



# LEAF ANATOMY

#### 6.1 Introduction

Leaves in the Hypoxidaceae are rosulate and three-ranked, the outer ones reduced to cataphylls. The foliage leaves are linear or lanceolate and hairy, seldom glabrous. In cross section, the leaves are V-shaped, inversely W-shaped (plicate) or crescentiform, with V-shape being the most common state in the family. In *Hypoxis*, the morphology of the foliage leaves provides a number of characters of taxonomic value. Characters such as leaf venation and indumentum were found to be particularly useful in demarcating species in the genus (Chapter 5).

To assess the potential value of anatomical characters, the leaves of 20 southern African species of *Hypoxis* (see Chapter 4 for a list of species) were studied using Light [LM] and Scanning Electron Microscopy [SEM]. The purpose of this chapter is to report on the leaf anatomical features of the species studied with reference to characters of taxonomic importance. As earlier contributions on the anatomy in the family formed an important framework for the interpretation of the results of the present study, a summary of the key ideas emanating from them is provided at the beginning of the Chapter. The results of the present study are integrated to provide a generic description of the leaf surface and internal anatomy for *Hypoxis*.

Leaf surface characters were found to be of little diagnostic value in separating species in *Hypoxis*. On the contrary, internal anatomy was taxonomically more useful and provided a few characters of diagnostic value. The mesophyll and arrangement of the sclerenchyma making up the inner sheath of the vascular bundles are useful for species identification. Figures depicting anatomical characters in leaves of *Hypoxis* and their variation are included within the generic description. Vouchers of all specimens cited are lodged at NH. A comparison of features from leaf cross sections are presented in Table 6.2. The Chapter concludes with a summary of anatomical characters of diagnostic value in *Hypoxis* and attempts to link them to the associated morphology.

#### 6.2 Literature review

Schnarf (1892) studied the comparative anatomy of roots, rhizomes, leaves and peduncles in the Hypoxidaceae. He used various species of *Hypoxis*, *Curculigo*, *Spiloxene* and *Pauridia* (Table 6.1)



Current names are indicated in brackets.

Author	Roots	Storage organ	Leaves	Peduncle
Schnarf (1892)	Curculigo sumatrana	Curculigo sumatrana	Curculigo sumatrana	
	Hypoxis sp.			Hypoxis sobolifera Hypoxis lanata (= H. stellipilis) Hypoxis microsperma (= H. villosa)
		Hypoxis linearis (= Spiloxene serrata var. serrata)	Hypoxis stellata (= S. capensis)	Spiloxene capensis
Nel (1914)			Hypoxis argentea Hypoxis dinteri (= H. argentea) Hypoxis gerrardii Hypoxis obtusa Hypoxis recurvata Hypoxis subspicata Hypoxis villosa	
Arber (1925)		Curculigo orchioides	<i>Curculigo</i> sp. <i>C. recurvata</i>	
	Hypoxis setosa	Hypoxis setosa	Hypoxis setosa	Hypoxis setosa
Thompson (1976)		Empodium plicatum	Empodium plicatum	
		Pauridia minuta	Pauridia longituba Pauridia minuta	
		Spiloxene aquatica Spiloxene capensis	Spiloxene aquatica Spiloxene capensis Spiloxene flaccida Spiloxene minuta Spiloxene ovata Spiloxene schlechteri Spiloxene serrata	
Heideman (1983)			Hypoxis acuminata Hypoxis argentea Hypoxis filiformis Hypoxis galpinii Hypoxis hemerocallidea Hypoxis interjecta Hypoxis multiceps Hypoxis neliana (=H. kraussiana) Hypoxis obtusa Hypoxis rigidula Hypoxis rooperi (= H.	



Table 6.1.—cont.

Author	Root	Storage organ	Leaves	Peduncle
Rudall et al.	Curculigo	Curculigo latifolia	Curculigo capitata	
(1998)	latifolia	Curculigo orchioides	Curculigo latifolia	
	Curculigo	Curculigo pilosa	Curculigo orchioides	
	villosa	Curculigo villosa	Curculigo pilosa	
			Curculigo recurvata	
			Curculigo villosa	
			Empodium elongatum	
			Empodium plicatum	
			Hypoxidia rhizophylla	
	Hypoxis	Hypoxis decumbens	Hypoxis angustifolia	
	decumbens	Hypoxis hygrometrica	Hypoxis decumbens	
	Hypoxis	Hypoxis sessiliflora	Hypoxis hygrometrica	
	hygrometrica	Hypoxis urceolata	Hypoxis obtusa	
	Hypoxis	Hypoxis villosa	Hypoxis rooperi (= H.	
	urceolata		hemerocallidea)	
	Hypoxis villosa		Hypoxis urceolata	
			Hypoxis villosa	
			Hypoxis sp.	
			Molineria latifolia	
			Pauridia longituba	
	Rhodohypoxis		Rhodohypoxis baurii	
	baurii		Rhodohypoxis	
			millioides	
			Spiloxene aquatica	
			Spiloxene schlechteri	
			Spiloxene stellata (= S.	
			capensis)	
			Spiloxene serrata	

and provided a detailed anatomical description of the root in *Hypoxis* and *Curculigo sumatrana*. Schulze (1893) mentions Hypoxis, Curculigo and Pauridia in a general discussion under Liliaceae and reports that Pauridia cannot be separated anatomically from Hypoxis. Schnarf (1892) and Schulze (1893) found that vascular bundles in the peduncle in Hypoxidaceae lie in a ring and are not scattered as in stems of most monocotyledons. Nel (1914) described the morphology of the corms in detail and used it as the main character in grouping species of *Spiloxene*. He found that the leaf anatomy in the genus was of little value in grouping species, as the vascular bundles lie close to the surface and they are hardly distinguishable from each other, appearing almost similar in cross section. In contrast, Nel (1914) reported that the internal leaf anatomy of *Hypoxis* provided good characters for classifying species. He explained the particular usefulness of number, distribution and size of vascular bundles especially those associated with the vein areas. These characters provided support for his infrageneric groupings. He did not, however, give prominence to the shapes of outlines of transverse sections of leaves, a character which is of diagnostic value in other petaloid monocotyledons, for example Kniphofia Moench (Baijnath 1980) and Bulbine Wolf (Baijnath & Cutler 1993). Figure 6.1 shows cross sections of leaves presented by Nel and a representative of sections he proposed for the African members of Hypoxis.

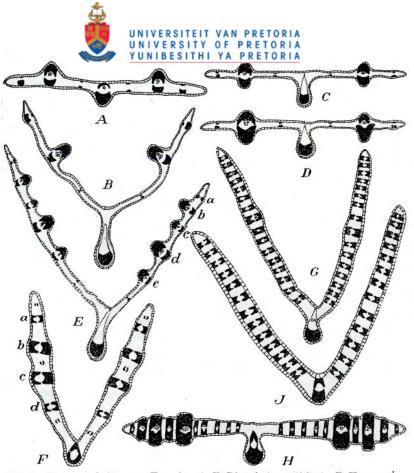


Fig. 2. Blattquerschnitte von Hypoxis. A H. Dinteri (Angustifoliae); B H. angustifolia (Angustifoliae); C H. argenteae (Argenteae): D H. Gerrardi (Argenteae); E H. villosa (Villosae); F H. recurrata (Recurvatae); G H. subspicata (Subspicatae); H H rigidula (Rigidulae); J H. obtusa (Obtusae). — Original.

Figure 6.1.—Cross sections of leaves in *Hypoxis* (sectional names in brackets). Taken from Nel (1914).

*H. dinteri* (A) and *H. angustifolia* (B) belonging to section *Angustifoliae*, as well as *H. argentea* (C) and *H. gerrardii* (D) of section *Argenteae* are similar in having linear leaves with one lateral vein on each side of the midvein close to the edge that is adaxially prominent. Nel selected *H. dinteri* to represent the two sections and described its anatomy in detail. Using the leaf type in *H. villosa* (E) to represent sections *Villosae*, *Orbiculatae* and *Nyassicae*, Nel described its appearance in cross section. The leaves in these groups are narrowly lanceolate and have about ten veins. Nel pointed out that the higher number of lateral veins (about five) on each half of the leaves, at least three of which are almost uniformly developed in members of these sections, differentiates them from sections *Angustifoliae* and *Argenteae*. Members of *Recurvatae*, represented by *H. recurvata* (F) have four unevenly thickened lateral veins on each half of a leaf. However, all veins are similarly thickened on both sides perpendicular to the leaf surface, with the inner veins *b* and *c* slightly thicker than those in position *a* and *d*. In comparison, the veins in sections *Villosae*, *Orbiculatae* and *Nyassicae* are thicker on the upper surface. In *Subspicatae* (*H. subspicata* = *H. polystachya*, G) and *Obtusae* (*H. obtusa*, J), the leaves have many lateral veins that are almost



uniformly thickened and differ frue from each other. There are grooves between the veins in these leaves. From Nel's illustrations and descriptions, the leaves in these sections appear very similar and therefore difficult to tell apart. The last leaf type recognised by Nel is that of *Rigidulae* (*H. rigidula*, H). This type differs from *Subspicatae* and *Obtusae* in its strong thickening of one to three lateral veins on each half of the leaf, close to the edge, noticeable to the naked eye. Nel also noted that the cuticle in *H. obtusa*, though variable, is usually thicker than in other species.

Arber (1925) included *Hypoxis* and *Curculigo* in her study of the structure of organs in monocots. She provided a plate (Figure 6.2) with illustrations of transverse sections of root, axis (rhizome), leaf and peduncle in *H. setosa* and uses the transverse sections of leaves in *Curculigo* sp. and *C. recurvata* Dryand. in leaf type comparisons. In trying to understand the mechanism by which root tissue contracts, Arber studied serial sections of the smooth and wrinkled zones in *H. setosa*. She concluded that it is the outer tissues that wrinkled while the central cylinder and inner cortex remained unaffected and she ascribed wrinkling of the whole outermost layer to its increased length in comparison to the inner tissues. With regard to the axis, Arber pointed out the cylindrical rhizome with a depressed growing point in *Hypoxis* (Figure 6.2, viiiB). She further confirmed the observations of Schnarf (1892) and Schulze (1893) that vascular bundles of the peduncle in *Hypoxis* are arranged in a ring (Figure 6.2, viiiE). Thompson (1972) noted that the arrangement was not unexpected as the peduncle is not the main stem or axis, but an axillary structure.

In 1976, Thompson published her studies in the Hypoxidaceae with emphasis on the vegetative morphology and anatomy in *Empodium*, *Spiloxene* and *Pauridia*. She found that the corms of all three genera are very similar anatomically, and that throughout the ground tissue of parenchyma cells, there are mucilage canals. These are not associated with the vascular bundles, but are surrounded by radially flattened cells as in the leaves. She compared the mucilage canals in these genera to Schnarf's (1892) account for *Hypoxis*, where the canals are described to arise schizogenously near the growing point and they have no membrane but are embedded in a ring of smaller cells. She further confirmed that the nature of the corm coverings in *Spiloxene* can be used to recognise the groups established by Nel (1914) for the genus. Using morphology and general anatomy, Thompson (1976) classified the leaves in the genera under study into four groups: carinate, canaliculate, terete and plicate. She found that leaf form is not a good basis for grouping



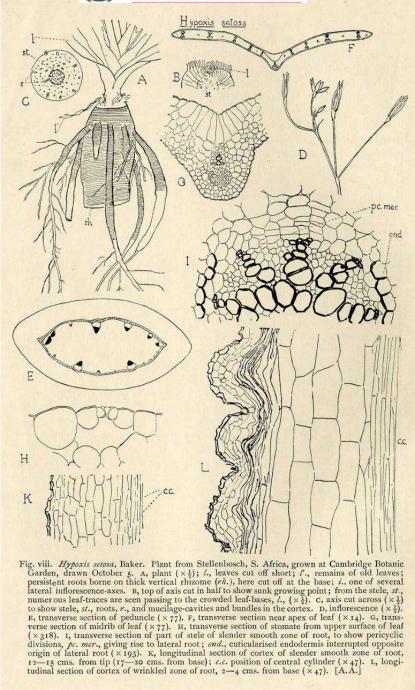


Figure 6.2.—Morphology and anatomy of *H. setosa*. Taken from Arber (1925).

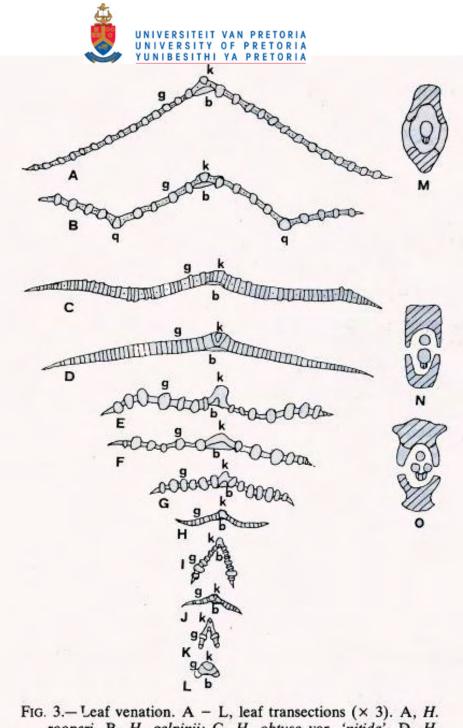


FIG. 3.— Leaf venation. A – L, leaf transections (× 3). A, H. rooperi, B, H. galpinii; C, H. obtusa var. 'nitida', D, H. obtusa var. obtusa; E, H. rigidula var. 'hemerocallidea'; F, H. rigidula var. pilosissima; G, H. rigidula var. rigidula; H. H. multiceps and H. interjecta; I, H. acuminata; J. H. neliana; K, H. argentea; L, H. filiformis. M–O, fibrous girders (× 30): M, crescent-shaped (in leaves A, B, H); N, crossbar-shaped (in leaves C, D, J, K, L); O, T-shaped (in leaves E, F, G, I).

Figure 6.3.—Cross sections of leaves in *Hypoxis*, classifying them according to girder types. Taken from Heideman (1983).



species in any of the three genera as very dissimilar species could have the same leaf form, and that leaf anatomy was of little taxonomic value.

After Nel (1914), Heideman (1983) paid attention to venation in *Hypoxis* leaves. She studied the leaf sections of eleven species of *Hypoxis* growing on the Witwatersrand and recognised three types of girders among the species: T-shaped, rectangular-(cross-bar) shaped and crescent-shaped (Figure 6.3). She indicated that T-shaped girders protrude as wide prominent ribs (veins) and that the other two types of girders protrude only slightly. According to her, T-shaped girders occur in *H. acuminata* and *H. rigidula*, rectangular girders in *H. argentea*, *H. filiformis*, *H. kraussiana* and *H. obtusa* and crescent-shaped girders in *H. galpinii*, *H. multiceps*, *H. interjecta* and *H. hemerocallidea* (*H. rooperi*). She also classified *H. colchicifolia* (*H. latifolia*) as having crescent-shaped girders. In the same publication, Heideman reported on SEM studies and illustrated leaf hairs in *H. kraussiana* and *H. multiceps* and the stomata in the latter species.

Nordal *et al.* (1985) studied the leaf indumentum in East African *Hypoxis* using LM. These authors broadly classified the hairs into six types based on colouration and distribution, as well as number and position of arms. In Nordal *et al.* (1985), 17 taxa are included as synonyms in the *H. obtusa* complex, and the constituent taxa representing four different hair types. Recent work on the genus in the region by Szymańska & Nordal (2006), however, does not maintain the complex.

Hitherto the most comprehensive study on leaf anatomy of Hypoxidaceae was by Rudall *et al.* (1998). The latter authors provided leaf anatomical descriptions of all genera except *Saniella* based on the study of a number of species (Table 6.1). They included eight species of *Hypoxis*, two being extra-African. In their publication, Rudall *et al.* (1998) presented leaf anatomical data for the asteloid genera and combined them with new rbcL sequence data in analysing the relationship of Asteliaceae and Hypoxidaceae. They confirmed the placement of *Pauridia* in the Hypoxidaceae. The rbcL analysis indicated similarity between *Rhodohypoxis* (a South African genus) and the North American *Hypoxis*, as well as *Spiloxene* (South Africa) and *Hypoxis glabella* (Australia), a species included in section *Ianthe* and has been proposed to belong to *Spiloxene* (Hilliard & Burtt 1978; Manning *et al.* 2002).



### 6.3 Results

#### 6.3.1 Leaf surface details

The description is based on observations using the SEM and follows the style used in the Anatomy of Monocot Series, Jodrell Laboratory, Kew:

*Surfaces* plane or ribbed, position of veins (ribs or costae) occupied by largest vascular bundles (Figure 6.4). *Epidermal cells* outline usually distinct, polygonal, mainly hexagonal or axially elongated (Figure 6.5A), rarely elongated transversely (Figure 6.5B); sometimes with distinct costal and intercostal cell arrangement, in general cells of the latter more elongated longitudinally. Position of anticlinal walls denoted by slight depressions, sometimes obscured by wax particles or sheets. *Hairs* bifurcate (Figure 6.6A–D) or stellate (Figure 6.7A–F and 6.8A–D) consisting of two or more unicellular unbranched arms; arms varying in length, central arm often longer and thicker than the rest. *Papillae* absent. *Striae* present in most species, both longitudinal and transverse (Figure 6.9A), often striae spreading transversely from flanks of outer stomatal lips over the subsidiary cells (Figure 6.9B,C). *Stomata* present on both surfaces, more plentiful on abaxial, paracytic; outer stomatal lip (cuticular ledges) level, elliptic in outline (Figure 6.9B). *Wax* usually as sheets on both surfaces, often flaking (Figure 6.9D,E) sometimes particles amorphous (Figure 6.9A) or distinct (Figure 6.9F).

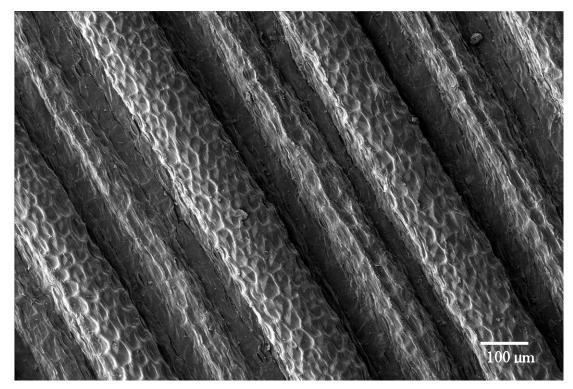


Figure 6.4.—Scanning electron micrograph of upper surface of leaf showing position of veins (ribs) in *H. obtusa* (*Singh* 277), adaxial surface.

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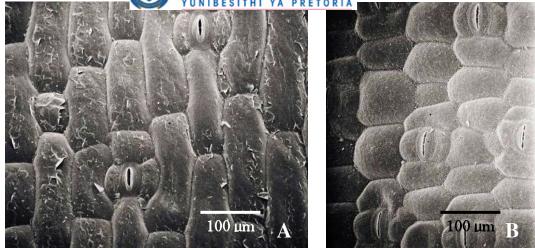


Figure 6.5.—Scanning electron micrographs of leaf surfaces showing epidermis. A, longitudinally elongated cells in *H. longifolia* (*Singh 502*), adaxial surface; B, transversely elongated cells in *H. angustifolia* var. *buchananii* (*Singh 535*), abaxial surface.

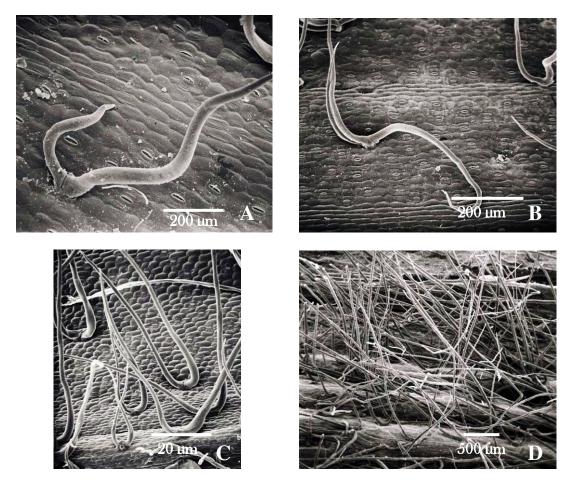


Figure 6.6.—Scanning electron micrographs of leaf surfaces showing bifurcate, ascending hairs. A, H. hemerocallidea (Singh 262); B, H. multiceps (Singh 322); C, H. parvifolia (Singh 470); D, H. rigidula var. pilosissima (Singh 318). A,B,D, adaxial surface; C, abaxial surface.



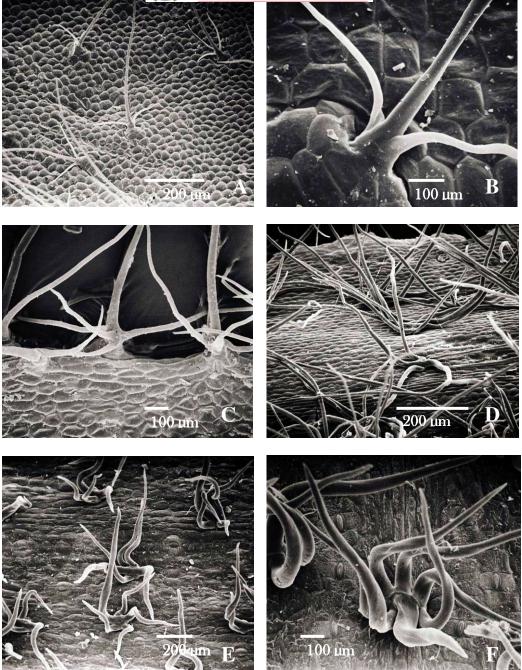


Figure 6.7.—Scanning electron micrographs of leaf surfaces showing stellate, ascending hairs: A–C, *H. parvula* var. *parvula* (*Singh 465*); D, *H. sobolifera* var. *sobolifera* (*Singh 233*); E,F, *H. multiceps* (*Singh 615*). A,B,D,E, abaxial surface; C,F, abaxial surface.



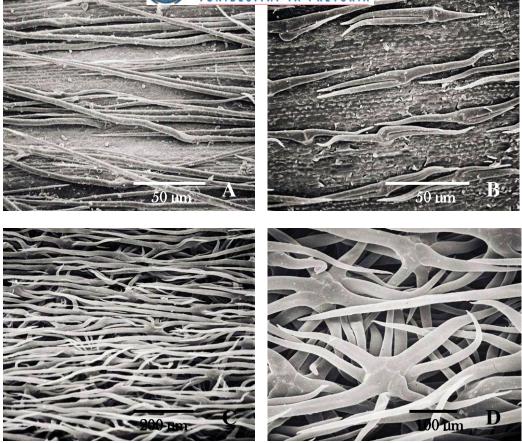


Figure 6.8.—Scanning electron micrographs of leaf surfaces showing stellate, adpressed hairs. A, H. rigidula var. rigidula (Singh 317); B, H. obtusa (Singh 330); C, D, H. stellipilis (Singh 621). A, adaxial surface; B–D, abaxial surface.



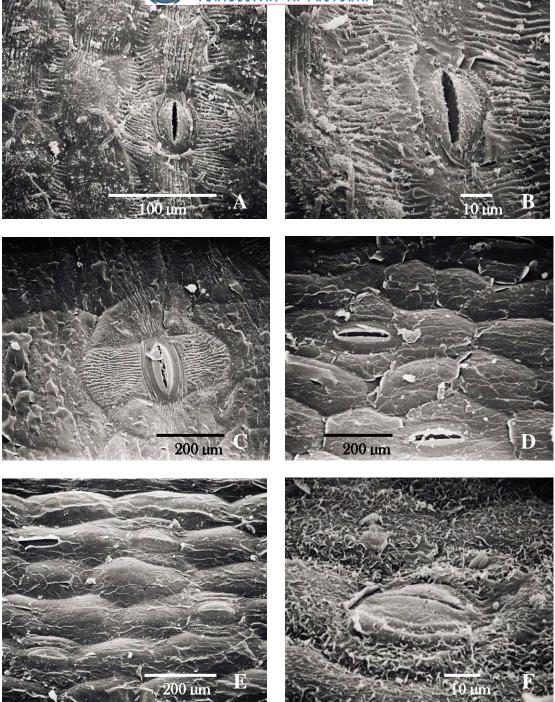


Figure 6.9.—Scanning electron micrographs of leaf surfaces showing cuticular striae, stomata and wax. A, longitudinal and transverse striae in *H. colchicifolia* (Singh 481); B, C, transverse striae radiating from flanks of outer stomatal lips over subsidiary cells in B, *H. colchicifolia* (Singh 481) and C, *H. multiceps* (Singh 615); D, wax as sheet in *H. argentea* var. sericea (Singh 295); E, wax as sheet in *H. filiformis* (Singh 418); F, wax particles in *H. rigidula* var. rigidula (Singh 317). A, adaxial surface; B–F, abaxial surface.



Outline dorsiventrally flattened, crescentiform, V-shaped or plicate. Cuticle thin on both surfaces, following outer walls of epidermal cells and exposed walls of guard cells. *Epidermis* cells taller than wide, or as wide as tall or wider than tall, cells on adaxial surface usually larger than abaxial (Figure 6.10A–D); bulliform cells present on one or both surfaces (Figure 6.11A, B), sometimes darkly staining due to tanniniferous substances (Figure 6.12A-F). Hairs arising from multicellular base; arms circular to square in outline (Figure 6.13A–D). Stomata present on both surfaces. Guard cells about <sup>1</sup>/<sub>3</sub> to <sup>1</sup>/<sub>2</sub> dimensions of subsidiary cells; outer, poral and inner walls more thickened than epidermal wall; outer stomatal lip (ledge) well-developed, often raised; inner stomatal lip (ledge) obscure or poorly developed; substomatal cavity well-developed; subsidiary cells as tall as or twice as tall as wide (Fig 6.14A–D). *Hypodermis* absent. *Mesophyll* chlorenchymatous, spongy type (Figure 6.15A, B) or sometimes differentiated into palisade (Figure 6.16A, B), 2–11 cells thick, shape and dimensions variable. Vascular bundles in a single row with bundle in midrib region usually the largest (Figure 6.17A–F), usually of three sizes (Figure 6.18A–F); large (Figure 6.19A– F), medium (Figure 6.20A–F) and small (Figure 6.21A–D); large bundles alternating with medium and small bundles, commonly with sclerenchyma forming girders (Figure 6.19A–F), occasionally linking to smaller lateral bundles (Figure 6.19B); medium bundles with sclerenchyma as girder and cap at opposite poles (Figure 6.20A–C) or caps at both poles (Figure 6.20D–F) and small bundles with sclerenchyma reduced (Figure 6.21A–D); randomly orientated tiny bundles lacking sclerenchyma present in spaces between large bundles, also single row of tiny bundles at the flanks of largest bundles partially or wholly surrounded by inner sclerenchyma bundle sheath. Phloem in abaxial position. Xylem in adaxial position. Margins without sclerenchyma. Bundle sheaths with outer sheath of single layer of parenchyma, inner sheath sclerenchymatous, variously developed. Crystals present as raphides in sacs in mesophyll (Figure 6.22A–D), styloids absent. Mucilaginous canals absent. Silica bodies absent. Tanniniferous substances present in epidermal and bulliform cells (Figure 6.12A–F).

#### 6.3.3 Anatomical characters of taxonomic value

The examination of leaf surface characters in this study was limited to SEM. Surface ornamentation in *Hypoxis* was found to be of little value for taxonomy in comparison to other monocot groups, for example Kniphofia (Baijnath 1980), Bulbine (Baijnath & Cutler 1993) and in Aloe and Gasteria (Cutler 1972) where striae and papillae provide valuable characters for separating species. Nevertheless, the results of this study proved useful in confirming the distribution and types of hairs discussed in Chapter 5.

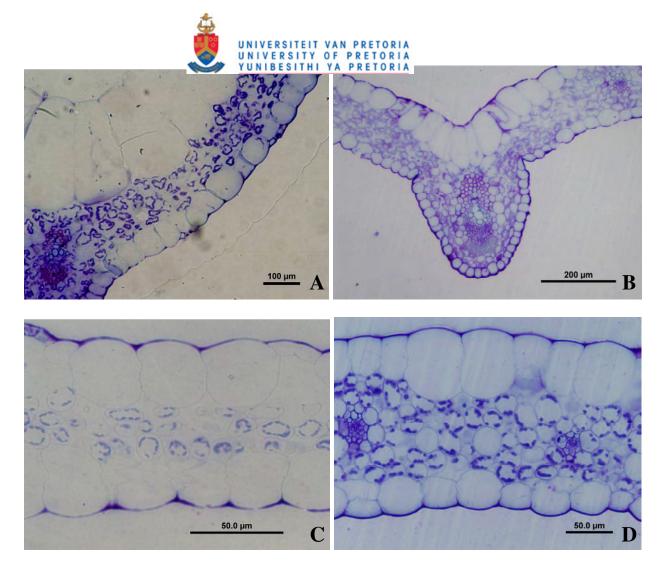


Figure 6.10.—Transverse sections of leaves showing narrow mesophyll. A, *H. parvula* var. *parvula* (Singh 556);
B, *H. angustifolia* var. *buchananii* (Singh 814);
C, *H. membranacea* (Singh 826);
D, *H. flanaganii* (Singh 807).

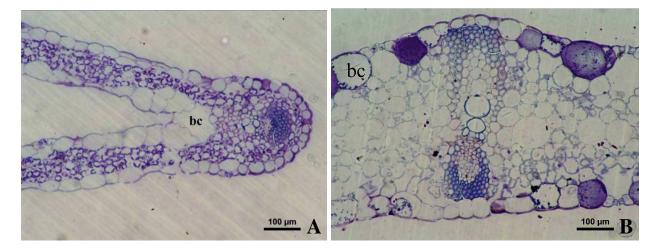
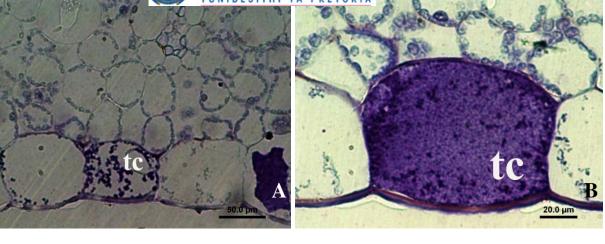
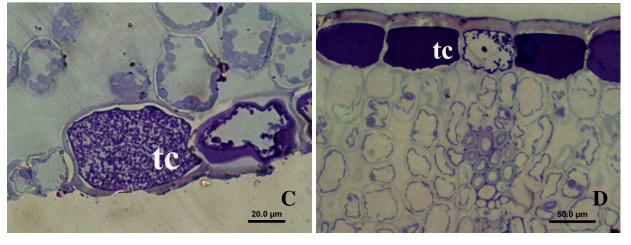


Figure 6.11.—Transverse sections of leaves showing bulliform cells (bc). A, *H. argentea* var. *sericea* (*Singh* 259); B, *H. hemerocallidea* (*Singh* 262).

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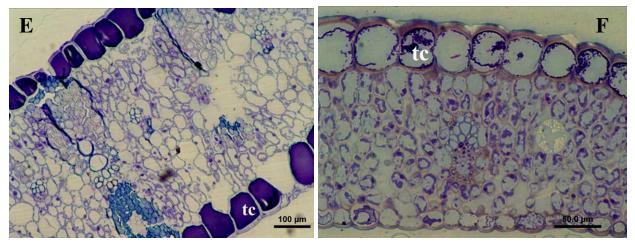


Figure 6.12.— Transverse sections of leaves showing cells with tanniniferous compounds (tc). A, B,
H. hemerocallidea (Singh 262); C, H. rigidula var. pilosissima (Singh 263); D, H. galpinii (Singh 334); E,
H. obtusa (Singh 337); F, H. stellipilis (Singh 621).



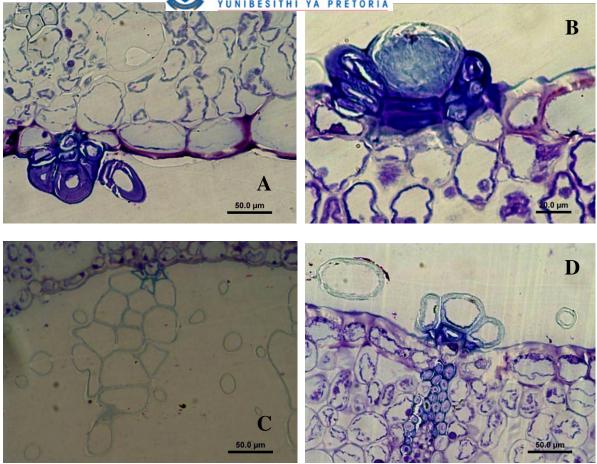


Figure 6.13.— Transverse sections of leaves showing hair attachment and outline. A, *H. multiceps* (*Singh* 279); B, *H. rigidula* var. *rigidula* (*Singh* 329); C, *H. stellipilis* (*Singh* 621); D, *H. obtusa* (*Singh* 283).



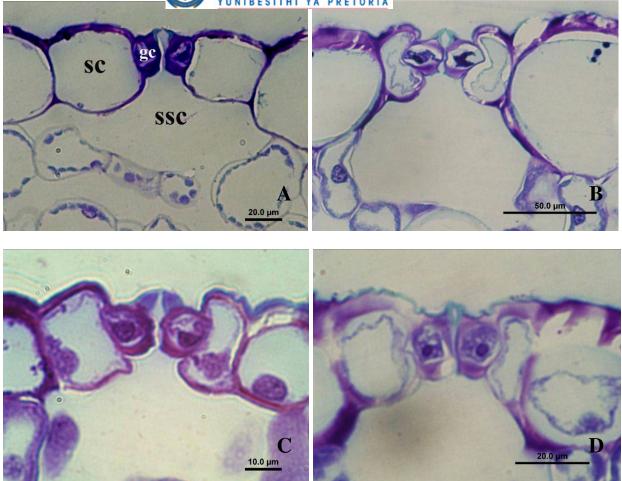


Figure 6.14.—Transverse sections of leaves showing outer stomatal lip. A, *H. costata* (*Singh* 803), sc = subsidiary cavity, ssc = substomatal cavity, gc = guard cell; B, *H. interjecta* (*Singh* 280); C, *H. acuminata* (*Singh* 286); D, *H. rigidula* var. *rigidula* (*Singh* 282).



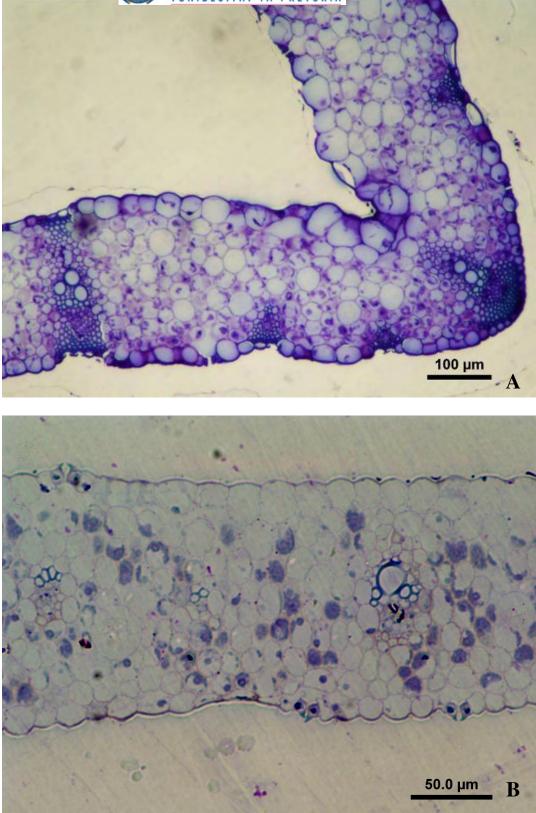


Figure 6.15.—Transverse sections of leaves showing spongy mesophyll. A, *H. filiformis* (*Singh 418*); B, *H. argentea* var. *argentea* (*Singh 626*).

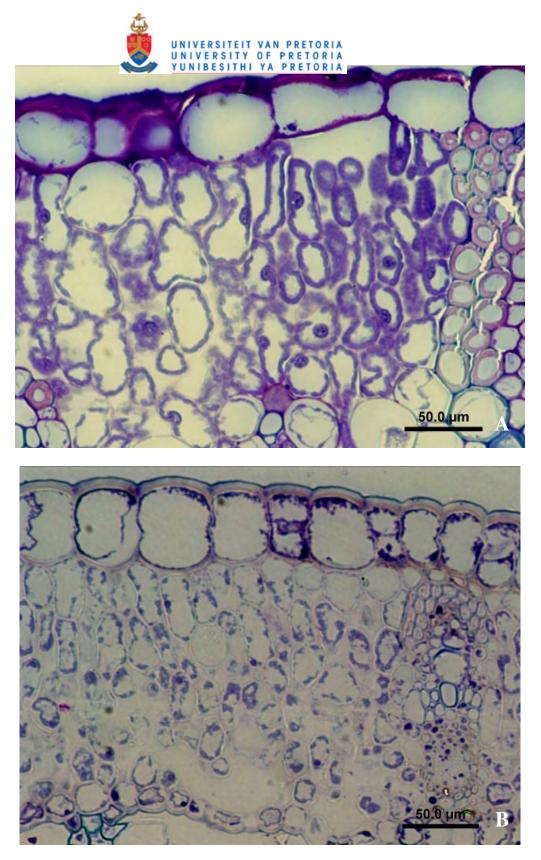
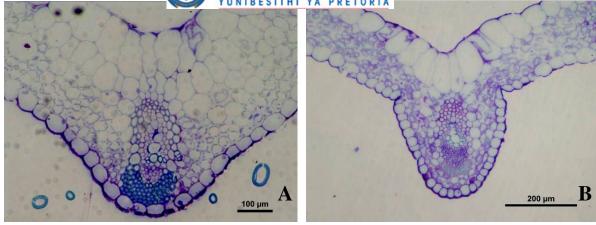
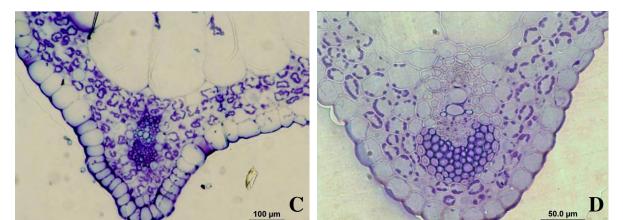


Figure 6.16.—Transverse sections of leaves showing palisade mesophyll. A, *H. obtusa* (*Singh 337*); B, *H. stellipilis* (*Singh 621*).

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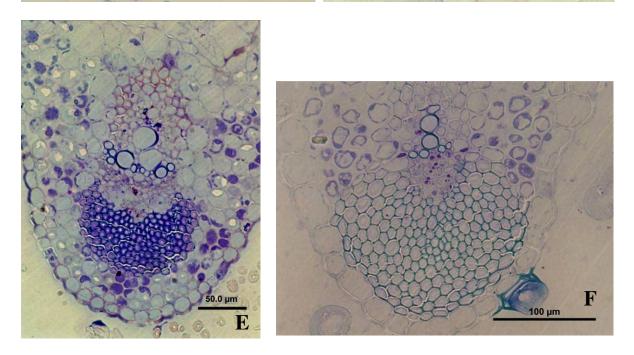
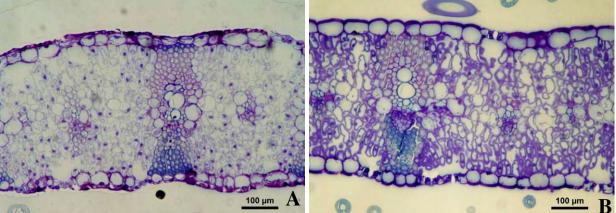
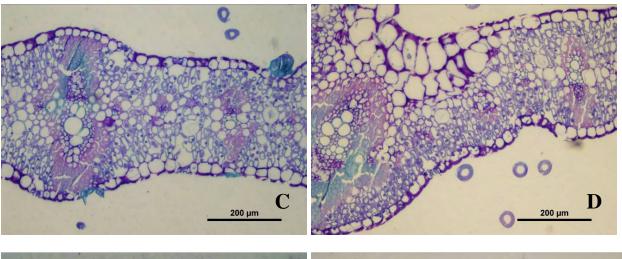


Figure 6.17.—Transverse sections of leaves showing midrib vascular bundle. A, H. costata (Singh 803);
B, H. angustifolia var. buchananii (Singh 814); C, H. parvula var. parvula (Singh 556); D, H. flanaganii (Singh 807); E, H. argentea var. argentea (Singh 626); F, H. parviflora (Singh 470).







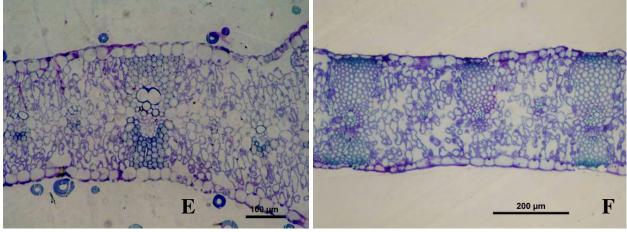


Figure 6.18.—Transverse sections of leaves showing vascular bundles of three sizes arranged in a single row.
A, H. obtusa (Singh 277); B, H. obtusa (Singh 337); C, H. rigidula var. pilossisma (Singh & Baijnath 318);
D, H. rigidula var. rigidula (Singh 317); E, H. obliqua (Singh 531); F, H.costata (Singh 300).