

Pharmacognosy of Indian Substitutes of Male Fern II

*Dryopteris barbiger*a and *Dryopteris cochleata*

By P. N. MEHRA and T. C. MITTAL

A macroscopic and microscopic description of the rhizome and leaf bases of *Dryopteris barbiger*a (Moore) O. Ktze. and *D. cochleata* (Don) C. Chr. is presented. Both the species compare very favorably in oleoresinous content and 'crude filicin' with the official male fern. The two species, described in this paper are differentiated from each other on the basis of the shape and size of the rhizome, structure of the ramenta present on the rhizomes, the presence and absence of dark sclerotic nests in the ground tissue, the number of cauline vascular bundles, and the shape and size of the stalk of internal secretory unicellular glandular hairs.

EXTRACTUM FILICIS has been extensively used as a teniacide for the last two centuries or so (1). Since the drug has been included in various pharmacopoeias and also the International Pharmacopoeia, its importance cannot be over-emphasized. Considerable quantities of the expensive male fern extract are imported into India for medicinal purposes, since both the official species, *Dryopteris filix-mas* (L.) Schott and *D. marginalis* (L.) Asa Gray are not met with in India. Search for an Indian substitute for this important drug has shown that there are some excellent substitutes that grow wild in the Himalayas, some abundantly enough to be commercially exploited (2, 3). The detailed pharmacognosy of two of them has already been published (4), while that of *D. barbiger*a (Moore) O. Ktze. and *D. cochleata* (Don) C. Chr. is described in this communication. The oleoresin content of both the species varies from 7 to 9% and the "crude filicin" in *D. barbiger*a is approximately 2.2% and in *D. cochleata* up to 1.8% (2).

The purpose of the study is to report the macroscopic and microscopic characteristics of these two species in order to establish satisfactorily their identity in commerce. The description of the plant has been provided to facilitate its collection, as no such appropriate description is available in previous literature on Indian ferns (5-11).

MATERIAL AND METHODS

The material for *Dryopteris barbiger*a was gathered from alpine meadows above Khillanmarg (Kashmir) and en route to Rohtang pass (Kulu Hills), between altitudes of 10,000-13,000 feet, where it grows in fair abundance. The sample of *D. cochleata* was collected from Raiwala forests, Dehradun (Western Himalayas), and Majitar bridge (Eastern Hima-

layas), between altitudes of 1,500-3,000 feet where it occurs gregariously. The technique employed was the same as described in an earlier communication (4).

INVESTIGATIONS

*Dryopteris barbiger*a (Moore) O. Ktze.

Plant.—The rhizome is thick, ascending, sometimes branched, copiously covered with chaffy, chestnut brown, large ramenta. The stipes are tufted, 15-30 cm. long, rachis as well as the stipes are densely covered with large, bright, rufo-brown, imbricate ramenta, and soft silky hairs; lamina 60-90 cm. long, ovate-lanceolate (Fig. 1, part 1). Pinnae are close, up to 20 pairs or more, 8-15 cm. long, 2.5-4 cm. broad, gradually reduced below, linear-lanceolate. Pinnules are usually distinct, ovate-oblong, double serrate, or deeply lobatoincised and each lobe again provided with copious aristate teeth, the edge often incurved; texture herbaceous, both the surfaces copiously fibrillose-scaly, more so the lower surface; midrib fibrillose-scaly; sori in two rows close to the midrib of the pinnules; indusium small, entire, pale colored, of thick texture with small and shallow sinus.

Macroscopy of the Rhizome.—Rhizome may be branched or unbranched, of variable shapes, ovoid, oblong, or elongated (Fig. 1, part 2), measuring up to 20 cm. long and up to 6 cm. or more thick at the crown. Externally reddish-brown, entirely covered by thick, stout, persistent, curved, or flattened, or angular frond bases which are up to 15 mm. thick and somewhat wrinkled. Long shreds of hypodermal sclerenchyma are observed attached to the frond base. The entire rhizome and stipe bases are densely clothed in rufus-castaneous, broadly lanceolate, hair-pointed scales which are 2-4 cm. in length. The rhizome breaks with hard and uneven fracture. Transversely cut surface, which in fresh drug is markedly pale green and in dried drug is brown, reveals 6-9 whitish patches of cauline meristeles arranged in a diffused circle and 4-5 leaf-traces, each composed of 10-13 smaller vascular strands, arranged in the form of closed 'C' at angular regions. Odor slight, taste at first sweetish, later becoming acrid and nauseous.

The ramenta are ovate to lanceolate with acuminate apex, each measuring 2-4 cm. or more in length,

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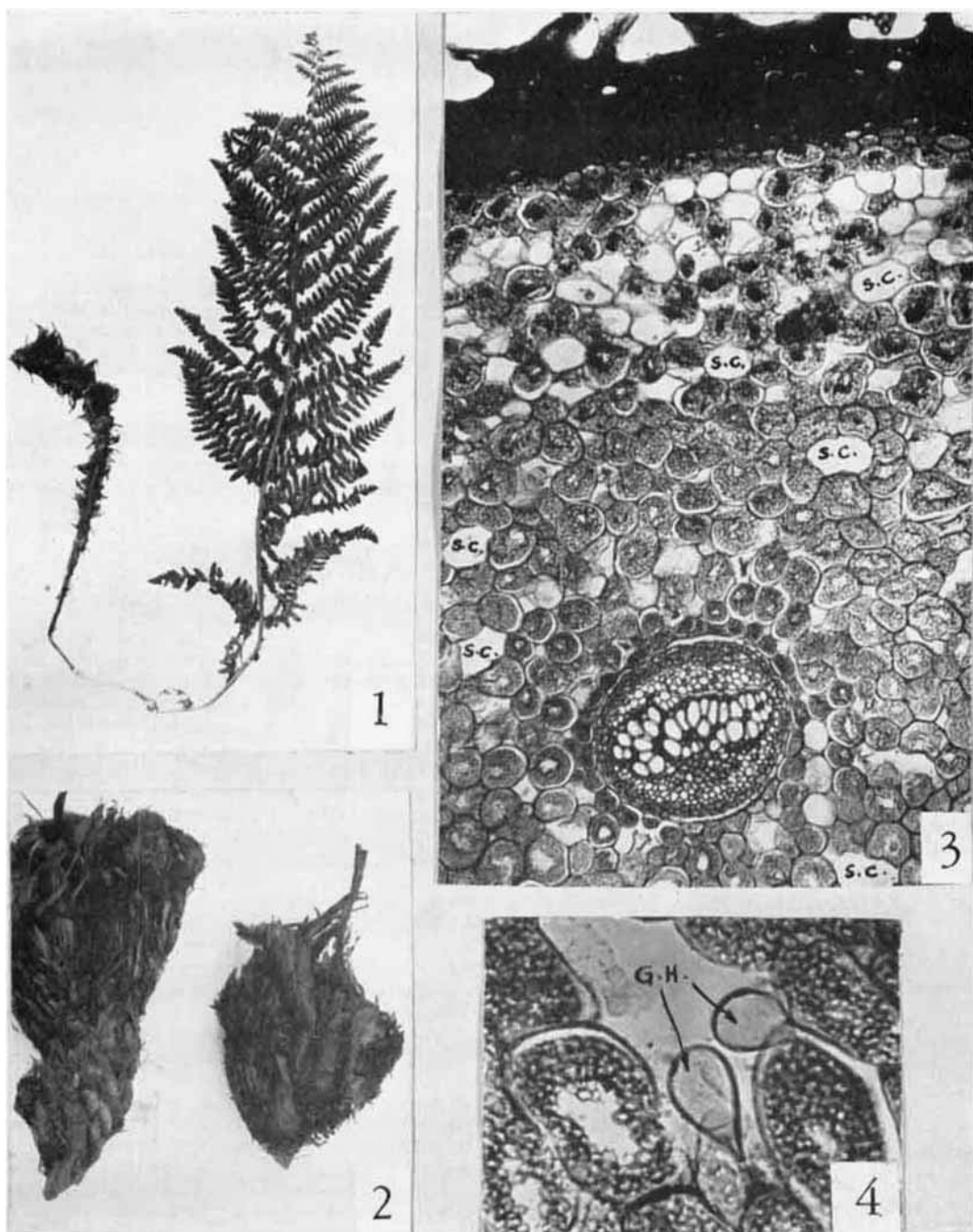


Fig. 1.—*Dryopteris barbiger* frond, $\times \frac{1}{5}$. 2, Rhizome, $\times \frac{1}{3}$. 3, Sector of T.S. of the rhizome magnified $\times 200$; S.C., schizogenous cavity. 4, L.S. of the rhizome showing schizogenous cavity containing unicellular secretory glandular hairs, $\times 250$; G.H., glandular hairs.

and 3–8 mm. in breadth. Each ramentum is composed of thin-walled, elongated cells, each measuring $500\text{--}1200 \times 20\text{--}30 \mu$. At places the marginal cells form peg-like projections, each consisting of two parallel and contiguous cells. No glandular hairs are observed on the margin or the surface of the ramenta.

Histology of the Rhizome.—A transverse section of the rhizome from the periphery to the center is

shown in Fig. 1, part 3. The epidermis is composed of a single layer of cells which vary in shape from oblong to rectangular. These cells are brownish in color and contain occasional tannin-resin masses. Some of the cells bear unicellular pyriform glandular hairs, and a few others the ramenta cut variously. Since the rhizome is profusely scaly a large number of cut ramenta are observed in a cross section.

The hypodermis is differentiated into two zones,

an outer one of 2-3 layers of cellulosic fibers, and an inner band of 5-8 layers of thick-walled lignified fibers. The cells of the latter appear yellowish to orange in unstained preparations due to the deposition of tannin-like material on the walls. In T.S. the cells of the inner zone appear polygonal and are more or less isodiametric, but in maceration they are 10-20 times as long as broad and possess flat or somewhat tapering ends (Fig. 2a). This region is compact and lacks intercellular spaces.

The cortex is composed of several layers of rather thick-walled parenchymatous cells, often filled with simple starch grains. Besides starch, the cells, particularly next to the hypodermis and around the meristemes, contain spherical masses or granule aggregates of tannins. The walls of such cells are cellulosic in nature while the remaining cells possess suberized walls. Some of the cells possess simple pits on their walls and such cells are devoid of inclusions. The cortical cells in maceration appear either isodiametric or slightly elongated (Fig. 2c), each measuring $70-90 \times 40-90 \mu$. In T.S. they number about $400/\text{mm}^2$.

The cortex shows numerous small intercellular air spaces and large schizogenous cavities. Within the latter project characteristic unicellular glandular hairs (Fig. 1, part 4), each surrounded by greenish-yellow oily secretion. The glands are $60-100 \mu$ long of which $20-25 \mu$ is the stalk. The head is oval or pyriform in shape and measures $35-56 \mu$ in diameter. The schizogenous cavities in the region of their maximum distribution number about $45/\text{mm}^2$, as observed in T.S. In longitudinal section, the cavities measure $160-350 \times 60-160 \mu$ and each contains 2-3 glands.

Sclerotic nests are absent both in the cortex and the pith. Pith cells are identical with the cortical cells.

Within the ground tissue is present a diffused ring of 6-9 large cauline meristemes. Numerous smaller strands arranged in the form of closed 'C' constituting leaf traces are present at angular regions of the rhizome. Occasionally root traces are also observed, passing out of the cortex.

Meristeme.—The cauline meristemes and the leaf trace bundles are oblong to elliptical in outline and amphicribal in nature. The leaf trace bundles are relatively much smaller and in longitudinal sections are observed to arise from the margin of the leaf gaps.

Each meristeme shows a single-layered endodermis of rectangular cells, the walls of which are suberized and the radial ones possess feebly developed Casparian thickenings. This is followed by 1-3 layers of thin-walled, nonlignified parenchymatous pericycle cells, filled with starch grains and amorphous tannin deposits. In cross section the cells appear polygonal to oblong in shape. Phloem is many layered and encircles the central xylem on all sides except at two lateral ends where the pericycle directly abuts on the xylem. It consists of sieve tubes and phloem parenchyma. The cells of the phloem parenchyma are filled with tannin deposits. Xylem is central in position and consists of tracheids and xylem parenchyma. The tracheids in cross section have typical polygonal outline. In maceration they appear elongated and the metaxylem elements show scalariform thickenings. The bundle, according to its size, may have 2-3 small protoxylem

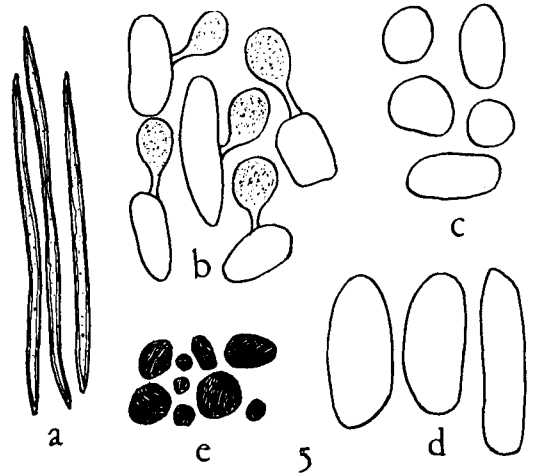


Fig. 2.—*Dryopteris barbigera*, tissue macerations. a, Hypodermal fibers, $\times 160$; b, gland bearing cortical cells, $\times 160$; c, ordinary cortical cells of the rhizome, $\times 160$; d, ordinary cortical cells of the stipe base, $\times 160$; e, starch grains, $\times 700$.

groups consisting of small annular or spiral tracheids; frequently one is found at each end of the xylem mass.

Histology of the Stipe.—In T.S. it is somewhat oval in outline. The hypodermis is well developed and formed of lignified fibers. Unlike the rhizome it is not differentiated into two zones. The cells of the cortex are 2-3 times larger than those of the rhizome (Fig. 2d), and are predominantly oblong or rectangular. The few layers that lie next to the hypodermis contain tannoid aggregates, but such contents are missing from other cells. The cortical cells number about $250/\text{mm}^2$ in contrast to nearly 400 in the rhizome. The number of schizogenous cavities per unit area is almost the same as in the rhizome. The number of vascular strands varies from 10-14. They are arranged in the form of closed 'C', and the two adaxial ones are larger with the xylem notched internally.

Dryopteris cochleata (Don) C. Chr.

Plant.—Rhizome is ascending, and clothed in golden yellow, ovate-lanceolate, or linear-subulate ramamenta. Fronds are strongly dimorphic (Fig. 3, part 6); the stipe is up to 40 cm. long, comparatively much longer and thicker in fertile fronds, tufted; main rachis glabrous; lamina up to 40 cm. long, deltoid or ovate-oblong with slightly cordate or rounded base. Pinnæ are up to 10 pairs, basal pinnæ not any longer than the ones next above, not much basiscopically produced, ovate-lanceolate, acuminate, the sterile basal ones are subsessile or sessile, up to 13 cm. long and 4 cm. broad, fertile ones distinctly though shortly petiolate, up to 8 cm. long and 2 cm. broad, the lower ones ascending, others patent; the rachis deeply canaliculated above, the underside of the rachis and rachises of fertile fronds fibrillose but of sterile fronds is setose hairy; sterile pinnules more or less adnate to a winged costule, oblong-obtuse, base truncate, serrate; fertile pinnules subsessile with serrate margin; texture subcoriaceous, green or pale green, veins inconspicuous; sori large, well below the middle of upper veinlet

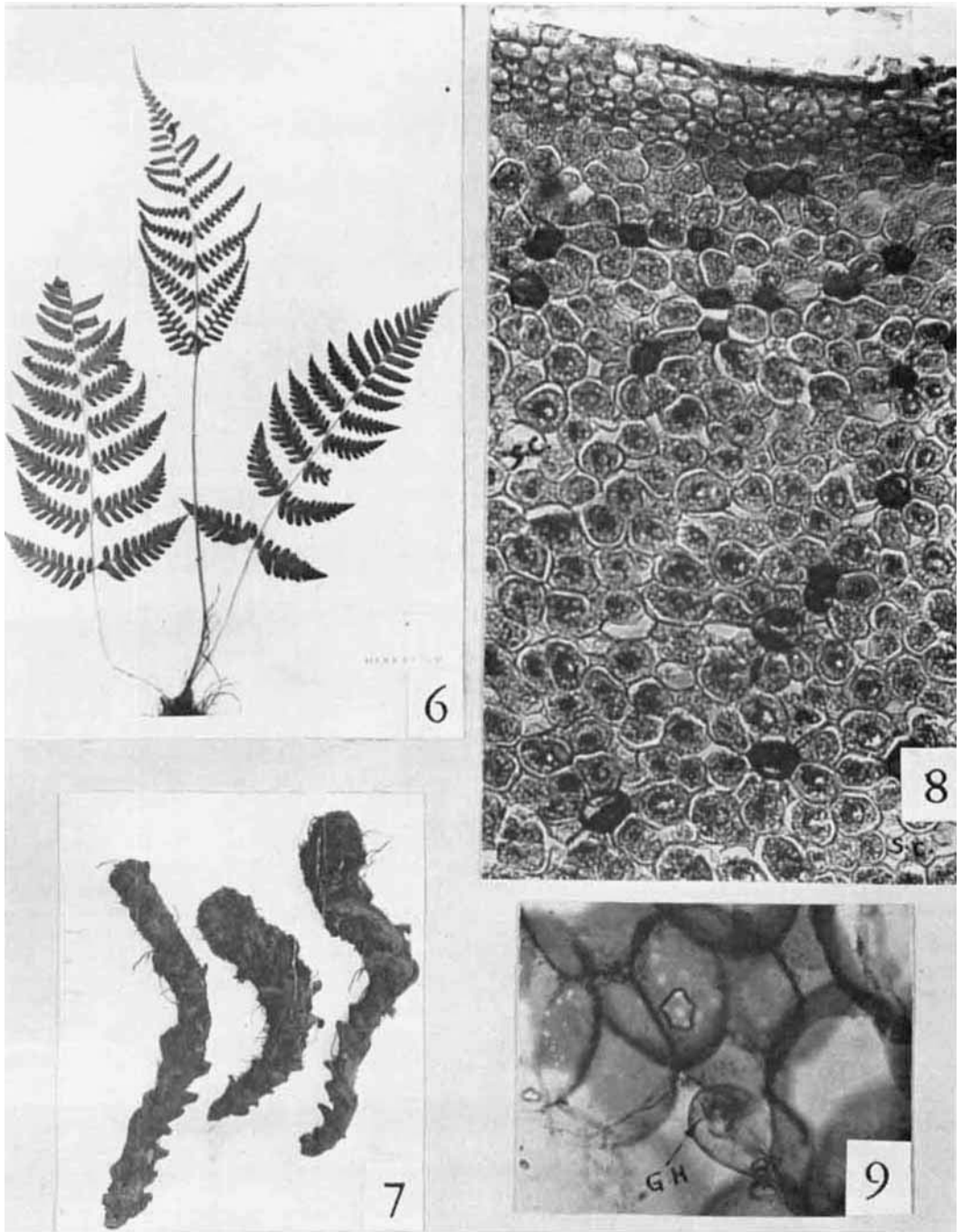


Fig. 3.—*D. cochleata*, plant with two sterile fronds and central fertile one, $\times \frac{1}{3}$. 7, Rhizome, $\times \frac{1}{2}$. 8, Sector of T.S. of the rhizome magnified, $\times 100$; S.C., schizogenous cavity. 9, L.S. of rhizome showing a gland in a schizogenous cavity, $\times 250$; G.H., glandular hair.

forming a row on each side of the pinnule near the midvein; indusium is reniform, and persistent.

Macroscopy of the Rhizome.—Rhizomes are flattened-cylindrical, tapering at the base (Fig. 3, part 7), 8–20 cm. long, 1.5–2.5 cm. thick, actual thickness without leaf bases varies from 1–1.5 cm. Externally rufo-castaneous or dull brown, covered by distantly situated frond bases which are 6–11 mm.

thick and wrinkled. The entire rhizome and the frond bases are profusely scaly. Fracture is short. Freshly cut surface is pale green, marked by 3–5 cauline meristemes and numerous smaller vascular strands constituting leaf traces. Dark spots of sclerotic nests are abundant in the rhizome and leaf bases. Odor is slight. Taste at first sweetish, later becoming acrid and nauseous.

The rammenta vary in shape from linear-lanceolate to ovate-lanceolate with acuminate apex, each measuring 1-2 cm. \times 1-6 mm. Unicellular glands are found all over the surface and the margin. In older scales the marginal cells are somewhat drawn out in groups to form multicellular, long fibrils. In addition, two-celled projections, characteristic of the genus *Dryopteris*, are also present on the margin. The unicellular glands measure 40-50 μ in length, and about 20 μ in diameter at the globular head.

Histology of the Rhizome.—A sector of the transverse section of the rhizome is represented in Fig. 3, part 8. The general histological pattern is the same as described in the earlier species except that numerous dark scattered sclerotic cells or groups of such cells forming sclerotic nests are observed in the ground tissue. In T.S. the number of cortical cells is about 275/mm.². These cells in maceration measure 56-210 \times 42-100 μ . Schizogenous cavities are fewer than in the previous species, being only about 25/mm.² as seen in T.S. In longitudinal section, the cavities measure 64-180 \times 35-60 μ . Into each cavity project one or two characteristic unicellular glandular hairs (Fig. 3, part 9). The latter are 38-80 μ long including the stalk which is 6-14 μ long with a somewhat swollen base. The diameter of the head measures 24-42 μ . Embedded in the ground tissue are numerous sclerotic cells or sclerotic nests, scattered all over but more frequent in the outer cortical region (Fig. 3, part 8), particularly where the frond bases are attached. The sclerotic nests in the outer zone consist of two macrosclereids but those in the pith region are broader and formed of a number of sclereids. The sclereids, which in T.S. appear isodiametric to polygonal in outline with narrow lumen, are elongated cells with pointed or blunt ends and highly pitted walls (Fig. 4d). The walls are impregnated with a coloring matter which is responsible for the dark appearance of the nests. Each sclereid measures 250-1275 \times 45-120 μ . The number of cauline meristemes varies from 3-5 and each leaf trace is comprised of 10-14 smaller vascular strands.

Histology of the Stipe.—The cortical cells are only slightly broader than those of the rhizome and they number about 200/mm.².

The sclerotic cell/nests are numerous and scattered all over the ground tissue. Each sclerotic nest consists of up to four sclereids. The vascular strands are 10-14, arranged in the form of a closed 'C'. As in the previous species, the two inner bundles are larger than the rest and their xylem possesses anterior hooks.

SUMMARY

The present investigation provides macroscopic and microscopic descriptions of the rhizome and stipe bases of *Dryopteris barbigera* (Moore) O. Ktze. and *D. cochleata* (Don) C. Chr.

D. cochleata is strongly dimorphic, but *D. barbigera* is monomorphic. The rhizome of the former is flattened-cylindrical with the frond bases distantly situated. That of *D. barbigera* is ovoid, oblong, or elongated in shape with numerous closely arranged frond bases.

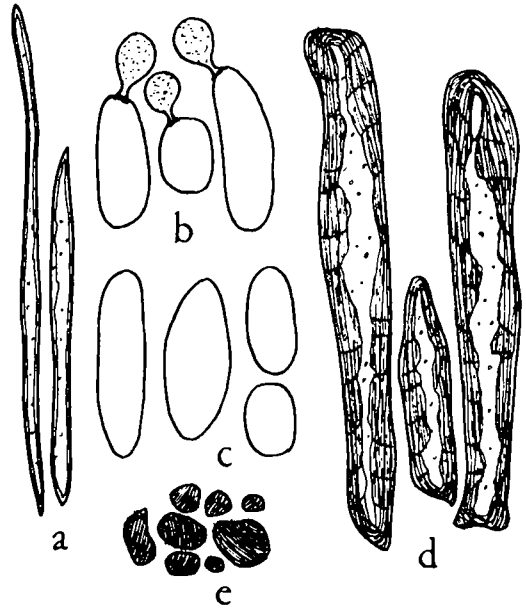


Fig. 4.—*D. cochleata*, tissue macerations. a, Hypodermal fibers, \times 160; b, gland bearing cortical cells, \times 160; c, cortical cells, \times 160; d, sclereids \times 160; e, starch grains, \times 700.

Rhizomes of both the species are covered all over with rammenta. Size and structure of the rammenta is characteristic in each species.

The histological pattern in both the species is similar except for the presence of scattered dark spots of sclerotic cells and nests in the ground tissue of the rhizome and stipe bases of *D. cochleata* which are absent in *D. barbigera*. Both the species possess schizogenous cavities distributed in the ground tissue, into which project characteristic unicellular secretory glandular hairs. The secretory glands possess longer stalks are *D. barbigera* than in *D. cochleata*. The base of the stalk is slightly swollen only in *D. cochleata*. The number of cauline meristemes and the leaf traces are less in *D. cochleata* than in *D. barbigera*.

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