

ANATOMICAL STUDIES ON THE GENUS *BLUMEA* DC. (ASTERACEAE, INULEAE, PANICULATAE) IN PENINSULAR INDIA

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The genus *Blumea* is one of the largest genera in the tribe Inuleae. In this study, stem and leaf anatomical features of the genus *Blumea* in the section *Paniculatae* DC. of *Randeria* are investigated. *Blumea laciniata* DC., *B. barbata* DC., *B. sessiliflora* Decne., *B. membranacea* DC., *B. axillaris* DC., *B. virens* Wall. ex DC., *B. lacera* (Burm.f.) DC., and *B. fistulosa* Kurz. are the eight species present in the Peninsular Indian region. The samples collected from different areas of Peninsular India were studied anatomically using the conventional method. The results revealed that generally the species share similar anatomical characteristics but there are some remarkable differences. With respect to the results, trichome morphology appears to be of taxonomic value. The species can be separated on the basis of trichome types, the presence of secretory canals, and the layer of palisade tissue. An identification key to the taxa is provided based on the anatomical features of the leaf and stem.

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مطالعات تشریحی جنس *Blumea* DC. (Asteraceae, Inuleae, Paniculatae) از شبه جزیره هند

موتالاولیتیل دیلشا: مرکز تحصیلات تکمیلی تحقیقات گیاهشناسی دانشکده سنت جوزف

نادوویلاک کان دییل انجالی: مرکز تحصیلات تکمیلی تحقیقات گیاهشناسی دانشکده سنت جوزف

جرج ساتیش: مرکز تحصیلات تکمیلی تحقیقات گیاهشناسی دانشکده سنت جوزف

جنس *Blumea* یکی از بزرگترین جنس‌های قبیله Inuleae است. در این مطالعه صفات تشریحی برگ و ساقه جنس *Blumea* در بخش *Paniculatae* در سیستم رده‌بندی *Randeria* مورد بررسی قرار گرفته است. گونه‌های *B. laciniata* DC., *B. barbata* DC., *B. sessiliflora* Decne., *B. membranacea* DC., *B. axillaris* DC., *B. virens* Wall. ex DC., *B. lacera* (Burm.f.) DC., *B. fistulosa* Kurz. هشت گونه‌ای هستند که در شبه جزیره هند حضور دارند. نمونه‌های جمع‌آوری شده از مناطق مختلف هند با استفاده از روش‌های معمول مورد بررسی تشریحی قرار گرفتند. نتایج نشان دادند که همه گونه‌ها دارای صفات تشریحی مشابهی هستند ولی تفاوت‌های جالب توجهی نیز در میان آنها وجود دارد. با توجه به نتایج مشخص شد که ریخت‌شناسی کرک‌ها دارای ارزش تاکسونومیکی است و همچنین گونه‌ها می‌توانند بر اساس انواع کرک، حضور کانال‌های ترش‌چی و لایه بافت نردبانی از یکدیگر تفکیک شوند. بر اساس صفات تشریحی، یک کلید برای شناسایی گونه‌ها ارائه می‌گردد.

INTRODUCTION

Asteraceae is an advanced and highly specialized family, distributed in the tropics and warm temperate

regions of South, South East, and East Asia, Africa including Madagascar and central South America. About 950 genera and 20000 species are representing

the family all over the globe (Rahman 2008). Herbaceous plants represent the greatest proportion of this family, and trees and shrubs include two percentages (Lawrence 1966).

The genus *Blumea* DC. (Asteraceae), belongs to the tribe Inuleae, subtribe Plucheinae. Natural habitats of this genus include disturbed areas such as roadsides, railway lines, forest edges, etc. and some taxa are present in evergreen forests (Daruwalla 1995). Traditional Chinese medicine uses many of the taxa of *Blumea*. Its highest diversity is seen in tropical Asia and also distributed in Africa and Oceania too.

Randeria (1960) divided the genus *Blumea* into 7 sections containing a total of 49 species over the entire geographical range. These sections are *Semivestitae* DC., *Macrophyllae* DC., *Sagittatae* Randeria, *Hieraciifoliae* Randeria, *Paniculatae* DC., *Oxyodontae* DC., and *Dissitiflorae* DC. The sections are separated on the basis of leaf, inflorescence, and receptacle characters as well on some anatomical features. The genus is represented by 36 taxa in India (Hooker 1882).

The characters defining the section *Paniculatae* DC. are herbaceous form, leaves rounded or tapering at the base, the margins never spiny toothed, and the upper surface variously pubescent. Upper epidermal cells with sinuate walls, the stomata on both surfaces. Capitula paniculate to glomerulate. Outer phyllaries linear. Receptacle alveolate, glabrous to pubescent. Corolla lobes of bisexual florets with glandular and unicellular hairs. Female florets with filiform, tubular, equally 2- to 4-lobed corollas, glabrous (except *B. manillensis*). Another tails are longer to shorter than the thickened portion of the filament. Achenes ribbed or not ribbed (Randeria 1960). *Blumea* section

Paniculatae is represented by eight species in Peninsular India (Hajra & al. 1995).

Anatomical studies of the family Asteraceae have been carried out in the past by many researchers (Lotocka & Geszprych 2004). Milan & al. (2016) studied the comparative morphology and anatomy of several genera of Asteraceae. Metcalfe & Chalk (1950) reported the anatomical diversity of the Asteraceae is commonly manifested in the structures of the leaves and secretory structures such as ducts, hydathodes, trichomes, and glandular appendices are of great taxonomical importance and their restricted distribution has an important diagnostic value. Schizogenous secretory ducts, resin canals, or resin cavities, are found in a few genera in the tribe Inuleae (Funk & al. 2009).

Several authors commented on the usefulness of anatomical characters for resolving the taxonomy of some plant groups. But anatomical features have not been fully explored in the genus *Blumea*.

The objective of this study is to investigate the stem and leaf anatomical characters of *Blumea* species in the section *Paniculatae* and also to prepare an identification key to the species.

MATERIALS AND METHODS

Fresh materials and preserved (formalin or FAA treated) specimens of 8 species of *Blumea* including *B. laciniata* DC., *B. barbata* DC., *B. sessiliflora* Decne., *B. membranacea* DC., *B. axillaris* DC., *B. virens* Wall. ex DC., *B. lacera* (Burm.f.) DC., and *B. fistulosa* Kurz. were used for this study. The voucher specimens are deposited in the herbarium of St. Joseph's College (Autonomous), Devagiri, Calicut (DEV). Information about the voucher specimens is listed in Table 1.

Table 1: Collection data of the examined species.

Species	Collection Data
<i>Blumea barbata</i> DC.	Kodikuthimala, Malappuram District, Kerala State, Dilsha M. V., 6942, DEV, 24/11/2018
<i>B. sessiliflora</i> Decne.	Nilambur, Malappuram District, Kerala State, Dilsha M. V., 8659, DEV, 10/11/2020
<i>B. membranacea</i> DC.	Kakkayam, Kozhikode District, Kerala State, Dilsha M. V., 8670, DEV, 30/11/2020
<i>B. axillaris</i> DC.	Pavna dam site, Maharashtra State, Dilsha M. V., 6985, DEV, 26/12/2019
<i>B. virens</i> Wall. ex DC.	Nilambur, Malappuram District, Kerala State, Dilsha M. V., 8673, DEV, 11/12/2020
<i>B. lacera</i> (Burm.f.) DC.	Shiradi Ghat, Karnataka State, Dilsha M. V., 8651, DEV, 02/02/2020
<i>B. fistulosa</i> Kurz.	Pulney hills, Tamil Nadu state, Dilsha M. V., 6954, DEV, 20/02/2019
<i>B. laciniata</i> DC.	Shiradi Ghat, Karnataka State, Dilsha M. V., 8649, DEV, 02/02/2020



Fig. 1. Habit of *Blumea* spp. A, *B. sessiliflora*; B, *B. membranacea*; C, *B. laciniata*; D, *B. axillaris*; E, *B. lacera*; F, *B. virens*; G, *B. fistulosa*; H, *B. barbata*.

The samples for the anatomical studies were collected from different areas of Peninsular India. The collected materials were cleaned using tap water. The stems and leaves of the collected materials were cut into suitable dimensions. Freehand sectioning of the material was done. Hand-cut sections of stems and leaves were taken manually using a razor blade. The leaves were cut into delicate sections using a pith. After free-hand sectioning of the materials, they were transferred into safranin or toluidine blue solutions for one minute for staining (Johansen 1940). The stained sections were washed using distilled water. Mounted the sections using a drop of glycerin and examined under a stereo microscope at a magnification of 4x for a ground plan of the stem, 10x for the epidermis, cortex, pith, and 20 x for epidermis, cortex, and pith for stems and 10x for leaves.

The photographs of the transverse sections of both stem and leaves were captured using Leica DFC 290, a camera attached to Leica DM 100 trinocular research microscope. Anatomical features were described according to the terminology of Esau (1960).

RESULTS

General Anatomical Characters of the Genus *Blumea*

Stem Anatomy

Epidermis is made up of single-layered, small cells with depositions. A thick cuticle is present. Multicellular trichomes were identified in all stems. The cortex is differentiated into the hypodermis, middle cortex, and inner cortex. Hypodermis is made up of collenchyma cells with depositions. The middle cortex is made up of chlorenchyma cells, and the inner cortex is made up of parenchymatous cells. Depositions are present in the hypodermis of the stems. Numerous vascular bundles are present. Sclerenchyma cells are seen just above the phloem. Phloem is made up of sieve cells, companion cells, fibers, and phloem parenchyma. Xylem is differentiated into metaxylem and protoxylem. Metaxylem is oriented towards the periphery and protoxylem vessels are oriented towards the pith, making the xylem end arch. Medullary rays are composed of 3-4 vertically arranged cells. Pith is made up of large, spherical, thin-walled parenchymatous cells with intercellular spaces. The cells just below the vascular bundles are small, thick-walled, and parenchymatous (Figs. 2-5).

Multicellular capitate glandular and non-glandular hairs are present in *Blumea fistulosa*, *B. sessiliflora*, *B. laciniata*, *B. axillaris*, *B. barbata*, and *B. membranacea*, but in *B. lacera*, only one type of hair is

present. Multicellular non-glandular hairs are absent in *B. lacera*. Both types of glandular and non-glandular hairs are absent in *B. virens*. Stem ridges are present in *B. fistulosa*, *B. sessiliflora* and *B. membranacea*. Numerous secretory epithelial canal cells are present in the parenchymatous inner cortex of the stems of *B. fistulosa*, *B. sessiliflora*, *B. membranacea*, and *B. laciniata*. Epithelial canal cells are absent in other stems. Yellow-colored depositions are present in *B. membranacea*, *B. fistulosa*, *B. sessiliflora*, and *B. laciniata*.

Leaf Anatomy

Epidermis is made up of single-layered, small, thick-walled cells with thick cuticles. Multicellular epidermal hairs are present on the leaves. Mesophyll is differentiated into palisade tissue and spongy tissue. Spongy cells are made up of 1-4 small, spherical, irregularly arranged cells with chloroplast. The hypodermis of the midrib is composed of 2-3 layers of collenchyma cells. Ground tissue is made up of large, irregularly shaped parenchymatous cells with intercellular spaces. Vascular bundles are few in the midrib portion, ranging from 3-7. Vascular bundles are composed of the xylem and phloem. The phloem is oriented towards the lower epidermis and the xylem is oriented towards the upper epidermis. Xylem is differentiated into metaxylem vessels and protoxylem vessels. Tracheary elements are also present. Sclerenchyma cells are present just above the xylem and just below the phloem (Figs. 6-7).

Both capitate multicellular glandular and non-glandular hairs are present in *B. sessiliflora*, *B. laciniata*, and *B. lacera*. Multicellular non-glandular hairs are present in *B. fistulosa*, *B. membranacea* and *B. barbata*, *B. virens*, and *B. axillaris*. Peltate glandular trichomes are present only in *B. axillaris*. The upper epidermis of the leaf blade is larger than the lower epidermis in the case of *B. barbata*. Leaf lamina is broad in *B. fistulosa*, and *B. laciniata*, in other cases it is narrow. Palisade tissue is a single layer in the case of *B. laciniata*, *B. lacera*, *B. barbata*, and *B. membranacea*. More than one layer of palisade tissue is seen in *B. fistulosa*, *B. axillaris*, and *B. virens*. Epithelial canal cells are seen just below the vascular bundles in the case of *B. fistulosa*, *B. sessiliflora*, *B. laciniata*, and *B. membranacea*. Epithelial cells were absent in *B. axillaris*, *B. barbata*, *B. lacera*, and *B. virens*. In *B. virens*, *B. axillaris*, and *B. fistulosa*, the number of vascular bundles in the midrib ranges from 3-5; but it ranges from 4-6 in *B. lacera* and *B. barbata*, and from 5-7 in *B. sessiliflora*, *B. laciniata*, and *B. membranacea*.

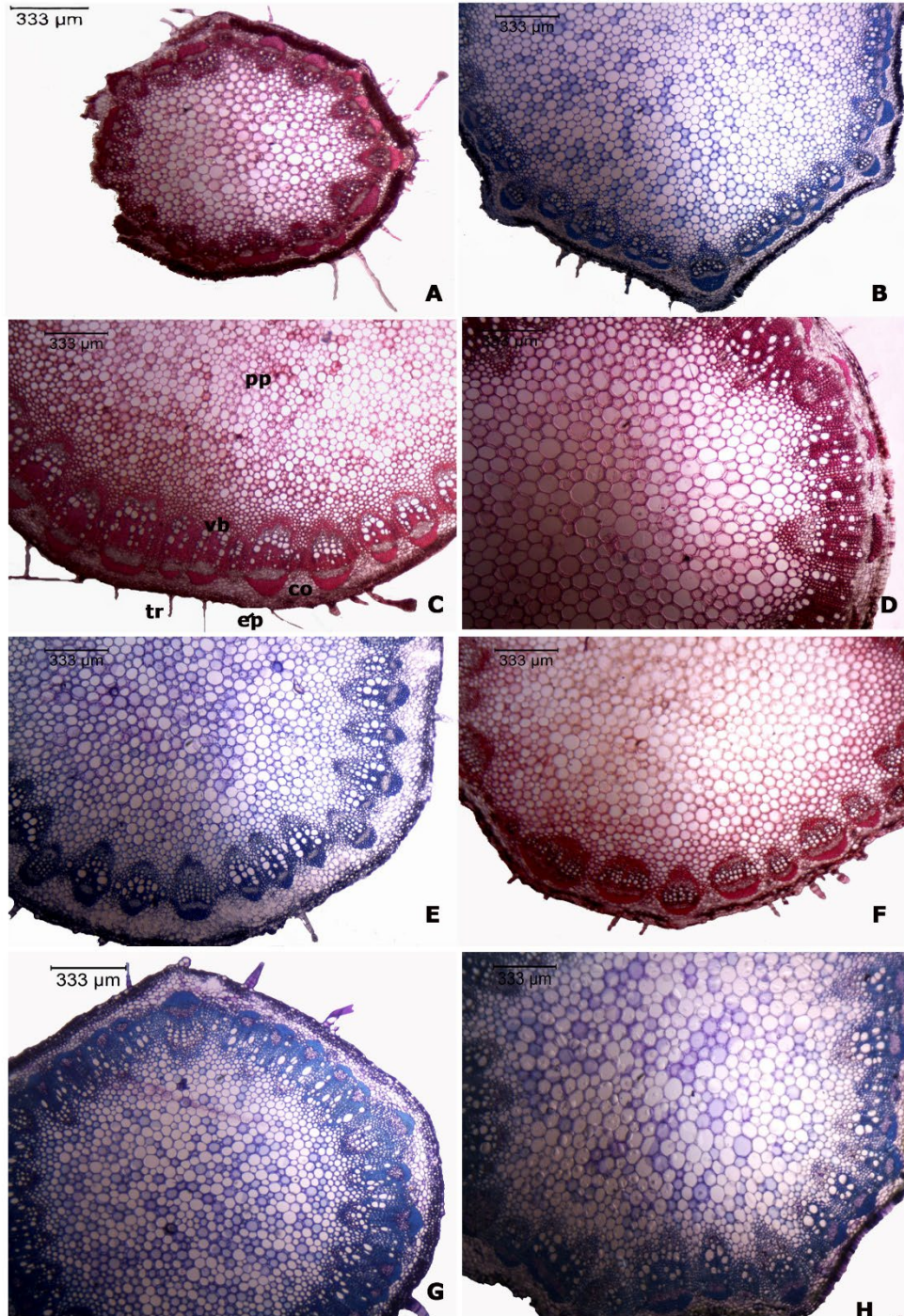


Fig. 2. Ground Plan of Stem (4X). A, *B. fistulosa*; B, *B. sessiliflora*; C, *B. laciniata*; D, *B. axillaris*; E, *B. lacera*; F, *B. barbata*; G, *B. membranacea*; H, *B. virens*; ep, epidermis; co, cortex; tr, trichome; vb, vascular bundle; pp, pith parenchyma.

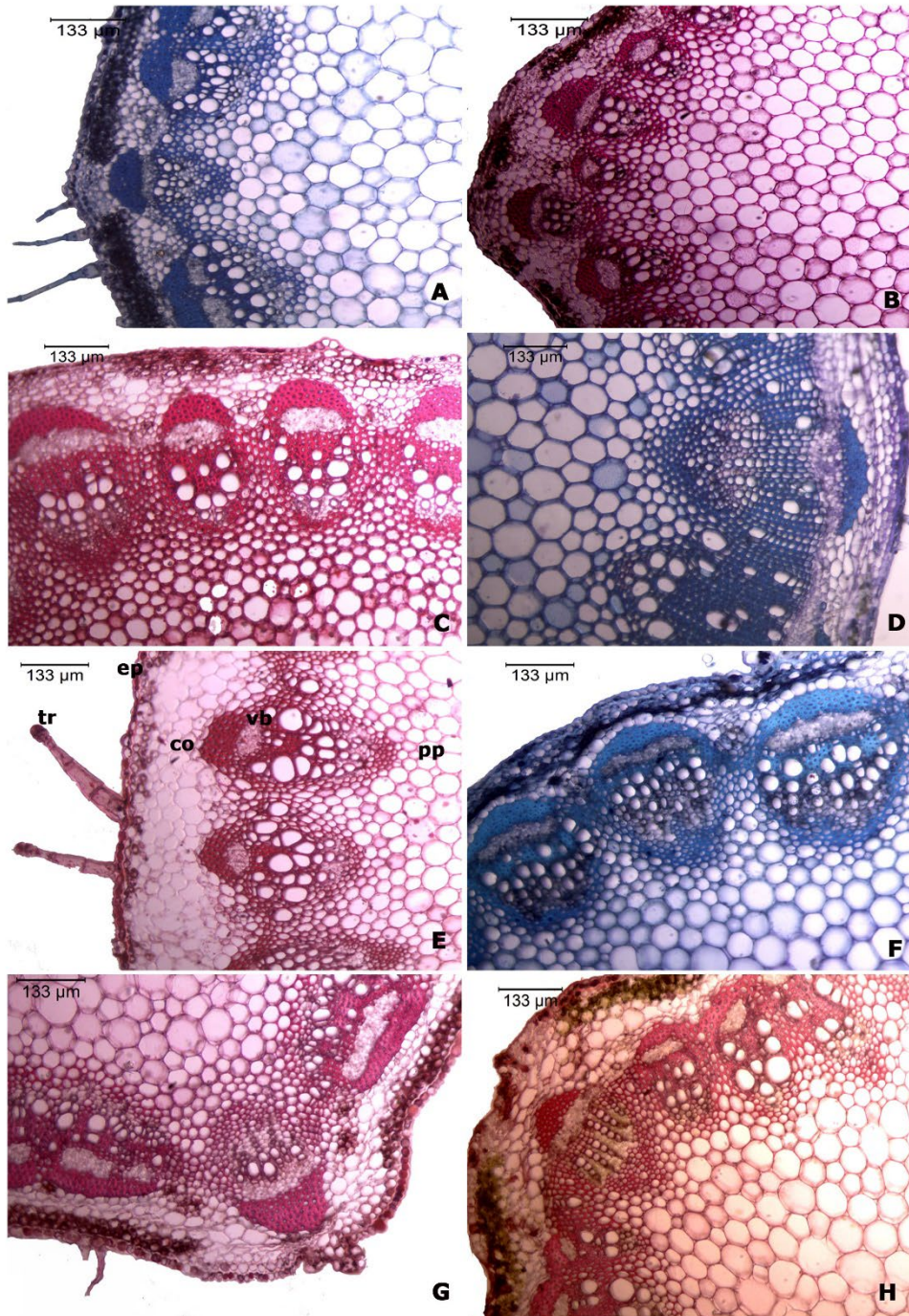


Fig. 3. Section showing the epidermis, cortex, and Vascular bundles (10X). A, *B. fistulosa*; B, *B. sessiliflora*; C, *B. laciniata*; D, *B. axillaris*; E, *B. lacera*; F, *B. barbata*; G, *B. membranacea*; H, *B. virens*; ep, epidermis; tr, trichome; co, cortex; vb, vascular bundle; pp, pith parenchyma.

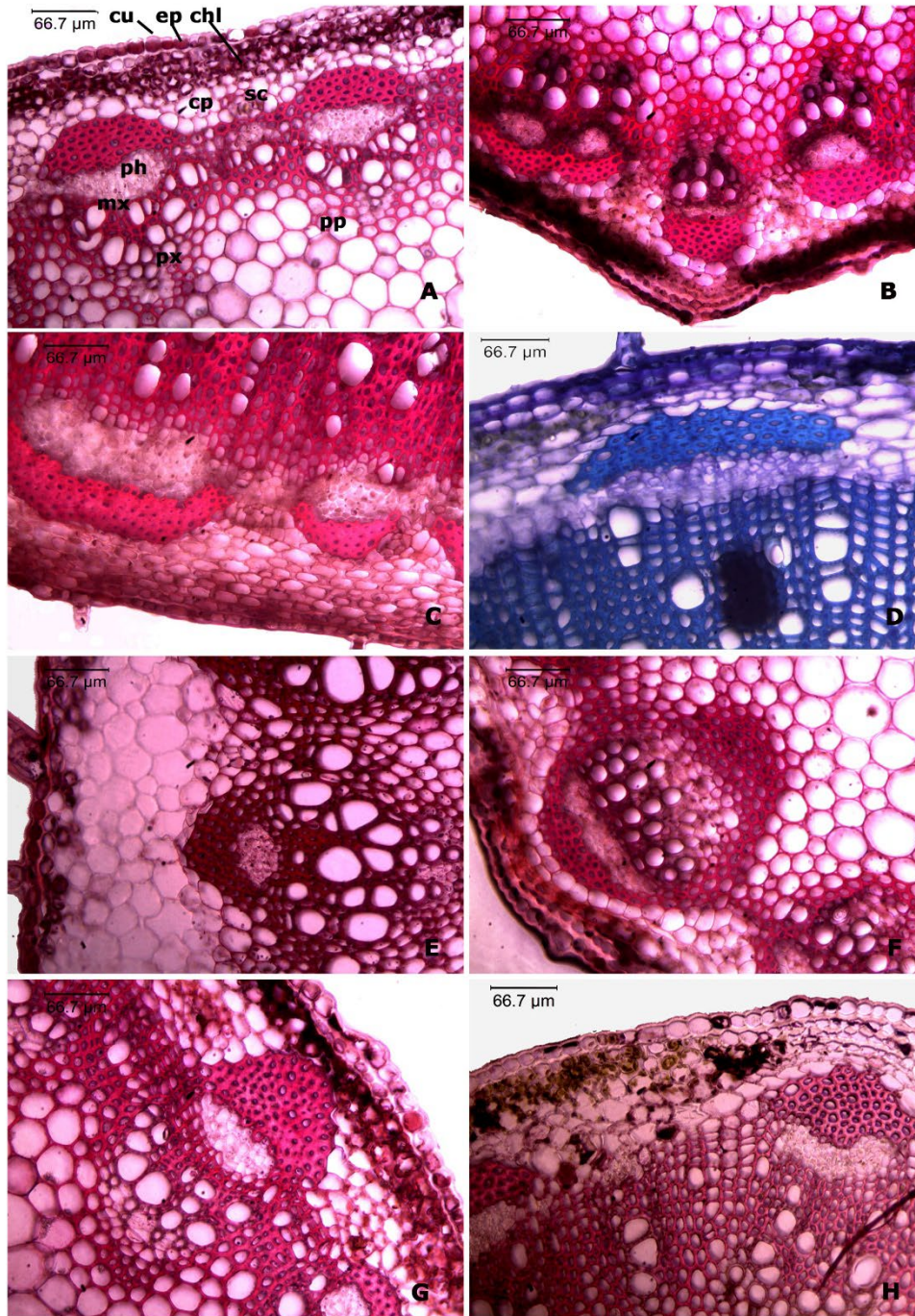


Fig. 4. Section showing epidermis, cortex, and vascular bundle (20X). A, *B. fistulosa*; B, *B. sessiliflora*; C, *B. laciniata*; D, *B. axillaris*; E: *B. lacera*; F, *B. barbata*; G, *B. membranacea*; H, *B. virens*; cu, cuticle; ep, epidermis; chl, chlorenchyma; cp, cortical parenchyma; sc, secretory canals; ph, phloem; mx, metaxylem; px, protoxylem; pp, pith parenchyma.

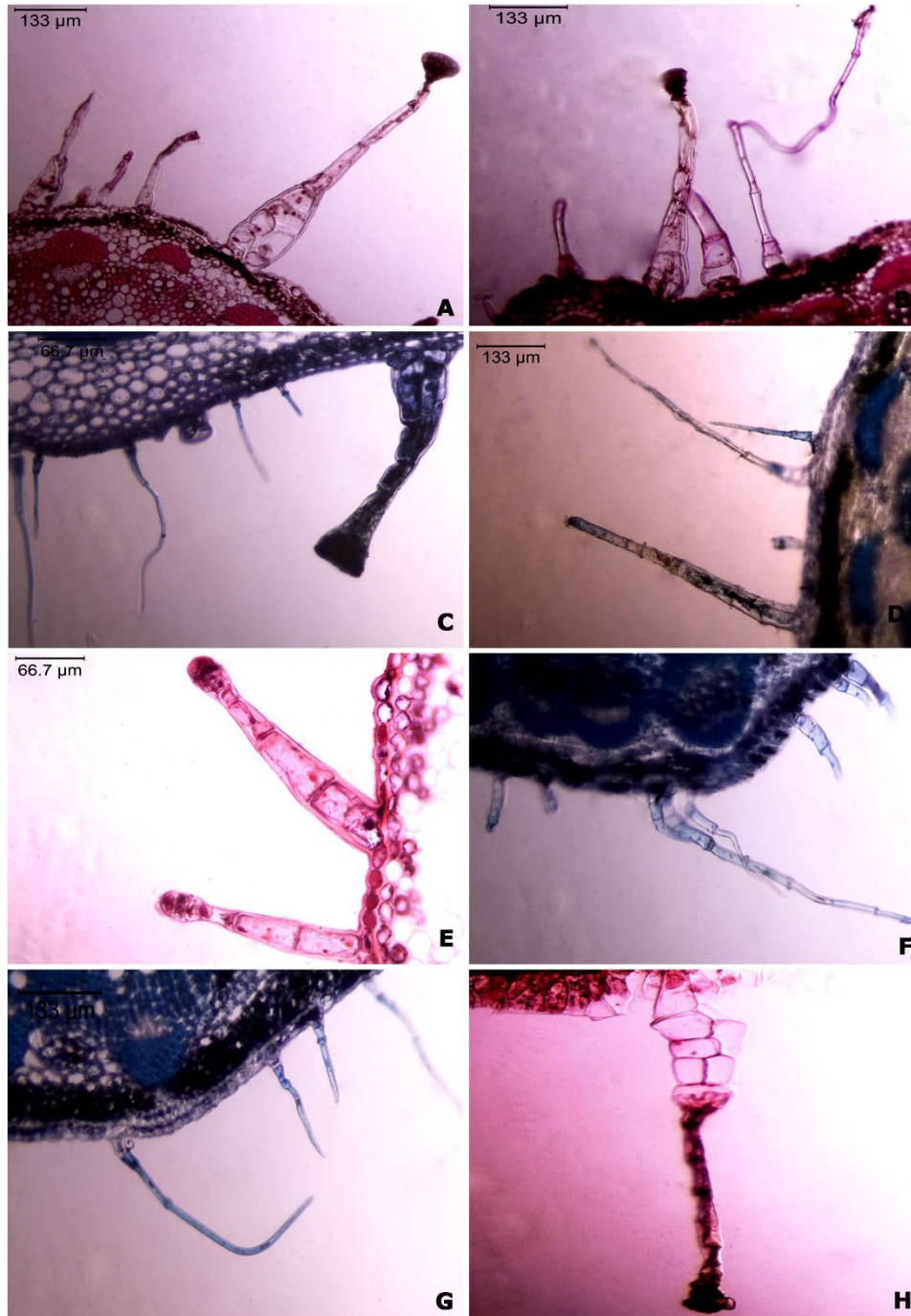


Fig. 5. Stem epidermal hair of *Blumea* spp. A, *B. fistulosa*; B, *B. sessiliflora*; C, *B. laciniata*; D, *B. axillaris*; E, *B. lacera*; F, *B. barbata*; G & H, *B. membranacea*.

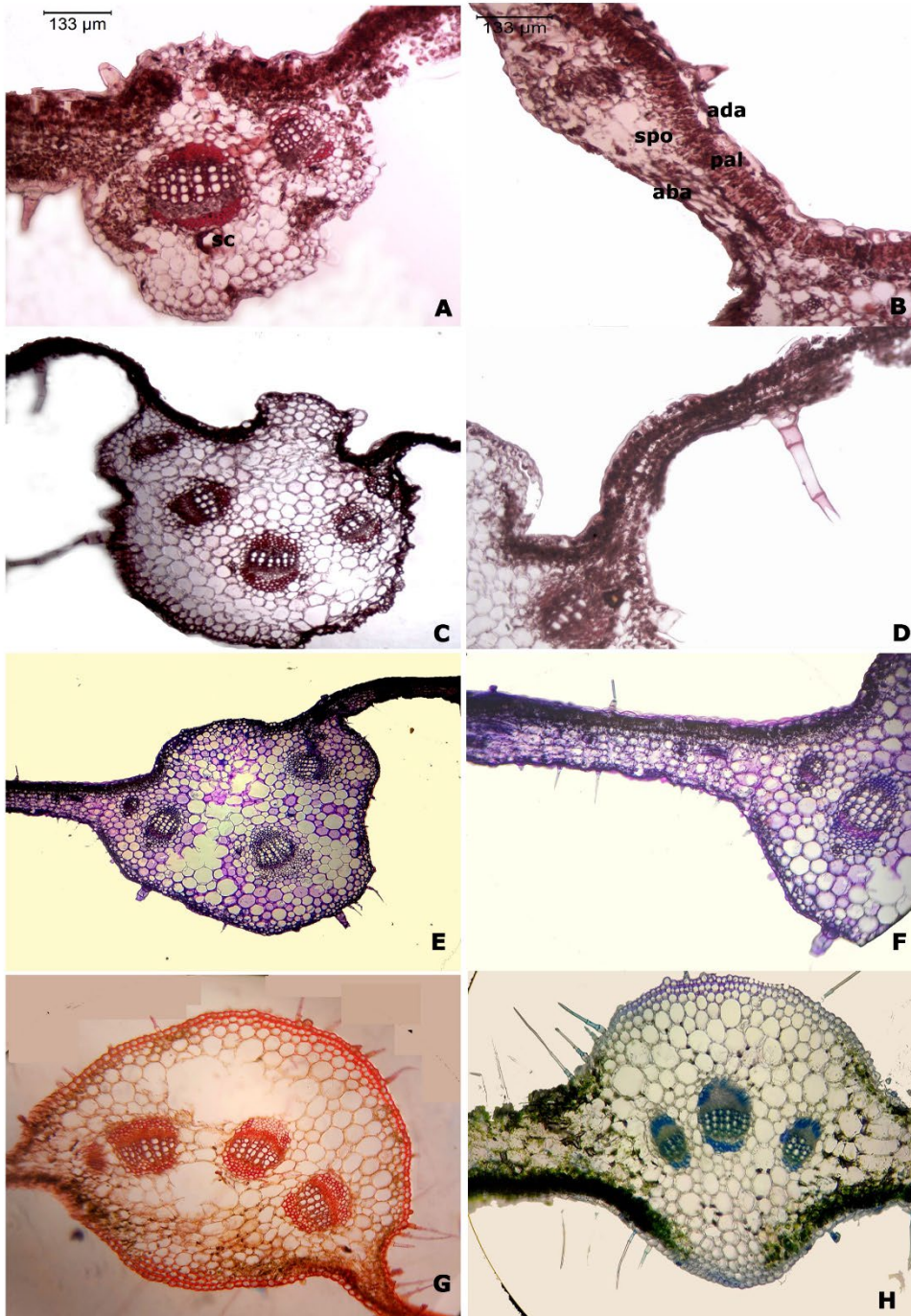


Fig. 6. Midvein and lamina of *Blumea* spp. A & B, *B. fistulosa*; C & D, *B. sessiliflora*; E & F, *B. laciniata*; G & H, *B. axillaris*; sc, secretory canals; ada, adaxial epidermis; pal, palisade tissue; spo, spongy tissue; aba, abaxial epidermis.

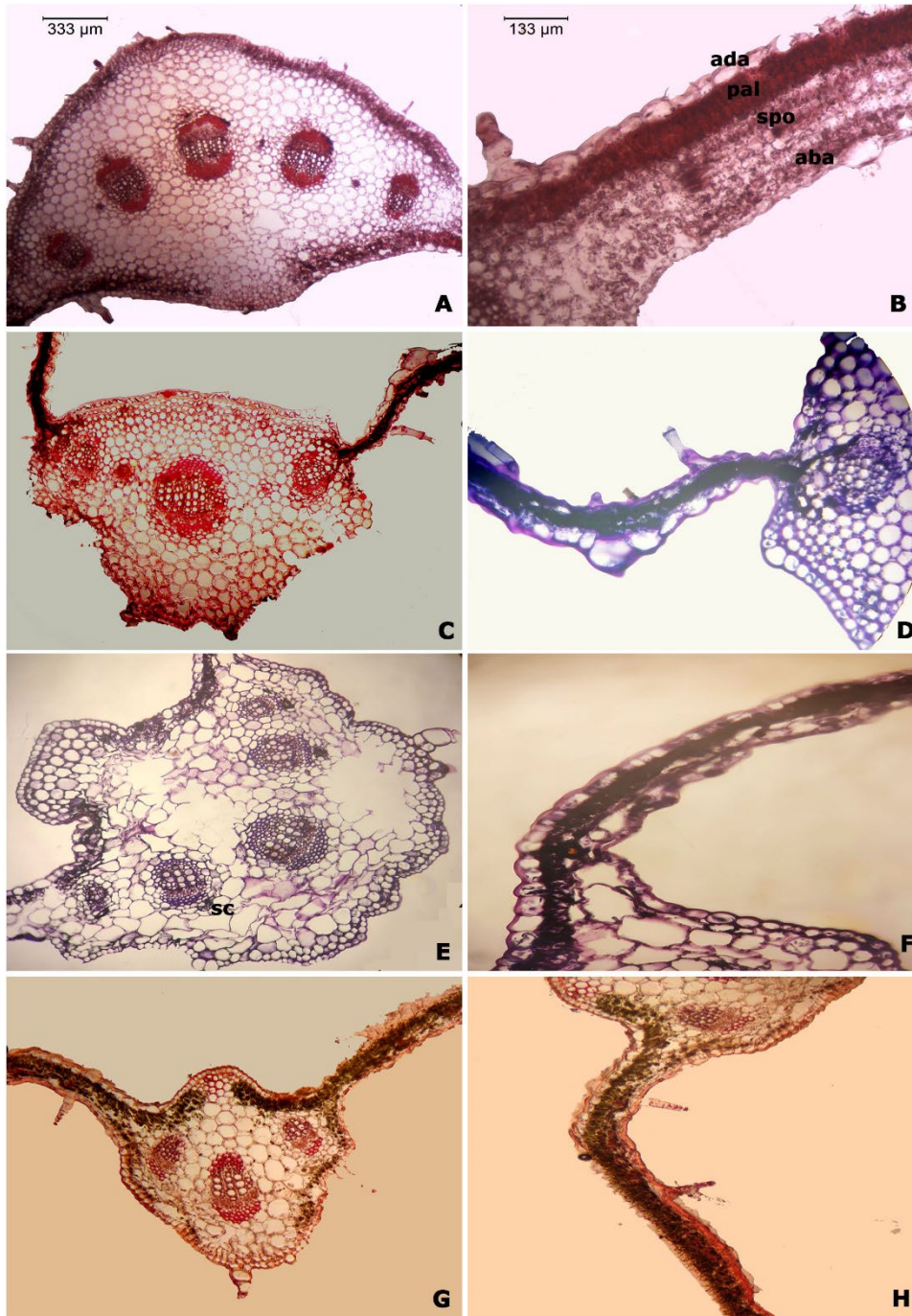


Fig. 7. Midvein and lamina of *Blumea* spp. A & B, *B. lacera*; C & D, *B. barbata*; E & F, *B. membranacea*; G & H, *B. virens*; sc, secretory canals; ada, adaxial epidermis; pal, palisade tissue; spo, spongy tissue; aba, abaxial epidermis.

Identification Key to the *Blumea* species, section *Paniculatae*

- 1. Stem trichomes absent *B. virens*
- 1. Stem trichomes present 2
- 2. Epithelial cells present at the cortex and just below the vascular bundles in leaves 3
- 2. Epithelial cells are absent at the cortex and just below the vascular bundles in leaves 6
- 3. Stem ridges absent *B. laciniata*
- 3. Stem ridges present 4
- 4. Multicellular capitate glandular hairs present in leaves *B. sessiliflora*
- 4. Multicellular capitate glandular hairs absent in leaves 5
- 5. Leaf lamina broad *B. fistulosa*
- 5. Leaf lamina narrow *B. membranacea*
- 6. Peltate glandular hairs present; Palisade tissue not single layered *B. axillaris*
- 6. Peltate glandular hairs absent; palisade tissue single-layered 7
- 7. Upper epidermal cells larger than lower epidermal cells; multicellular non-glandular hairs are present on stem..... *B. barbata*
- 7. Upper epidermal cells not larger than lower epidermal cells; multicellular non-glandular hairs absent on stem *B. lacera*

DISCUSSION

Tomblinson (1961) reported the taxonomic significance of vegetative anatomy in taxa delimitation. Carlquist (1961a, 1961b) has pointed out the taxonomic relevance of the trichome complement for the members of Asteraceae. Investigation of trichomes can be useful at a specific level (Faust & Jones 1973; Rollins and Shaw 1973; Adedeji 2004). Metcalfe & Chalk (1979) have reported the significance of the particular type of trichomes for delimiting species, genera, or particular families. Leaves of many members of Asteraceae possess secretory canals, glandular trichomes, cavities, and idioblasts (Milan & al. 2006; Duarte & al. 2011; Budel & al. 2012). Rahman (2013) reported non-glandular multicellular trichomes in *B. lacera* (Burm. f) DC and *B. laciniata* DC. The results of the present study showed the presence of both multicellular glandular and non-glandular trichomes in *B. laciniata* and multicellular capitate glandular trichomes in *B. lacera*.

Certain anatomical features like the shape of leaf epidermal cells of both laminar surfaces, the position of stomata, the type and morphology of trichomes (rather than their relative abundance), and the structure of wood were used by Randeria (1960) for the taxonomic revision of *Blumea* for continental South East Asian

species. In this study, we found that in the case of *B. barbata*, the upper epidermis of the leaf blade is larger than the lower epidermis. In *B. fistulosa* and *B. laciniata*, the leaf lamina is broad; in other species, it is narrow. According to Metcalfe & Chalk (1950), mesophyll cells are very variable in Asteraceae. We found that the palisade parenchyma of mesophyll of *B. laciniata*, *B. lacera*, *B. barbata*, and *B. membranacea* is a single layer. Instead, it has more than one layer in *B. fistulosa*, *B. axillaris*, and *B. virens*.

Different members of Asteraceae possess secretory ducts which are frequently related to the endoderm or vascular system (Pagni & Masini 1999; Budel & Duarte 2010; Souza & al. 2011). *Blumea laciniata* DC., *B. sessiliflora* Decne, *B. fistulosa* Kurz, and *B. membranacea* DC exhibited epithelial secretory canals.

Adedeji & Jewoola (2008) well-documented the leaf epidermal characteristics of Asteraceae. Their studies emphasized that the distribution of trichomes in Asteraceae is amazingly complex. The complexity is due to the diversity in the type of trichomes, differences in the density of trichomes, and also due to the differences in the distribution of trichome types on different parts of a given plant. Our studies on *Blumea* exactly show the same results. In *Blumea* species different types of trichomes can be seen, but *Blumea virens* lack trichomes on the surface.

In the family Asteraceae, the variations in the vascular bundles and leaf midrib are characteristic anatomical traits (Inceer & Ozcan 2011; Aydin & al. 2013; Özcan & al. 2014; Ekeke & Mensah 2015). According to our research, different species of *Blumea* can be distinguished by the number of vascular bundles in the midrib. The number of vascular bundles in the midrib varies from 3-5 in *B. virens*, *B. axillaris*, and *B. fistulosa*, from 4-6 in *B. lacera* and *B. barbata*, and from 5-7 in *B. sessiliflora*, *B. laciniata*, and *B. membranacea*.

In conclusion, it was noticed that some anatomical characters can be used for species delimitation in the genus *Blumea*. Trichome characters and the presence of secretory canals can serve as useful characters for separating the species.

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