp but in *Regnellidium* the sporangia are attached on the radial walls of soral chamber.