# A comparison of the leaf anatomy of *Ficus subpuberula*, *F. atricha* and *F. brachypoda* (Moraceae)

## Dale J. Dixon

Tropical Plant Sciences, School of Tropical Biology, James Cook University,
Townsville, Queensland, Australia 4811.

Address for correspondence: Herbarium of the Northern Territory, PO Box 496,
Palmerston, Northern Territory, Australia 0831.

### Abstract

Dixon, D.J. A comparison of the leaf anatomy of *Ficus subpuberula*, *F. atricha* and *F. brachypoda* (Moraceae). *Nuytsia* 15(1): 27–32 (2002). The leaf anatomy of *Ficus subpuberula* Corner, *F. atricha* D.J.Dixon, and *F. brachypoda* (Miq.) Miq. are compared in order to facilitate identification of these partly sympatric species. *Ficus subpuberula* was found to possess distinctly thinner (267.6  $\pm$  4.3  $\mu$ m), isobilateral leaves compared to the much thicker (313.3  $\pm$  12.1  $\mu$ m and 425.6  $\pm$  17.8  $\mu$ m), dorsiventral leaves of *F. atricha* and *F. brachypoda* respectively. Tanniniferous cells were present in *F. atricha* and *F. brachypoda*, but absent in *F. subpuberula*. The upper epidermis and the palisade parenchyma of *F. brachypoda* were about twice as thick as in *F. atricha*.

# Introduction

The affinity of *Ficus subpuberula* Corner to *Ficus platypoda sensu* Chew (1989) (*Urostigma* sect. *Malvanthera*) has long been debated. Chew (1989) considered *F. subpuberula* to be closely related to *F. platypoda* commenting that it may be a variant with thicker, more rigid leaves. In contrast, Wheeler (1992) described *F. subpuberula* as a species with thinly coriaceous leaves. Dixon (2001) defined the discontinuities between the taxa associated with the "F. platypoda" complex and recognised the following taxa: *F. brachypoda* and *F. atricha*. The application of types to these taxa, discussed in Dixon (2001), revealed that the name *F. platypoda* did not apply to these taxa but actually applied to the species previously known as *F. leucotricha*. Further, the two names synonymous with *F. brachypoda viz.*, *F. platypoda* var. *minor* Benth. and *F. platypoda* var. *lachnocaulos* (Miq.) Benth., are the taxa previously confused with *F. subpuberula* with respect to leaf texture.

Although, morphologically, *Ficus subpuberula* can be distinguished from *F. brachypoda* and *F. atricha* by a suite of characters when reproductive material is present (Dixon, in press), the potential for confusion remains as all three species have a sympatric distribution for part of their range and vegetative material is often difficult to identify. In habitat, *F. subpuberula* is a spindly upright lithophyte with pendulous blue-green foliage. The leaves of *F. subpuberula* can either be glabrous or (minutely) pilose, but never with ferruginous hairs as found in *F. brachypoda*. *Ficus atricha*, as the name suggests, is completely glabrous in all its parts. A previous study on the anatomy of *Ficus* leaves by van Greuning *et al.* (1984) revealed that anatomical characters have been useful for identification purposes. In this

study of 24 African *Ficus* species van Greuning *et al.* (1984) found that, the number of epidermal layers, the structure of the spongy parenchyma, and the ratio of palisade parenchyma to the spongy parenchyma were all important in the delimitation of taxa. The aim of this research was to compare the leaf anatomy of *F. subpuberula*, *F. brachypoda*, and *F. atricha* to further substantiate the differences between the three taxa.

## **Methods**

# Sample preparation

Six individuals of *F. subpuberula* and *F. brachypoda* and five individuals of *F. atricha* were selected to represent the species across their respective distributional ranges (Appendix 1).

Fresh leaf material was fixed in FAA and then transferred to 70% ethanol. Samples from herbarium material were rehydrated in 70% ethanol. The central portion of the lamina including the midvein was then excised, dehydrated through a series of graded baths up to 100% ethanol, and infiltrated with paraffin wax. The samples were then imbedded in paraffin and sectioned at 5 and 10 micrometres on a rotary microtome. Haupt's adhesive was used to attach the sections to the microscope slides which were then dried overnight in a 60°C oven. Each section was stained in Alcian Blue and Safranin and permanently mounted in DPX. The slides have been lodged with DNA.

### **Section evaluation**

Measurements of the total leaf thickness, width of the upper and lower cuticle, the upper and lower epidermis, the hypodermis, and the palisade parenchyma, were made at three sites along the section for each individual examined. Measurements were made adjacent to the midvein, at the central portion of the lamina, and just inside the margin of the leaf.

Eighteen measurements for each tissue type were made on *F. subpuberula* and *F. brachypoda*, and 15 measurements on *F. atricha*.

## Results

The leaf anatomy of *F. subpuberula*, *F atricha* and *F. brachypoda* is described below and the relevant statistical data are summarised in Table 1.

# Ficus subpuberula (Figures 1A & B)

- upper (B) and lower epidermis of small unevenly sized parenchyma cells
- hypodermis of large uniformly sized parenchyma cells (B).
- upper palisade parenchyma of two to three layers, the third layer becoming indistinct (A).
- lower palisade parenchyma of one to two layers, may be undeveloped in places, leaf isobilateral (A).
- taniniferous cells absent.
- midvein often surrounded by a one to four layered fibre sheath (not shown).

Table 1. Comparison of leaf anatomy summary statistics in *Ficus subpuberula*, *F. atricha* and *F. brachypoda*.

Character	Summary statistics		
(thickness)	F. subpuberula	F. atricha	F. brachypoda
upper cuticle	$2.8 \pm 0.1 \; \mu m$	$3.7\pm0.4~\mu m$	$4.8\pm0.7~\mu m$
upper epidermis	$6.2\pm0.5~\mu m$	$6.8 \pm 0.7 \ \mu m$	$11.5 \pm 1.6  \mu m$
hypodermis	$49.3 \pm 3.1 \ \mu m$	$73.1\pm2.6~\mu m$	$86.7\pm6.0~\mu m$
palisade parenchyma	$60.6 \pm 4.0 \ \mu m$	$76.6 \pm 7.3 \ \mu m$	$143.0\pm10.5~\mu m$
lower epidermis	$27.7 \pm 1.5 \ \mu m$	$32.8\pm1.6~\mu m$	$50.5 \pm 4.3~\mu m$
lower cuticle	$3.8\pm0.3~\mu m$	$4.0\pm0.4~\mu m$	$5.3\pm0.9~\mu m$
leaf total thickness	$267.6\pm14.3~\mu m$	$313.3 \pm 12.1 \ \mu m$	$425.6 \pm 17.8 \ \mu m$

## Ficus atricha (Figure 1C & D)

- upper (D) and lower epidermis of small elongated evenly sized parenchyma cells in two layers.
- hypodermis of larger unevenly sized parenchyma cells (D).
- upper palisade parenchyma of one to two layers, leaf dorsiventral (C), some lower palisade parenchyma cells but never as well developed as in *F. subpuberula* (A).
- taniniferous cells interspersed with the palisade parenchyma (D).
- mid vein surrounded by a one to four layered fibre sheath (not shown).

# Ficus brachypoda (Figure 1E & F)

- upper epidermis of small elongated evenly sized parenchyma cells (F).
- hypodermis of larger rounded to elongated irregular parenchyma cells (F).
- lower epidermis of irregularly sized parenchyma cells.
- palisade parenchyma cells of irregular length sometimes up to five layers, mostly of two layers, leaf dorsiventral (E), lower palisade parenchyma absent.
- taniniferous cells interspersed with the palisade parenchyma (F).
- midvein surrounded by a one to three layered fibre sheath (not shown).

### Discussion

Leaf anatomical differences has proved reliable in the delimitation of the three taxa investigated in this study. The leaves of *F. subpuberula* are isobilateral unlike those of *F. atricha* and *F. brachypoda* which are dorsiventral. *Ficus subpuberula* has long slender petioles that allow the leaves to 'weep' or hang. This habit may account for the isobilateral development of the palisade parenchyma. The results also show that *F. subpuberula* has much thinner leaves than both *F. atricha* and *F. brachypoda* (Table 1) supporting Wheeler's (1992) assertion that *F. subpuberula* has thinly coriaceous leaves. The thicker leaves of both *F. brachypoda* and *F. atricha* can be rigid to such a degree that fresh material will snap if bent too far. However, when *F. atricha* is found in the more mesic areas of its distribution, the leaves can become more coriaceous. *Ficus brachypoda* has a wide distributional range, and specimens

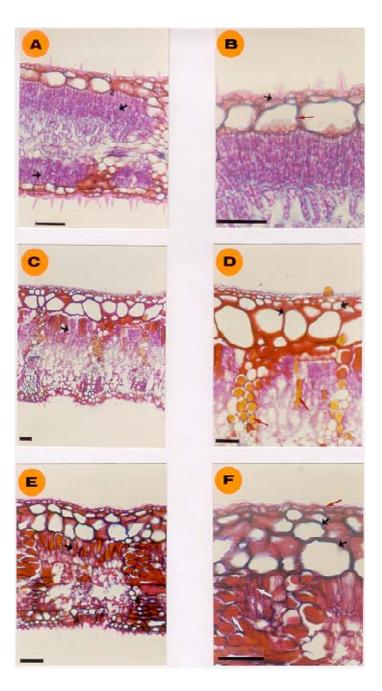


Figure 1. The light micrographs of the leaf transverse sections. Scale bars in each case equal 50 μm. A – the isobilateral leaf of *Ficus subpuberula* with the development of adaxial and abaxial palisade parenchyma (black arrows); B – the upper epidermis of unevenly sized cells (short black arrow), and the hypodermis of larger uniformly sized cells (long black arrow) of *Ficus subpuberula*; C – the dorsiventral leaf of *F. atricha* with the development of adaxial palisade parenchyma only (black arrow); D – the upper epidermis of evenly sized cells (white arrow), the hypodermis of larger unevenly sized cells (short black arrows), and the taniniferous cells (long black arrows) dispersed throughout the palisade parenchyma of *Ficus atricha*; E – the dorsiventral leaf of *F. brachypoda* with the development of adaxial palisade parenchyma only (black arrow); F – the upper epidermis of evenly sized cells (long black arrow), the hypodermis of larger unevenly sized cells (short black arrows), and the taniniferous cells (white arrows) dispersed throughout the palisade parenchyma of *Ficus brachypoda*.

collected from central Australia have leaves that are much narrower and quite rigid compared to specimens from northern Australia. *Ficus brachypoda* and *F. atricha* also differ from *F. subpuberula* by the presence of taniniferous cells distributed throughout the palisade parenchyma. These cells may also contribute to the rigid nature of the leaves. Each species has a distinct multilayered epidermis and a hypodermis which is in contrast to the single layered epidermis reported by van Greuning (1984) for African species in the *Ficus* subgen. *Urostigma*.

The anatomical characters revealed in this study, together with the morphological characters described for *F. brachypoda* and *F. atricha* in Dixon (2001), provide compelling data supporting the distinctiveness of *F. subpuberula*, *F. brachypoda* and *F. atricha*. *Ficus subpuberula* can be differentiated on the presence of adaxial and abaxial palisade parenchyma and the absence of taniniferous cells. The leaves of *F. atricha* and *F. brachypoda* are comparatively similar having only one adaxial layer of palisade parenchyma but differ in thickness. The greatest differences between these two species were in the upper epidermis and palisade parenchyma, both of which were about twice as thick in *F. brachypoda* as in *F. atricha*. With the addition of morphological characters *F. atricha* and *F. brachypoda* are easily separated. *Ficus atricha* is the only species in the *Urostigma* sect. *Malvanthera* to be completely glabrous in all its parts. In comparison *F. brachypoda* has a combination of hyaline and ferruginous hairs present on all its parts. Hyaline hairs are sometimes found on the leaves of *F. subpuberula*; however, it is more common for the leaves of this species to be minutely pubescent, that is, almost pubescent. Ferruginous hairs are never found on *F. subpuberula*.

# Acknowledgements

I wish to express my gratitude for the financial support provided by the Rainforest CRC, the Doctoral Merit Research Scheme, and the School of Tropical Biology at James Cook University. Without this support this research would not have been possible. I am especially thankful for Sue Riley's help while using the microtome. I thank the Directors of DNA, JCT, MEL, and PERTH for allowing the removal of leaf material for sectioning. Finally I wish to express my thanks to Dr Betsy Jackes and Dr Leone Bielig for their guidance, encouragement, support, patience and comments on the manuscript.

## References

- Chew, W.L. (1989). Moraceae. *In:* George, A.S. (ed.) "Flora of Australia." Vol. 3, pp. 15–68. (Australian Government Printing Service: Canberra.)
- Dixon, D.J. (2001). A chequered history: the taxonomy of Ficus platypoda and F. leucotricha (Moraceae: Urostigma sect. Malvanthera) unravelled. Australian Systematic Botany 14(4): 535–563.
- Dixon, D.J. (in press). A taxonomic revision of the Australian *Ficus* species in the section *Malvanthera* (*Ficus* subg. *Urostigma*:Moraceae). *Telopea*.
- Van Greuning, J.V. Robbertse, P.J. & Grobbelaar, N. (1984). The taxonomic value of leaf anatomy in the genus *Ficus. South African Journal of Botany* 3(5): 297–305.
- Wheeler, J.R. 1992. Moraceae. *In:* Wheeler, J.R. *et al.* "Flora of the Kimberley Region." (Department of Conservation and Land Management: Como, Western Australia.)

# **Appendix**

The collection details for the specimens used in the anatomical study. Herbarium acronym where specimen is held is indicated in parentheses.

Ficus atricha: King Creek Gorge, 15 km SW of Bedford Downs Homestead, 65 km W of Great Northern Highway, 102 km NNW of Halls Creek, Kimberley, 23 June 1976, A.C. Beauglehole ACB53670 (PERTH); c. 40 km SSW of Nathan River Homestead, 15°56'S, 135°20'E, 27 Aug. 1985, P.K. Latz 10108 (DNA); c. 3 km E of Gallen Well on Catamaran Bay, Dampierland Peninsula, 16°31'S, 122°59'E, 20 Aug. 1985, K.F. Kenneally 9463 (PERTH); Boomerang Bay, W side of Bigge Island, Bonaparte Archipelago, W Kimberley coast, 14°33'S, 125°09'E, 28 May 87, K.F. Kenneally 10014 (PERTH); Brunswick 8.7 km NE of Cape Brewster on unnamed island, site 2, 15°03'S, 124°57'E, 12 June 1987, K.F. Kenneally & B.P.M. Hyland 10365 (PERTH).

Ficus brachypoda: Kununurra, off road N of Hidden Valley Caravan Park, 15°43'S, 128°44'E, 21 Oct. 1997, *D. Dixon* PHD444 & *I. Champion* (DNA; JCT); off Great Northern Highway, 95.5 km NE of Fitzroy Crossing, 18°44'S, 126°05'E, 25 Oct. 1997, *D. Dixon* PHD457 & *I. Champion* (DNA, JCT); off Great Northern Highway, 92 km NE of Fitzroy Crossing, 18°45'S, 126°04'E, 25 Oct. 1997, *D. Dixon* PHD459 & *I. Champion* (DNA, JCT); Victoria Highway, 100 km E of Kununurra, 16°00'S, 129°29'E, 27 Oct. 1997, *D. Dixon* PHD460 & *I. Champion* (DNA, JCT); Ross Highway E of Alice Springs, Mt Benstead Creek crossing, 23°35'S, 134°26'E, 5 Nov. 1997, *D. Dixon* PHD485 & *I. Champion* (DNA, JCT); Ross Highway E, of Alice Springs, Mt Benstead Creek crossing, 23°35'S, 134°26'E, 5 Nov. 1997, *D. Dixon* PHD486 & *I. Champion* (DNA, JCT).

Ficus subpuberula: Edith Falls, 16 Oct. 1997, D.J. Dixon PHD419 & I. Champion (DNA, JCT); Edith Falls, 16 Oct. 1997, D.J. Dixon PHD418 & I. Champion (DNA, JCT); 13 km W of Kununurra off highway on track to Blackrock waterhole, 15°39'S, 128°39'E, 20 Oct. 1997, D.J. Dixon PHD442 & I. Champion (DNA, JCT); Cannon Hill, 12°22'S, 132°57'E, 17 July 1975, M. Parker 650 (DNA); Edith Falls Reserve, 14°12'S, 132°11'E, 5 Oct. 1977, M.O. Parker 1123 (DNA); 2 km N of Nabarlek Airstrip, 12°17'S, 133°19'E, 26 Apr. 1979, M.O. Rankin 2187 (DNA, MEL).