# Petiolar anatomy of some hitherto unstudied Acanthaceae

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# Abstract

Anatomy of petioles of 20 species belonging to 12 genera of the family Acanthaceae is investigated. The observations pertain to outline of petiole in T. S., the cell wall contours of epidermis, hypodermis, conjunctive tissue, besides the vascular tissue and cell inclusions, if any. The vascular tissue is generally resolved in an central arc and few vascular bundles. The various anatomical features differ taxon to taxon and hence found useful for their identification.

Keywords: Petiolar anatomy, Acanthaceae.

# INTRODUCTION

The family Acanthaceae consist of 240 genera and 2200 species (Lawrence, 1951). However Airy-Shaw (1973) informed about 250 genera for the family. A perusal of past literature indicates that while palynological, embryological, cytological and phytochemical investigations are fairly well in the Acanthaceae, studies in the vegetative anatomy in still relatively few and far between. According to Carlquist (1961) leaf is perhaps anatomically the most varied organ of angiospermic plants. He further stated that it is not surprising that this organ possesses many anatomical features of taxonomic significance. The present authors, from this view point, extended anatomical observations to hitherto unstudied taxa of the Acanthaceae. The results of our studies are being presented in this paper.

# MATERIALS AND METHODS

The plants were collected from various places like Tropical Botanic Garden and Research Institute, Palode, Thiruvanthapuram District (Kerala); Malbar Botanical Garden, Kozhikode (Kerala); Munnar, Idukki District (Kerala); Forest Research Institute, Peechi, Trichur (Kerala); Calicut University, Botanical Garden, Kozhikode (Kerala); Lal Bag Garden, Bangalore (Karnataka); Government Batanic Garden, Ootacamund (Tamilnadu) and Charanmal, District Dhule (Maharashtra). They were preserved in F.A.A. solution. For free hand transections middle part of petiol was selected. They were stained in safranin (1%) and fast green (1%) and mounted in D. P. X. after the customary method of dehydration. The unusual paraffin embedding method of Johansen (1940) has been followed especially for transactions of petioles. They were mounted in glycerin and made semi-permanent slides by ringing with nail paints. The cellular sketches were drawn using prism type of camera lucida. They were inked by using Camligraph or Rotring isographs technical pens with

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# 0.1, 0.2, 0.3 points.

## Abbreviations

Col : Collenchyma, Ct : Cystolith, Scl : Sclerenchyma, Sph :Sphaeraphides, Tc : Tannin cell.

# **OBSERVATIONS**

## Adhatoda beddomei C. B. Clarke

In T. S., it is circular in outline. Epidermis is the outermost delimiting layer. It is single layered and composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and have thick cuticle outside. Few multicellular trichomes are present on it. The epidermis is followed by six layered collenchymatous hypodermis. Few cells of collenchyma contain cystoliths. The vascular tissue is in the form of prominent central lunar-shaped arc. It is collateral. The pair of vascular bundles is present at the two corners towards adaxial side. They are collateral. The vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are rounded, thin walled and parenchymatous. The thickness of cell walls of conjunctive tissue gradually decreases towards the center. Some cells of it contain tannin (Fig.1).

# Andrographis elongata (Vahl ) T. And.

In T. S., it is more or less circular in outline and winged adaxially forming a shallow channel. Epidermis is the outermost delimiting layer. It is single layered and composed of rounded, compactly arranged cells. Their outer walls are thicker than the inner ones and with thick cuticle outside. Few multicellular trichomes are present on it. The epidermis is followed by three layered collenchymatous hypodermis. The vascular tissue is in the form of prominent central lunar-shaped arc. It is collateral. The pair of vascular bundles is present at the two corners towards adaxial side. They are collateral. The vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are rounded, thin walled and parenchymatous. Their thickness gradually decreases towards the center (Fig.2).

## Andrographis macrobotrys Nees

In T. S., it is more or less circular in outline with wings adaxially forming a shallow channel. Epidermis is the outermost delimiting layer. It is single layered and composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and with thick cuticle outside. Some cells of epidermis shows cystoliths. The epidermis is followed by four layered collenchymatous hypodermis. The vascular tissue is in the form of prominent central lunar-shaped arc. It is collateral. The pair of collateral vascular bundles is present at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are rounded, thin walled and parenchymatous. The thickness of cells gradually decreases towards the center. Some cells contain tannin (Fig.3).

## Andrographis stellulata C. B. Clarke

In T. S., it is more or less circular in outline and winged adaxially forming a shallow channel. Epidermis is the outermost delimiting layer. It is single layered and composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and have thick cuticle outside. Few cells of it contain cystoliths. The epidermis is followed by five layered collenchymatous hypodermis. The vascular tissue is in the form of a conspicuous central horse shoe-shaped arc. It is collateral and capped outside by two layers of sclerenchyma. The pair of collateral vascular bundles is present at the two corners towards adaxial side. The vascular tissue lies embedded in the conjunctive tissue. The cells of latter rounded, thin walled and parenchymatous. The thickness of their cell walls gradually decreases towards the center. Some cells contain tanin(Fig.4).

# Andrographis wightiana Arn.ex Nees

In T. S., it is more or less circular in outline and winged adaxially forming a shallow channel. The outermost delimiting layer is epidermis, which is single layered and composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and thickly cuticled outside. Some cells of it contain cystoliths. The three layered collenchymatous hypodermis follows the epidermis from inner side. The vascular tissue is resolved in a prominent central lunar-shaped arc. It is collateral and capped outside by two layered sclerenchyma. The pair of collateral vascular bundles is associated at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are rounded or polygonal, thin walled and parenchymatous. The thickness of cell walls of conjunctive tissue gradually decreases centripetally (Fig.5).

# Barleria prattensis Santapau

In T. S., it is more or less circular in outline, winged adaxially and channeled shallowly. Epidermis is the outermost delimiting layer. It is single layered and composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and with thick cuticle outside. Few cells of epidermis contain cystoliths. The epidermis is followed by three to four layered collenchymatous hypodermises. The vascular tissue extends as a prominent central lunar-shaped arc. It is collateral and capped outside by two layered sclerenchyma. The pair of vascular bundles is present at the two corners towards adaxial side which are collateral. The vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are rounded thin walled and parenchymatous. The thickness of cell walls of conjunctive tissue gradually decreases centripetally (Fig.6).

## Beloperone comosa Nees

In T. S., it is circular in outline. The outermost delimiting layer is epidermies. It is single layered and composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and with thick cuticle outside. Few multicellular and glandular trichomes are present on it and some cells of it contain cystoliths. The epidermis is followed from within by three layered collenchymatous hypodermis. The vascular tissue is resolved to a prominent central lunar-shaped arc. It is collateral. The pair of collateral vascular bundles is associated at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are polygonal, thin walled and parenchymatous. The thickness of cell walls of it gradually diminishes towards the center (Fig.7).

#### Beloperone nemorosa Nees

In T. S., it is circular in outline. Epidermis is the outermost delimiting layer. The squarish, compactly arranged cells compose the epidermis. Their outer walls are thicker than the inner ones and thick cuticle outside. Few multicellular trichomes are present on it. The epidermis is followed from within by four layered collenchymatous hypodermis. The vascular tissue is in the form of a conspicuous central lunar-shaped arc. It is collateral and capped outside by two layers of sclerenchyma. The pair of collateral vascular bundles is present at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are polygonal or rounded, thin walled and parenchymatous. The thickness of cell walls of conjunctive tissue gradually decreases towards the center (Fig.8).

## Dicliptera foetida (Forsskal) Blatter

In T. S., it is more or less circular in outline with two adaxial wings. Epidermis is the outermost delimiting layer which is single layered and composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and thick cuticle outside. Few multicellular trichomes are present on it and some cells shows cystoliths. The epidermis is followed from within by six to eight layered collenchymatous hypodermises. The vascular tissue is in the form of a prominent central lunar-shaped arc. It is collateral and capped outside by two layers of sclerenchyma. The pair of collateral vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are rounded, thin walled and parenchymatous. The thickness of cell walls of conjunctive tissue gradually diminishes towards the center (Fig.9).

#### Fittonia gigantea Linden ex Andre.

In T. S., it is circular in outline. The outermost delimiting layer is the epidermis. It is solitary and consists squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and with thick cuticle outside. Few multicellular trichomes are present on it. The epidermis is followed by four to five layered collenchymatous hypodermises. The vascular tissue is in the form of a conspicuous central lunar-shaped arc. It is collateral. The pair of collateral vascular bundles is present at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The cells of parenchymatous conjunctive tissue are rounded and thin walled. The thickness of cell walls of it gradually decreases towards the center (Fig.10).

## Goldfussia anysophylla (G. Lodd) Nees

In T. S., it is more or less circular in outline and winged adaxially forming a shallow channel. Epidermis is the outermost delimiting layer. It is single layerd and contains squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and thick cuticle outside. Some cells of it contain cystoliths. The epidermis is followed from inside by four layered collenchymatous hypodermis. The vascular tissue is in the form of a prominent central lunar-shaped arc. It is collateral and capped outside by two layers of sclerenchyma. The pair of collateral vascular bundles is associated at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are rounded, thin walled and parenchymatous. Some cells of it shows sphaeraphides. The thickness of cell walls of conjunctive tissue gradually diminishes towards the center (Fig.11).

# Graptophylum pictum (L.) Griffith

In T. S., it is circular in outline. The single layerd outermost epidermal layer is composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and have thick cuticle outside. Few multicelluer trichomes are present on it and some cells of it contain cystoliths. The epidermis is followed from within by five to six layered collenchymatous hypodermises. The vascular tissue is in the form of a prominent central horse shoeshaped arc. It is collateral and capped outside by two layers of sclerenchyma. The pair of collateral vascular bundles is present at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The cells of latter rounded, thin walled and parenchymatous. The thickness of their cell walls gradually decreases towards the center. Some cells of collenchymas and conjunctive tissue contain sphaeraphides (Fig.12).

## Hygrophila schulli (Buch – Ham.) M.R. Almeida and S.M. Almeida

In T. S., it is more or less circular in outline with winged adaxially forming a deeply channel. Epidermis is the outermost delimiting layer. It is single layered and composed of rounded, compactly arranged cells. Their outer walls are thicker than the inner ones and have thick cuticle outside. The epidermis is followed inside by three layered collenchymatous hypodermis. The vascular tissue is in the form of a prominent central lunar-shaped arc. It is collateral. The four collateral vascular bundles are present at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are rounded, thin walled and parenchymatous. The thickness of cell walls of it gradually decreases towards the center (Fig.13).

## Hypoestes sanguinolenta Hook.

In T. S., it is circular in outline. The outermost delimiting layer is epidermis, which is single layered and composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and thick cuticle outside. Some cells of epidermis contain cystoliths and few multicellular trichomes are present on it. It is followed from within by three to five layered collenchymatous hypodermises. The vascular tissue is in the form of a prominent central lunar-shaped arc. It is collateral and capped outside by two layers of sclerenchyma. The pair of collateral vascular bundles is present at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The cells of latter rounded, thin walled and parenchymatous. The thickness of cell walls of it gradually decreases towards the center (Fig.14).

## Justicia carnea Edward F. Gilman

In T. S., it is circular in outline. The outermost delimiting layer is epidermis. It is single layered and composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and have thick cuticle outside. Few multicellular trichomes are present on it. Some cells of epidermis shows cystoliths. The epidermis is followed within by five layered collenchymatous hypodermis. The vascular tissue is in the form of a prominent central lunar-shaped arc. It is collateral. The pair of collateral vascular bundles is present at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The parenchymatous cells of conjunctive tissue are rounded and thin walled. The thickness of cell walls of the conjunctive tissue gradually decreases towards the center (Fig.15).

## Justicia trinervia Vahl

In T. S., it is more or less circular in outline and with adaxial wings Epidermis is the outermost delimiting layer. It is single layered and composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and have thick cuticle outside. The epidermis is followed from inside by three to four layered collenchymatous hypodermis. The vascular tissue is in the form of a prominent central lunar-shaped arc. It is collateral. The four collateral vascular bundles are present at the two corners towards adaxial side. The vascular tissue is embedded in the conjunctive tissue. The cells of conjunctive tissue are polygonal or rounded, thin walled and parenchymatous. The thickness of cell walls of the conjunctive tissue gradually decreases towards the center (Fig.16).

## Justicia wynaddensis (Nees) Heyne ex T. Ander.

In T. S., it is circular in outline. The single layered epidermis is composed of barrel-shaped, compactly arranged cells. Their outer walls are thicker than the inner ones and with thick cuticle outside. Few multicellular trichomes are present on it. The epidermis is followed internally by three layered collenchymatous hypodermis. The vascular tissue is in the form of a prominent central lunar-shaped arc. It is collateral and capped outside by two layered sclerenchyma. The pair of collateral vascular bundles is present adaxially. The vascular tissue is embedded in the conjunctive tissue. The parenchymatous cells of the conjunctive tissue are polygonal and thin walled. The thickness of cell walls of the conjunctive tissue

gradually decreases towards the center (Fig.17).

## Libonia floribunda K. Koch

In T. S., it is circular in outline. The outermost delimiting layer is epidermis, which is single layered and contains rounded compactly arranged cells. Their outer walls are thicker than the inner ones and have thick cuticle outside. Few unicellular trichomes are present on it. The epidermis is followed internally by three layered collenchymatous hypodermis. Few cells of it contain cystoliths and sphaeraphides. The vascular tissue is in the form of a prominent central lunar-shaped arc. It is collateral and capped outside by one layered sclerenchyma. The pair of vascular bundles is present at two corners towards adaxial side which are collateral. The vascular tissue is embedded in the conjunctive tissue. The cells of the conjunctive tissue are rounded or polygonal, thin walled and parenchymatous. The thickness of cell walls of the conjunctive tissue gradually decreases towards the center. Some cells of conjunctive tissue contain sphaeraphides (Fig.18).

# Mackenzia integrifolia (Dalz.) Bremek.

In T. S., it is more or less circular in outline. The outermost delimiting layer is epidermis. It is single layered and composed of squarish and elongated unequal-sized compactly arranged cells. Their outer walls are thicker than the inner ones and have thick

cuticle outside. Few unicellular trichomes are present on it and some cells of it contain cystoliths. The epidermis is followed by four to five layered collenchymatous hypodermises. The vascular tissue is in the form of a prominent central lunar-shaped arc. It is collateral. The four vascular bundles are present adaxially, which are collateral. The vascular tissue is embedded in the conjunctive tissue. The cells of the conjunctive tissue are rounded, thin walled and parenchymatous. The thickness of cell walls of the conjunctive tissue gradually diminishes towards the center (Fig.19).

# Neuracanthus sphaerostaschys (Nees) Dalz.

In T. S., it is more or less circular in outline. It is winged and deeply channeled adaxially. The single layered epidermis is the outermost delimiting layer which is composed of squarish, compactly arranged cells. Their outer walls are thicker than the inner ones and thick cuticle outside. Few cells of it contain cystoliths. The epidermis is followed within by four layered collenchymatous hypodermis. The vascular tissue is in the form of a prominent central lunar-shaped arc. It is collateral and capped outside by two layers of sclerenchyma. The four pair of collateral vascular bundles are present at the two corners towards adaxil side. The vascular tissue is embedded in the conjunctive tissue. The cells of the conjunctive tissue are rounded, thin walled and parenchymatous. The thickness of cell walls of the conjunctive tissue gradually decreases towards the center (Fig.20).







## DISCUSSION

In T.S., the petioles are usually circular or broadly circular in outline. The petioles in all the species investigated are adaxially channeled. The channels are generally shallow. They are deep in *Hygrophila schulli* and *Neuracanthus sphaerostaschys*.

Epidermis is the outermost delimiting layer. It is generally single layered containing small cells. The outer cell walls of epidermal cells are generally thicker than the inner ones. Moderately thick to thick cuticle is overlaid on the outside. The shape of epidermal cells differs from species to species. In majority of taxa, they are squarish in outline. They are rounded in certain species e.g. *Andrographis elongata, Hygrophila schulli* and *Libonia floribunda*. They are clearly barrel-shaped e.g. *Justicia wynaddensis*. The epidermal layer is interrupted at some places due to the presence of cystoliths.

Usually, the epidermis is followed from within by the hypodermis. The hypodermal cells are collenchymatous in all the species. They are generally rounded in shape and have obviously thick walls. The hypodermal layers are two to ten forming continues layer throughout. Out of 20 species studied, about 06 species exhibit

two to three layered hypodermis. They are four to five layered in 12 species. The hypodermis consists of six to eight layers in 02 species. Occurrence of collenchymatous hypodermal layers in the petiolar region can be obviously conceived from the stand point of mechanical function.

The petiole in Acanthaceae generally receives a solitary vascular strand from the nodal vasculature as in most of the sympetalous taxa (cf. Metcalfe and Chalk, 1950). This vascular strand during its traverse through the petiolar region is resolved differently. In all species the petiolar vasculature is mostly resolved into a conspicuous or medium-sized central arc and a few to many vascular bundles. Out of 20 species studied, in the petioles in 16 species the vascular tissue is comprised of a prominent central vascular arc and pair of adaxial vascular bundles. There are certain species wherein the petiolar vasculature is resolved into four vascular bundles, besides the central arc e.g. Hygrophila schulli, Justicia trinervia and Mackenzia intergrifolia. In case of Neuracanthus sphaerostaschys, the vasculature consists of a central prominent arc and eight vascular bundles. The vascular bundles in all species are collateral. The central vascular arc assume generally two types of shapes viz., lunar-shaped and horse shoe-shaped. In

about 18 taxa, the former condition prevails, whereas in rest others it is horse shoe –shaped. In case of 10 species, the central vascular arc is capped abaxially by sclerenchyma. The sclerenchyma is usually two to three continuously layered. It is single layered in case of *Libonia floribunda*. The vascular and other tissues are generally surrounded by conjunctive tissue. The cells of conjunctive tissue are parenchymatous and thin walled. They are rounded or oval and polygonal in shape. The larger parenchymatous cells are generally located in the central region of the petiole. They decrease in size gradually towards the outside. Thickness of their cells walls generally diminishes towards the centre of the petiole. The cells are polygonal throughout in case of *Beloperone comosa* and *Justicia carnea*.

As stated earlier, the occurrence of cystoliths is noteworthy in the family Acanthaceae. However, they are wanting in petiolar region of some taxa of the present account e.g. *Andrographis elongata, Beloperone comosa, Fittonia gigantea, Hygrophila schulli, Justicia trinervia* and *J. wynaddensis.* When present, they are distributed usually in the epidermal and hypodermal layers. Another notable occurrence of cell inclusions is the sphaeraphides. They are usually located in the cells of conjunctive tissue and hypodermis. Of the present account, they are present in 03 species e.g. *Goldfussia anysophylla, Graptophylum pictum* and *Libonia floribunda*. The present investigator also noted tannin cells in the conjunctive tissue of some taxa e.g. *Adhatoda beddomei, Andrographis macrobotrys* and *A. stellulata*.

Petiole is one of the organs in the acanthaceous taxa which received a little attention of the anatomists. Metcalfe and Chalk's work (1950) is probably only report on its anatomy. They gave an account of petiole anatomy with particular emphasis on vascular system. They also illustrated nodal anatomy although topographically. They noted petioles in the various genera of the Acanthaceae containing a central vascular arc with few or none vascular bundles. They also pointed out occurrence of a continuous vascular cylinder in the petiole.

Although the leaves provide many anatomical characters of taxonomic significance, they are derived from the blade (lamina). The petioles are generally neglected in anatomical studies. Petiole anatomical data, as with most all other types of data, can also be used. This was indicated well by Metcalfe and Chalk (1950). They pointed out importance of vascular pattern in the petiole of some genera of Acanthaceae. However, there are also anatomical data of the petioles which can be fruitfully employed in systematic thinking. This aspect of anatomy of petiole has been largely overlooked. Schofield (1968) conveniently employed petiolar anatomy of the Guttiferae and related families as their diagnostic features. The present investigator investigated petiolar anatomy of 20 species in the family. The features such as shape of epidermal cells, number of hypodermal layers, occurrence of petiole wings, pattern of vascular supply, cell contours of conjunctive tissue, presence or absence sclerenchyma associated with vascular tissue and cell inclusions (sphaeraphides, cystoliths, etc.) appeared as taxonomic significance specially at specific levels. Their diagnostic utility is already explained earlier Tayade and Patil (2008) and Shisode and Patil (2008) also found these features useful to distinguish the taxa of family Convolvulaceae and the order Celastrales respectively.

Thakur and Patil (2009) also employed them similary in the euphorbiaceous taxa. The literature survey indicated no petiolar anatomical study in the Acanthaceae. It appears that this study should be taken up which will bring round the role of petiole anatomy and taxonomy of other disputed taxa in the Acanthaceae and its related families.

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