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RESEARCH ARTICLE

Seedling leaf anatomy of three species of *Dendrocalamus* Nees

Naziya K[1, 2]. V. B. Sreekumar[1, 3]. E. J. Vincent[2] N. Anjana[3, 4*]. Prabhat Kumar[5]

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Abstract: In early stages, seedlings of Bamboo grows slowly and species identification at this stage is difficult and demanding because of absence of distinctly separable morphological features. In this study, seedling leaf samples of three species of bamboo, *Dendrocalamus stocksii*, *D. longispathus*, and *D. brandisii*, were gathered, sectioned, stained, and examined under a light microscope. Images were captured and anatomical features of the species such as venation pattern, plicate cells (pc); crystals (cry); fusoid cells (fc); micro hairs (mh); long cells (lc); stomata (st); trichomes (tr); prickles (pr); bulliform cells (bc); silica bodies (sb); and vascular bundles (vb) were considered. The study highlights the importance of the leaf structure of bamboo seedlings, along with their anatomical features to distinguish species which are closely related. The findings deliver valuable insights into taxonomic uncertainties among the species of *Dendrocalamus* and lay the foundation for future investigations of bamboo seedling anatomy. We focus to highlight the significance of leaf anatomy of seedling as a reliable tool for bamboo seedling identification.

Keywords: Bamboo leaf anatomy, *Dendrocalamus*, Diaphanization, Franklin's solution, Bamboo taxonomy

[1] Department of Forest Botany, KSCSTE - Kerala Forest Research Institute, Thrissur, Kerala, India

2 Department of Botany, Christ College (Autonomous), Thrissur, Kerala, India

3 Department of Wood Science and Technology, KSCSTE- Kerala Forest Research Institute, Thrissur, Kerala, India

4 University of Calicut, Thenhipalam, Malappuram, Kerala, India

5 Horticulture Commissioner and Mission Director, National Bamboo Mission, New Delhi

Correspondence should be addressed to N. Anjana
Email: anjana.nkp@gmail.com

Intoduction

For over a century, anatomical characters that are stable and conserved have been aiding in taxonomic studies (Radford *et al*., 1979). In bamboo, the morphological classification system, by using culm and leaf sheath, is unstable and often subject to changes due to ecological factors. Therefore, taxonomists investigated anatomical research to assess the possibility of developing the classification system in accordance with anatomical characters. For bamboos, the leaf and its epidermal characters are especially effective for identifying and classifying different species (Soderstrom *et al*.,1987; Van Tien *et al*., 2014; Refulio-Rodriguez *et al*., 2025). In the species of Asian tropics, the shape, and patterns of distribution of papillae in woody bamboos are recognized as reliable diagnostic features for identification up to the genus level (Yang *et al*., 2008). Conservative characters that are less impacted by external factors are considered in anatomy and micromorphology.

The epidermis of the leaves and culms is examined for diagnostic features. Calderón and Soderstrom (1973) identified three main epidermal appendage types in the subfamily Bambusoideae: prickles, micro -hairs, and papillae. The presence of fusoid cells, as reported by Renvoize (1985), Soderstrom *et al*., (1987) and Soderstrom and Ellis (1988) is an important trait to define the type of bamboo leaf anatomy. Another prominent feature of bamboo leaf is the papillae seen on the lower epidermis (Judziewicz *et al*., 1999). Phytoliths and trichomes serve as additional diagnostic traits.

The lower epidermis frequently has stomata and guard cells, typically smaller in appearance than epidermal cells (Rao *et al*., 1985). The forms and distribution pattern of silica cells are also used in

taxonomic identification. The present study is an attempt to elucidate the anatomical features of seedling leaves of three *Dendrocalamus* species, giving a reliable method for their identification at the initial stage.

Materials and Methods

Leaf samples of three species of *Dendrocalamus* seedling were collected from the bamboo nursery of Kerala Forest Research Institute, Peechi and 70% ethanol was used to preserve the samples. The selection of seedlings was based on proper healthy growth and steady age (5-7 weeks). For each species, samples from six separate individual seedlings were collected and anatomical observations were conducted. Freehand sections were prepared from the leaf margin, lamina, and midrib. Sections were washed in distilled water, cleared with sodium hypochlorite, stained using safranin, and mounted in 50% glycerol. The leaves were cleared (diaphanization) to acquire clear pictures of the leaf venation and epidermal surfaces. After being chopped into square pieces, the materials were cleaned to rid of excess fixatives and placed in Franklin's solution, which contains equal amounts of 35% hydrogen peroxide and glacial acetic acid, up to 24 hours at room temperature. The formation of large air bubbles between two epidermises indicated the complete removal of veins. Cleared leaves were cleaned with water and submerged in a sodium hypochlorite solution for 20 to 30 minutes to make the epidermis transparent. Samples were again washed and stained for 5 to 10 minutes using 0.01% aqueous safranin solution. Excess stain was removed by washing and the samples were mounted in glycerol. The sections were examined using light microscope and the images captured using Leica DFC 280 camera with image analysing software. Quantitative anatomical parameters are measured with ImageJ software.

Results

Leaf anatomy of three *Dendrocalamus* species examined were found to share characteristics with other bamboo species as reported by Metcalfe (1956), Ellis (1976) and Vieira *et al*., (2002). The epidermis is divided into zones when viewed from the surface, similar to the members of Poaceae. The epidermis is similarly structured in the three species under examination, aligning with Metcalfe's (1956) observation on the anatomy of bamboo leaves, particularly the presence of short and long epidermal cells in their intercostal and costal zones, micro-hairs and saddle shaped silica bodies. Metcalfe (1956) noted that various kinds of long cells are helpful for taxonomy but not entirely reliable since intermediate forms could exist. Long cells can vary in shape, from short with thin walled which are non-sinuous to long thicker, sinuous walls. The observations consistent with Vieira *et al*., (2002), as all three species exhibit long cells with sinuous thin walls on both leaf surfaces. Short cells, either in pairs or solitary with silicaceous cells, are found in the epidermis and are referred as cork (cc) or suberous cells, as per Metcalfe (1956, 1960), Calderón and Soderstrom (1973), Renvoize (1985) and Vieira *et al*., (2002). The detailed anatomical descriptions of the three *Dendrocalamus* species are provided below:

Dendrocalamus longispathus (Kurz) Kurz

Synonym: *Bambusa longispatha* Kurz

Morphology

The leaf-blades are lance-shaped; 10-13 cm in length and 25-45 mm wide, with a short petiole-like connection to the sheath and conspicuous midrib; secondary veins are 16-20 in number. Leaf with scabrous surfaces and margins with acuminate apex.

Epidermis

Oval or cubical shaped; single-layered; tightly packed without intercellular space; thick cuticle at abaxial and adaxial side (Fig. 1. D).

Long cells: 1-3 rows of globose or elliptical-shaped; narrow papillae; with highly sinuous straight and oblique end walls at the costal (cz) and intercostal regions in abaxial, non-papillate in the adaxial (Fig. 1. D, F, G).

Stomata and interstomatal cells: Amphistomatic; dumb-bell shaped guard cells; abaxial intercostals with stomata in 3 horizontal alternating rows surrounded by 4 papillae on the corners of the stomatal complex. Adaxially scarce; with an average stomatal density of 26.75 ± 5.51 stomata/mm². (Fig. 1. F, G).

Short cells: Silica cells are solitary at costal and paired with cork cell at intercostal region in adaxial and abaxial. Saddle-shaped silica cell with large cylindrical/ irregular cork cell at abaxial and intended dumb-bell shaped silica cell with short cylindrical cork cell at adaxial (Fig. 1. F, G).

Bulliform cells: Raised and sunken type; 2-3 horizontal rows at the intercostal region of adaxial side; 4-5 celled near midrib; 2-3 celled at lamina; 4-5 near margin; abaxially absent. (Fig. 1. A, B, C, D, G).

Large crystals: At bulliform cells (Fig. 4. A).

Trichome: Long and short unicellular; non-glandular; filiform and papillary type at both costal and intercostal region of abaxial side; 2-3 trichomes within a bulliform range; Adaxially absent (Fig. 1. E).

Prickle: Short-spined; oval to round shaped base associated with cork cells at the costal and intercostal region of the abaxial side. Adaxially absent/ scarce (Fig. 1. F).

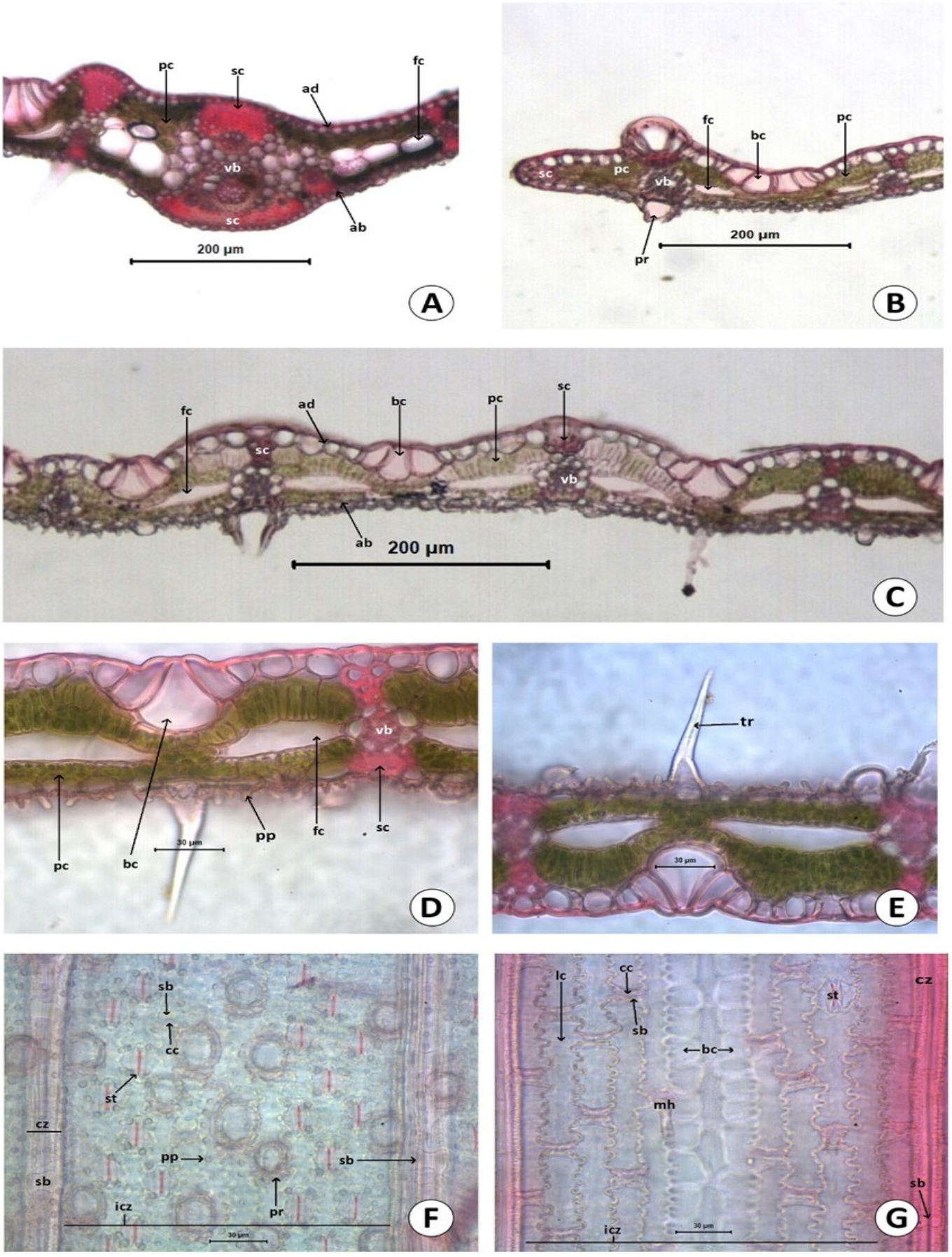

Fig 1. *D. longispathus*. A: Transverse section of the midrib at median level (bar=200µm), B: Leaf margin (bar =200µm), (C), (D); Leaf lamina (bar=200µm and 30µm) E: Trichome (bar=30µm); F: Abaxial surface, G: Adaxial surface of leaf epidermis (bar=30µm). Abbreviation: ad: adaxial; ab: abaxial; bc: bulliform cells; fc: fusoid cells; pc: plicate cells; vb: vascular bundle; sc: sclerenchyma; pp: papilla; tr: trichome; cz: costal zone; icz: intercostal zone; sb: silica bodies; cc: cork cells; pr: prickle; mh: micro hair; st: stomata; lc: long cells

Micro hairs: 2 celled (thick walled basal and thin-walled distal cells) at the intercostal region of both abaxial and adaxial side (Fig. 1. G).

Ribs and furrows close to one of the two margins and one side of the midrib (Fig. 1. A, B).

Arm cells: Polygonal-shaped; chlorophyllous; palisade-like plicate cells; 1-2 layered at adaxial, 1 layered at abaxial side (Fig. 1. D).

Fusoid cells: 2 cells perpendicular to the vascular bundles; abaxial; translucent; achlorophyllous; thin walled; separated by arm cells (Fig. 1. D).

Vascular Bundle

Collateral*;* 5-7 small vascular bundles alternating with large ones; mean distance between vascular bundles is 170.836 ± 50µm.

Endodermis: Outer achlorophyllous parenchymatous (abaxially interrupted), inner sclerenchymatous pericycle (absent in smaller bundles) and sclerenchymatous sheath extensions (Fig. 1. C, D).

Midrib vascular bundle: Abaxially projected; 1 large central vascular bundle; abaxial side - 2 small vascular bundles; adaxial side - 1 vascular bundle which is also small; parenchymatous cells are seen between the 3 vascular bundles and the sides. Presence of protoxylem and lacuna (Fig. 1. A); sclerenchyma sheath extension to both epidermal sides (Fig. 1. A). Patch of sclerenchyma at the margin tip (Fig. 1. B).

Dendrocalamus brandisii (Munro) Kurz

Synonym: *Bambusa brandisii* Munro, *Arundarbor brandisii* (Munro) Kuntze

Morphology

The leaf-blades are oblong or lanceolate; 20-30 cm in length and 25-50 mm width; connected to the sheath by a petiole; 10-12 secondary veins, and pubescent and hairy surfaces with an acuminate apex.

Epidermis

Oval or cubical shaped; single-layered; tightly packed without intercellular space; thick cuticle at abaxial and adaxial side (Fig. 2. D).

Long cells: 1-2 rows of globose-shaped papillae; narrow with highly sinuous straight and oblique end walls at the costal and intercostal regions at adaxial and abaxial sides. Highly papillate at abaxial, non-papillate at adaxial (Fig. 2. G).

Stomata and interstomatal cells: Amphistomatic; with 2 dumb-bell shaped guard cells; abaxial intercostal with 2-3 horizontal alternating rows; surrounded by 4-5 papillae on the corners of the stomatal complex; adaxial- scarce; average stomatal density is 15.59 ± 1.95 stomata/mm² (Fig. 2. D, F).

Short cells: Solitary; saddle shaped silica cells at costal region and paired (narrow dumb bell-shaped silica cells with smaller cylindrical or irregular cork cells at adaxial and narrow saddle shaped silica cells with larger cylindrical or irregular cork cells at abaxial) at intercostal region of both adaxial and abaxial side (Fig. 2. F, G).

Bulliform cells: 2-3 horizontal rows of sunken type and rarely raised type of bulliform cells are present at the intercostal region of adaxial side; 4-5 celled near midrib; 2-3 celled at lamina and 6-7 near to margin; abaxially absent (Fig. 2. A, B, C, D, G).

Large crystals: At bulliform cells (Fig. 4. B).

Trichomes: Long and short unicellular; non-glandular; filiform and papillary type at both costal and intercostal region of abaxial side; 1-3 trichomes within a bulliform range; adaxially absent (Fig. 2. E).

Prickle: Short spined; oval to round shaped base at the intercostal region of abaxial side; adaxially absent/ scarce (Fig. 2. F).

Micro hairs: 2-celled, intercostal region of both abaxial and adaxial side (thick-walled basal and thin-walled distal cell) (Fig. 2. G, F).

Ribs and furrows close to one of the two margins and one side of the midrib (Fig. 2. A, B).

Plicate cells: Polygonal shaped; chlorophyllous; palisade-like (arm cells); 1-2 layered at adaxial side and 1 layered at abaxial (Fig. 2. D).

Fusoid cells: Abaxial; translucent; achlorophyllous; thin walled; 2 cells which is perpendicular to the small vascular bundles; separated by the arm cells (Fig. 2. D).

Vascular bundle

Collateral; 5-6 small vascular bundles between large ones; mean distance between vascular bundles is 287.254 ± 70µm.

Endodermis: Outer achlorophyllous parenchymatous (abaxially interrupted); inner sclerenchymatous pericycle (absent in smaller bundles) and sclerenchymatous sheath extensions (Fig 2. C, D).

Midrib vascular bundle: Abaxially projected; 1 large central vascular bundle; 2 small vascular bundle towards

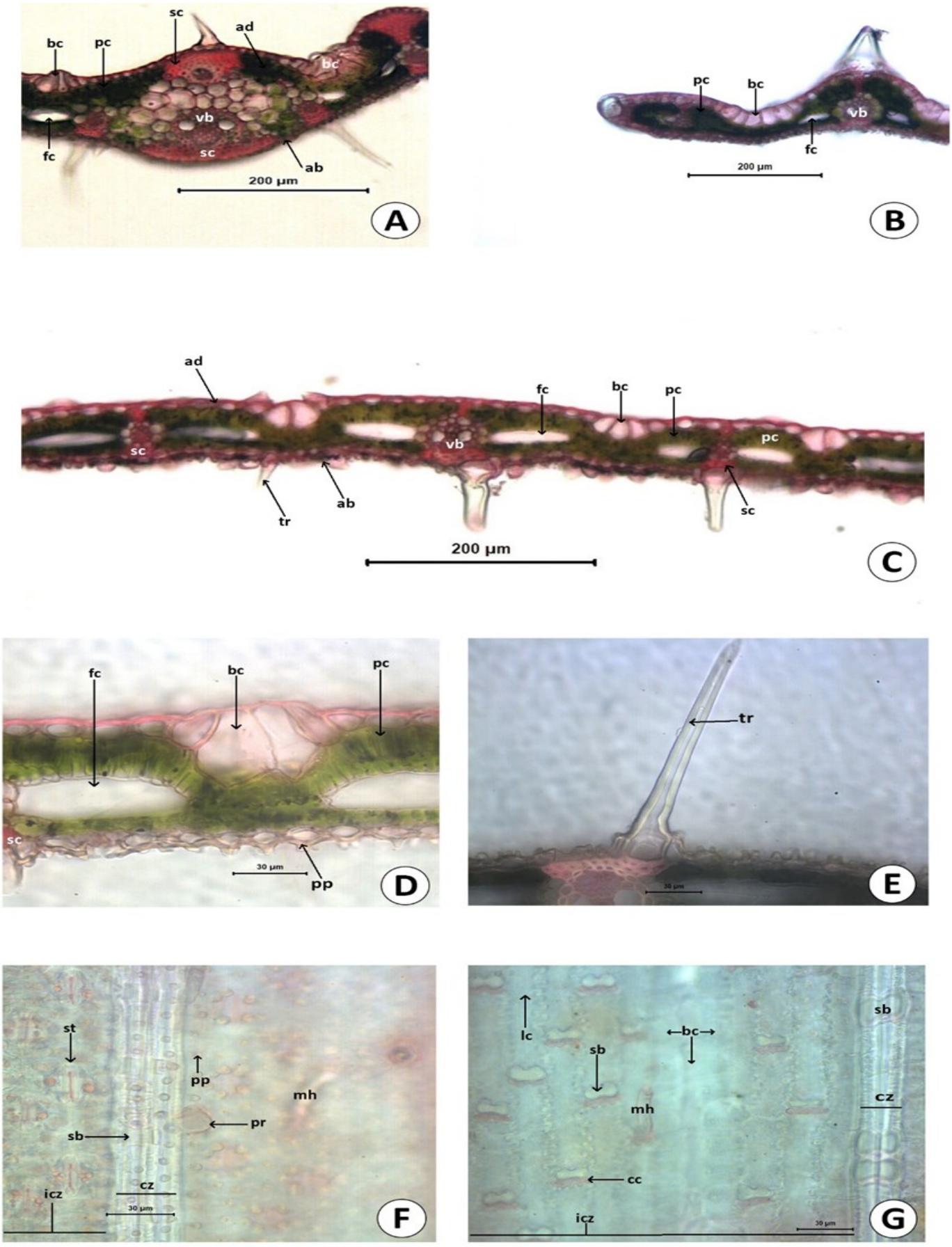

Fig 2. *D. brandisii*. A: Transverse section of the midrib at median level (bar=200µm), B: Leaf margin(bar=200µm), (C), (D); Leaf lamina (bar=200µm and 30µm) E: Trichome (bar=30µm); F: Abaxial surface, G: Adaxial surface of leaf epidermis (bar=30µm). Abbreviation: ad: adaxial; ab: abaxial; bc: bulliform cells; fc: fusoid cells; pc: plicate cells; vb: vascular bundle; sc: sclerenchyma; pp: papilla; tr: trichome; cz: costal zone; icz: intercostal zone; sb: silica bodies; cc: cork cells; pr: prickle; mh: micro hair; st: stomata; lc: long cells

abaxial side; 1 small vascular bundle towards adaxial side interrupted by parenchymatous cells; Protoxylem present without lacuna; sclerenchyma sheath extension to both epidermal sides (Fig. 2. A). Sclerenchymatous patch at margin tip (Fig. 2. B).

Dendrocalamus stocksii (Munro) M. Kumar, Remesh and Unnikrishnan

Synonyms: *Oxytenanthera stocksii* Munro, *Pseudotenanthera stocksii* (Munro) R.B. Majumdar; *Pseudoxytenanthera stocksii* (Munro)

Morphology

Leaf-blades are lanceolate; 10-20 cm in length and 10-20 mm in width; with a 0.2 cm petiole-like attachment to the sheath; smooth surfaces; with 10-12 secondary veins; and scabrous margins with attenuate apex.

Epidermis

Oval or cubical-shaped; single-layered; without intercellular space; thick cuticle at both abaxial and adaxial side (Fig. 3. D).

Long cells: 1-3 rows; with globose-shaped papillae at abaxial side; narrow with highly sinuous, straight, and oblique end walls at the costal and intercostal regions of both abaxial side and adaxial side (Fig. 3. D, F, G).

Stomata and interstomatal cells: abaxial side: Amphistomatic; 2 dumb-bell shaped guard cells; abaxial intercostals with 3 horizontal alternating rows of stomata surrounded by 4-6 papillae on the corners of the stomatal complex; scarce in adaxial; average stomatal density is 19.55 ± 2.59 stomata/mm² (Fig. 3. F).

Short cells: paired (irregular-shaped silica cell with large cylindrical cork cell) and rarely solitary (cylindrical cork cell) at intercostal region; solitary (saddle shaped silica cell) and rarely paired (saddle-shaped silica cell with cork cell) at costal region of both adaxial and abaxial side (Fig. 3. F, G).

Bulliform cells: 2-3 horizontal rows; sunken at the intercostal region of adaxial side; 4-5 celled near midrib; 2-3 celled at lamina; 5-6 near to margin (Fig. 3. A, B, C, D, G).

Large crystals: At bulliform cells (Fig. 4. C).

Trichome: Long and short; unicellular; non-glandular; filiform; rarely papillary type at both costal and intercostal region of abaxial side; 1-2 trichomes within a bulliform range. Adaxially absent (Fig. 3. E).

Prickle: Short spined; oval to round shaped base at the adaxial side prickles are seen only in the intercostal region; associated with cork cells at the intercostal region (Fig. 3. F, G).

Micro hairs: 2-celled (thick walled basal and thin-walled distal cells) at intercostal regions of both abaxial and adaxial sides (Fig. 3. F, G).

Ribs and furrows close to one of the two margins and one side of the midrib (Fig. 3. A, B).

Plicate cells: Polygonal shaped; chlorophyllous; palisade like; 2-3 layered at the adaxial side and 1-2 layered at the abaxial side (Fig. 3. D).

Fusoid cells: abaxial; translucent; achlorophyllous; thin walled; perpendicular to the vascular bundle; separated by arm cell (Fig. 3. D).

Vascular bundle

Collateral; 7-8 small vascular bundles between each large one; mean distance between vascular bundles is 147.832 ± 38µm.

Endodermis: Outer achlorophyllous parenchymatous (abaxially interrupted); inner sclerenchymatous pericycle (absent in smaller bundles) and sclerenchymatous sheath extension to one or both the epidermis (Fig. 3. C, D).

Midrib vascular bundle: Abaxially projected; one large central vascular bundle with protoxylem; sclerenchyma sheath extension to both epidermal sides (Fig. 3. A). Sclerenchymatous patch at margin tip (Fig. 3. B).

Discussion

The leaves of Bambusoideae have tiny globose structured papillae, micro hairs (trichome, often consisting two cells, apical cell having thin wall and the basal one having a thick wall) and prickles as epidermal appendages in both long cells and short cells, particularly in the intercostal zones (icz) and can range in number from one to many (Calderón and Soderstrom, 1973; Ellis, 1976; Renvoize, 1985; Vieira, 2002;). In intercostal zones, stomata is usually seen as well-defined bands and are protected by papillae. According to Ellis (1976), bulliform cells, a component of the epidermis, can be distinguished from other epidermal components as they are larger and more inflated. Bulliform cells turn flaccid when there are high water losses, which causes the plant to infold or bend and reduce the transpiration surface of leaf. On both the abaxial side (ab) and adaxial side (ad), the mesophyll of Bambusoideae comprises infolded chlorophyllous parenchymatous tissue, known as arm cell or plicate cells, which divide them into tubular compartments (Brandis, 1907; Calderón and Soderstrom, 1973; Vieira *et al*., 2002).

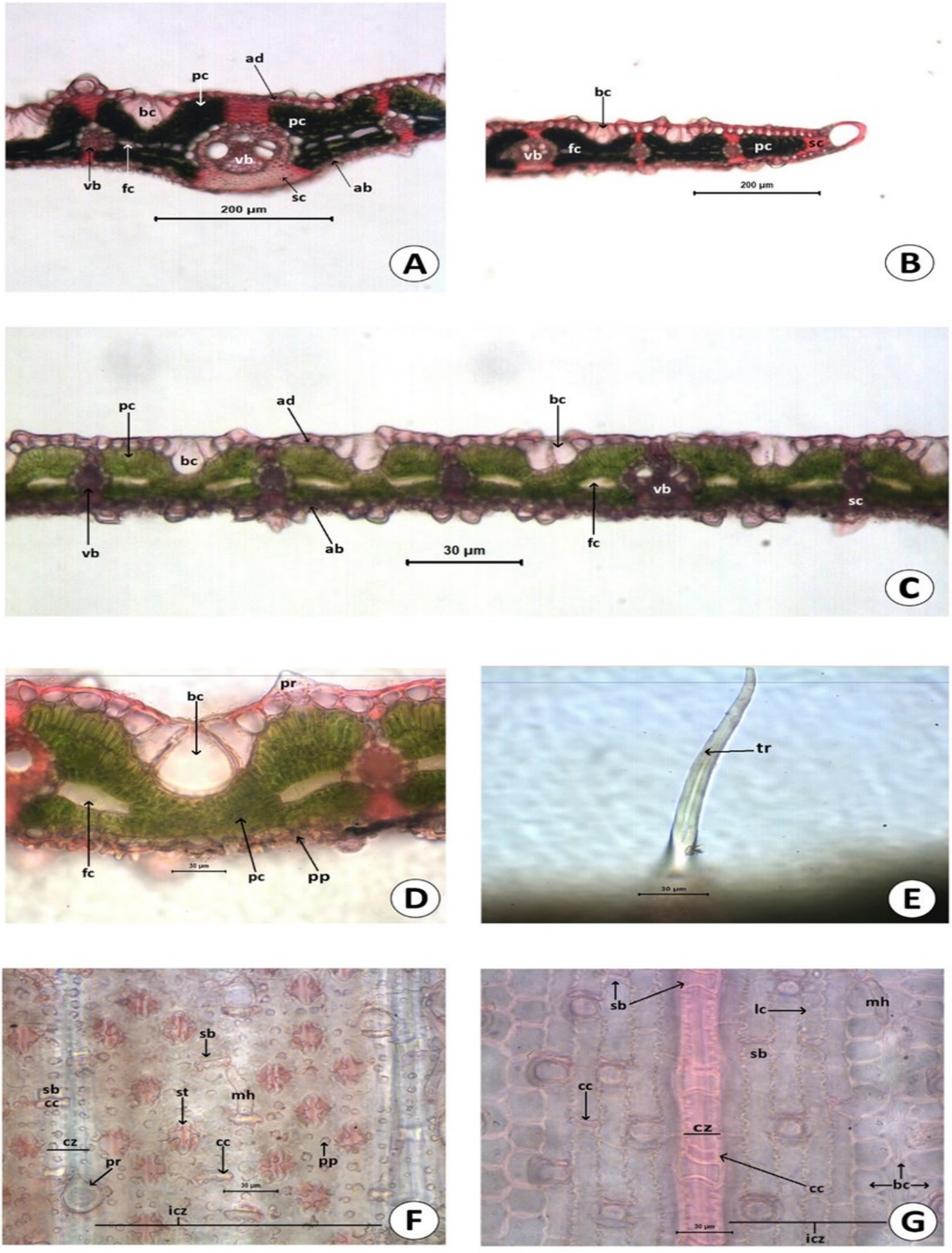

Fig 3. *D. stocksii*. A: Transverse section of the midrib at median level (bar=200µm), B: Leaf margin(bar=200µm), (C), (D); Leaf lamina (bar=200µm and 30µm) E: Trichome (bar=30µm); F: Abaxial surface, G: Adaxial surface of leaf epidermis (bar=30µm). Abbreviation: ad: adaxial; ab: abaxial; bc: bulliform cells; fc: fusoid cells; pc: plicate cells; vb: vascular bundle; sc: sclerenchyma; pp: papilla; tr: trichome; cz: costal zone; icz: intercostal zone; sb: silica bodies; cc: cork cells; pr: prickle; mh: micro hair; st: stomata; lc: long cells

The presence of fusoid cells which are large air spaces in the leaf blades is consistent with assertions of Vieira *et al*., (2002) and Ellis (1976) that these structures are essential for distinguishing the leaves of the Bambusoid type. Another peculiar characteristic is their unique midrib (Brandis, 1907), which consists of one to three minor side bundles and a large median vascular bundle (Calderón and Soderstrom, 1973). The present study also confirms that some species have double sheaths, as mentioned by Renvoize (1985). The outer achlorophyllous parenchymatous endodermis is abaxially disrupted when both bundles are present. According to Vieira *et al*., (2002) and Calderón and Soderstrom (1973), the pericycle or inner sheath is composed of sclerenchymatous elements that are absent in smaller bundles. The species under investigation have sclerenchyma at the leaf margin and close to vascular bundles. Sclerenchymatous sheath extensions can reach one or both epidermises.

These species have crystals in their epidermal bulliform cells, a characteristic unique to the genera. Sunken and raised bulliform cells were found in the adaxial epidermal cells of *D. longispathus* and *D. brandisii*, while raised cells were found in *D.* adaxial epidermal cells of *D. longispathus* and *D. brandisii*, while raised cells were found in *D. stocksii.* There are 2-3 bulliform cells at the lamina and 4-5 near the midrib vascular bundle in *Dendrocalamus* species. *Dendrocalamus* species differ in the number of bulliform cells close to the margin. On the abaxial surface of *D. brandisii, D. longispathus* and *D. stocksii,* trichomes are abundant and widely dispersed. The species studied exhibited papillary trichomes and micro hairs on both epidermal sides. Vascular bundles of both simple and complex forms are observed on the lamina and midrib. In the species under study, typically 5-7 small vascular bundles are present between the larger bundles in *D. longispathus*, while 5-6 small vascular bundles were observed in *D. brandisii* and 7-8 were observed in *D. stocksii*. Stomata were arranged in 2-3 alternating horizontal rows in *D. brandisii,* while 3 alternating rows were seen in *D. stocksii* and *D. longispathus.* Seedlings of *D. longispathus* and *D. brandisii* were found to have prickles on the abaxial epidermal side, whereas *D. stocksii* had prickles on both epidermal sides. There are distinct saddle and dumbbell-shaped silica cells in the intercostal and coastal regions of the abaxial and adaxial surfaces, either individually or paired with cork cells (Table 1).

Bamboo species pose considerable challenges for identification at the seedling stage due to their morphological uniformity. Seedlings of many bamboo species exhibit highly similar external features such as

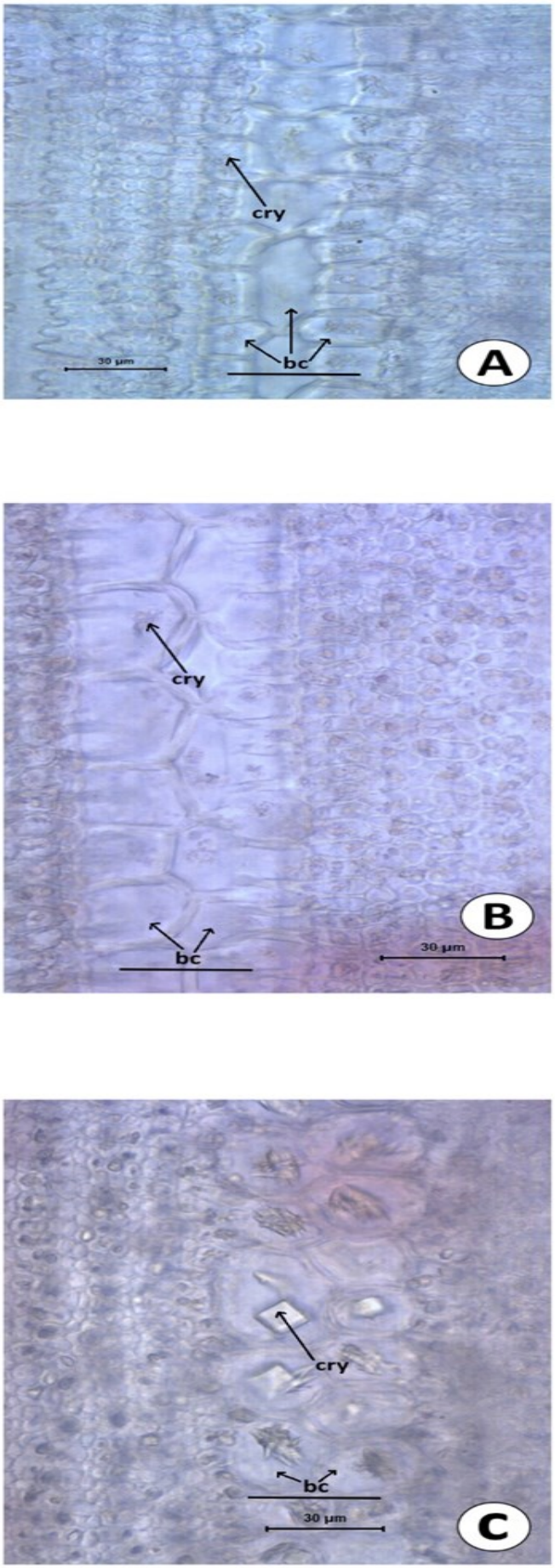

Fig 4. Silica crystals (cry) in bulliform cells (bc) of (A) *D. longispathus*; (B) *D. brandisii*; (C) *D. stocksii* (bar=30µm). Abbreviation: bc: bulliform cells

Table 1. Anatomical comparison of *Dendrocalamus sp.* leaves

Species seedling	Cell wall and Papillae	Epidermal cells	Bulliform cells	Stomata	Micro hairs	Prickles	Trichomes	Silica bodies	Crystal	Plicate cells	Vascular Bundles at lamina	T.s of midrib
Dendrocalamus lon ispathus	Present at both sides; wavy; abaxial papillae	Oval; Same size from middle to sides of BC	Both raised and sunken types. 2-3 cells at the lamina,4-5 near margin, 4 -5 near midrib	3 alternating horizontal rows; surround by 4 papillae	Present at both sides	Present at abaxial side	Short and long unicellular non glandular filiform and papillary types, 2-3 trichomes within one BC range	Saddle and dumb bell shaped	Present	adaxial side 1-2 layers; abaxial side single layered	5-7 small V. B between each large ones	1 large central V. B and 2 small V. B below the central at abaxial side and1 small V. B at adaxial side. Protoxylem and lacuna is present
Dendrocalamus brandisii	Present at both sides; wavy; abaxial papillae	Oval; Same size from middle to sides of BC	Sunken type and rarely raised type. 2-3 cells at the lamina, 6-7 near margin, 4-5 near midrib	2-3 alternating horizontal rows; surround by 4-5 papillae	Present at both sides	Present at abaxial side	Tough long and short unicellular non glandular filiform and papillary types. 1-3 trichomes within one BC range	Saddle and dumb bell shaped	Present	adaxial side 1-2 layers; abaxial side single layered	5-6 small V. B between each large ones	1 large central V. B and 2 small V. B towards abaxial side and 1 small V. B towards adaxial side. Presence of protoxylem is also seen
Dendrocalamus stocksii	Present at both sides; wavy; abaxial papillae	Oval; Same size from middle to sides of BC	Sunken type, 2-3 cells at the lamina, 5-6 near margin, 4-5 near midrib	3 alternating horizontal rows; surround by 4-6 papillae	Present at both sides	Present at both side	Long and short unicellular non glandular filiform and sometimes papillary types and rarely at adaxial side,1-2 trichomes within one BC range	Saddle and irregular shaped	Present	adaxial side 2-3 layers; abaxial side 1-2 layered	7-8 small V. B between each large ones	1 large central vascular bundle with protoxylem is seen

BC - Bulliform cells, V. B - Vascular Bundles

leaf size, shape, venation patterns, and coloration, which often overlap among genera and species. These characteristics are usually insufficient for precise identification, as seedlings lack completely developed diagnostic features such as culm sheath, type of rhizome, flowers and branching patterns. Moreover, the lengthy irregular flowering cycles of bamboo prevent relying only on flowers for classification functions during the initial stages of development. Phenotypic plasticity in vegetative traits due to environmental factors often result in misleading morphological characters.

Considering such limitations, for the species-level identification, a valuable and consistent alternative is anatomical analysis of leaves, especially during initial stages of growth. Anatomical features are likely to be less impacted by environmental variability and within the taxonomic lineage they are often more conservative, which makes them ideal for distinguishing species when morphology is unsure. Key diagnostic features include the structure and distribution of silica bodies, type and arrangement of vascular bundles, presence of fusoid cells and bulliform cells, epidermal structures, density and type of stomata, and the absence or presence of papillaes and trichomes (Vieira *et al*., 2002; Wysocki *et al*., 2016). The present study focuses very delicate differences in the leaf anatomy of *Dendrocalamus* species.

In practical fields, such as germplasm conservation, afforestation, nursery management, and selective breeding, for accurate selection of species anatomical studies act as an essential tool particularly when managing juvenile materials. In addition, alternative to molecular techniques, anatomical analysis is more accessible and cost-effective. Leaf anatomy give strengthens taxonomy and provides deeper understanding of diversity and evolution of bamboo by giving functionally meaningful and consistent characters. It also provides insights into the physiological and ecological adaptations of species of bamboo.

Conclusion

This present comparative study helps in the bamboo taxonomy by analysing the seedling anatomy of *Dendrocalamus* species. Identification of species of bamboo is complicated because of the similarities in morphology, genetic variability, long and synchronised flowering cycles, and phenotypic plasticity. These factors create the external characters unreliable for precise classification. This problem is greater in seedling stage. Anatomical research addresses these difficulties by means of reliable and consistent species identifiers, like prickles, silica crystals, bulliform cells, vascular bundles, micro hairs, influences. Along with taxonomic classification, structural differences identified in this study may hint at ecological roles, for example, silica cells provide mechanical strength, while trichomes and bulliform cells are involved in regulation of water. However, functional testing is needed to confirm these possibilities. Beyond taxonomy, the present work offers practical applications for sustainability and conservation. As bamboo varieties are used in commercial industries, forestry and in control of erosion, accurate species identification is essential. By complementing molecular techniques such as genome sequencing or DNA barcoding, anatomical investigations provide an economical and powerful approach to comprehend bamboo species, thereby ensuring precise species identification. Benefiting the conservation of biodiversity and nursery operations, the anatomical markers seen here gives a valuable tool for early-stage grouping. To summarize, by developing a reliable anatomical framework for identifying the *Dendrocalamus* species, this study contributes to conservation efforts and botanical research. A better knowledge of bamboo's taxonomy and adaptation techniques will be crucial for its sustainable use and preservation as it continues to gain significance on a worldwide scale in both ecological and economic contexts.

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