

PETIOLE ANATOMICAL STUDIES OF SOME RONDELETIA L. SPECIES (RUBIACEAE)

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A comparative study on the petioles of four species of section *Pedicellares* in genus *Rondeletia* (Rubiaceae) was undertaken in order to investigate anatomical variations which may be useful in species identification. The cells of the cortical parenchyma are collenchymatously thickened in each species and contain antrachinon. The vascular tissue is horseshoe-shaped and open to the adaxial side. The abaxial side of the xylem is bordered by phloem. The species differ in presence or absence of trichomes, crystals, bundle size and number of lateral bundles.

Key words: Rubiaceae, *Rondeletia*, *Pedicellares*, petiole TS

INTRODUCTION

The Neotropical genus *Rondeletia* L. includes a group of trees and shrubs with opposite, entire and evergreen leaves and interpetiolate stipules. The species of section *Pedicellares* are distributed in Haiti and Cuba. They are characterised by triangular or widening stipules and short petioles (0.5–3 mm long). The blades are oval, oval-oblong or oblong-elliptic, middle-sized (2–4 cm long, 0.5–2 cm wide) and usually lacking trichomes (Fernandez 1994).

There are several classifications of the vascular pattern types of the petiole. Hare (1942) distinguished three main types in transverse section: the U-, O- and I-shaped. Each type consists either of separate vascular bundles or of a solid mass of vascular tissue. In the family Ericaceae and Rhamnaceae there is a range of different forms, each characteristic of a particular genus. In some families (for example Proteaceae) the petiole has a high diagnostic value at the generic level.

Metcalfé and Chalk (1950) have devised a classification, in which nine types of steles are recognised, varying from the arc to the open or fused ring of xylem and phloem.

Superficial characters and vascular pattern of the petiole can be of descriptive and taxonomic value, whose systematic level does vary from one taxon to another. In some cases families can be recognised, in other cases genera, species, or varieties can be distinguished. The most complete system of

classification of major venation patterns in the petiole of woody dicotyledons is that of Howard (1962). He established 40 categories. His work has shown that the petiole classification can be applied successfully to the majority of the vascular system patterns found in all dicotyledons. His classification emphasises the importance of examining sections throughout the length of the petiole to completely understand the changes that occur in the vascular configuration.

Dickinson (1969) carried out comparative investigations in the family Dilleniaceae. The diversity present at the species level indicates that the petiole may be of taxonomic significance in the family.

Cells and tissues of the petiole will give several characteristics of immediate value in determining or limiting the classification. The nature and distribution of sclerenchyma as stone cells or fibres in the cortex of the petiole, the presence and distribution of latex or resin cells, canals, chlorenchyma, collenchyma, hypodermis or crystals are all of taxonomic value (Howard 1962).

MATERIAL AND METHODS

Species used in this study were collected from herbarium specimens. Hand cross-sections and sections with sliding microtome were made throughout the length of the petiole (at its proximal, median and distal parts, micrographs were taken of the median transverse sections). Herbarium specimens were boiled gently before fixation, and the petioles, together with a small part of the leaf, were embedded using a traditional method (Sárkány and Szalai 1966).

The antrachinon in the cortical cells was identified by the Borntraeger reaction and by ferrichlorid staining (Verzárné Petri 1979). The $\text{Ca}(\text{COO})_2$ content of the cells was detected by sulphuric acid reaction (Sárkány and Szalai 1966).

Samples were taken from 2–3 leaves for each species. Micrographs were taken using a HFX-DX microscope attached to a Nikon FX–35DX camera.

Sections were investigated with Nikon SE 102 microscope, the measurements were carried out with a ZEISS binocular microscope. The number of cell layers and the parameters of the transverse section were determined on the basis of 10 slides.

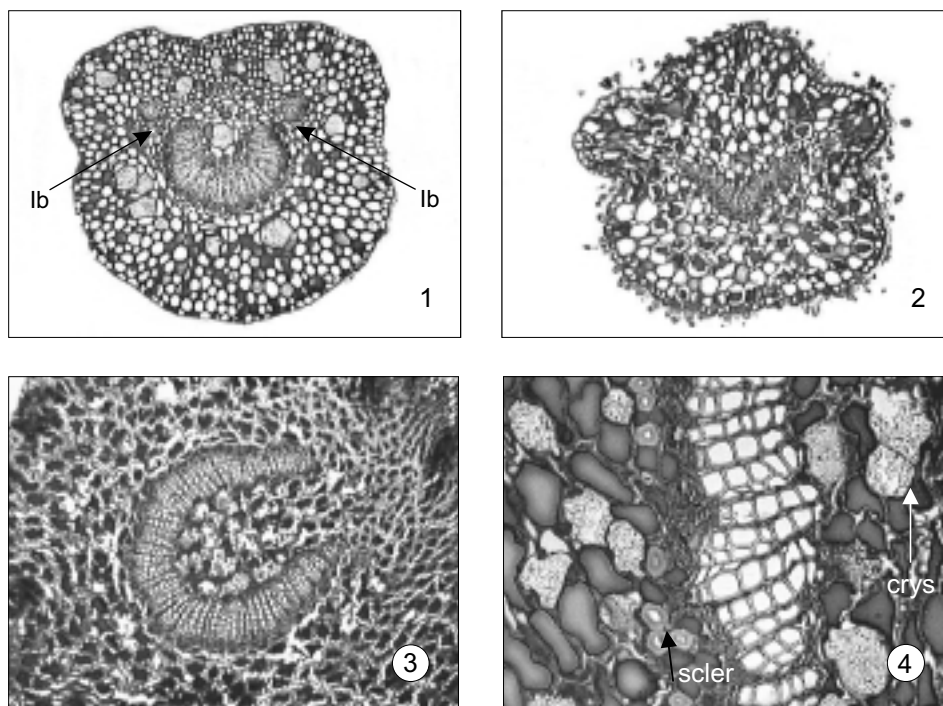
Specimens examined: *Rondeletia filisepala* Borhidi, coll. Buch 1165, Haiti. – *Rondeletia microphylla* Griseb., Prov. Pinar del Río. – *Rondeletia pachyphylla* Krug. et Urb. subsp. *pachyphylla*, coll. Wright 007593, Holguín. – *Rondeletia pycnophylla* Urb., coll. Ekman 6791, Holguín, Sierra Cristal.

RESULTS AND DISCUSSION

The species were characterised on the basis of the following criteria: outline of the petiole, features of epidermal and parenchyma cells, characteristics of the bundles, presence or absence of trichomes, cuticle, sclerenchyma and crystals.

Rondeletia filisepala (Fig. 1): Trichomes and adaxial hollow of the petiole are lacking. The epidermal cells are isodiametric or twice as high as wide and there is a thick cuticle on the epidermis.

The cortical cells are arranged in 8–9 layers, which were counted in the axis of petiole from the abaxial epidermis to the bundle. The cells contain antrachinon. $\text{Ca}(\text{COO})_2$ crystals are present mainly near the central vascular bundle.



Figs 1–4. Petiole anatomy in species of section *Pedicellares* (*Rondeletia*). – 1 = *R. filisepala*. Transverse section of the petiole, note the lateral bundles (lb). – 2 = *R. microphylla*. Transverse section of the petiole. – 3 = *R. pachyphylla*. The central vascular bundle. – 4 = *R. pycnophylla*. Transverse section of the petiole showing part of the central vascular strand, note crystal-containing cells (crys.) and sclerenchymatous fibres (scler.). (1, 2, 3: $\times 100$; 4: $\times 400$)

There are sclerenchymatous elements around the bundle. The proportion of xylem and phloem is 4:1 (thickness of xylem and phloem was measured in the middle region of the bundle). A pair of smaller lateral strands is present. The bundle shape does not change through the length of the petiole.

Rondeletia microphylla (Fig. 2): The petiole has trichomes, but no hollow. The epidermal cells are isodiametric, they are smaller in size to that of parenchyma cells under them. The cuticular layer is thin.

The cortical region consists of 4–6 layers of cells, which contain antrachinon. Crystals cannot be observed.

The vascular bundle is small, the ratio of xylem and phloem is 3:1, and there are no sclerenchymatous elements. Lateral bundles are absent.

Rondeletia pachyphylla subsp. *pachyphylla* (Fig. 3): Petiole is lacking trichomes and hollow. The epidermal cells are as high as wide, they are a bit smaller than the first layer of cortical cells. A thin cuticle is present on the epidermis.

Crystals are absent, the cortical parenchyma is made up by 6–7 layers of cells, the cells contain antrachinon.

The central bundle is middle-sized, the xylem is bordered by a narrow band of phloem. There are 1 or 2 lateral bundles surrounded by thick-walled sclerenchymatous elements. The central bundle can fuse with the lateral bundles towards the leaf blade, and both ends of the U-shaped bundle bending outwards.

Rondeletia pycnophylla (Fig. 4): The petiole has a small hollow, without trichomes. The epidermal cells are isodiametric. In transverse section their width and height is similar to that of parenchyma cells. A thin cuticle is present.

The parenchyma consists of 7–8 cell layers, containing antrachinon. Crystals are present everywhere in the cortical region.

The vascular tissue is of a medium size. The xylem, bordered by a thin phloem, contains several layers of thick-walled tracheary elements. Sclerenchymatous elements encircle the vascular system incompletely.

CONCLUSIONS

As a summary it can be stated that the wall of the cortical parenchyma cells is collenchymatously thickened and contain antrachinon. The central vascular tissue is horseshoe-shaped and open to the adaxial side. The abaxial side of xylem is bordered by phloem. The species differ in presence or absence of trichomes, crystals, in size, and number of lateral bundles. The results of this study show that certain characters are present in all of the four species but there is also substantial variation, which can help to distinguish them.

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