A MORPHOLOGICAL STUDY OF THE HAIRS OF *MARSILEA* L.

by

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In memory of my father Malama Samu Mkozokele, mother Tiza Mwanida Chinunda and son Chuba Mkozokele.

ABSTRACT

A morphological study of hairs of *Marsilea* L. was undertaken. Methods involving scanning electron microscopy, histology and light microscopy were employed. Examined material based on 24 African species, 3 New World and 3 Australian species. The hairs are dorsiventrally flattened; sculptured with numerous papillae on dorsal surface, but very few or lacking on ventral surface. Hairs in all species are all anvil-shaped, elaborated into a two-armed, asymmetrical, uniseriate body. *Marsilea farinosa* is an exception for possessing two hair kinds, the anvil-shaped and the simple type. Traditional classification of *Marsilea* species is based on sporocarp characters; but existence of interspecific variation in hairs can lend support to a stable classification of the genus.

CONTENTS

1. INTRODUCTION

1.1 The Rationale

The development of the scanning electron microscope and its introduction on the market since 1965 (Grimstone, 1979) has aroused great interest in the study of plant epidermal structures. Notable structural features of the plant surfaces are the hairs and papillae which are, for the sake of convenience, collectively termed the trichomes. However, the papillae as small epidermal outgrowths are considered to be morphologically different from the trichomes (Cutler, 1978; Theobald *et al.*, 1979) whereas the hairs are simply described as epidermal appendages. The significance of 'trichomes' (this term will be used interchangeably for hairs) in plant systematics has long been recognised in the classification of angiosperms (Davies and Heywood, 1973; Cutler, 1978; Payne, 1978; Seithe, 1979; Theobald *et al.*, 1979; Edmonds, 1982; Fahn, 1982). In Pteridophytes, studies of trichomes have been made in some leptosporangiate ferns (Inamdar *et al.*, 1971) and in the family Polypodiaceae (Mitsuta, 1982).

In the genus *Marsilea* L. early anatomical studies have been concentrated on the nature of the sporocarp structures of some species. Important contributions have been reported on the vascular anatomy of the sporocarp of *Marsilea minuta* L. (Puri and Garg, 1953) with a brief mention of the sporocarp hairs. In recent years Mickel and Votava (1971) and Bhardwaja *et al.* (1977)

have reported accounts of their work on stomatal structure and their variation in *Marsilea* spp. and *M. coromandelina* Willd. respectively. Bilderback (1978) in his studies on the development of the sporocarp in *M. vestita* Hook & Grev. touched on the sporocarp hairs in relation to the overall anatomy of the reproductive organ, and his work was aided by illustrations from light microscope micrographs. In *Marsilea* the reigning tenet has been that both the leaf and sporocarp hairs are uniform throughout the whole range of species within this taxon.

1.2 Systematic background and objectives

a) Systematic background

Marsilea L. is the type genus of one of the heterosporous fern families, Marsileaceae. Its taxonomic status as provided by Tryon and Tryon (1982) is as follows:

Marsileaceae Mirbel. Hist. Nat. Veg. (Lam. & Mirb.) 5: 126, 1802. Type: *Marsilea* L.

Marsilea L. Sp. Pl. 1099. 1753: Gen. Pl. ed. 5 485. 1754. Type: *Marsilea quadrifolia*.

Syn. *Spheroidea* Dulac. Pl. Dept. Haut-Pyrenees 39. 1867.

Zalusianskya Necker (Acta Theod. Palat. Phys. 3: 303, 1775.

It should be noted that towards the end of the 19th century O. Kuntze (1891) had persistently adopted the name *Zalusianskya*; but in accordance with the rules governing the International Code of Botanical Nomenclature (ICBN) the latter two names have been rendered non-superfluous. Braun (1839) and Bower (1926) have provided

excellent early systematic accounts on the Marsileaceae and both considered this taxon to be related to Schizaeaceae purely on the basis of gross anatomical and morphological evidence; both families possess similar features such as the solenostelic rhizome, dichotomous venation in leaflets and - an important feature for the purposes of this account - the stem trichomes described merely as simple hairs "as is commonly the case in primitive ferns" (Bower, 1926; Holtum, 1968; Tryon and Tryon, 1982). However, in Marsileaceae, *Marsilea* is distinct from other members of the family by the possession of four leaflets borne terminally on the stipe; *Regnellidium* Lindm. bears two leaflets whereas *Pilularia* L. has one filiform leaf arising from the rhizome.

Marsilea comprises 65 species some of which are distributed throughout the tropical regions; most of the species, however, are restricted to Africa and Australia; while three occur in southern Europe. A very detailed account on the taxonomy and geographical distribution of the African species has been given by Launert (1968) in his monographic survey of the genus. The African continent is inhabited by about 24 species - representing approximately 37% of all the species. The taxonomic treatment of this intriguing amphiphytic genus has been rendered very difficult due to the great range of variation in vegetative parts, and the nature of such phenotypic plasticity is clearly expressed in diverse shapes and sizes of the leaflets (Launert, 1968). This high degree of variation in leaves is often encountered within any one population as well as within a single plant. The polymorphic nature of the vegetative parts is

essentially correlated to seasonal variation in the water levels found at the fringe of shallow open pools often subject to periodic inundation (Page, 1979). When the plants are partly submerged, they bear large glabrous leaflets and are strictly vegetative, but when plants attain a terrestrial habit the reproductive phase sets in with the simultaneous production of relatively smaller, often crenate to deeply lobed leaflets. Because of this greater diversity in vegetative form within any one taxon, very few taxonomists have attempted a systematic classification of the species of this genus.

The taxonomic treatment of the African species, as elucidated by Launert (1968) shows a number of synonyms appended to several validly described taxa. This in itself suggests (or confirms) the difficulties encountered in the classification of *Marsilea* and therefore giving rise to dynamic changes in the systematic position of the taxa at species level. Nonetheless, the delimitation of *Marsilea* species is strictly based on the discontinuous variation of the most conservative reproductive organ, the sporocarp (Stace, 1980).

b) Objectives

As indicated above, the structure of trichomes can be of great value in the classification of plants, or at least may support other characters which have been used. The aim of this project is to give an account of the morphology of the *Marsilea* hairs on the thesis that any evidence of interspecific variation in trichomes could

lend support in delimitating closely related species. This report - judging from current available information - is the first treatment on the micromorphology of *Marsilea* hairs with special reference to African species.

2. MATERIALS AND METHODS

2.1 Materials

Studies for this project were based on herbarium materials made available by the Botany Department at the British Museum (Natural History). A total of 24 African species were examined. The non-African species included for comparative purposes comprised 3 of Australian origin, 2 South American and one Caribbean species. A list of specimens examined is given in Appendix 1. Fresh living materials of *Marsilea drummondii* were supplied for histological preparation by Chelsea Physic Garden.

2.2 Methods

a) Scanning Electron Microscopy

Using a fine pair of forceps, dissecting blades and needles, the dry hairs were mounted on stubs. Sporocarp and leaf hairs were mounted separately, but some sporocarp hairs were mounted with their ventral surface facing upwards. Both sides of the dorsiventrally flattened hairs were so arranged in order to detect any variation in hair sculpturing on ventral and dorsal surfaces. Scanning was performed using the JSM T2 Scanning Electron Microscope at an accelerating voltage of 20 kV and scanning angle of 45^{o}.

b) Light Microscopy

For the purpose of measurements, several sporocarp and leaf hairs were mounted on separate microscope slides in a drop of lactophenol. About 1-2 minutes was allowed

for hairs to thoroughly soak before placing a cover glass. Microscopic examinations and measurements were performed with the use of a Reichert Student microscope fitted with an eyepiece graticule. From each sporocarp sample and each leaf sample, 10 hairs from each organ were picked at random across the microscope field and measured for hair length, hair width at the widest point, the length of the proximal arm (proximal cell); the number of cells per hair and the diameter of the hair's contact face with the foot cell. All measurements are given in μm.

c) Histology

In order to determine the mode of hair development and attachment relative to the epidermis, histological preparations were carried out. The fresh material of young leaves and sporocarps were fixed in F.A.A. (Formalin: Acetic acid:70% EtOH; 5:5:90), cleared in chloroform and embedded in paraffin wax. Thin sections of 12-15 μm were cut using a Reichert Rotary microtome. Differential staining involved the use of the Safranin and Fast Green staining schedule (Purvis *et al.*, 1966). Photomicrographs were taken using the Wild Photomicroscope. Some microscopic drawings were carried out with an aid of a camera lucid

3. RESULTS

3.1 Scanning Electron Microscopy

A representative number of the SEM micrographs is appended in Plates 1-12. A survey made on the species reveals the trichome morphology that is unique to the genus, *Marsilea* L. Since the material was obtained from dried herbarium specimens the characteristics so revealed on the SEM micrographs represent the dry state of the hairs. The hairs of all the species examined are dorsi-ventrally flattened; the dorsal surface is sculptured with numerous papillae. The anterior part (i.e. the proximal arm) of the hairs are often appressed to the sporocarp or leaf surface, while the greater part (or the distal arm) lie raised from the surface, with the terminal end sticking out at acute angles. In some species the entire length of the hair is adpressed to the organ surface (see Plate 5a). In leaves of some species, such as *Marsilea capensis*, *M. botryocarpa* and *M. macrocarpa*, the hairs also have their whole length adpressed to the epidermal surface (see Plates 1a, 6b and 8c).

The widest part of the trichomes is on the proximal arm, usually at the point immediately above the hair contact face. Septation of the hairs is externally marked by transverse lines of abjunctions which are posteriorly followed by the progressive reduction of the next segment giving the shape of tapered feature of a fine narrow wedge. In some species the hairs are nearly moniliform (see Plates 2b, 3c, 12a); some have lingulate proximal

arms (see Plate 2b). The anvil-shape of the trichomes is a common feature in all species. *Marsilea farinosa* is an exception in that its indumentum consists of two types of hairs, the anvil-shaped and the simple form and both are uniseriate (see Plates 9a, b, c). The simple hairs are more concentrated on edges of the sporocarps and leaves; they appear collapsed in dry state and are generally anfractuose (or curly), with a muticous (blunt) apex (see Fig. 1).

3.2 Light Microscopy

The hairs examined under the light microscope are illustrated in Plates 13-20, and measurements made are appended in Appendix II and summarised in Table 1. Notes on the range of some parameters will be given species for species below.

Hair Description

The description is based on the sporocarp hairs only and includes the range of measurements taken for each species, but not included in the tables.

1. *Marsilea aegytiaca.* Hair length 412.5-850 μ, 48-63 μ wide (note the widest point being at the proximal cell); contact fact diameter 16-19.2 μ, 3-7 cells per hair. Hair tapers gently.
2. *M. apposita.* Hair gently tapering both anteriorly and posteriorly; proximal cell with prominent protruded stalk. 425-900 μ long; 32-64 μ wide, contact face diameter 16 μ; 5-8 cells per hair.

3. *M. berhautii.* Hair wavy. 2187-2425 μ long; 48-76 μ wide; contact face diameter 32-48 μ; 5-8 cells per hair.

4. *M. botryocarpa.* Hair length 337.5-887 μ; 48-73.6 μ wide; contact face diameter 16 μ; 3-6 cells per hair.

5. *M. burchellii.* Hair length 350-825 μ; 48-73.6 μ wide; contact face diameter 12.8-16 μ; 4-7 cells per hair.

6. *M. capensis.* Hair gradually tapering. Hair length 550-1337.5 μ; 38.4-102.5 μ wide; contact face diameter 19.2-25.6 μ; 3-8 cells per hair.

7. *M. ethiopica.* Hair with characteristic tailing of the distal arm, anterior end aduncate (hooked). Hair length 712-1387 μ; 96-115 μ wide; contact face diameter 22.4-25.6 μ; 4-8 cells per hair.

8. *M. coromandelina.* Hair length 300-537.5 μ; 32-64 μ wide; contact face diameter 12.8-16 μ; 3-6 cells per hair.

9. *M. distorta.* Proximal cells of hair aduncate anteriorly distal arm tailing to fine narrow end. Hair length 925-1612.5 μ; 51-115 μ wide; contact face diameter 25.6-38.4 μ; 5-8 cells per hair.

10. *M. ephippiocarpa.* Hair gently tapering with straight anterior end. 35-800 μ long, 38.4-96 μ wide, contact face diameter 12.8-19.2 μ; 2-7 cells per hair.

11. *M. fadeniana.* Material lacking hairs.

12. *M. farinosa*. Comprises two hair types; the anvil type and the simple type. Anvil type: 350-550 μ long; 32-64 μ wide; contact face diameter 12.8-16 μ; 3-5 cells per hair. Simple type: 350-475 μ long; 32-41.5 μ wide; contact face diameter 35.2-54 μ; 7-8 cells per hair.
13. *M. fenestrata*. Hair length 587.5-750 μ; 32.7-36 μ wide; contact face diameter 16 μ; 4-6 cells per hair.
14. *M. gibba*. Hair length 487.5-912 μ; 32-70 μ wide; contact face diameter 16 μ; 3-6 cells per hair.
15. *M. macrocarpa*. Hair with short intermediate cells. Hair length 750-1762.5 μ; 32-102 μ wide; contact face diameter 22.4-25.6 μ; 4-9 cells per hair.
16. *M. megalomanica*. Anterior end of hair aduncate. Hair length 437.5-700 μ; 48-73.6 μ wide; contact face diameter 16-19.2 μ; 4-6 cells per hair.
17. *M. minuta* var. *incurva*. Anterior end of hair slightly obtuse. Hair length 387-950 μ; 25.6-76.4 μ wide; contact face diameter 12.8-16 μ wide; 3-5 cells per hair.
18. *M. minuta* var. *minuta*. Anterior end of hair slightly obtuse. Hair length 350-775 μ; 22.4-73.6 μ wide; contact face diameter 9.6-16 μ; 3-5 cells per hair.
19. *M. nubica* var. *gymnocarpa*. Hair length 187.5-375 μ; 32-48 μ wide; contact face diameter 9.6 μ; 3-5 cells per hair.
20. *M. nubica* var. *nubica*. Hair length 475-775 μ; 48-67.2 μ wide; contact face diameter 16 μ;

21. *M. schelpiana.* Hair length 875-1387 μ; 64-102 μ wide; contact face diameter 16-22.4 μ; 4-8 cells per hair.

22. *M. strigosa.* Hair contact face exceptionally protruded, anterior end sharply tapering. Hair length 812.5-2437.5 μ; 44.6-76.8 μ wide; contact face diameter 19.2-22.4 μ; 3-8 cells per hair.

23. *M. subterranea.* Hair length 575-1412.5 μ; 48-96 μ wide; contact face diameter 16-25.6 μ; 3-5 cells per hair.

24. *M. unicornis.* Hair length 275-1612.5 μ; 48-105 μ wide; contact face diameter 16 μ; 3-8 cells per hair.

25. *M. vera.* Hair length 1912.5-2500 μ; 108-128 μ wide; contact face diameter 16-22.4 μ; 4-11 cells per hair.

26. *M. villifolia.* Hair with prominently protruded contact face, with conspicuous streaks of a brown yellow substance; proximal cell anteriorly upturned. Hair length 1550.5-2850 μ; 67.2-128 μ wide; contact face diameter 22.4-32 μ; 5-8 cells per hair.

27. *Marsilea* spp. Hair with characteristic tailing of distal arm. 1712.5-3012.5 μ long; 112-160 μ wide; contact face diameter 25.6-48 μ; 8-10 cells per hair.

3.3 Histology

Transverse sections of young leaves and sporocarps prepared to show tissue relationships have revealed interesting information. Micrographs taken from the prepared slides are presented in Plates 21-22. The mode in which the hair is attached to the epidermis is illustrated in Plate 21 and sketched in Fig. 2.

The hair arises from a more or less trapeziform epidermal foot cell which is comparatively smaller than the neighbouring epidermal cells and slightly lower resulting in the hair dipping below the general level of the epidermis (see Plate 12e and Fig. 2c). The stages of the hair development is shown in Plate 22b, c. The foot cell divides periclinally giving rise to the initial hair cell which later divides anticlinally to form a two-armed, asymmetrical, uniseriate structure oriented parallel to the surface of the leaf or sporocarp. In this particular Australian species, *M. drummondii*, the hairs occur only on what later becomes the lower side of the leaf when the leaflets are fully open in the mature state. As shown on Plate 22a the hairs can be seen to develop on the outermost surface of the folded leaflets, and that the most external leaflets and their hairs mature much earlier than the two innermost leaflets.

3.4 General description for hairs of the genus

This description refers to examined specimens of the African species only. The hairs are very variable within the genus. Usually, the hair is anvil-shaped but elaborated into a two-armed asymmetrical, uniseriate

Fig. 1. *Marsilea farinosa* Launert. Showing microscopic impression of the two hair types.

a) Anvil-shaped sporocarp hair. x 125

b) Simple sporocarp hair. Note blunt apex. x 125

c) Anvil-shaped leaf hair. x 125

d) Simple leaf hair. x 125

FIG. 1. Marsilea farinosa Launert

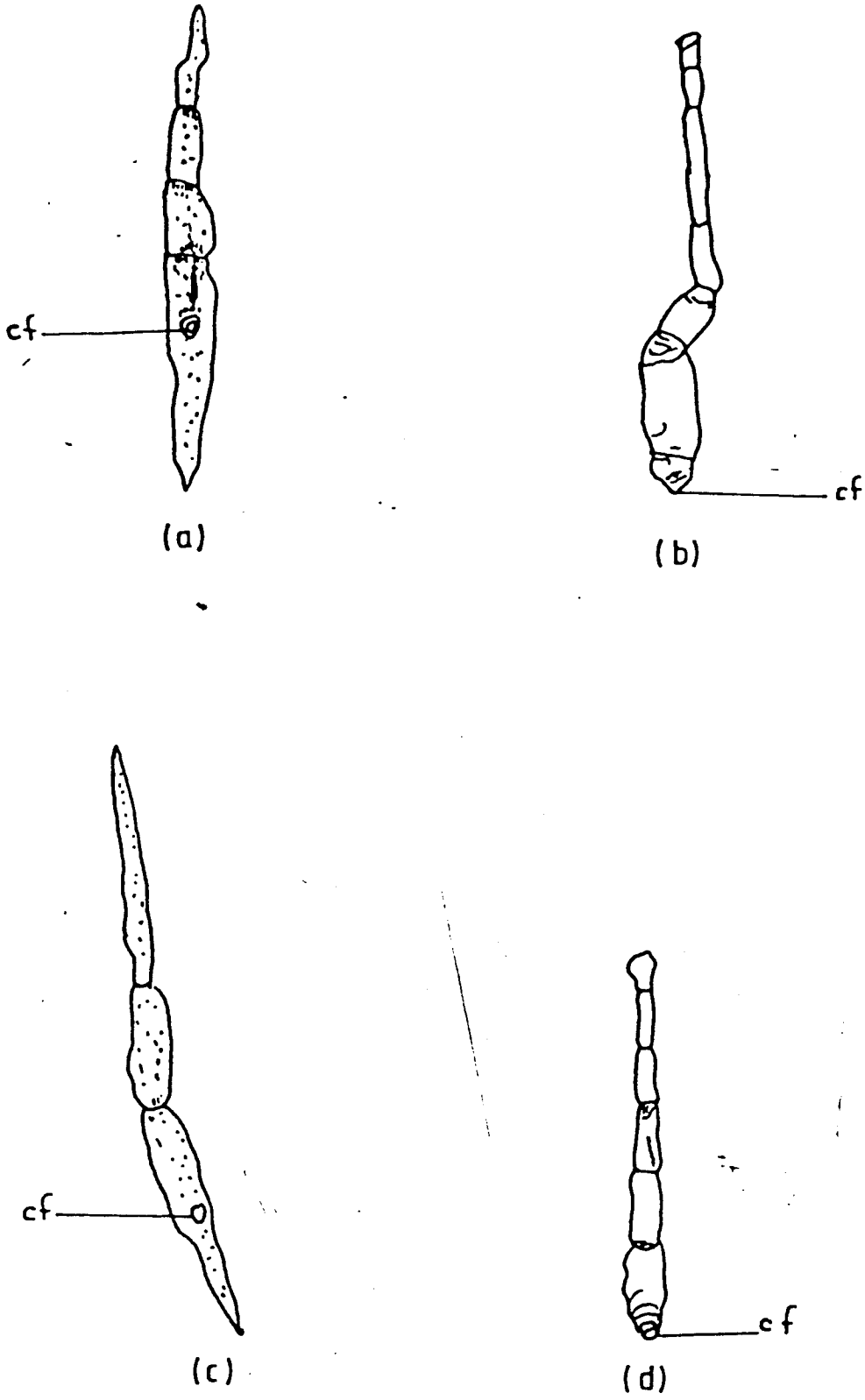


Fig. 2. *Marsilea drummondii* A. Braun

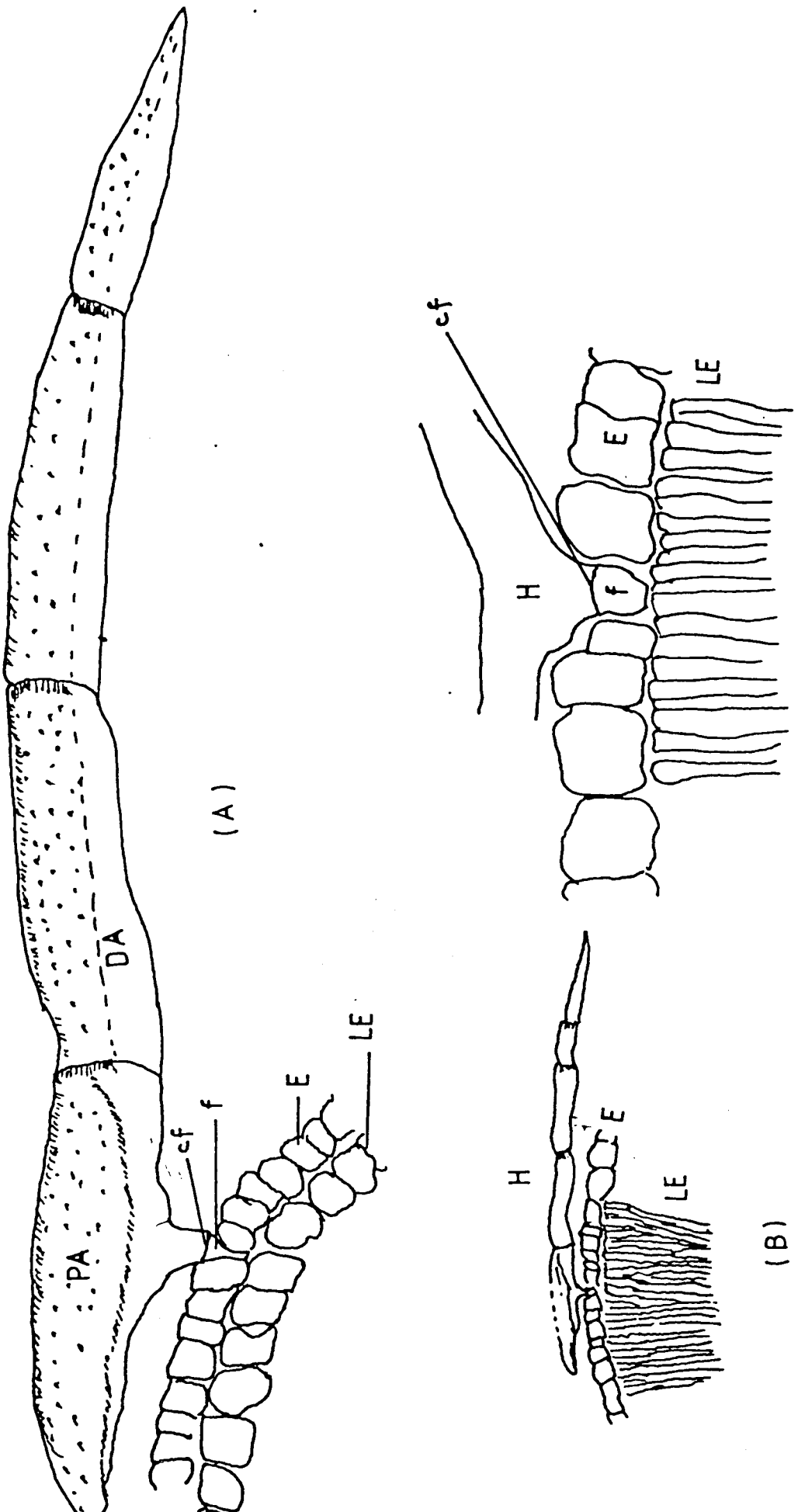
a) Stipe hair. x 700

b) Sporocarp hair. x 300

c) Sporocarp hair showing mode of attachment to the epidermis. x 1500

Key to labelling: cf. contact face; DA distal arm; E epidermis; f foot cell; H hair; LE hypodermis; PA proximal arm (= proximal cell)

FIG. 2.



body forming a shorter proximal arm (cell) and a longer distal arm, both oriented in opposite direction parallel to the surface of the leaf or sporocarp. Sporocarp hairs with proximal arm adpressed to the organ surface, the distal arm raised; leaf hairs with either entire hair body appressed or with distal arm raised. Widest part of hair occurs on proximal arm directly above the hair's contact face. Sporocarp hair length 300-3012.5 μ; 32-160 μ wide; contact face diameter 12.8-48 μ; cell number per hair, 3-10. Leaf hairs 210-2087.5 μ long; 28.6-80 μ wide; contact face diameter 9.6-16 μ; cell number per hair 2-8.

The dorsal surface sculptured with numerous papillae; papillae very few to absent on ventral surface. The development of papillae is attributable to environment, particularly light intensity.

3.5 Analysis

Sample means and means of sample means per species were calculated as measures of central tendency; and measures of dispersion expressed as standard deviation (Campbell, 1967; Rowntree, 1981). The calculated data are presented in Append ix II and summarised in Table 1. The scatter diagram shown in Fig. 3 was based on data from Table 1. Similarly, histograms for such parameters as hair length versus number of species, and number of cells per hair versus species numbers for both the sporocarp and leaf hairs, were derived from Table 1. The scatter diagram shows a positive correlation between

Table 1. Summary: Calculated means of hair measurements taken from 3-4 samples of each species

	SPOROCARP HAIRS							LEAF HAIRS						
Species	No. of cells/ hair	PA_1 μ	PA_w μ	$\frac{PA_1}{PA_w}$	Stalk dia. μ	Total hair length μ	$\frac{PA_1}{H_1}$	No. of cells/ hair	PA_1 (μ)	PA_w (μ)	$\frac{PA_1}{PA_w}$	Stalk dia. (μ)	Total hair length μ	$\frac{PA_1}{H_1}$
aegyptiaca	3-7	250.00	58.02	4.308	17.07	583.33	0.428	2-4	295.83	39.46	7.496	11.73	562.91	0.525
apposita	5-8	255.63	59.59	4.289	16.00	660.00	0.387	2-4	272.86	41.76	6.534	9.60	612.88	0.445
berhautii	5-8	464.38	79.14	5.867	29.60	2263.16	0.205	-	-	-	-	-	-	-
botryocarpa	3-6	254.58	58.88	4.323	16.00	627.50	0.405	2-4	203.31	38.72	5.250	10.66	442.92	0.459
burchellii	4-7	159.00	59.44	2.674	14.70	541.88	0.293	2-4	211.93	38.00	5.577	9.60	373.75	0.567
capensis	3-8	363.46	79.92	4.547	22.16	1051.38	0.345	2-6	235.16	37.37	6.292	10.66	518.61	0.453
ethiopica	4-8	394.55	104.88	3.762	24.98	1232.08	0.320	2-5	340.00	50.24	6.727	16.00	855.40	0.397
coromandelina	3-6	165.00	47.17	3.490	13.51	400.83	0.411	2-4	181.25	33.60	5.394	9.60	371.25	0.488
distorta	5-8	444.58	79.64	5.582	32.82	1400.25	0.317	2-4	227.21	36.00	6.311	9.60	510.81	0.444
ephippiocarpa	2-7	232.63	61.10	3.807	16.00	565.10	0.411	2-5	252.35	38.81	6.58	14.75	555.79	0.454
fadeniana	4	525.00	92.80	5.657	16.00	737.50	0.711	2	116.25	42.88	2.711	9.60	210.00	0.553
farinosa (i)	3-5	218.47	56.06	3.897	13.81	483.15	0.452	2-3	182.50	44.13	4.135	10.66	438.31	0.416
farinosa (ii)	7-8	48.62	52.26	0.930	16.00	393.74	0.133	4-6	25.00	23.04	1.085	12.80	262.50	0.095
fenestrata	4-6	238.75	65.28	3.657	16.00	673.75	0.354	2-3	237.50	43.73	5.431	9.60	377.08	0.704

Table 1. (continued)

	SPOROCARP HAIRS								LEAF HAIRS						
	Species	No. of cells/ hair	PA_l μ	PA_w μ	$\frac{PA_l}{PA_w}$	Stalk dia. μ	Total hair length μ	$\frac{PA_l}{H_l}$	No. of cells/ hair	PA_l (μ)	PA_w (μ)	$\frac{PA_l}{PA_w}$	Stalk dia. (μ)	Total hair length μ	$\frac{PA_l}{H_l}$
	macrocarpa	4-9	442.48	87.15	4.077	25.44	1327.50	0.333	2-4	287.85	36.58	7.869	9.60	599.01	0.480
	megalomanica	4-6	243.75	56.48	4.315	17.60	599.38	0.406	2-5	265.63	27.70	9.589	9.60	662.38	0.401
	minuta var. *incurva*	3-5	287.66	67.63	4.253	12.95	667.92	0.430	2-3	144.43	33.48	4.314	9.60	276.69	0.522
	minuta var. *minuta*	3-5	234.58	60.77	3.860	14.13	569.69	0.411	2-3	177.42	31.68	5.600	9.60	340.02	0.521
	nubica var. *gymnocarpa*	3-5	115.55	43.44	2.659	9.60	285.00	0.405	2-3	141.66	26.66	5.314	6.40	312.00	0.454
	nubica var. *nubica*	3-5	213.75	61.42	3.480	16.00	608.13	0.351	2	133.33	23.47	5.680	9.60	291.66	0.457
	schelpiana	4-8	446.88	81.12	5.755	20.27	1210.62	0.369	2-5	313.50	44.79	6.999	11.04	726.23	0.432
	strigosa	3-8	384.58	60.26	6.382	20.62	1497.08	0.256	2-5	252.91	33.33	7.588	10.66	789.00	0.321
	subterranea	3-5	407.92	72.75	5.607	18.13	1040.68	0.391	-	-	-	-	-	-	-
	unicornis	3-8	244.58	63.75	3.836	16.00	791.25	0.309	2-5	215.00	34.61	6.212	10.27	495.42	0.433
	vera	4-11	488.47	156.25	3.126	24.53	1934.03	0.252	3-5	317.5	80.01	3.968	13.56	913.75	0.347
	villifolia	5-8	559.15	96.78	5.776	22.81	2012.92	0.277	2-8	357.5	32.10	11.137	12.26	1264.85	0.282
	Marsilea sp.	8-10	356.25	127.04	2.804	36.62	2537.00	0.140	2-5	237.5	39.82	5.96	12.16	626.25	0.379

Table 1. (continued)

	SPOROCARP HAIRS								LEAF HAIRS						
	Species	No. of cells/ hair	PA_1 μ	PA_w μ	$\frac{PA_1}{PA_w}$	Stalk dia. μ	Total hair length μ	$\frac{PA_1}{H_1}$	No. of cells/ hair	PA_1 (μ)	PA_w (μ)	$\frac{PA_1}{PA_w}$	Stalk dia. (μ)	Total hair length μ	$\frac{PA_1}{H_1}$
	drummondii	4-8	322.50	104.00	3.100	29.60	1648.75	0.195	4-6	515.00	60.48	8.515	16.00	2082.50	0.247
	hirsuta	5-8	305.00	85.70	3.558	24.40	1307.50	0.233	3-6	402.50	76.80	5.240	16.00	1557.50	0.258
	macra	6-8	307.50	93.76	3.279	27.20	1388.75	0.221	2-6	450.00	65.28	6.893	17.92	1761.25	0.255
	minuta	3-4	182.50	61.12	2.985	12.80	380.00	0.480	-	-	-	-	-	-	-
	polycarpa	3-6	196.56	55.62	3.533	16.00	622.65	0.315	3-4	182.50	35.20	5.185	-	588.00	0.310
	vestita	4-6	256.25	48.32	5.303	12.80	692.50	0.370	2-3	235.00	32.96	7.129	9.60	445.00	0.528

to abbreviations: PA_1 = length of proximal arm; PA_w = width of proximal arm; H_1 = hair length

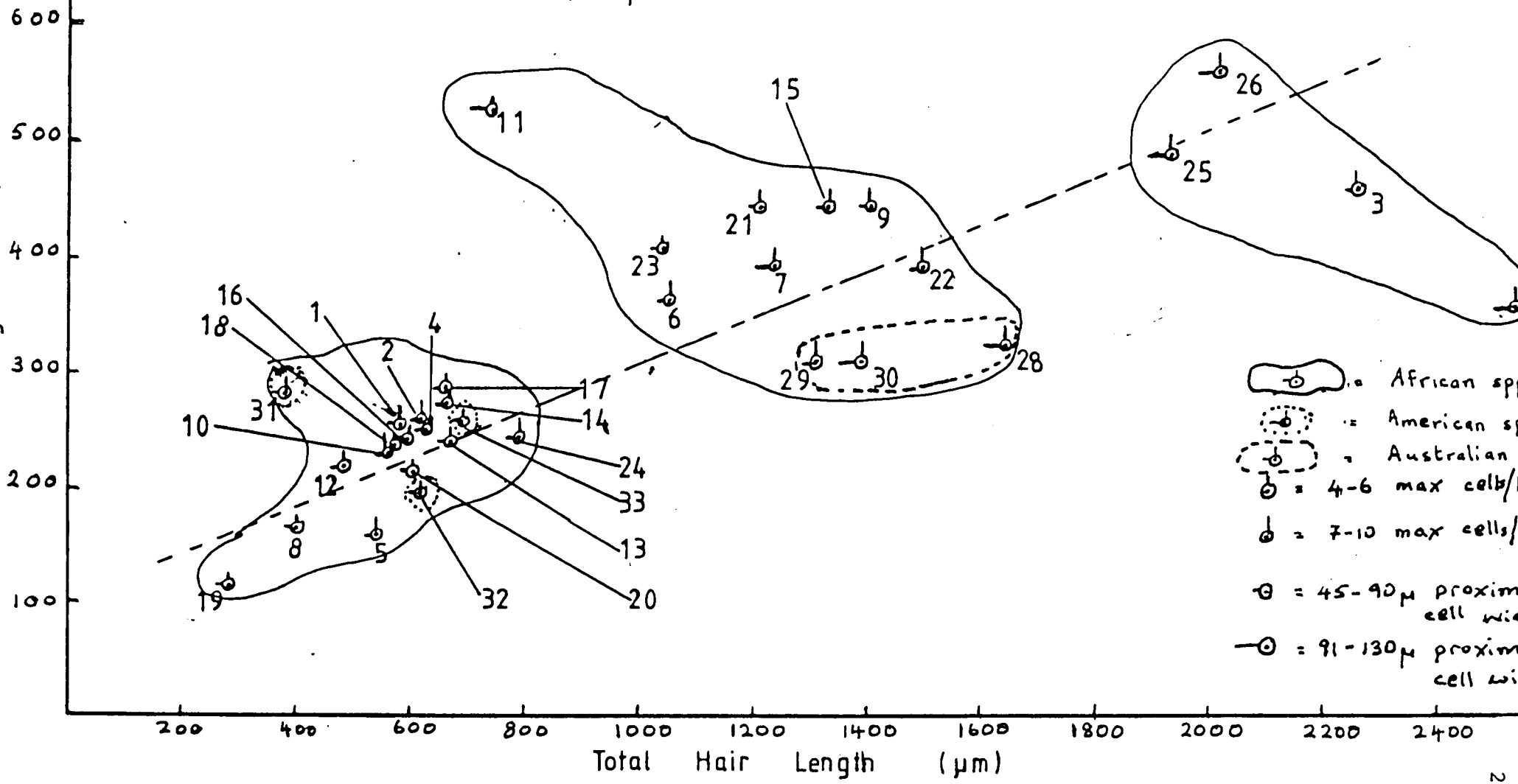
N.B. Stalk dia. is expressed as "Contact face" in text.

FIG 3 SCATTER DIAGRAM FOR 33 SPECIES OF MARSILEA

(Key: Refer to Appendix I for species names)

For Sporocarp hairs

Scal



FIG, 4. HISTOGRAMS OF HAIR LENGTHS vs NUMBER OF SPECIES

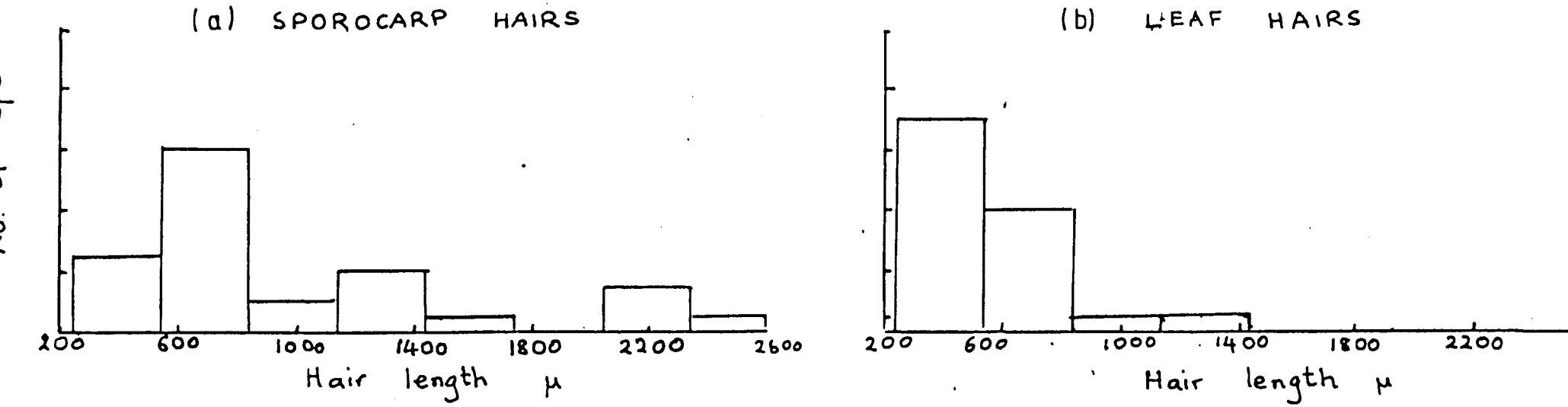
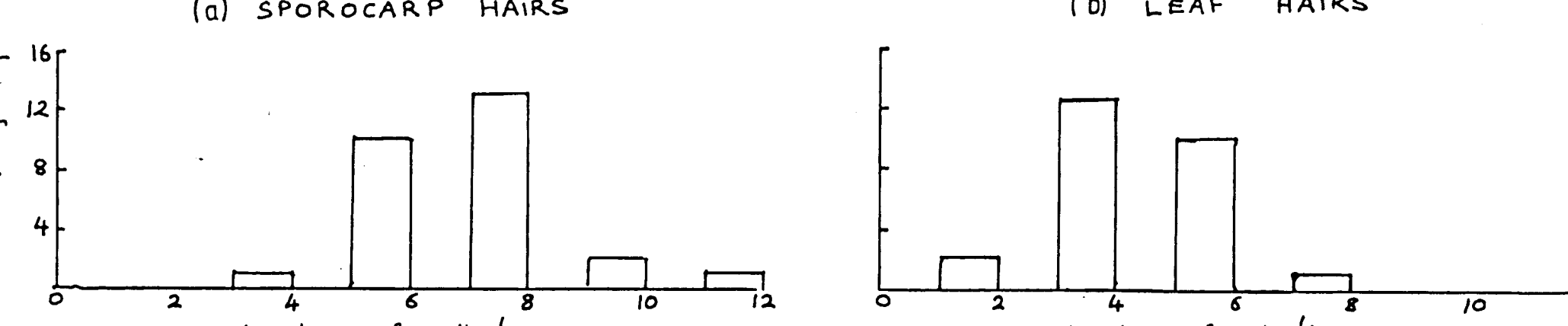


FIG. 5: HISTOGRAMS OF NUMBER OF CELLS vs NUMBER OF SPECIES



the hair length and the proximal cell length, with species clustered in three main groups. The 15 African species and the 3 New World species score much lesser dimensions, 4 species (all African) registering much higher values, and 8 African species and 3 Australian ones taking the intermediate position. In Fig. 4a the three peaks correspond to the three groups in the scatter diagram. As for leaf hairs in Fig. 4b the histogram is skewed towards species with hair lengths in the range of 200-600 μ, emphasising that in general the leaf hairs are shorter than sporocarp hairs. Fig. 5a and b show a normal distribution of cell numbers per hair. For many species the maximum cell number is 8 in sporocarp and 4 in leaf hairs.

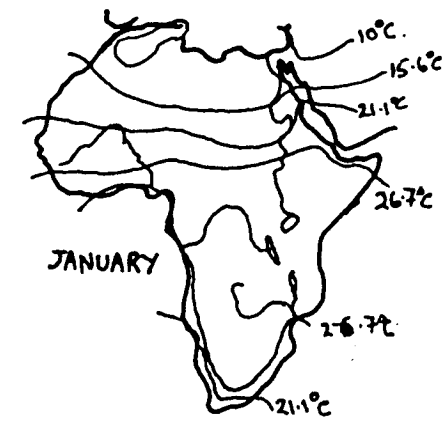
3.6 Phytogeography

The distribution of the species examined is given on a map (see Fig. 6). The highest number of species is recorded from Namibia. Maps shown in Fig. 7 have been included to show some correlation between species distribution and some of the climatological elements, such as monthly isotherms for peak seasons and the annual precipitation. Most species are distributed along the Sudano-Zambezian, the Karroo Namib and the Cape phytogeographical regions. Few, if any have been recorded in the Guinea-Congolian Region. The largest species diversity occurs within the Karroo-Namib Region.

FIG 6 Distribution of examined Marsilea spp.

KEY TO SPECIES

1. M. aegyptiaca
2. M. apposita
3. M. berhautii
4. M. burchellii
5. M. botryocarpa
6. M. capensis
7. M. ethiopica
8. M. coromandelina
9. M. distorta
10. M. ephippiocarpa
11. M. fadeniana
12. M. farinosa
13. M. fenestrata
14. M. gibba
15. M. macrocarpa
16. M. megalomanica
17. M. minuta var. incurva
18. M. minuta var minuta
19. M. nubica var gymnocarpa
20. M. nubica var nubica
21. M. schelpiana
22. M. strigosa
23. M. subterranea
24. M. unicornis
25. M. vera
26. M. villifolia



Mean Isotherms

Mean annual precipitation (inches)

Fig. 7. Maps showing climatological elements
(Adapted from Kendrew, W.G., 1963)

4. DISCUSSION

4.1 Morphology and Anatomy

A number of reports have been made on anatomical accounts of some species of *Marsilea*. This present study merely adds further information on yet a small area involving this genus. The evidence obtained from the scanning electron microscopy and histological procedures reveals new information. The nature of the hairs of *Marsilea* was never fully understood before. Bower (1926) earlier gave an account of the entire family, Marsileaceae, but his emphasis lay in the anatomical features of the related genus *Pilularia*. He described the hairs of *Pilularia globulifera* L. as simple hairs and an illustration of the median longitudinal section to show the nature of the indumentum of the sporocarp. About 27 years later, Puri and Garg (1953) reported anatomical accounts on the sporocarps of *Marsilea minuta* L. The trichomes were merely referred to as "unbranched multicellular hairs"; and also included an illustration of the developmental stages of the sporocarp hairs. The hairs were then interpreted as upright structures commonly found in hairs of most angiosperms. Indeed Puri and Garg account was an excellent contribution to the evolutionary aspect of the sporocarp in one of the heterosporous ferns; their work was a vital start that triggered further investigation into *Marsilea* as a group. However, their interpretation of the mature state of the hair gives an impression of an upright ordinary hair; but this could have been a hair appearing from behind the epidermal tissue out of field

microscopic focus. Hairs of *Marsilea* in all species including *M. minuta* do not conform to the shape illustrated by Puri and Garg. Later Bilderback (1978) described the sporocarp hairs of *M. vestita* as multicellular trichomes, but his studies were restricted to histochemistry in relation to the development of the sporocarp.

The hairs of *Marsilea* bear some resemblance to the hairs found in Malpighiaceae as described by Herberlandt (1914) and Payne (1978). But in Malpighiaceae the hairs are essentially unicellular. As indicated above, in the hairs of *Marsilea* the intial stages of development begins wit the foot cell dividing to give rise to an anvil-shaped hair body, this continues to divide and produce a uniseriate structure but still retaining the same Malpighian-like posture. The hairs of *Marsilea* are thus a very distinct entity and typical of the Marsileaceae.

4.2 Importance of hairs in taxonomy

The core substance of this report is to determine whether trichomes of *Marsilea* can be used as supporting characters in the classification of this genus. It has now been established that, although similar in shape to those in the Malpighiaceae and few other families, for example, Cruciferae, the hairs of *Marsilea* are uniquely different from those of other taxa in morphological and anatomical characters. In addition, it has also been shown that variation exists in the nature of hairs at interspecific level - sufficiently adequate to warrant the inclusion of hair characters in delimiting the species of *Marsilea*.

The analysis shown in both the scatter diagram and the histograms (see Fig. 3, 4 and 5) provide evidence for discontinuous variation. The morphological characters exhibited in SEM and light microscope micrographs confirm differences in the nature of hairs between different species. To illustrate this point, there arose a situation where, according to Launert (1968), *M. capensis* contained few specimens from South Africa and Ethiopia, but later research based on more material showed that the specimen from Ethiopia could be specifically separated from the South African species. The revised species are *M. capensis* A. Braun and *M. ethiopica* Launert (1983). This investigation shows that the sporocarp and leaf hairs of *M. capensis* are shorter with a narrower width compared to those of *M. ethiopica*; morphological distinctions give a straight anterior end of the proximal cell found in *M. capensis* whereas *M. ethiopica* has a hooked anterior end; and other differential characters are found in the number of cells per hair.

It must be noted here that the analytical technique of using scatter diagrams is a useful tool in indicating some obvious variation in parameters of different taxa. In some few cases two or more taxa which cluster in one group on the scatter diagram may not necessarily imply a phylogenetic relationship; just as well that two different taxa occupying different positions with a wide distance between them may not entail phylogenetic dissimilarity. *Marsilea berhautii* and *M. polycarpa* are widely separated on the scatter diagram, thus giving an

impression that the two species are only remotely related. On the contrary, *M. polycarpa* bears a remarkable resemblance to *M. berhautii* as both possess sporocarps that are arranged in a single row along the stipe. This relationship was realised by Tardeue and later supported by Alston to a point where the West African species, *M. berhautii* was proposed as a synonym of the New World species, *M. polycarpa* (Launert, 1968). Plate 17 illustrates and confirms Launert's (1968) contention that the two species are quite distinct. The sporocarp hairs of *M. berhautii* are wavy and give a mean length of 2263.6 μ compared to hairs of *M. polycarpa* which are shorter and stout with a mean length of 622.65 μ. The point to be stressed here is that hair characters can strongly support the delimitation of species but cannot, by themselves, be employed to show any phylogenetic relationship between related taxa. It requires a whole range of characters from as many organs as possible if phylogenetic trends are to be realised.

The hairs of the undescribed species (ref. no. 27), Merxmüller and Giess 30,377, collected from Ovamboland in Namibia, exhibits hair characters very distinct from the rest of the species examined in this study. This species scores the longest mean sporocarp hair length of 2537.5 μ. A critical examination of this plant's sporocarps as well as other attributes such as its ecology and breeding system may reveal interesting results.

4.3 Phytogeography

The genus *Marsilea* is a vascular hydrophyte adapted to open water edges and often occurring in areas subject to periodic flooding. The examined specimens are shown to have been collected in localities distributed right across the Sudano-Zambezian domain as well as the Cape and the Karroo-Namib regions. The greatest diversity of the species of *Marsilea* is recorded from Namibia. The cause for such high diversity in species in the Namib region is not clear. It is only tempting to speculate that steep gradients of isotherms and incoming radiation more or less permanently stationed in this region could create sharp environment differences over small distances and set up mechanisms for speciation in plant taxa occurring in the region. This hypothesis requires more evidence from field studies.

5. CONCLUSIONS

There is clear evidence that hair variation exists in species of *Marsilea* as indicated in this study on the African species. The hairs form a unique structural pattern known only in this genus and never reported in any other plant groups. The interspecific variation exhibited is sufficient evidence for using the hair characters in lending support to the overall systematic classification of the species. The existence of variation in the epidermis of *Marsilea* species was earlier recognised by Nickel and Votava (1971). This report offers further evidence, and agrees with the proposed thesis, that the hairs as epidermal appendages do exhibit variation of taxonomic importance to the classification of the species of *Marsilea*. The sporocarps still remain the major organ that provides characters in the classification, but the hairs can now consolidate and help stabilise the classification system for the genus. Hairs alone cannot provide adequate characters in a classification since the Adamsonian approach of using as many plant characters as possible is the best course in an effort to produce a stable classification.

Further investigations with regard to the bio-systematics of the genus is required before a comprehensive revision of *Marsilea* can be embarked upon. Integrated world-wide studies on such specific areas as phytogeography, ecology, cytogenetics and breeding systems of the species

supported by field and experimental studies could unravel valuable data important in the understanding of this intriguing genus.

ACKNOWLEDGEMENTS

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REFERENCES

ALSTON, A.H.G. 1959. The Ferns and Fern-allies of West Tropical Africa. C.A.O.G. London.

ARONA, K., GRACE, J. and STEWART, F. 1982. Epidermal features of *Heraclean mantegazzianum* Somm. & Lev., *H. sphondylium* L. and their hybrid. Bot. J. Linn. Soc. 85: 169-177.

BHARDWAJA, T.N. and SHARMA, P. 1977. Distribution, Intraspecific Variation and Epidermal Studies of *Marsilea coromandelina* Willd. in India. Nova Hedwigia 28 (2 + 3): 451-462.

BILDERBACK, D.E. 1978. The Development of the Sporocarp of *Marsilea vestita*. Amer. J. Bot. 65(6): 629-637.

BOLD, H.C., ALEXOPOULOS, C.J. and DELEVORYAS, T. 1980. Morphology of Plants and Fungi, 4th Ed. Harper & Row.

BOWER, F.O. 1926. The Ferns (Filicales). Vol. II. Cambridge University Press.

CAMPBELL, R.C. 1967. Statistics for Biologists. Cambridge University Press.

CUTLER, D.F. 1978. Applied Plant Anatomy. Longmans.

DAVIS, P.H. and HEYWOOD, V.H. 1973. Principles of Angiosperm Taxonomy. Robert E. Krieger.

EDMONDS, Jennifer M. 1982. Epidermal Hair Morphology in *Solanum* L. section *Solanum*. Bot. J. Linn. Soc. 85: 153-167.

FABBRI, F. 1963. Tables of Chromosome Numbers in Pteridophytes. Caryologia 16: 237-332.

FAHN, A. 1982. Plant Anatomy. Third Ed. Pergamon Press.

GRIMSTONE, A.V. 1979. The Electron Microscope in Biology. Arnold.

HABERLANDT, G. 1914. Physiological Plant Anatomy. Macmilland & Co. Ltd.

HOLTTUM, R.E. 1968. Flora of Malaya. Vol. II. Government Printer, Singapore.

INAMDAR, J.A., BHATT, D.C. and PATEL, R.C. 1971. Epidermal Structure and Ontogeny of Stomata in some Leptosporangiate Ferns. Israel J. Bot. 20: 111-123.

KAUR, Surjit. 1972. Contribution to the Morphology of *Dennstaedtia*. Proc. Indian Acad. Sc. Sect. B. 75(1): 32-39.

KENDREW, W.G. 1963. The Climates of the Continents. Oxford.

LAUNERT, E. 1968. A Monographic Survey of the Genus *Marsilea* Linnaeus. Senckenberg. Biol. 49: 273-315.

LAUNERT, E. 1975. Marsileaceae. In: Flore d'Afrique Centrale (Zaire-Rwanda-Burundi), Pteridophytes. Jard. Bot. Natl. de Belgique, Meise. pp. 1-7.

LAUNERT, E. 1983. Personal communication.

MICKEL, J.T. and VOTAVA, F.V. 1971. Leaf Epidermal Studies in *Marsilea*. Amer. Fern J. 61: 101-109.

MITSUTA, S. 1982. Venation and Trichomes of Drynorioid Ferns (Polypodiaceae). Acta. Phytotaxon Geobot. 33(0): 264-275.

PAGE, C.N. 1979. The Diversity of Ferns. An Ecological Perspective. In: The Experimental Biology of Ferns, by Dyer, A.F. (Editor). Acad. Press.

PAYNE, W.W. 1978. A Glossary of Plant Hair Terminology. Brittonia 30(2): 239-255.

PURI, V. and GARG, M.L. 1953. A Contribution to the Anatomy of the Sporocarp of *Marsilea minuta* L. with

a Discussion of the Nature of the Sporocarp in the Marsileaceae. Phytomorph. 3: 190-209.

PURVIS, M.J., COLLIER, D.C., WALLS, D. 1966. Laboratory Techniques in Botany. Butterworths.

ROWNTREE, D. 1981. Statistics Without Tears. Penguin.

SEITHE, A. 1979. Hair Types as Taxonomic Characters in *Solanum*. In: J.G. Hawkes, R.E. Lester and A.D. Skelding (Eds.) The Biology and Taxonomy of the Solanaceae. Acad. Press.

SPORNE, K.R. 1975. The Morphology of Pteridophytes. Hutchinson.

STACE, C.A. 1980. Plant Taxonomy and Biosystematics. Arnold.

THEOBALD, W.L., KRAHULIK, J.L. and ROLLINS, R.C. 1979. Trichome Description and Classification. In: Anatomy of the Dicotyledons, Vol. I, 2nd Ed. by C.R. Metcalfe and Chalk (Eds.).

TIMES, THE. 1967. The Times Atlas of the World. Times Newspapers Ltd.

TRYON, R.M. and TRYON, Alice F. 1982. Ferns and Plant Allies. Springer-Verlag.

WERGER, M.J.A. 1978. Biogeography and Ecology of Southern Africa. Monographiae Biologicae. Vol. 31:

APPENDIX I. LIST OF SPECIMENS EXAMINED

Ref. No.	Species	
1 a.	*Marsilea aegyptiaca* Willdenow 1810	Egypt, Nilum Abur Zabel, Jan., 1835. Schimper et Wiest 33 (BM)
b.	*M. aegyptiaca* Willdenow 1810	Egypt, Ghizeh, Jan., 1926, Elliot 3290 (BM)
c.	*M. aegyptiaca* Willdenow 1810	Namibia, Omaruru, 18.2.63, Kers 968 (BM)
2 a.	*M. apposita* Launert 1968	Zimbabwe, Matopo Hills X.1905 Gibbs 289 (BM, holotype)
b.	*M. apposita* Launert 1968	Zimbabwe, Bulawayo, 30.6.53 Schelpe 3915 (BM)
3 a.	*M. berhautii* Tardieu 1955	Nigeria, Borno State, 27.10.77 Kornas 6282 (BM)
b.	*M. berhautii* Tardieu 1955	Nigeria, Kaduna, 3.11.55 Jordan s.n. (BM)
4 a.	*M. botryocarpa* F. Ballard 1962	Kenya, Taita Dist., 1.6.71 Evans & Mwaikeki 62 (BM)
b.	*M. botryocarpa* F. Ballard 1962	Kenya, Taita Dist., 23.1.72 Faden 72/102 (BM)
c.	*M. botryocarpa* F. Ballard 1962	Kenya, Taita Dist., 22.1.72 Faden 72/85 (BM)
5 a.	*M. burchellii* A. Braun 1864	South Africa, Aberdeen, 11.2.54 Schelpe 4562 (BM)
b.	*M. burchellii* A. Braun 1864	Namibia, Fish River Stn. XIII 1935 White s.n. (BM)
c.	*M. burchellii* A. Braun 1864	South Africa, Bloemfontain 1875-80 Rehmann 3760 (BM)
d.	*M. burchellii* A. Braun 1864	Botswana, Nata R. 13.8.30 Van Sow 1804c (BM)
6 a.	*M. capensis* A. Braun 1864	South Africa, Grahamstown II 1955 Pocock s.n. (BM)
b.	*M. capensis* A. Braun 1864	South Africa, Cape Province, 8.11.61 Barker 9660 (BM)
c.	*M. capensis* A. Braun 1864	Regio occidentalis, 21.1.1901 Prater Kloof 10889 (BM)

APPENDIX I. (continued)

Ref. No.	Species	
7 a.	*Marsilea ethiopica* Launert 1983	Ethiopia, Eritrea, 25.1.02 Pappi 2549 (FL)
b.	*M. ethiopica* Launert 1983	Ethiopia, Eritrea, 19.1.09 Friori 821 (FL)
c.	*M. ethiopica* Launert 1983	Ethiopia, Eritrea, 3.1.43 Baldrati 4533 (FL)
8 a.	*M. coromandelina* Willd 1810	Kenya, Taita, 22.1.72 Faden 72/84 (BM)
b.	*M. coromandelina* Willd 1810	Burundi, Bujumbura, 10.2.68 Lewalle 2802 (BM)
c.	*M. coromandelina* Willd 1810	Namibia, Haribes, 11.4.56 Volk 12331 (BM)
9 a.	*M. distorta* A. Braun 1864	Tanzania, Moshi Dist., VIII 1928 Haarer 1468 (BM)
b.	*M. distorta* A. Braun 1864	Nigeria, Borno State, Mafa 24.11.77 Kornas 6419 (BM)
c.	*M. distorta* A. Braun 1864	Zimbabwe, Chipinga, 1.6.72 Russell 2084 (BM)
10 a.	*M. ephippiocarpa* Alston 1930	Zimbabwe, Ndanga, 15.5.63 Whellan 2034 (BM)
b.	*M. ephippiocarpa* Alston 1930	Zimbabwe, Fort Victoria, 9.8.29 Rendle 307 (BM)
c.	*M. ephippiocarpa* Alston 1930	Botswana, Ngamiland, 5.5.30 Van Sow 1802c (BM)
d.	*M. ephippiocarpa* Alston 1930	Namibia, Amaruru, 26.3.65 Tolken & Hardy 825 (BM)
11	*M. fadeniana* Launert	Kenya, Kwale, 29.5.71 Evans & Maikeveki 55 (BM)
12 a.	*M. farinosa* Launert	South Africa, Waterburg Dist., 18.5.1905 Burtt-Davy 4081 (BM)
b.	*M. farinosa* Launert 1968	Namibia, Grootfontein, 13.7.34 Dinter 7688 (BM)
c.	*M. farinosa* Launert 1968	Namibia, Lothlekane, 1965 Wild & Drummond 7263 (BM)

APPENDIX I. (continued)

Ref. No.	Species	
13	*Marsilea fenestrata* Launert 1960	South Africa, Zululand, 7.2.54 Ward 2458 (Type, BM)
14 a.	*M. gibba* A. Braun 1871	Tanzania, Serengeti (no date) Greenway 9878 (BM)
b.	*M. gibba* A. Braun 1871	Kenya, Laikipia, 26.1.70 Evans 9 (BM)
c.	*M. gibba* A. Braun 1871	Rwanda, Kagera, 30.12.67 Lewalle 2612 (BM)
15 a.	*M. macrocarpa* Presl 1845	South Africa, Roxburg, 3.10.25 Roux 17157 (BM)
b.	*M. macrocarpa* Presl 1845	Kenya, Kitui, 26.12.70 Gillet 19269 (BM)
c.	*M. macrocarpa* Presl 1845	Botswana, Joverega, 15.5.77 Smith 2014 (BM)
d.	*M. macrocarpa* Presl 1845	Namibia, Ovamboland, 29.3.74 Merxmuller & Giess 30643 (BM)
e.	*M. macrocarpa* Presl 1845	Kenya, Kajiado, 15.1.72 Faden 72/78 (BM)
16 a.	*M. megalomanica* Launert	Kenya, Isiolo, 6.2.70 van Swinderen Is/83 (BM)
b.	*M. megalomanica* Launert	Kenya, Taita, 22.1.72 Faden 72/80 (BM)
17 a.	*M. minuta* var. *incurva* (L.) Launert	Ghana, Accra, 1941. Scholes s.n. (BM)
b.	*M. minuta* var. *incurva* (L.) Launert	Ghana, Gonja, 16.12.53. Morton 25015 (BM)
c.	*M. minuta* var. *incurva* (L.) Launert	Ghana, Dawa, 28.10.50. Adams 436 (BM)
18 a.	*M. minuta* var. *minuta* (L.) Launert	Nigeria, Badagri, 6.10.56. Thorold 2023 (BM)
b.	*M. minuta* var. *minuta* (L.) Launert	Tanzania, Iringa, 22.8.33. Greenway 3591 (BM)

APPENDIX I. (continued)

Ref. No.	Species	
19 a.	*Marsilea nubica* var. *gymnocarpa* (A. Braun) Launert	Nigeria, Borno, 30.9.77, Kornas 6162 (BM)
b.	*M. nubica* var. *gymnocarpa* (A. Braun) Launert	Chad, Northern, VIII 1959, Popov s.n. (BM)
c.	*M. nubica* var. *gymnocarpa* (A. Braun) Launert	Senegal, S. Bayonne, 8.1.66 Audru 3074 (BM)
20 a.	*M. nubica* var. *nubica* (A. Braun) Launert	Sudan, White Nile, 6.10.39, Arasch 1841 (BM)
b.	*M. nubica* var. *nubica* (A. Braun) Launert	Kenya, Taita, 28.12.71 Faden & Smeek 71/967 (BM)
c.	*M. nubica* var. *nubica* (A. Braun) Launert	Mozambique, 29.3.74 Correia et Marques 4073 (BM)
21 a.	*M. schelpiana* Launert 1960	South Africa, Uilenheyr (undated) Harvey C3 (BM)
b.	*M. schelpiana* Launert 1960	South Africa, Summerset, XII 1874 MacOwan 1535 (BM)
22 a.	*M. strigosa* Willd 1810	Atlas, Ajuilmon, 24.6.76 Lewalle 8801 (BM)
b.	*M. strigosa* Willd 1810	D'Alger 1854, Durando s.n. (BM)
c.	*M. strigosa* Willd 1810	Algeria, Balansa, 10.6.52 Christensen 1090 (BM)

APPENDIX I. (continued)

Ref. No.	Species	
23 a.	*Marsilea subterranea* Leprieur ex A. Braun	Nigeria, Kojo, 19.1.78 Kornas 6581 (BM)
b.	*M. subterranea* Leprieur ex A. Braun	Tanzania, Musoma Greenway 9976 (BM)
c.	*M. subterranea* Leprieur ex A. Braun	Nigeria, Ngala, 30.1.78 Kornas 6611 (BM)
24 a.	*M. unicornis* Launert 1968	Namibia, Ohopoho, 13.6.63 Kers 1359 (BM)
b.	*M. unicornis* Launert 1968	Namibia, Tsumebi, IV 1934 Dinter 7585 (Type, BM)
c.	*M. unicornis* Launert 1968	Namibia, Grootfontein, 21.3.50 Schweickerdt 2080 (BM)
25 a.	*M. vera* Launert 1960	Namibia, Grootfontein, 23.4.50 Schweickerdt 2200 (BM)
b.	*M. vera* Launert 1960	Botswana, Ngamiland, 11.1.75 Smith 1231 (BM)
c.	*M. vera* Launert 1960	Namibia, Grootfontein, 16.4.50 Schweickerdt 2179 (BM)
26 a.	*M. villifolia* Bremekamp & Obermeyer ex Alston & Schelpe 1952	Botswana, 7.5.30 Van Sow s.n. (BM)
b.	*M. villifolia* Bremekamp & Obermeyer ex Alston & Schelpe 1952	Botswana, 19.10.77 Hansen 3232 (BM)
c.	*M. villifolia* Bremekamp & Obermeyer ex Alston & Schelpe 1952	Botswana, 16.5.77 Smith 2007 (BM)
27	*Marsilea* sp.	Namibia, Ovamboland, 18.3.74 Merxmüller & Giess 30377 (BM)

APPENDIX I. (continued)

Ref. No.	Species	
28	*Marsilea drummondii* A. Braun	Australia, Braun 23 (Sept. 1870) (BM)
29.	*M. hirsuta*	Australia, 3.8.62 Kuchel 259 (BM)
30	*M. macra* A. Braun	Australia, September 1870, Braun 19 (BM)
31	*M. minuta* L.	Trinidad, 10.6.78 Rameharan 440 (BM)
32	*M. polycarpa* Hook	South America, Amazonia, 26.4.52 Black et al. 52-14264 (BM)
33	*M. vestita* Hook & Grev	Texas, 22.4.30 Jones 26034 (BM)

APPENDIX II. HAIR MEASUREMENTS

	SPOROCARP HAIRS					LEAF HAIRS				
.	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total hair length μ	No. of cells per hair	PA_l (μ)	PA_w (μ)	*Stalk dia. (μ)	Total hair length μ
.	4-6	256.25	63.36	19.20	563.75	2-4	280.00	44.80	12.80	498.75
.	3-5	236.25	48.96	16.00	562.50	2-3	337.50	35.20	9.60	641.25
.	4-7	257.50	61.76	16.00	623.75	2-4	270.00	38.4	12.80	548.75
	3-7	250.00 ± 9.73	58.026 ±6.44	17.066 ±1.508	583.33 ±28.58	2-4	295.83 ±29.74	39.46 ±3.99	11.73 ±1.50	562.91 ±59.03
.	5-7	260.00	64.80	16.00	667.50	2-4	338.25	36.48	9.60	797.00
.	5-8	251.25	54.38	16.00	652.5	2-4	207.50	47.04	9.60	428.75
	5-8	253.625 ± 4.375	59.50 ±5.21	16.00	660.00 ± 7.50	2-4	272.875 ±65.375	41.76 ±5.28	9.60	612.875 ±184.125
.	5-8	366.25	59.28	40.00	2458.33	-	-	-	-	-
.	6	562.50	99.00	19.20	2068.00	-	-	-	-	-
	5-8	464.375 ±98.25	79.14 ±19.86	29.60 ±10.40	2263.165 ±195.165					
.	4-5	262.50	59.84	16.00	623.75	2-4	178.75	48.00	9.60	383.75
.	3-6	257.50	54.08	16.00	572.50	2-4	226.25	40.64	12.80	465.00
.	4-6	243.75	62.72	16.00	686.25	2-4	204.95	27.52	9.60	450.00
	3-6	254.583 + 7.927	58.88 +3.591	16.00	627.50 ±46.51	2-4	203.31 ±19.42	38.72 ±8.47	10.96 ±1.50	442.916 ±42.28

*Stalk dia. is expressed as Contact face dia. in the text.

APPENDIX II (continued)

	SPOROCARP HAIRS					LEAF HAIRS				
f.	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total hair length μ	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total hair length
a.	4-6	262.50	59.84	16.00	633.75	2-4	207.75	31.68	9.60	330.00
b.	4-5	305.00	64.64	12.80	656.25	2-4	212.50	35.84	9.60	390.00
c.	5-7	211.25	57.28	14.80	511.25	2-3	242.50	37.12	9.60	323.25
d.	3-5	158.75	56.00	15.20	366.25	2-3	185.00	47.36	9.60	451.75
	4-7	159.00 +96.98	59.44 ±3.305	14.70 ±1.178	541.875 ±115.439	2-4	203.31 ±19.42	38.72 ±8.47	10.60 ±1.50	442.916 ±42.28
a.	7-8	425.40	97.35	25.24	1169.15	2-5	305.90	40.12	12.80	624.09
b.	5-9	350.00	67.84	19.20	1073.75	2-3	210.00	36.16	9.60	492.50
c.	3-8	315.00	74.56	21.866	911.25	2-6	190.00	35.84	9.60	439.25
	3-8	363.46 +46.06	79.916 ±12.628	22.155 ±2.53	1051.38 ±106.46	2-6	235.16 ±50.39	37.373 ±1.95	10.66 ±1.50	518.61 ±77.687
a.	4-7	410.00	106.31	25.60	1196.25	3-4	355.00	39.68	16.00	877.50
b.	4-8	403.75	104.96	24.22	1292.50	2-4	323.75	56.00	16.00	762.50
c.	6-8	369.95	103.38	25.142	1207.50	3-5	341.25	55.04	16.00	962.25
	4-8	394.55 ±17.59	104.88 ±1.19	24.98 ±0.57	1232.08 ±42.96	2-5	340.00 ±12.788	50.24 ±7.477	16.00	855.40 ±68.65
a.	4-6	160.00	48.71	12.80	401.25	2-4	181.25	33.60	9.60	371.25
b.	3-4	205.00	46.08	14.40	466.25	-	-	-	-	-
c.	4-5	130.00	46.72	13.33	335.00	-	-	-	-	-
	3-6	165.00 ±30.82	47.17 ±1.119	13.51 ±0.665	400.83 ±53.58	2-4	181.25 ±36.76	33.60 ±4.35	9.60	371.25 ±65.44

APPENDIX II (continued)

	SPOROCARP HAIRS					LEAF HAIRS				
	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total hair length μ	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total hair length
.	6-8	340.00	77.08	25.60	1390.00	2-4	298.65	37.68	9.60	681.05
.	7-8	351.25	93.58	34.48	1289.95	2	234.375	33.06	9.60	437.50
.	5-6	462.50	68.26	38.40	1520.83	2-4	148.611	37.28	9.60	413.88
	5-8	444.583 ±79.09	79.64 ±10.49	32.82 ±5.35	1400.25 ±94.53	2-4	227.21 ±61.46	36.00 ±2.08	9.60	510.81 ±120.763
.	4	192.50	44.80	12.80	387.50	3	216.50	39.52	16.00	411.50
.	5-7	278.00	79.94	19.20	691.40	2-5	251.785	48.45	15.466	544.61
.	2-6	243.75	52.16	16.00	590.00	2-4	288.75	28.48	12.80	711.25
.	4-7	216.25	67.50	16.00	592.50	-	-	-	-	-
	2-7	232.629 ±31.862	61.10 ±13.615	16.00 ±2.26	565.10 ±110.24	2-5	252.35 ±29.49	38.81 ±8.16	14.75 ±1.399	555.79 ±122.62
	4	525.00	92.80	16.00	737.50	2	116.25 ±14.84	42.88 ±13.72	9.60	210.00
.	3-4	212.50	52.26	16.00	480.00	2-3	210.00	43.53	9.60	492.50
.	4	229.16	64.40	12.80	481.94	2-3	165.00	54.30	12.80	412.44
.	3-5	213.75	51.52	12.80	487.50	2-3	172.00	34.56	9.60	410.00
	3-5	218.47 ± 7.57	56.06 ±5.60	13.866 ±1.50	483.146 ± 3.178	2-3	182.50 ±19.685	44.13 ±8.01	10.60 ±1.50	438.31 ±38.32
	4-6	238.75	65.28	16.00	673.75	2-3	237.50 +30.33	43.73 +16.88	9.60	377.08 +48.23

APPENDIX II (continued)

	SPOROCARP HAIRS					LEAF HAIRS				
E.	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total hair length μ	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total root length μ
a.	4-5	220.00	55.82	16.00	515.02	2-3	193.75	35.16	9.60	353.50
b.	4-6	287.50	66.88	16.00	671.25	2-3	201.25	36.48	9.60	430.00
c.	3-6	311.25	67.20	16.00	766.25	-	-	-	-	-
	3-6	272.91 ±38.65	63.30 ±5.29	16.00	650.84 ±103.57	2-3	197.50 ± 3.75	35.82 ±0.68	9.60	391.25 ±38.75
a.	6-9	350.00	94.00	28.80	1218.75	2-3	267.50	34.48	9.60	538.70
b.	4-7	588.75	73.95	22.85	1743.25	2-3	437.50	30.40	9.60	912.50
c.	4-8	562.50	73.92	27.73	1580.00	2-3	224.31	45.09	9.60	461.36
d.	4-6	334.95	79.40	23.04	1001.25	2-4	246.25	26.24	9.60	506.75
e.	4-6	376.20	114.46	24.80	1094.25	3-4	243.70	46.70	9.60	574.75
	4-9	442.48 ±109.82	87.146 ±15.50	25.44 ±2.425	1327.50 ±286.10	2-4	287.85 ±75.35	36.58 ±8.05	9.60	599.01 ±161.19
a.	5-6	260.00	54.72	19.20	630.00	3-5	327.50	23.40	9.60	962.25
b.	4-6	227.50	58.24	16.00	568.75	2	203.75	32.00	9.60	362.50
	4-6	243.75 ±16.25	56.48 ±1.76	17.60 ±1.60	599.38 ±30.60	2-5	265.63 ±61.88	27.70 ±4.30	9.60	662.38 ±299.88
a.	3-4	253.75	72.32	13.25	538.75	2	142.50	27.52	9.60	262.50
b.	3-5	283.75	70.40	12.80	716.25	2	147.73	27.05	9.60	263.63
c.	3-4	325.50	60.16	12.80	748.75	2-3	143.05	45.86	9.60	303.94
	3-5	287.66 ±29.42	67.63 ±5.33	12.95 ±0.21	667.92 ±92.29	2-3	144.43 ± 2.35	33.48 ±8.76	9.60	276.69 ±19.27

APPENDIX II (continued)

SPOROCARP HAIRS					LEAF HAIRS				
No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total root length μ	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total root length μ
3-5	327.50	64.87	15.64	724.09	2-3	191.25	26.56	9.60	373.75
3-4	157.50	56.32	12.80	367.50	2-3	152.27	41.29	9.60	317.05
4-5	218.75	61.12	16.00	617.50	2	188.75	27.20	9.60	371.25
3-5	234.58 ±70.299	60.77 ±3.49	14.13 ±1.43	569.69 ±149.45	2-3	177.42 ±17.81	31.68 ±6.79	9.60	340.015 ±26.16
3-5	105.00	38.72	9.60	290.00	2-3	141.66	26.16	9.60	312.50
3	112.50	45.74	9.60	265.00		-	-	-	-
3-4	129.17	45.86	9.60	300.00		-	-	-	-
3-5	115.55 ±10.09	43.44 ±3.33	9.60	285.100 ±14.72	2-3	141.66	26.16	9.60	312.56
3-5	208.75	60.80	16.00	573.75	2	133.33	23.46	9.60	291.66
4-5	218.75	62.04	16.00	642.50	-	-	-	-	-
3-5	213.75 ± 5.00	61.42 ±0.62	16.00	608.13 ±34.75	2	133.33	23.46	9.60	291.66
5-6	490.00	85.76	22.40	1247.5	2-3	327.50	41.25	12.48	613.75
4-8	403.75	76.48	18.13	1173.75	2-5	299.50	48.32	9.60	838.70
4-8	446.875 ±43.13	81.12 ±4.64	20.27 ±2.13	1210.62 ±36.87	2-5	313.50 ±14.00	44.79 ±3.54	11.40 ±1.44	726.23 ±112.48

APPENDIX II (continued)

	SPOROCARP HAIRS					LEAF HAIRS				
f. .	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total root length μ	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total hair length μ
a.	5-7	371.25	61.12	19.20	1452.50	2-4	236.25	35.52	12.80	640.00
b.	5-8	412.50	67.84	23.46	1727.50	4-5	233.75	31.64	9.60	912.00
c.	3-6	370.00	51.82	19.20	1311.25	3-4	288.75	32.96	9.60	815.00
	3-8	384.58 ±19.75	60.26 ±6.57	20.62 ±2.01	1497.08 ±172.83		252.91 ±25.35	33.33 ±1.61	10.66 ±1.50	789.00 ±112.55
a.	3-4	308.75	69.76	22.40	812.50	-	-	-	-	-
b.	4-5	432.50	77.44	16.00	1182.05	-	-	-	-	-
c.	3-4	482.50	71.04	16.00	1127.50	-	-	-	-	-
	3-5	407.92 ±73.03	72.13 ±3.35	18.13 ±3.02	1040.68 ±162.87					
a.	6-8	307.50	76.04	16.00	1351.25	2-5	205.00	36.80	9.60	561.25
b.	4-5	237.50	57.28	16.00	586.25	2-4	240.00	30.08	9.60	562.50
c.	3-5	188.75	57.92	16.00	436.25	2-3	200.00	36.95	11.60	362.50
	3-8	244.58 ±48.73	63.75 ±8.69	16.00	791.25 ±400.10	2-5	215.00 ±17.79	34.61 ±3.20	10.26 ±0.94	495.42 ±93.98
a.	6-8	626.25	132.04	22.4	2108.75	3-5	317.50	80.08	13.56	913.75
b.	8-11	310.00	117.76	28.8	2247.50	-	-	-	-	-
c.	4-6	529.17	218.98	22.4	1445.83	-	-	-	-	-
	4-11	488.47 ±132.28	156.25 ±44.73	24.53 ±3.02	1934.03 ±349.82	3-5	317.50	80.08	13.56	913.75

APPENDIX II (continued)

SPOROCARP HAIRS					LEAF HAIRS				
No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total root length μ	No. of cells per hair	PA_l (μ)	PA_w (μ)	Stalk dia. (μ)	Total root length μ
6-8	478.00	96.96	23.11	2078.75	3-8	250.00	37.76	11.20	1240.80
6-8	550.00	74.44	20.27	1671.25	2-5	303.75	27.20	12.80	977.50
5-8	652.45	115.93	25.07	2288.75	3-5	518.75	31.36	12.80	1546.25
5-8	559.15 ±72.73	96.78 ±15.72	22.81 ±1.97	2012.92 ±256.35	2-8	357.50 ±116.11	32.10 ±4.34	12.26 ±0.75	1254.85 ±232.40
8-10	356.25	127.04	36.62	2537.5	2-5	237.50	39.82	12.16	626.25 ±144.96
4-8	322.50	104.00	29.60	1648.75	4-6	515.00	60.48	16.00	2082.5
5-8	305.00	85.70	24.4	1307.5	3-6	402.50	76.8	16.94	1557.50
6-8	307.50	93.76	27.20	1388.75	2-6	450.00	65.28	17.92	1761.25
3-4	182.5	61.12	12.8	380.0	-	-	-	-	-
3-6	196.56	55.62	16.00	622.65	3-4	182.5	35.2	-	588.00
4-6	256.25	48.32	12.80	692.50	2-3	235.00	32.96	9.60	445.00

to abbreviations: PA_l = proximal arm length; PA_w = proximal arm width

Plate 1. *Marsilea capensis* A. Braun

a) Leaf hairs showing the dorsal surfaces. x 200

PLATE 1

1a

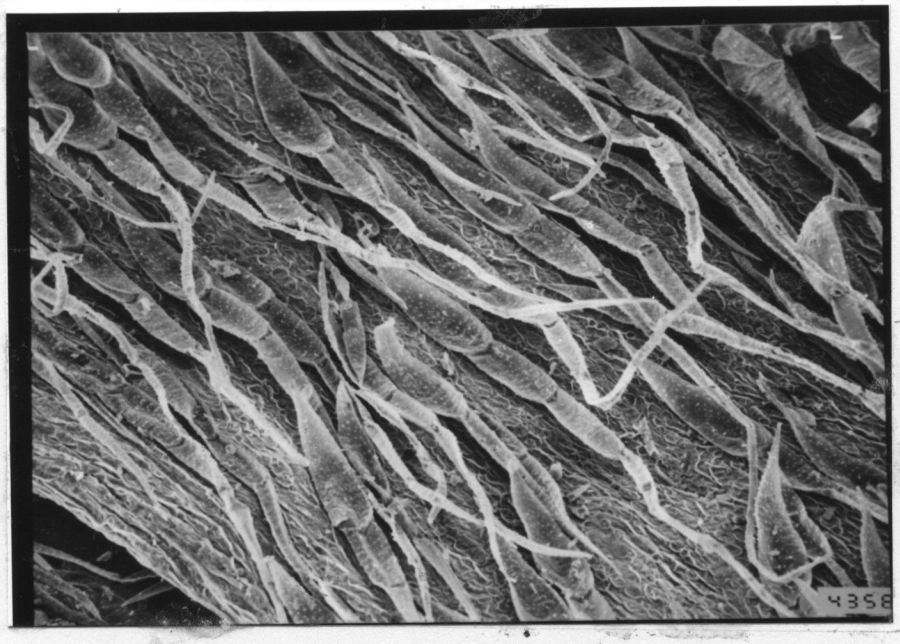
Plate 2. *Marsilea vera* Launert

a) Sporocarp hairs. x 75

b) Leaf hairs. Note the lingulate form of the proximal arm, and the moniliform nature of the distal arm. x 100

PLATE 2

2a



2b

Plate 3. *Marsilea villifolia* Bremekamp & Obermeyer ex Alston & Schelpe

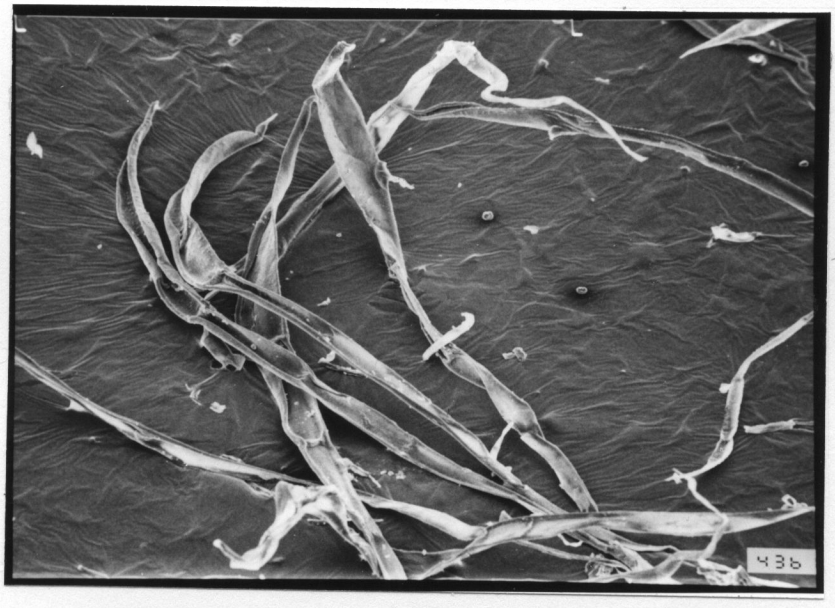
a) Sporocarp hairs. x 100

b) Sporocarp hairs ventral view. x 75

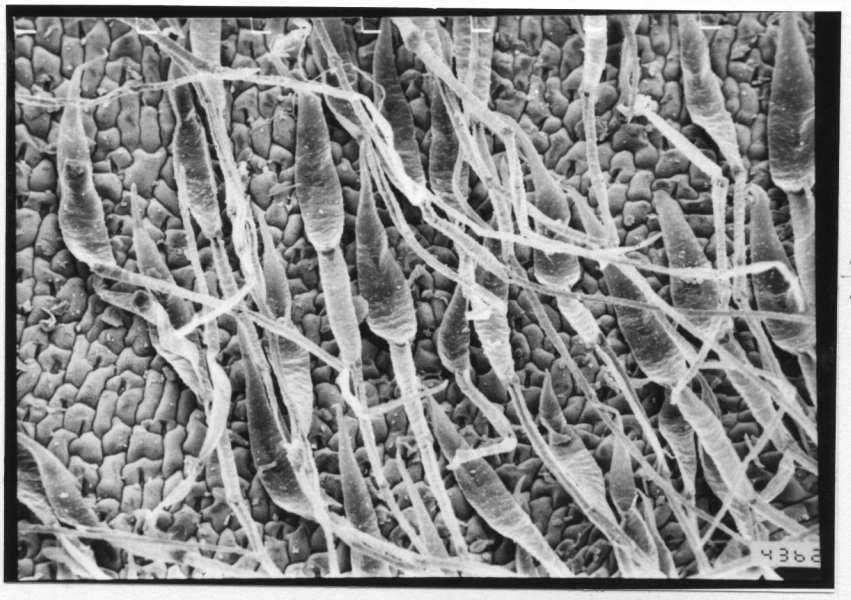
c) Leaf hairs. Note characteristic tailing of distal arm. x 150

PLATE 3

3a



3b



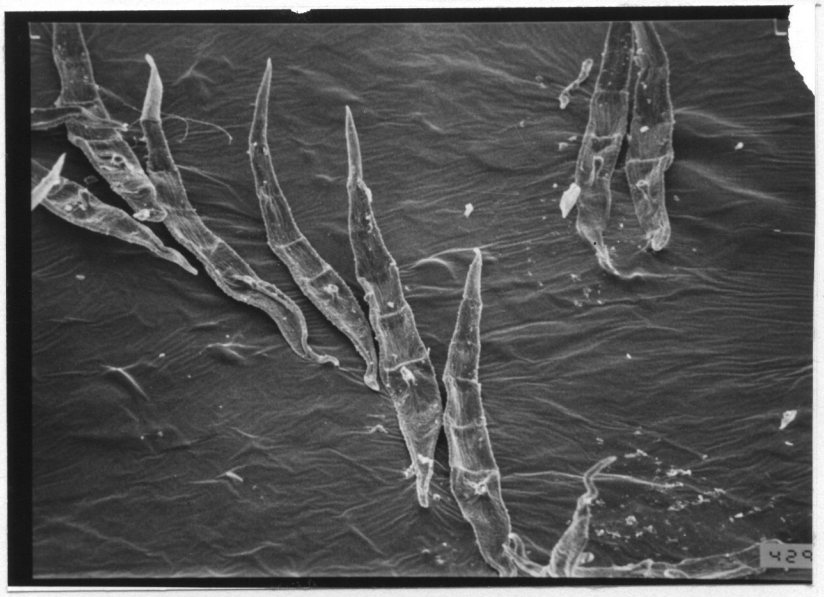
3c

Plate 4. *Marsilea aegyptiaca* Willdenow

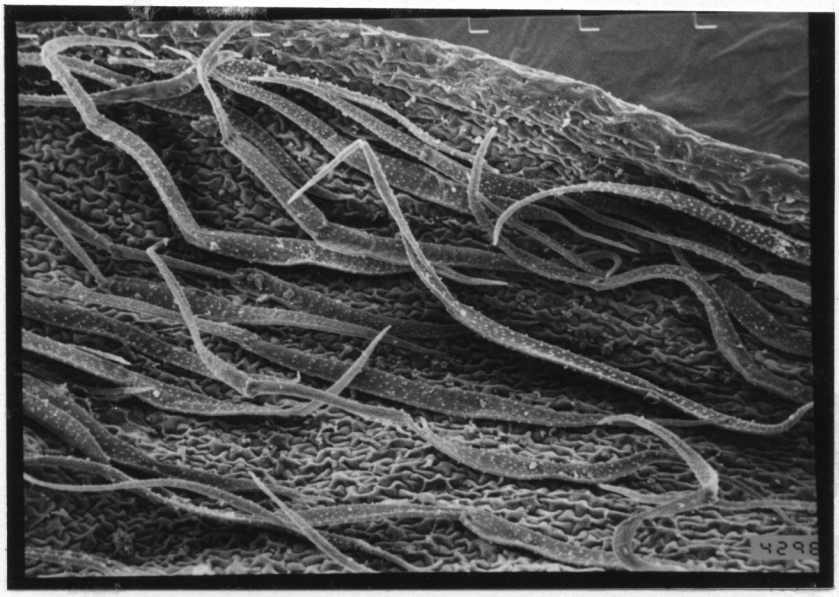
a) Ventral view of sporocarp hairs. Note transverse lines of abjunction. x 100

b) Leaf hairs. Note that adpressed proximal arm and raised distal arm. x 150

PLATE 4



4a



4b

Plate 5. *Marsilea minuta* var. *minuta* (L.) Launert

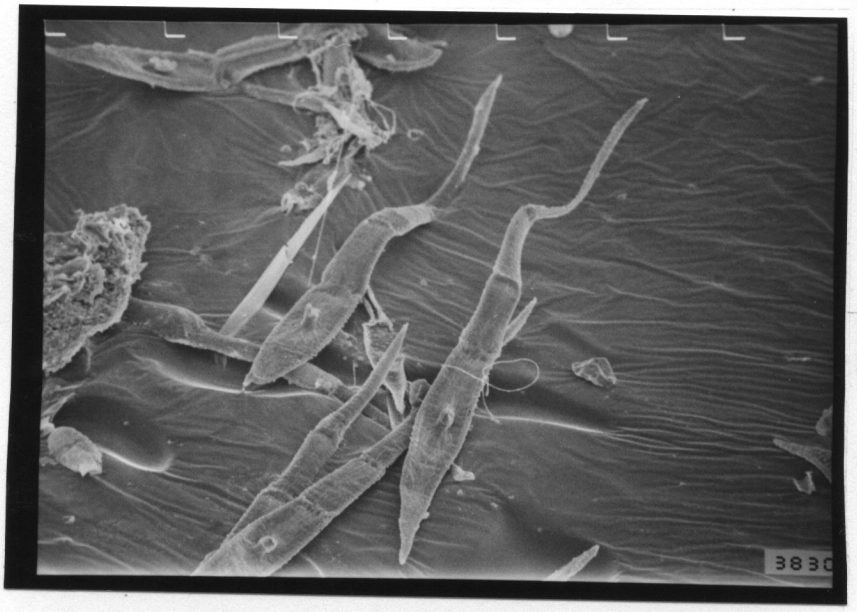
a) Sporocarp hairs with entire length appressed to the surface. x 200

b) Ventral view of sporocarp hairs. x 150

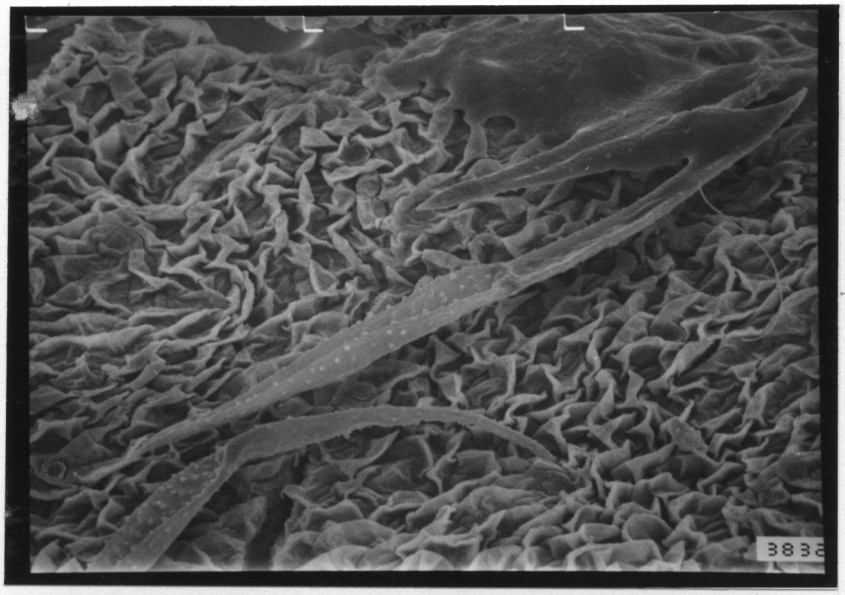
c) Leaf hairs. Note sculpturing of leaf surface. x 350

PLATE 5

5a



5b



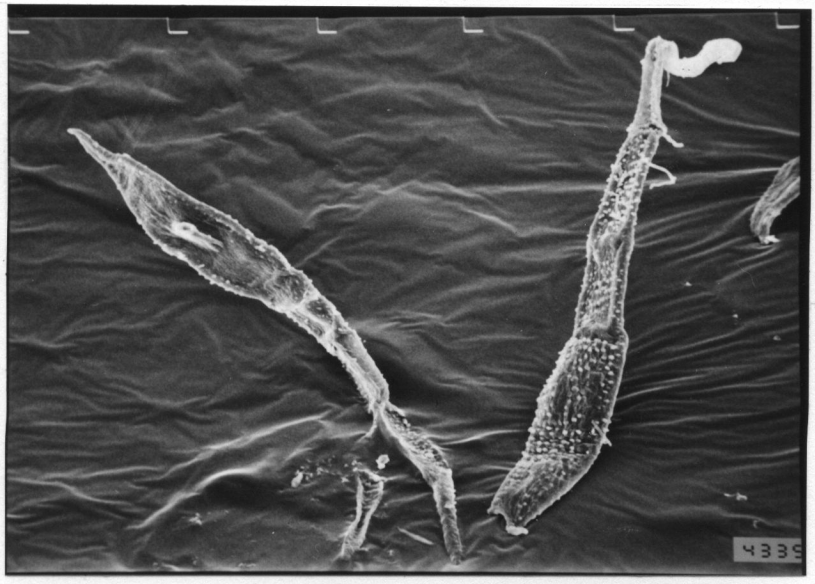
5c

Plate 6. *Marsilea botryocarpa* F. Ballard

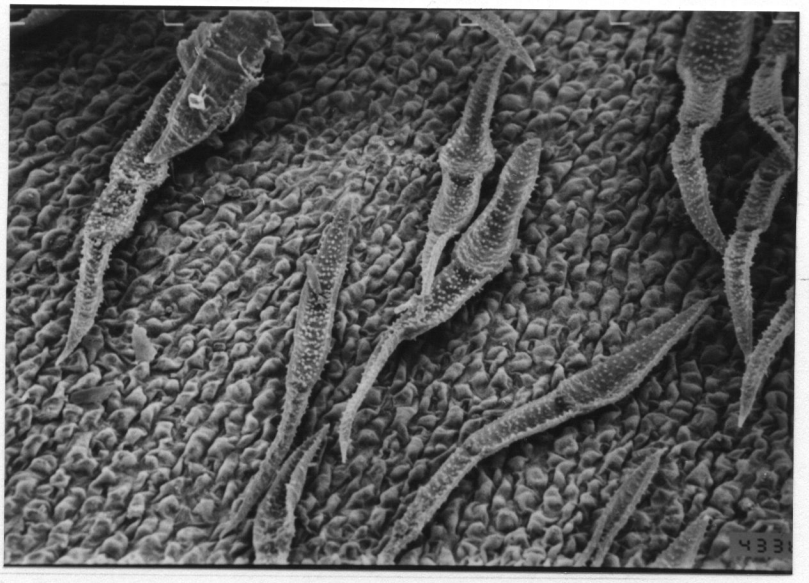
a) Sporocarp hairs. Note numerous papillae on dorsal face of hair on the right, and much less papillae concentrated on edges of ventral face of hair on the left. x 200

b) Leaf hairs. Note lack of papillae on ventral face of an upturned proximal arm of hair on left top corner.

PLATE 6



6a



6b

Plate 7. *Marsilea megalomanica* Launert

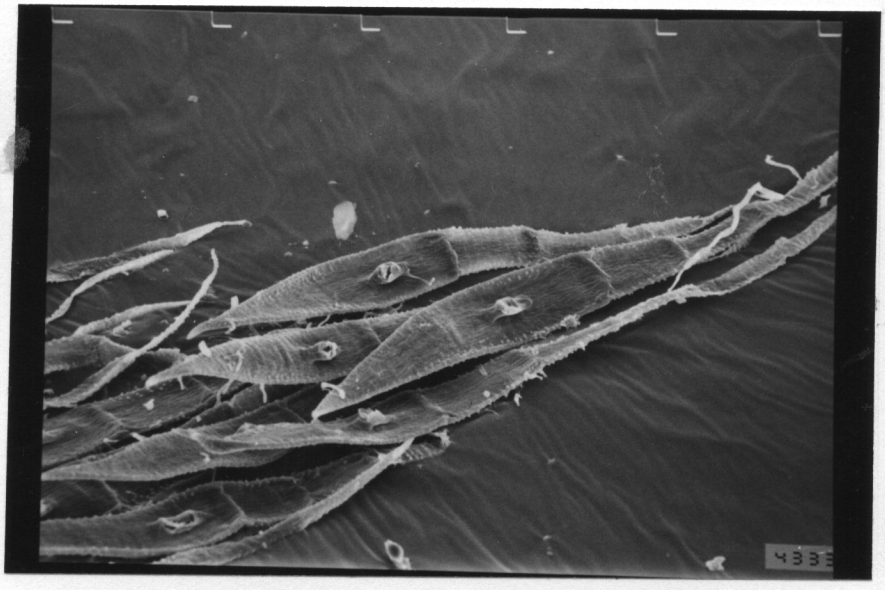
a) Sporocarp hairs, dorsal view. x 150

b) Ventral view of sporocarp hairs. x 200

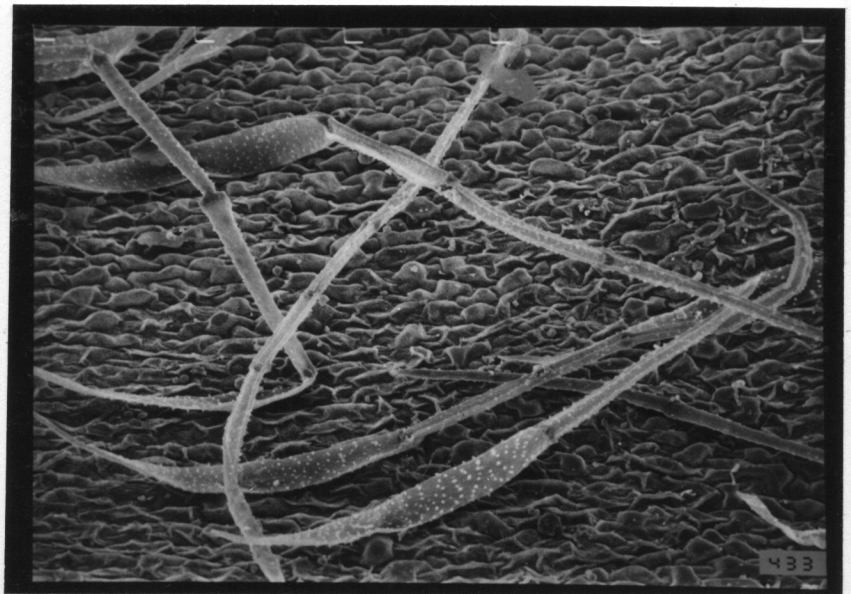
c) Leaf hairs. x 200

PLATE 7

7a



7b



7c

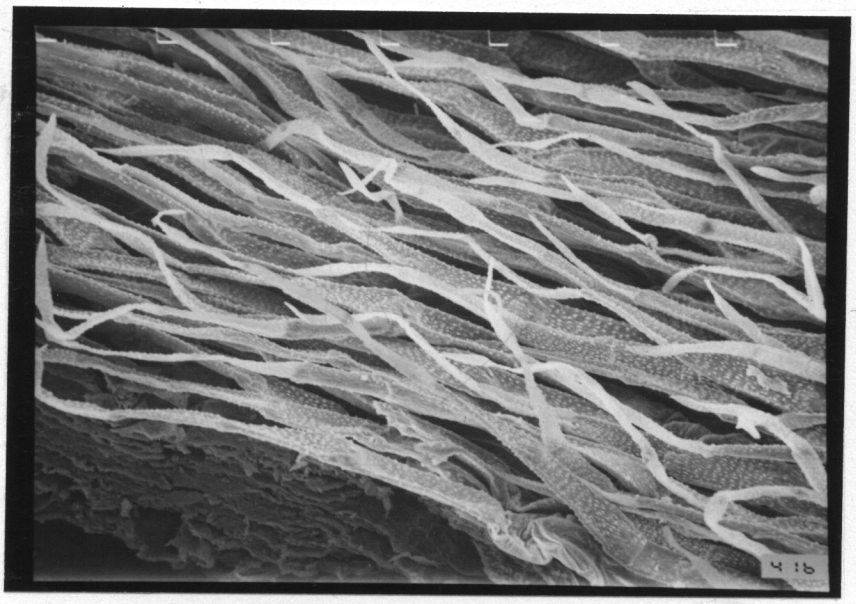
Plate 8. *Marsilea macrocarpa* Presl

a) Sporocarp hairs, dorsal view. x 150

b) Sporocarp hair, ventral view. x 200

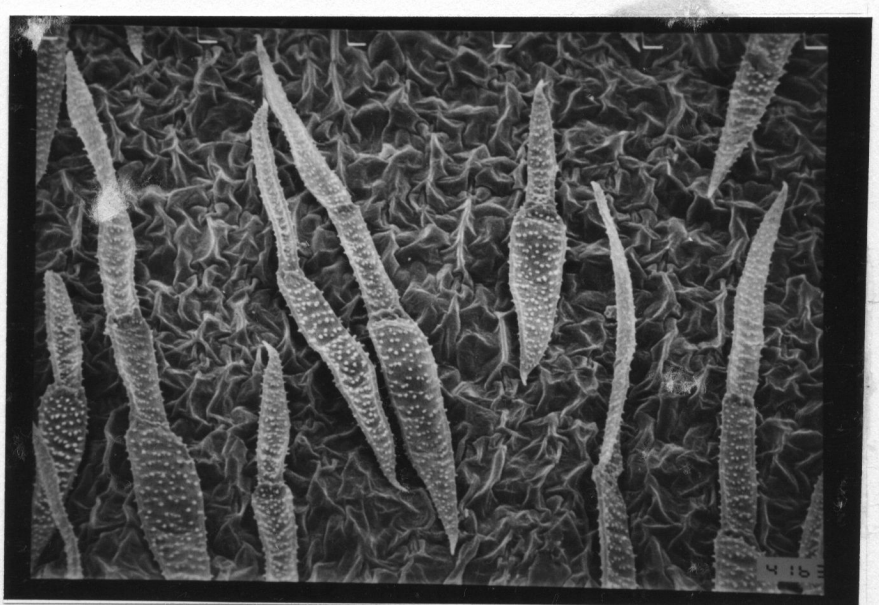
c) Leaf hairs. Note variation in number of cells per hair in the range of 2-4. x 200

PLATE 8



8a

8b



8c

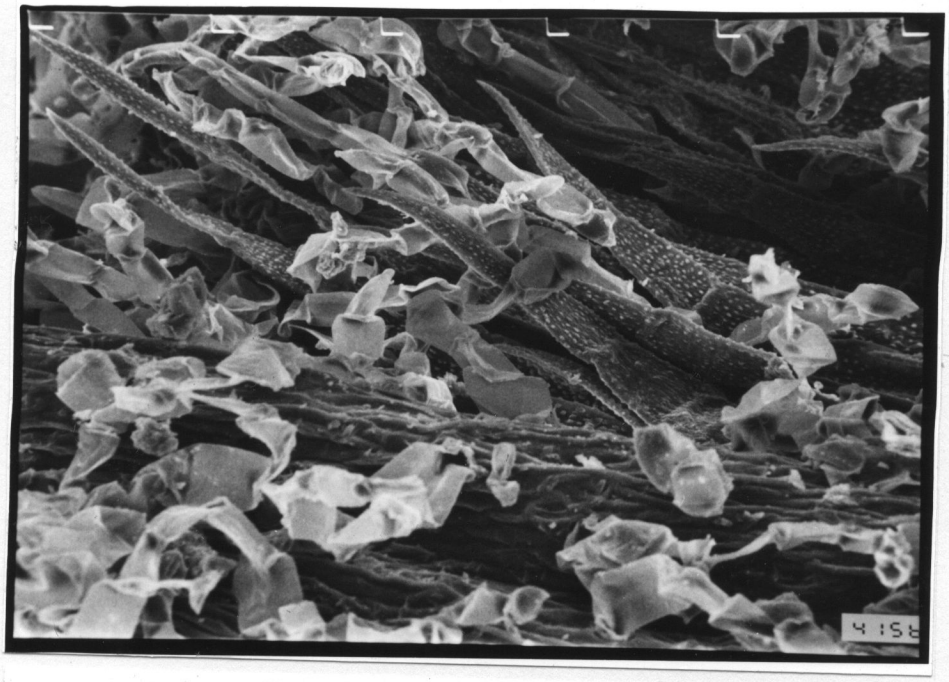
Plate 9. *Marsilea farinosa* Launert

a) Sporocarp hairs. Note presence of two hair kinds, the anvil-shaped and the simple form. x 200

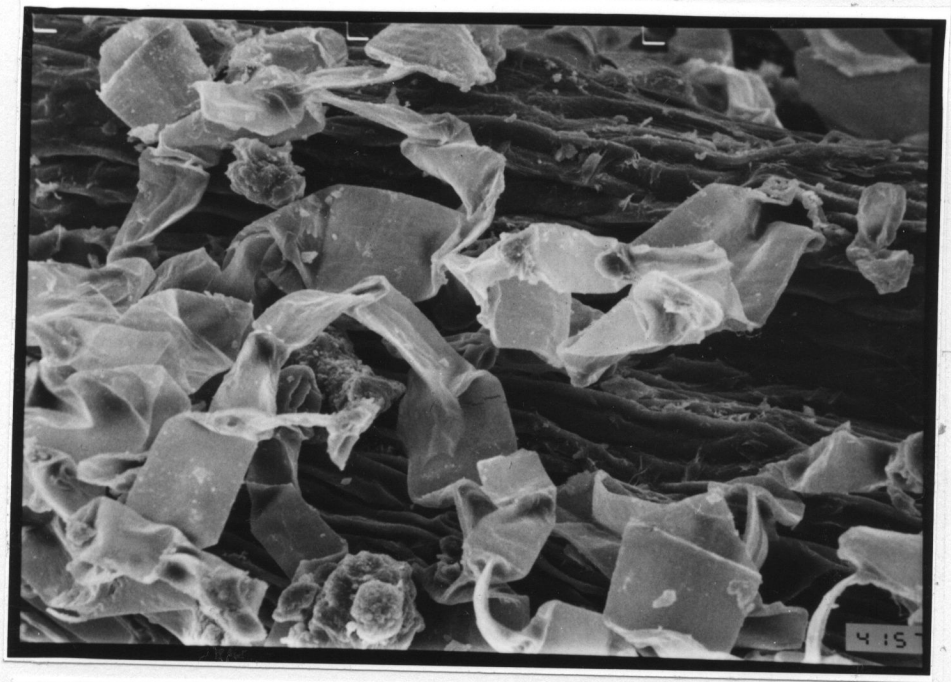
b) Sporocarp hairs, showing the simple forms only. Note the collapsed state of this wavy hair, and complete lack of papillae. x 350

c) Simple hairs on leaf surface. Note mode of contact to epidermis. x 350

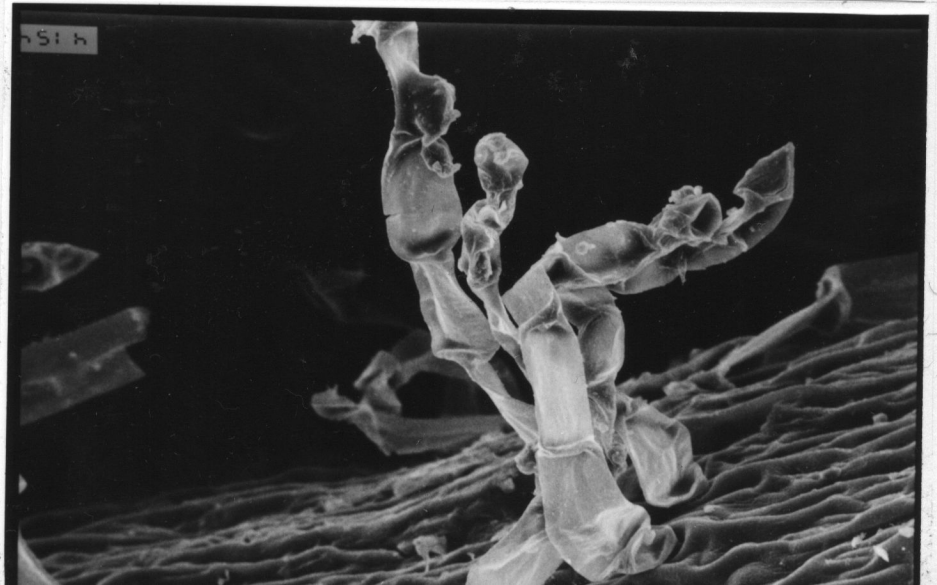
PLATE 9



9a



9b



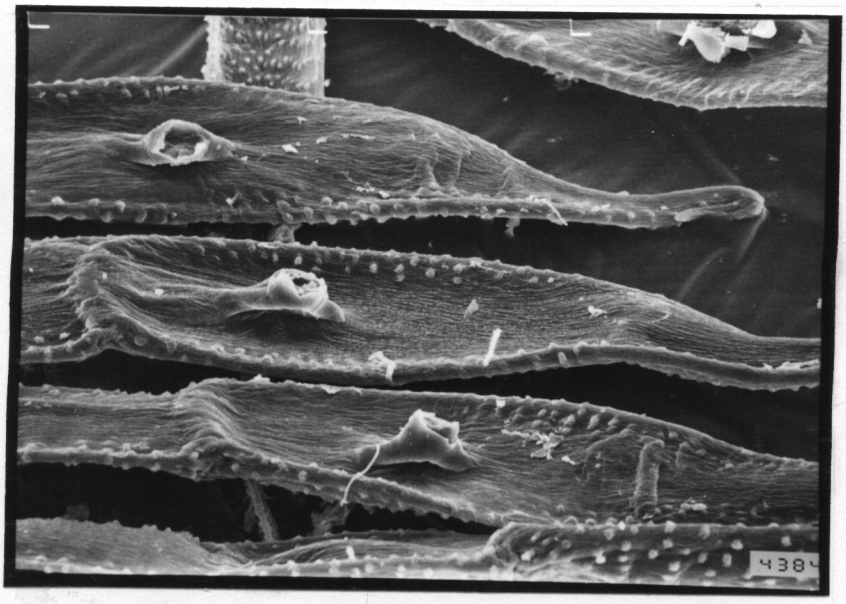
9c

Plate 10. *Marsilea ethiopica* Launert

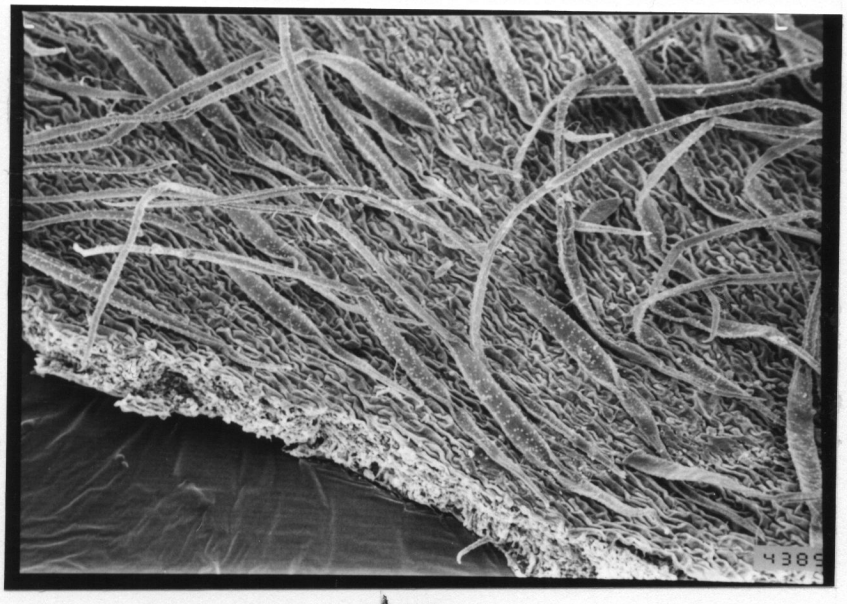
a) Ventral view of proximal arms.
Note the projected contact face.
x 350

b) Leaf hairs. x 100

PLATE 10



10a

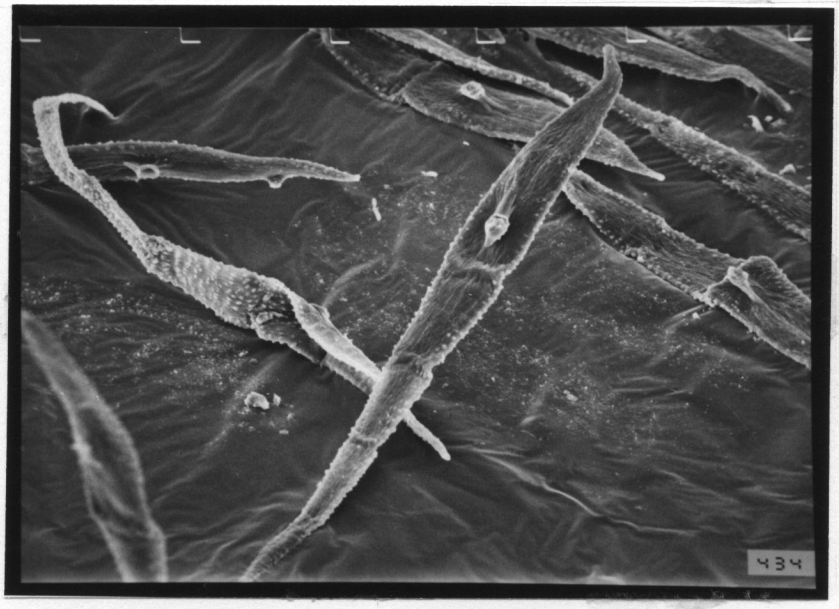


10b

Plate 11. *Marsilea gibba* A. Braun

a) Sporocarp hairs showing ventral view and one hair showing dorsal face. X 200

PLATE 11



11a.

Plate 12. *Marsilea* spp.

a) Sporocarp hairs. Note characteristic tailing of the distal arm. x 50

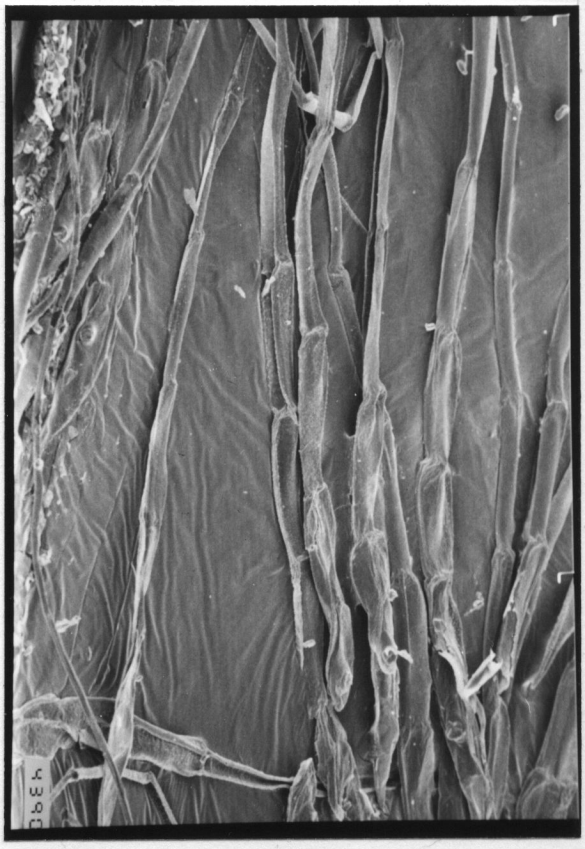
b) Ventral view of sporocarp hairs. x 75

c) Sporocarp surface. Note the sunken foot cell on top right corner. x 350

d) Leaf hairs. x 150

PLATE 12

12a



12b

Plate 13.

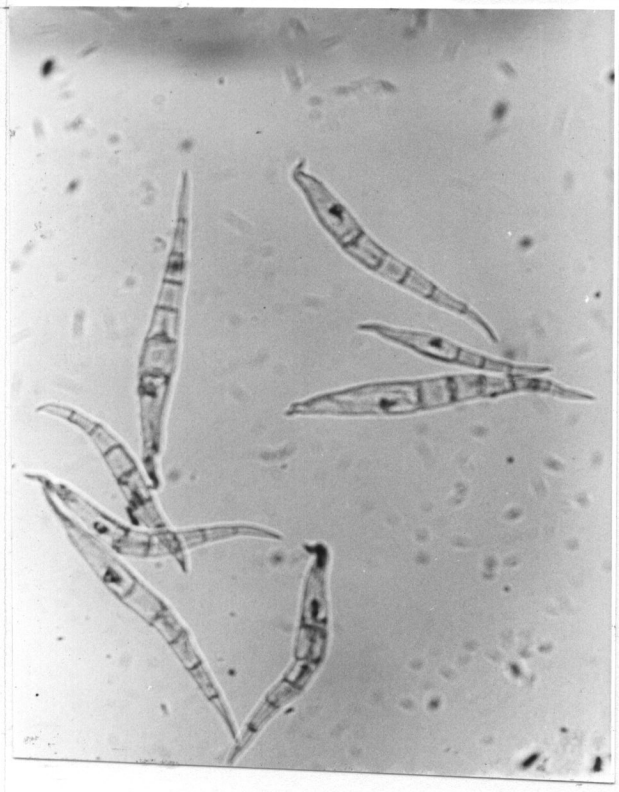
a) *Marsilea aegyptiaca* Willd.
Note hooked anterior end of sporocarp hairs. x 32

b) *Marsilea macrocarpa* Presl
Note the straight anterior end. x 32

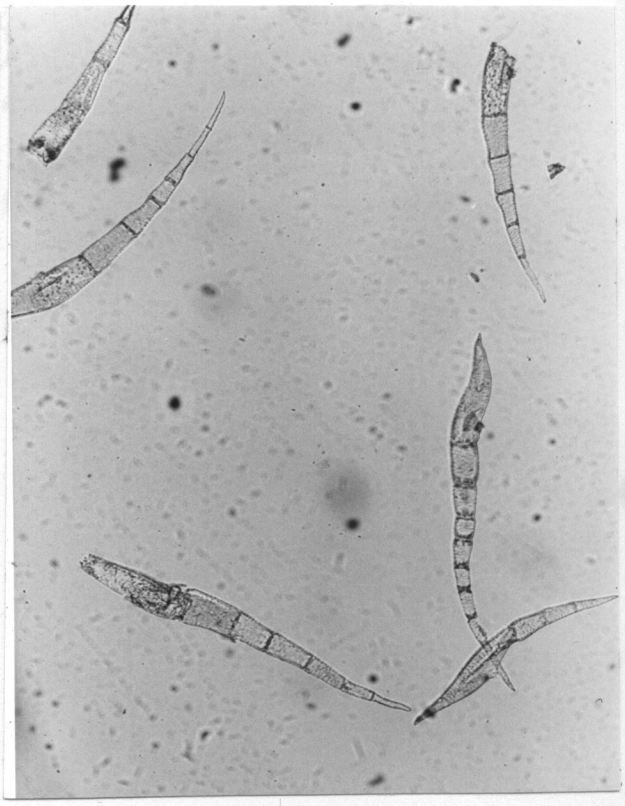
c) *Marsilea subterranea* Leprier ex A. Braun
Sporocarp hairs. x 32

d) *Marsilea unicornis* Launert
Sporocarp hairs. x 32

PLATE 13



13 a.



13 b.

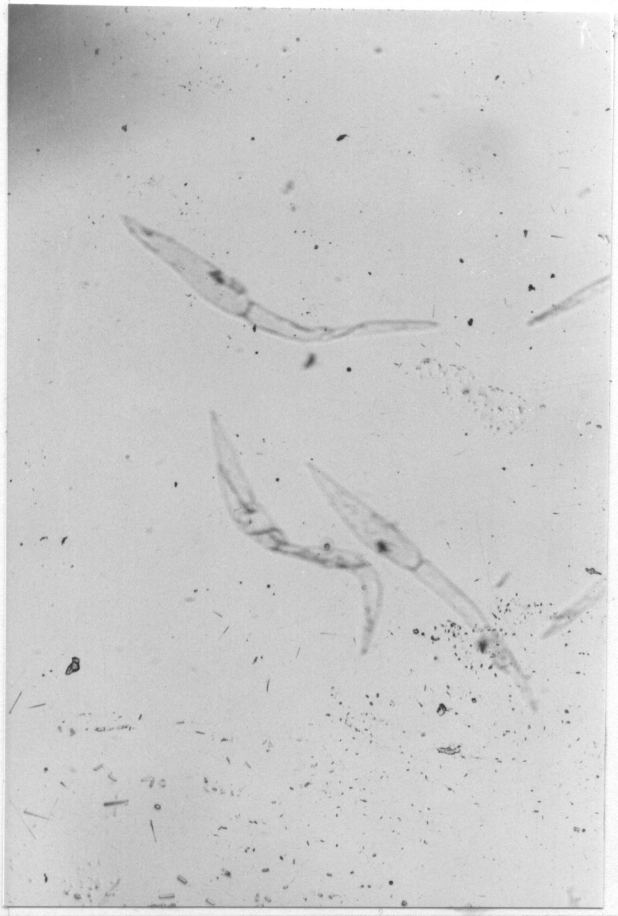


Plate 14.

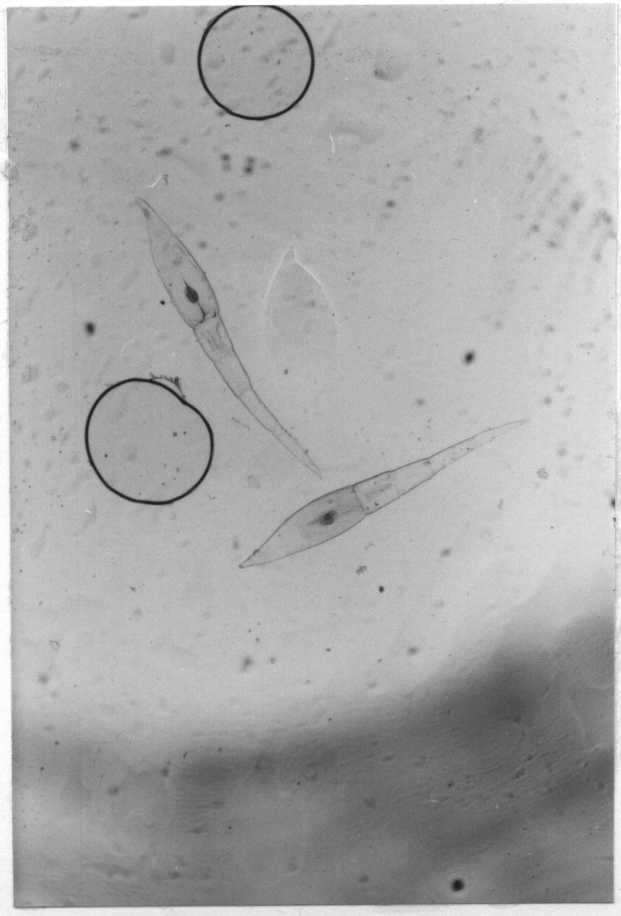
a) *Marsilea ephippiocarpa* Alston
Note gradual tapering of the sporocarp hair. x 32

b) *Marsilea apposita* Launert
Sporocarp hairs. x 32

c) *Marsilea botryocarpa* F. Ballard
Sporocarp hairs. x 32

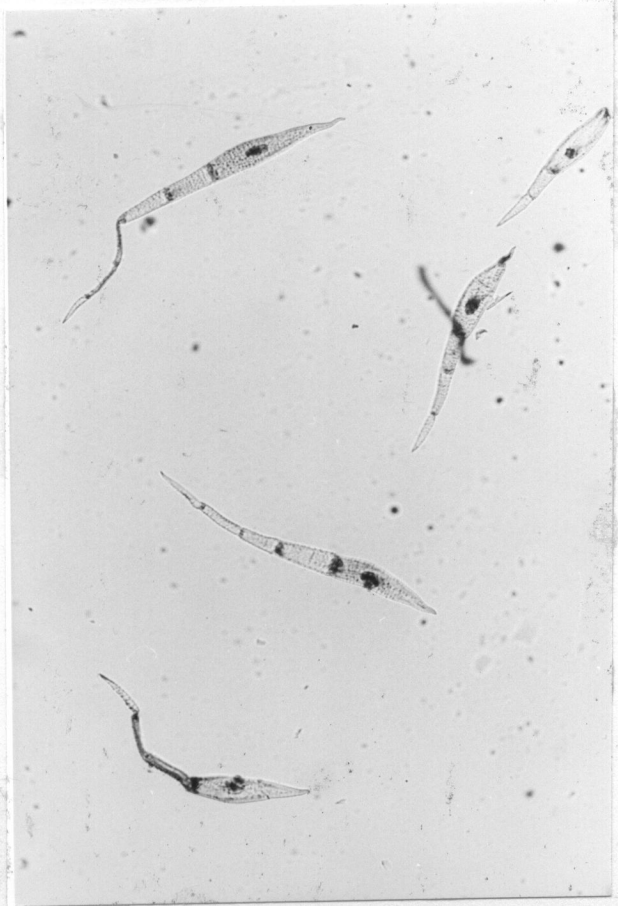
d) *Marsilea burchellii* A. Braun
Sporocarp hairs. x 32

PLATE 14



14a

14b



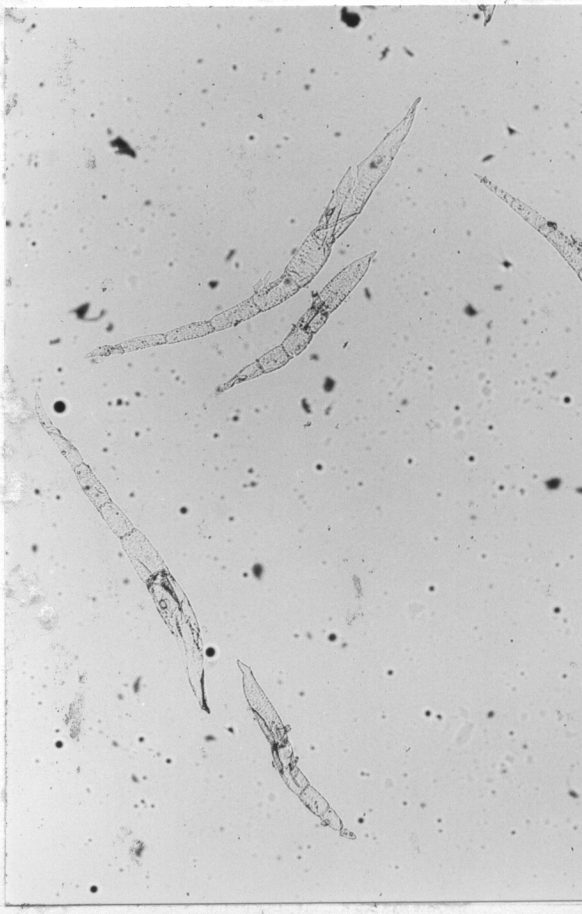


Plate 15.

a) *Marsilea fadeniana* Launert

Sporocarp hairs. x 32

b) *Marsilea nubica* var. *nubica* (A. Braun) Launert

Sporocarp hairs. The L-shaped structure is an artefact. x 32

c) *Marsilea minuta* var. *incurva* (L.) Launert

Sporocarp hairs. x 32

d) *Marsilea minuta* var. *minuta* (L.) Launert

Sporocarp hairs. x 32

PLATE 15

15a

15b

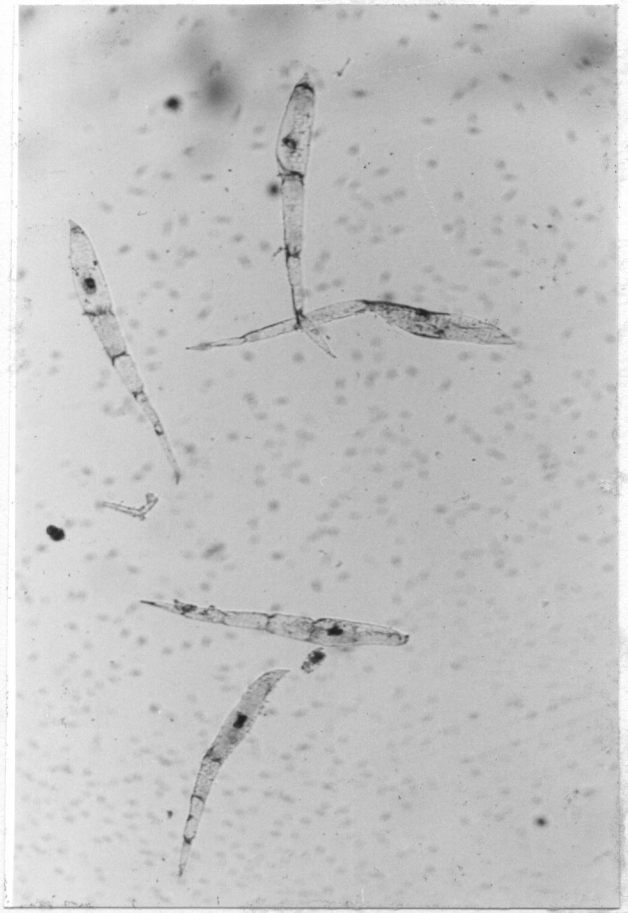


Plate 16.

a) *Marsilea farinosa* Launert

Leaf hairs. A simple form on the left and an anvil-shaped on the right. x 32

b) *Marsilea farinosa* Launert

Note symmetrical tapering of these sporocarp hairs. x 32

c) *Marsilea coromandelina* Willd.

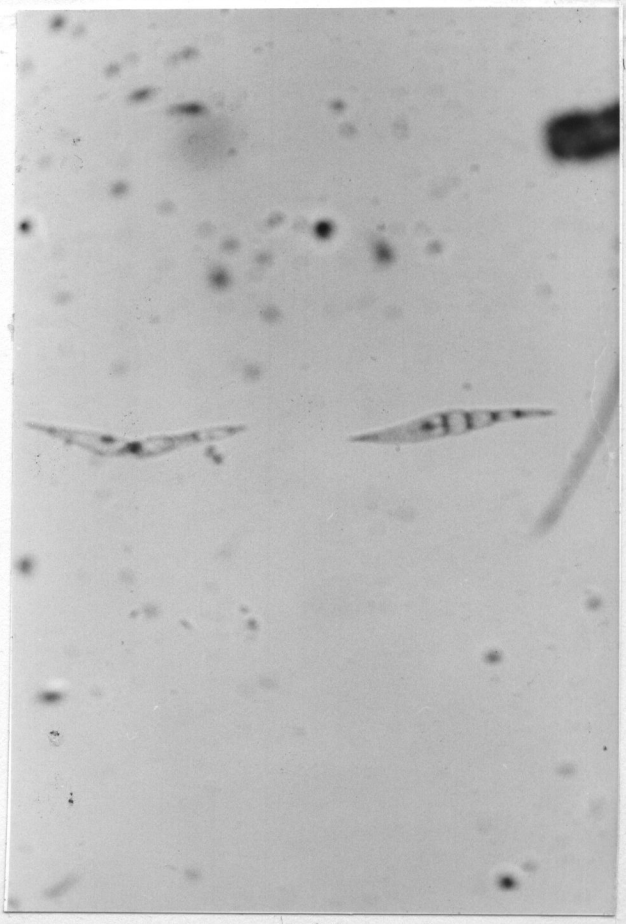
Sporocarp hairs. x 32

d) *Marsilea fenestrata* Launert

Sporocarp hairs. x 32

PLATE 16

16a



16b

Plate 17.

a) *Marsilea berhautii* Tardieu

Anterior end of sporocarp hair.

x 20

b) *Marsilea berhautii*

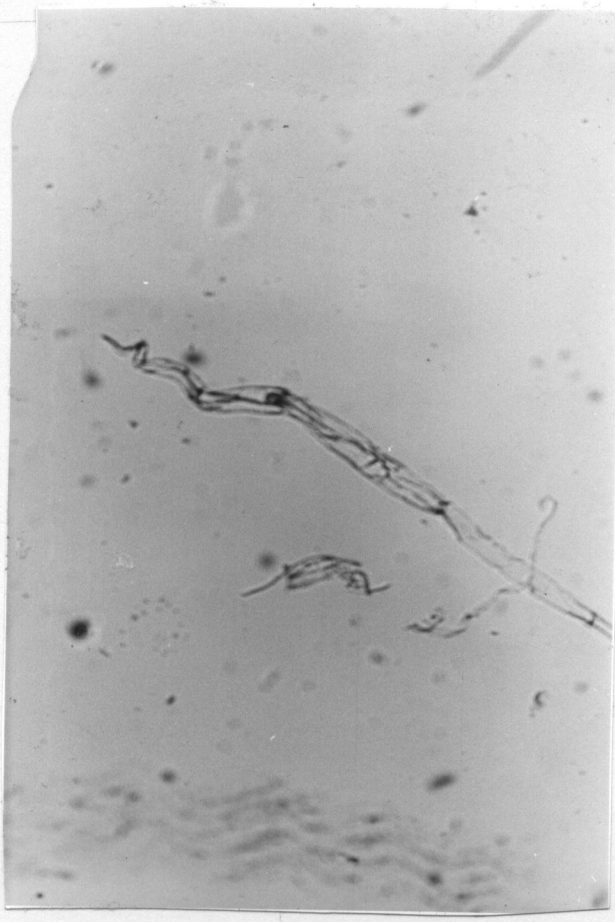
Posterior end of sporocarp hair shown in 17 a. Note the characteristic wavy hair.

x 20

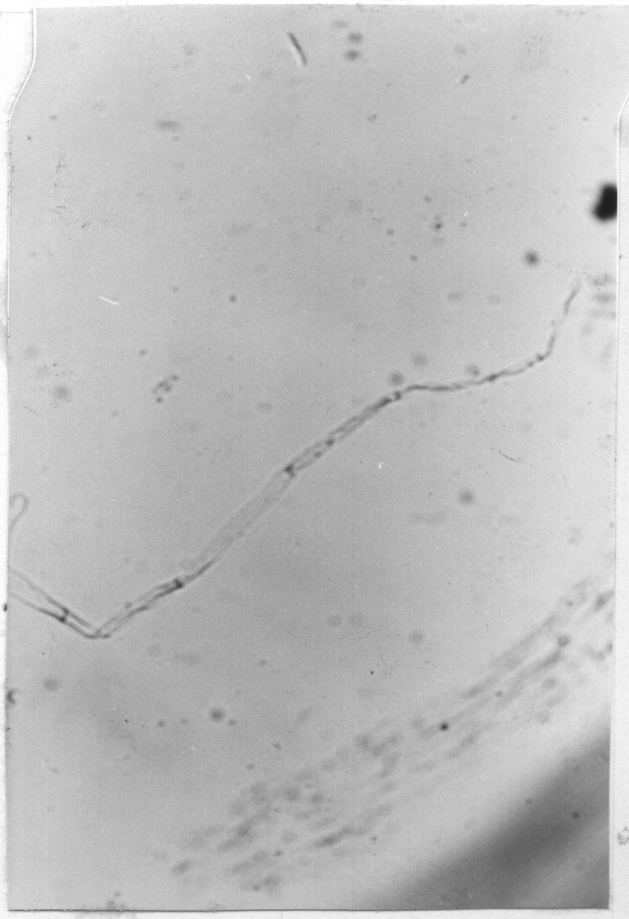
c) *Marsilea polycarpa*

Sporocarp hair. x 32

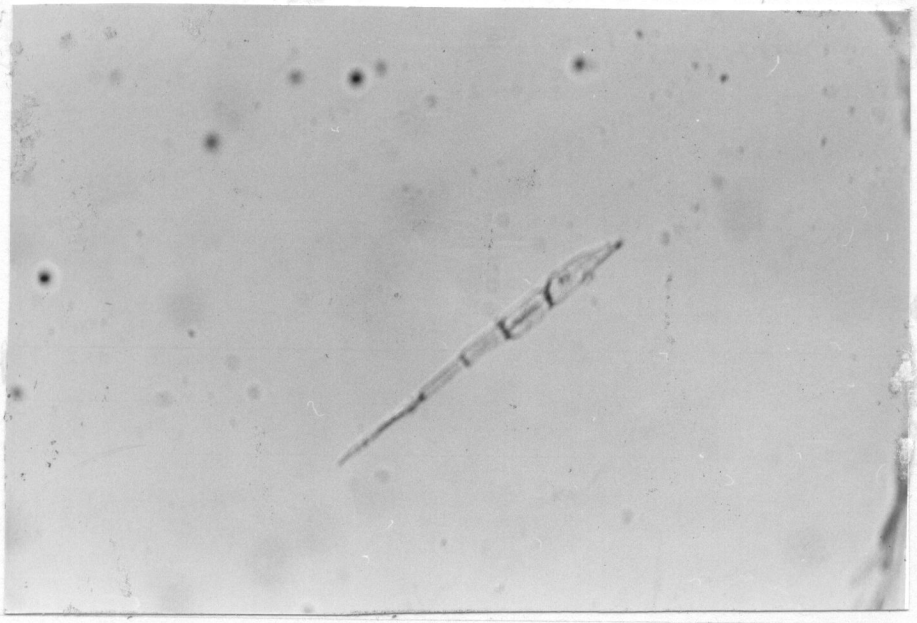
PLATE 17



17a



17b



17c

Plate 18.

a) *Marsilea villifolia* Bremekamp & Obermeyer ex Alston & Schelpe
Sporocarp hairs. Note prominently protruded contact face and conspicuous streaks of brownish-yellow substance. x 32

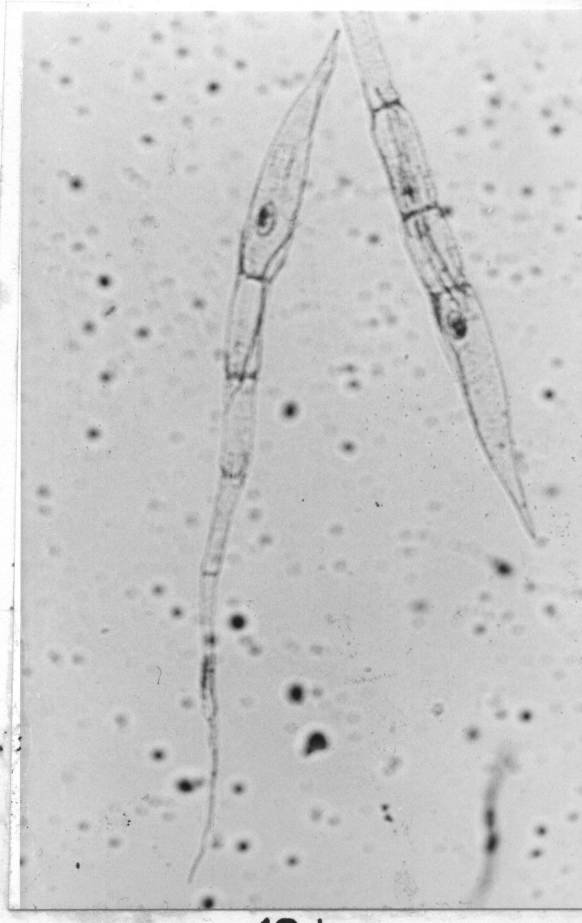
b) *Marsilea vera* Launert
Sporocarp hairs. x 32

c) *Marsilea schelpiana* Launert
Sporocarp hairs. x 32

d) *Marsilea stigosa* Willd.
Sporocarp hairs. x 32

PLATE 18

18a.



18b.

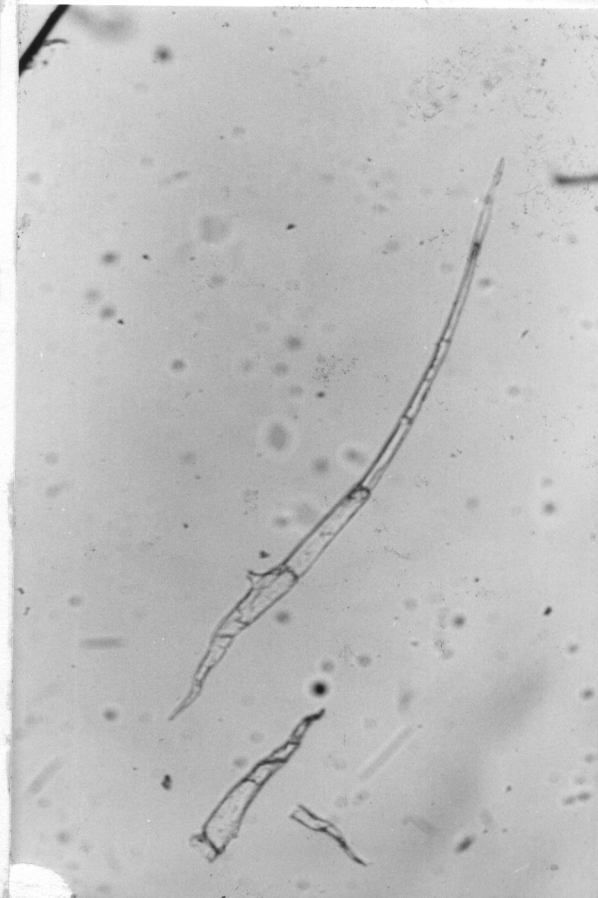


Plate 19.

a) *Marsilea megalomanica* Launert Sporocarp hairs. Note hooked anterior end. x 32

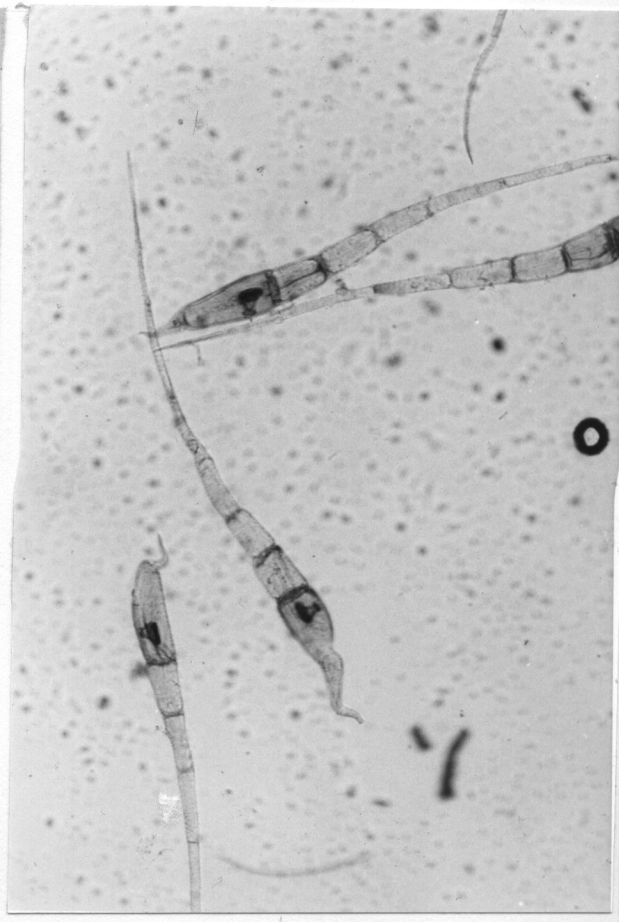
b) *Marsilea distorta* A. Braun Sporocarp hairs with hooked anterior end and tailing of distal arm. x 32

c) *Marsilea ethiopica* Launert Sporocarp hairs with tailing of distal arm, aduncate anterior end. x 32

d) *Marsilea capensis* A. Braun Sporocarp hairs. Note gradual tapering of distal arm. x 32

PLATE 19

19a



19b

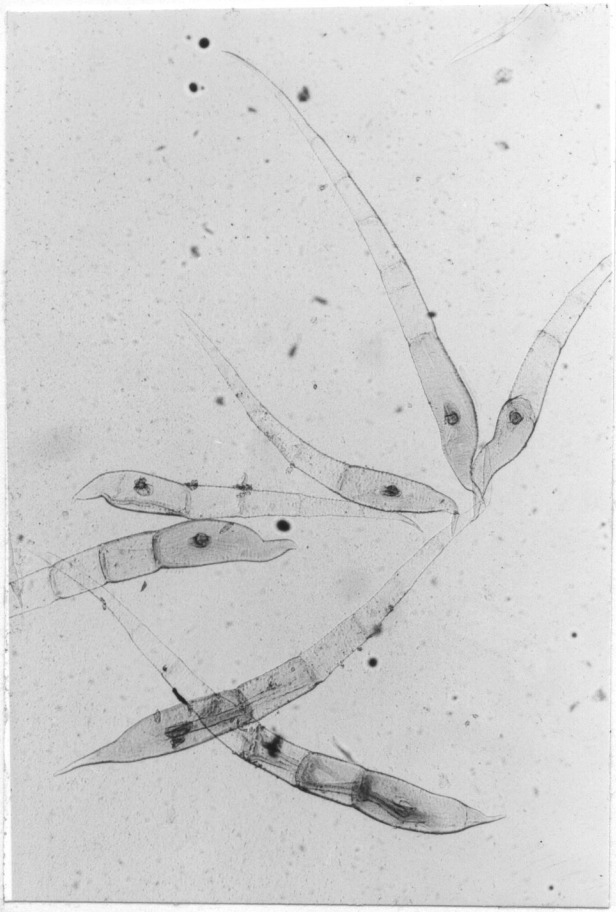


Plate 20.

a) *Marsilea* sp.

Note characteristic tailing of sporocarp hair. x 20

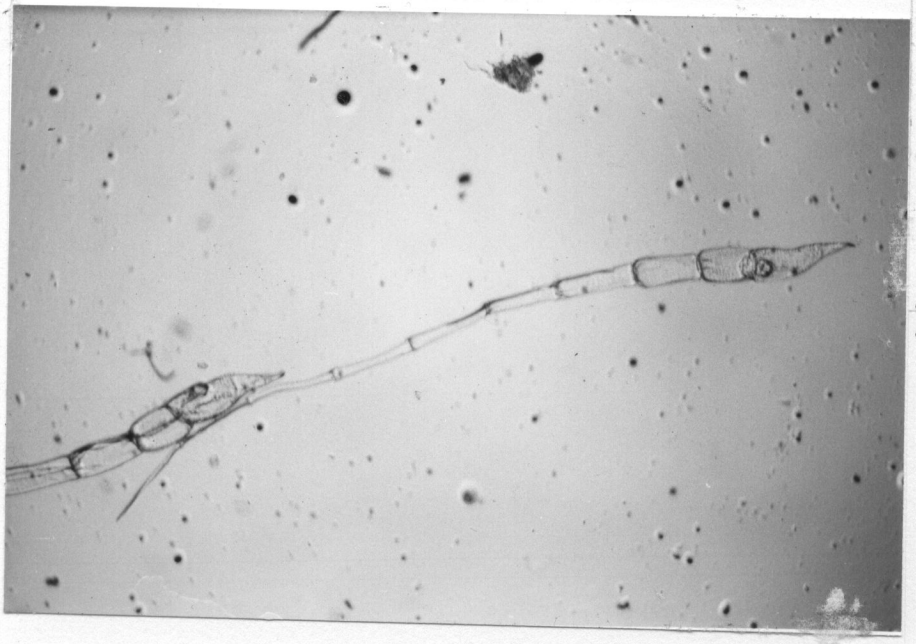
b) *Marsilea drummondii* A. Braun

Sporocarp hairs. Note some hair similarity of this Australian species to that of the hair of the Namibian species in 20 a. x 20

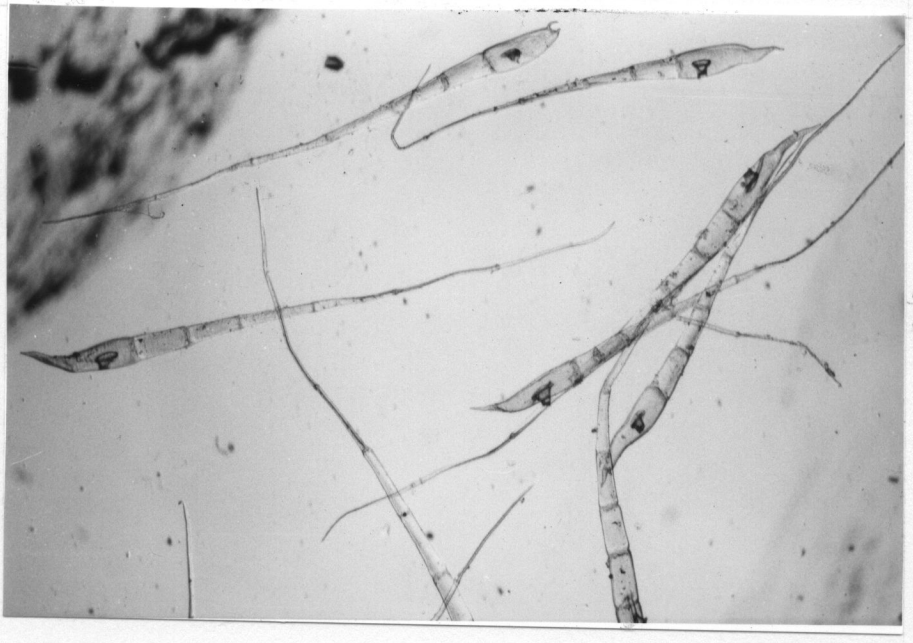
c) *Marsilea hirsuta*

Sporocarp hairs. Note the wide black band is a trapped air bubble. x 20

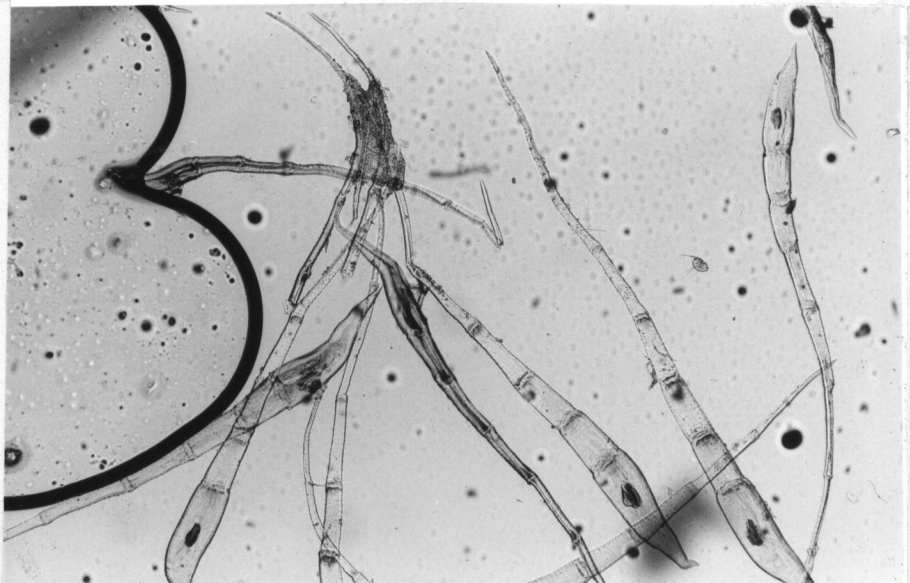
PLATE 20



20a



20b



20c

Plate 21. *Marsilea drummondii*

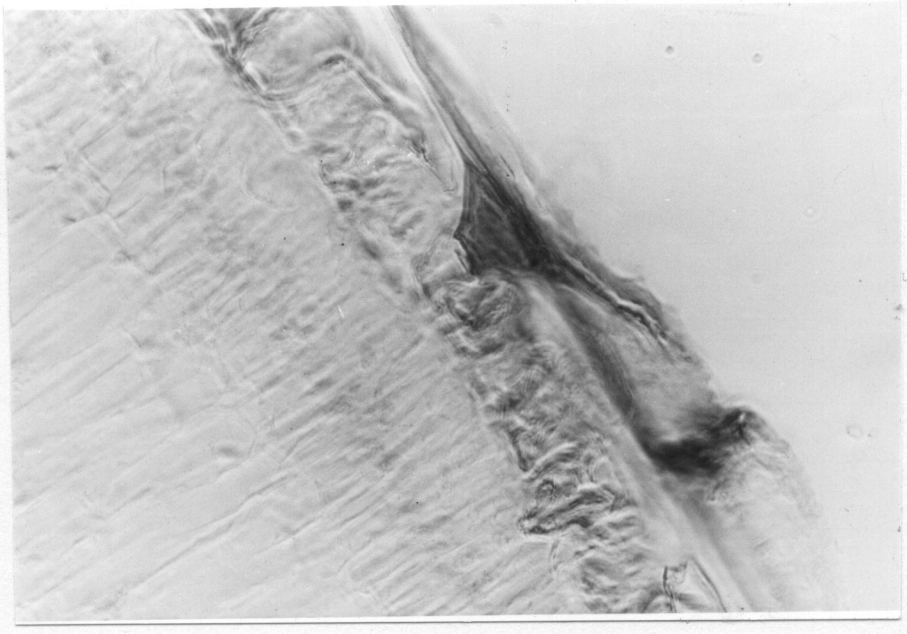
a) Stipe hair in longitudinal section. Note hair attachment to the epidermis. x 175

b) Sporocarp hair in longitudinal section. Note the sunken level of the hair contact face with the foot cell. x 320

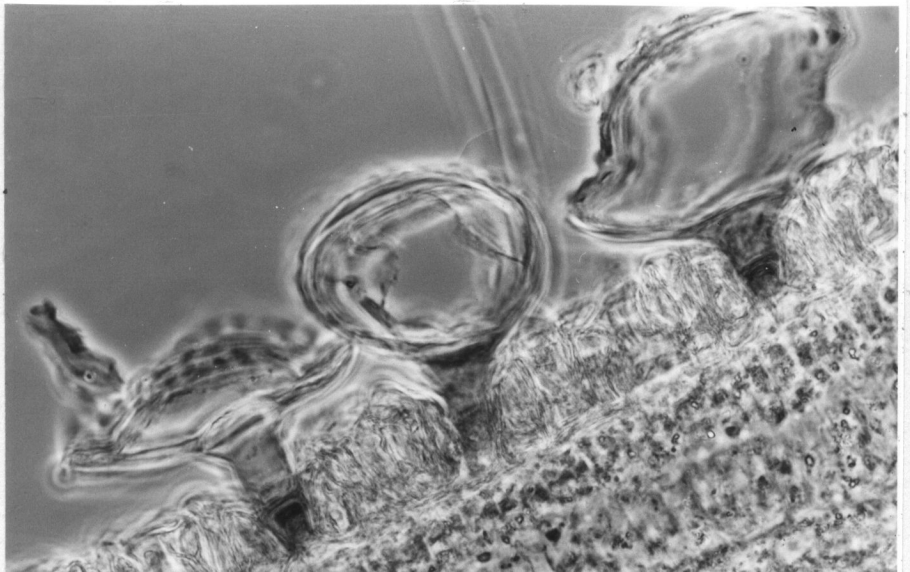
c) Sporocarp hairs in transverse section. x 320

PLATE 21

21a



21b



21c

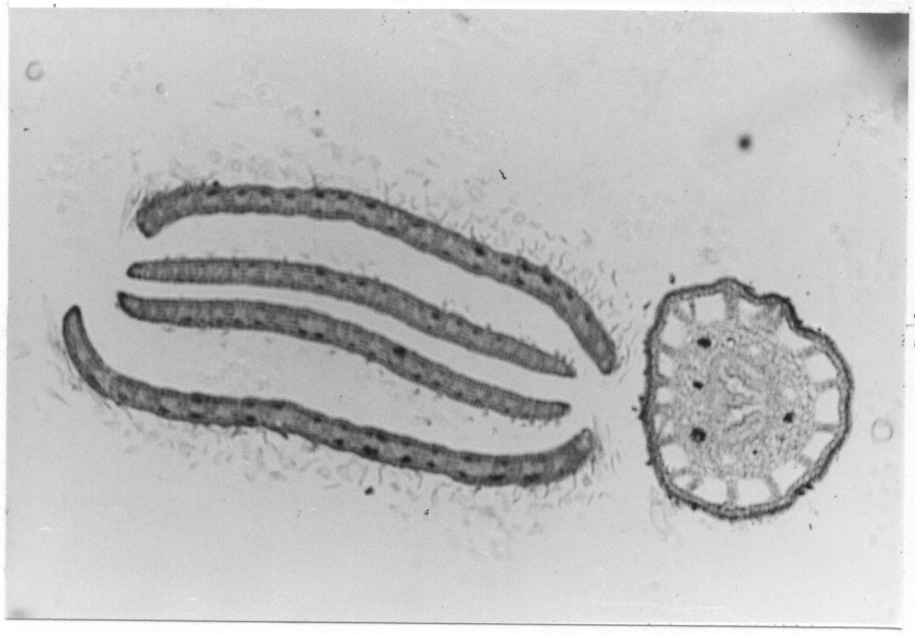
Plate 22. *Marsilea drummondii*

a) Four young leaflets with stipe on the right. Note that hairs develop on outermost surface of leaflet with inner surface devoid of hairs. x 32

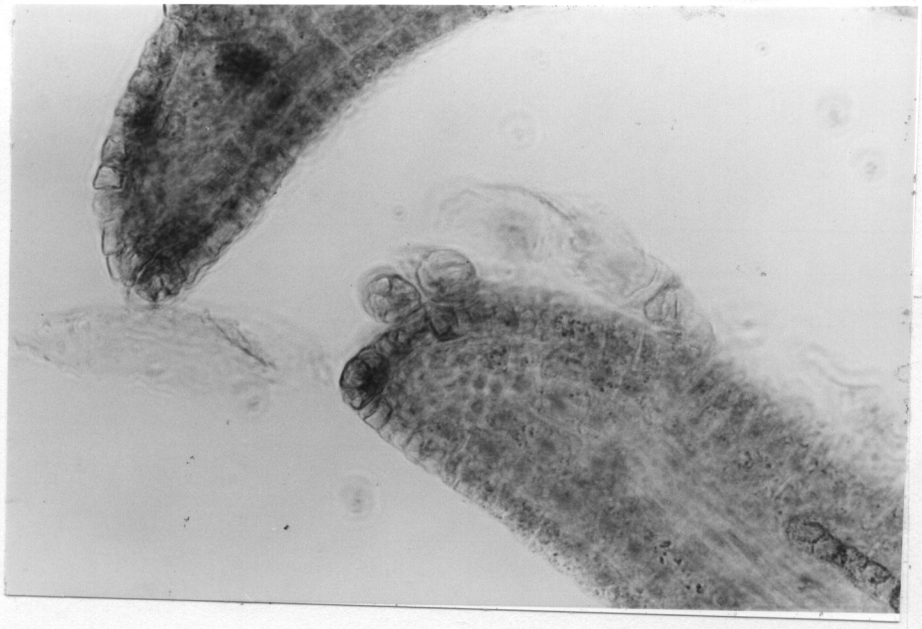
b) Young hairs developing at tip of leaflet. x 128

c) Young hairs at the tip of second leaflet from above shown at intermediate stage of development. Note the anvil-shaped form. x 128

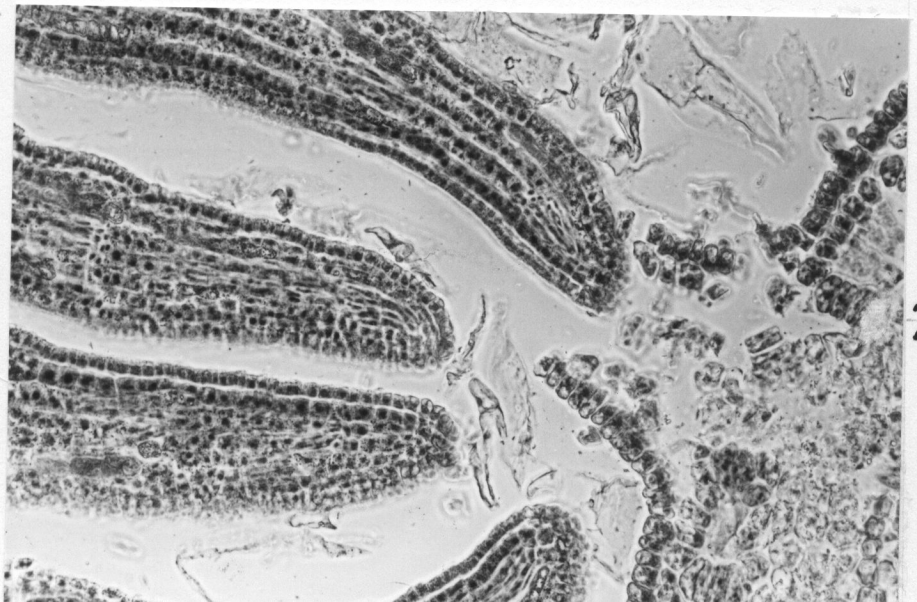
PLATE 22



22a



22b



22c