

# Morphoanatomy of *Guadua acreana*, a giant bamboo from the amazon

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## **TECHNOLOGY OF FOREST PRODUCTS**

#### **ABSTRACT**

**Background:** *Guadua* is a bamboo genus widely distributed in the state of Acre and holds significant potential for various applications; however, it remains largely underexplored. Bamboo morphology and anatomy are fundamental for accurate botanical identification, providing valuable insights for bioprospecting and influencing its functional properties. This study aimed to describe the morphology and perform an anatomical characterization of mature culms of *Guadua acreana*.

**Results:** Mature culms were collected from a bamboo grove located on a private lot adjacent to FUNTAC (Fundação de Tecnologia do Estado do Acre), at geographic coordinates Latitude 9°56'46.01" South and Longitude 67°52'8.86" West. A total of 10 culms were analyzed at FUNTAC. Anatomical sections were prepared and examined using optical microscopy, revealing size variations in the vascular bundles, with increasing metaxylem and phloem dimensions toward the inner culm region, which may influence mechanical properties or water conduction efficiency. The vascular bundles of *G. acreana* were classified as type V, characterized by a central vascular strand surrounded by condensed sclerenchyma sheaths and fiber cords.

**Conclusions:** This study enhances the anatomical understanding of *G. acreana* and highlights its potential applications in reforestation, construction, furniture making, and pulp and paper production. Future research should focus on the mechanical properties and chemical composition of this species to improve its industrial utility.

**Keywords:** Bambuseae; Botanical identification; Functional morphology; Anatomical features; Vascular Bundles.

## **HIGHLIGHTS**

A description of morphology and anatomy of mature *Guadua acreana* culms is provided. *G. acreana* has darker green leaves than *Bambusa vulgaris*, with distinct ligules and auricles. Vascular bundles showed size variations, with metaxylem and phloem increasing inward. Vascular bundles classified as type V, with a central strand and condensed sclerenchyma. Illustrations of vascular bundles are provided enhancing clarity and didactic value.

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#### INTRODUCTION

There are approximately 1,642 scientifically catalogued bamboo species worldwide, distributed across 119 genera (Clark et al., 2015; INBAR, 2019). However, new species and varieties continue to be discovered regularly. In Brazil, 258 native bamboo species have been identified, belonging to 35 genera, including the genus *Guadua* (Filqueiras and Viana, 2017; Drumond and Wiedman, 2017).

The genus *Guadua* Kunth (subtribe Guaduinae) is a group of tropical bamboos endemic to the Americas. It includes some of the largest and most economically important bamboo species in tropical America (Londoño and Clark, 2002). Comprising approximately 30 species, *Guadua* is distributed from Mexico to northern Argentina and Paraguay, with its center of diversity located in the Western Amazon and the Orinoco Basin (Shirasuna, 2023).

*Guadua* is characterized by its arborescent growth habit, with sizes ranging from medium to large. Its culms and branches bear thorns, facilitating anchorage to other plants during growth (Londoño and Peterson, 1991). Due to their rapid growth and pachymorph rhizome system, *Guadua* species can quickly colonize extensive areas of the forest understory and persist for long periods (Hechenberger et al., 2022).

Guadua species are semelparous, meaning they reproduce sexually only once in their lifetime; they are also monocarpic, dying after flowering (Silveira, 2005). This flowering pattern leads to synchronized, large-scale blooming and fruiting events, followed by mass mortality of the population (Hechenberger et al., 2022).

In Brazil, 22 species of *Guadua* have been described, of which six species and one subspecies are considered endemic: *G. calderoniana*, *G. maculosa*, *G. magna*, *G. refracta*, *G. tagoara*, *G. tagoara* subsp. *glaziovii*, and *G. virgata* (Reflora, 2023). The state of Acre holds the largest natural reserves of *Guadua* (Silveira, 2005; Pereira and Beraldo, 2007; Silva, 2019).

Located in the southwestern Brazilian Amazon, Acre is a Brazilian state renowned for its extensive open bamboo forests (Silva et al., 2019; Silva et al., 2020). These forests cover an area of approximately 165,000 to 180,000 km², extending into southeastern Peru and northern Bolivia (Silveira, 2005).

The most common bamboo species in Acre are *G. sarcocarpa* Londoño & P.M. Peterson and *G. weberbaueri* Pilg. (Carvalho et al., 2013). The Amazonian species *G. superba* Huber, *G. glomerata* Munro, and *G. paniculata* Munro are also present in Acre's flora (Reflora, 2023). Recently, four additional species have been described for the state of Acre: *Guadua chaparensis* Londoño & Zurita, *Guadua* aff. *chaparensis* Londoño & Zurita, *Guadua lynnclarkiae* Londoño, and *Guadua* aff. *lynnclarkiae* Londoño (Silva et al., 2020).

*G.* aff. *lynnclarkiae*, commonly known as "taboca, tabocão, or taboca gigante," is an arboreal bamboo species that grows up to 27 m tall. It is a woody, thorny species characterized by pachymorph rhizome and long reproductive cycles. Its culms are cylindrical and hollow,

with diameters ranging from 9 to 17 cm (Silva, 2019). In 2017, the natural occurrence of this species was recorded in vegetation patches in Vila do V, in the municipality of Porto Acre, Acre.

The classification of bamboos primarily relies on morphological characteristics (Clark et al., 2015). However, the documentation of many bamboo species is often incomplete due to the challenges in taxonomic studies, which also depend on information about inflorescence and floral morphology (Shalini et al., 2013). This complicates the use of morphological characteristics for precise species identification, as bamboo species typically flower only once in their lifetime. For many species, information about the timing of flowering is lacking, and some species have life cycles that include flowering after 20 to 60 years of age (Benton, 2015).

Accurate identification of plant species is a significant challenge in bioprospecting studies. Recognizing a specific specimen corresponding to an existing description or another identified plant is critical, as incorrect or imprecise identification can compromise the entire bioprospecting effort (Matos, 2009).

Genetic differentiation studies use morphoanatomical characterization through observable, distinguishable, and measurable qualitative and quantitative descriptors. Investigating the genetic divergence of bamboos is essential for understanding their evolutionary processes, speciation, conservation, and the optimal use of resources from the most promising species. By integrating both morphological and anatomical analyses, this approach offers several advantages, including practicality, cost-effectiveness, ease of data management, and greater efficiency in quantifying genetic divergence among accessions (Pereira, 2010).

This study hypothesizes that the morphology and anatomy of *G. acreana* reveal unique structural characteristics that can aid in the identification and differentiation of native Amazonian bamboo species. By focusing on *G. acreana*, a recently reclassified species endemic to the state of Acre (Afonso, 2024), this research introduces innovation to the field of forest resources.

Specifically, the objectives of this study are to: (1) describe the morphological characteristics of G. acreana, highlighting distinguishing features from other bamboo species; (2) analyze the anatomical structure of its culms; (3) compare the morphological and anatomical traits of G. acreana with those of other bamboo species to establish clear criteria for identification; and (4) provide a comprehensive overview of its morphoanatomy, thereby contributing valuable insights for future species identification and bioprospecting efforts. Consequently, this study significantly advances our understanding of Amazonian bamboo diversity, particularly in a region that remains underexplored (Hopkins, 2007; Carvalho et al., 2023). Notably, this work represents one of the first integrated analyses of the morphology and culm anatomy of Guadua acreana Londoño, Afonso & P.L. Viana.

#### **MATERIALS AND METHODS**

#### **Collection and Identification of Plant Material**

For the study of the morphology and anatomy of the culms of *G. acreana* and its botanical identification, plant material was collected in October 2021 from an area established in 2009 by the research group of the Acre State Technology Foundation (FUNTAC), as documented by Sola et al. (2023a) and Sola et al. (2023b). The materials were collected from a bamboo grove located on a private lot adjacent to FUNTAC, at the following geographic coordinates: Latitude 9°56′46.01″ South and Longitude 67°52′8.86″ West, and an altitude of 143 m (Figure 1). The regional climate is humid equatorial, with an annual precipitation of approximately 2,200-2,500 mm and an average annual temperature ranging from 22°C to 26°C (ACREBIOCLIMA, 2022).

According to reports from Mr. Manoel Rodrigues de Souza Sobrinho, a long-time employee of FUNTAC, the bamboo seeds were collected along the banks of the Purus River and planted in 2009 near the FUNTAC nursery, in degraded soil and without specific spacing. The area has since served as a bamboo germplasm bank in the field close to the FUNTAC location.

The plant material was randomly collected using a destructive sampling method. A total of ten bamboo culms from one clump were sectioned with a portable chainsaw

and further divided with a machete to remove leaves and small branches, preparing the specimens for drying. Voucher specimens were prepared from the population sample and deposited in the Herbarium of the Zoobotanical Park at the Federal University of Acre under the identification code UFACPZ 22247. A free search for the specimen registered in the herbarium can be conducted through the website: https://specieslink.net/search/. At the time, the specimens were identified as *Guadua* aff. *lynnclarkiae* Londoño, with the assistance of photographs that were sent to the botanist Ximena Londoño. However, according to Afonso (2024), *Guadua* aff. *lynnclarkiae* has been reclassified as a new species endemic to Acre, now named *Guadua acreana* Londoño, Afonso & P.L. Viana.

## Morphological Characterization of Guadua acreana

Morphological characterization was conducted based on qualitative and quantitative descriptors, following the methodology of Generoso et al. (2016). A total of 14 qualitative descriptors were established to evaluate the culm, branching, and leaves, including habit, shape and color of the culm, node characteristics, thorns, pubescence, internode filling, color and pubescence of the culm sheath, as well as the position, pubescence, and color of the sheath blade, and the development of auricles and ligules. Additionally, three quantitative descriptors for the culm were used: plant height, diameter, and wall thickness.



Figure 1: Localization of bamboo collection area.

To assess the color of the leaf blade, the green intensity scale proposed by Generoso et al. (2016) was used to determine the intensity of green shades in the leaf blades of the branches. For quantitative characterization, the known height of an individual was used as a reference for measuring the height of bamboo culms using trigonometric methods. The fundamental principle involves the formation of similar triangles, in which the ratio of the corresponding sides (including the heights of the bamboo and the individual) remains constant. The diameter and wall thickness were measured with a tape measure after harvesting. Samples from the base of the culms were collected and then dried in a forced-air oven at 100 °C for 48 hours.

## **Microscopic Anatomical Characterization**

The bases of the internodes of the culms were prepared into blocks through longitudinal cuts made parallel to the fibers, using a wide scalpel blade with approximately 1 cm spacing between each cut. Subsequently, a transverse cut was made across the fibers, producing blocks measuring 2 cm in height and 1 cm in width. The samples were preserved in 70% ethanol until analysis. For processing, these samples underwent microtomy using a Leica SM 2010R sliding microtome (Figure 2 A-C), which produced transverse sections with a thickness ranging from 35 to 40 µm for further evaluation.

The qualitative characterization of the vascular bundles followed the methodologies proposed by Grosser

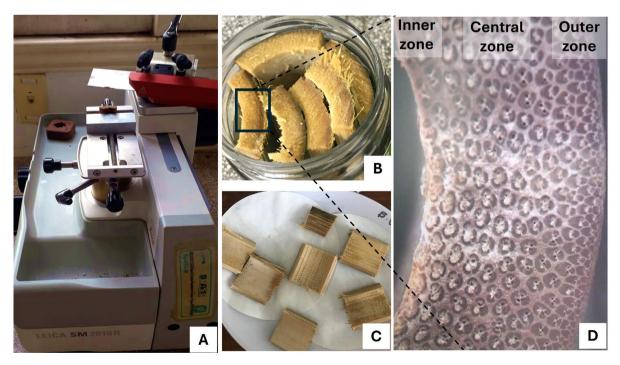
and Liese (1971), Liese (1985), and Rusch et al. (2018). The slide preparation followed the modified Franklin method described by Berlyn and Miksche (1976). The transverse sections, 35-40  $\mu m$  thick, were immersed in a 1% sodium hypochlorite solution for 2 min, followed by washing with distilled water. The sections were then stained with Basic Red Safranin for 2 min and subjected to sequential alcohol immersions at 50%, 70%, and 96% concentrations. Finally, the slides were rinsed with distilled water to remove excess stain.

A total of 50 images were captured from four histological slides using a Leica Application Suite camera attached to a microscope (LAS EZ Leica Microsystems version 3.4). The slides were divided into three zones of the *G. acreana* culm: the outer zone, central zone, and inner zone (Figure 2D). The vascular bundles were compared with the classification established by Liese (1985). Images were captured using TCapture software. Illustrations were created using Ibis Paint X software to enhance clarity and make the visualized microscopic structures more comprehensible and didactic.

#### **RESULTS**

#### Morphological characterization of Guadua acreana

The classification of bamboo is primarily based on morphological characteristics, which, although influenced by environmental factors, are deemed reliable and crucial for studies on diversity (Das et al., 2008; Silva, 2019). In our



**Figure 2:** Sample preparation and analysis. (A) Leica SM 2010R sliding microtome with a 1 x 2 cm sample. (B) Fragmented bamboo stored in glass jars with 70% alcohol until analysis. (C) Fragmented bamboo used for analysis. (D) Representation of the anatomical zones in the cross-section of the bamboo culm *Guadua acreana*.

study, we observed that *G. acreana* individuals exhibit an arborescent habit (Figure 3A), characterized by an upright base and a little curved apex. The culm internodes are cylindrical and hollow, green and free of spots on the internodes, when young. The nodes of the young culm are white, adorned with a distinct band of trichomes (Figure 3B). In contrast, the mature culm is distinguished by prominent horizontal nodal lines and a darker hue (Figure 3C). Mature culm exhibits a dull brownish-green color, marked by spots caused by lichen growth. Additionally, solitary, spiny primary branches were observed, with two to five thorns per node (Figure 3D).

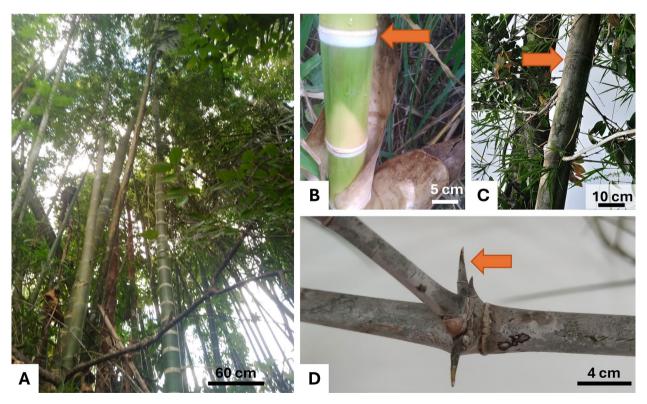
The culms of the species feature an erect, coriaceous culm leaf sheath, triangular shape, and exhibit a distinctive brownish-red coloration (Figure 4A). The culm leaf sheath is characterized by the presence of conspicuous pubescence, consisting of small dark-toned trichomes that may cause skin irritation. We observed that the culm leaf sheath measures between 36.5 cm (in young culms) and 45 cm (in mature culms) in length, and between 25 cm (in young culms) and 35 cm (in mature culms) in width.

The leaves of the branches are dark green, elliptic-lanceolate, and measuring an average of 21 cm in length for young culms and up to 40 cm for mature culms. The width ranges from 1.5 cm for young culms to 2.8 cm for mature ones. During pruning, the green leaves fold longitudinally, adopting a linear appearance. Compared

with *Bambusa vulgaris* Schrad. ex J.C.Wendl., *G. acreana* shows darker green leaves, similar in hue to those of *G. weberbaueri* (Figure 4B).

Upon analyzing the presence of pubescence on the adaxial and abaxial surfaces, as well as along the margins of the leaf blades, it was noted that the species under investigation exhibits pubescence on both surfaces and along the margins of the blades (Figure 4C). Regarding auricle development, the species was found to possess discrete auricles (Figure 4D). Additionally, at the junction between the blade and the sheath, an external ligule was identified on the abaxial surface of the leaf (Figure 4E). In our study, we observed branches with 8 green leaves, ligules measuring up to 3 mm long, and auricles measuring 1.5 mm long.

Each bamboo species exhibits quantitative morphological traits, such as height, diameter, wall thickness, and growth rates, which are distinct and vary with age (Nunes et al., 2021). At 12 years old (at the time of measurement), mature bamboo reaches 14 m in height, with internodes length of 30 cm, diameter of 9 cm, and a wall thickness of 1.9 cm. In contrast, young bamboo reaches a height of 12 m, with internodes length of 28 cm, diameter of 11 cm, and a wall thickness of 2.3 cm. *G. acreana* has culms ranging from 20 to 27 m in height and 9 to 17 cm in diameter, as described by Silva (2019). According to Afonso (2024), *G. acreana* can exceed 30 m in height, with internode diameters reaching up to 22 cm.



**Figure 3:** Characteristics of *Guadua acreana*: (A) General aspect of the clump. (B) Young culm, with an arrow indicating the node with white coloration. (C) Mature culm, with an arrow indicating the absence of coloration at the node. (D) Portion of the branch highlighting the thorns present at each node, varying from 2 to 5 thorns per node.

These morphological differences, along with anatomical characteristics, provide a reliable basis for distinguishing *G. acreana* from other species of the genus as shown in Table 1.

### **Microscopic Anatomical Characterization**

The anatomical structure of *G. acreana* was analyzed from cross sections of the culm wall. The proportion of vascular bundles and parenchymatous tissue varies across the culm wall, influencing the mechanical properties of the bamboo.

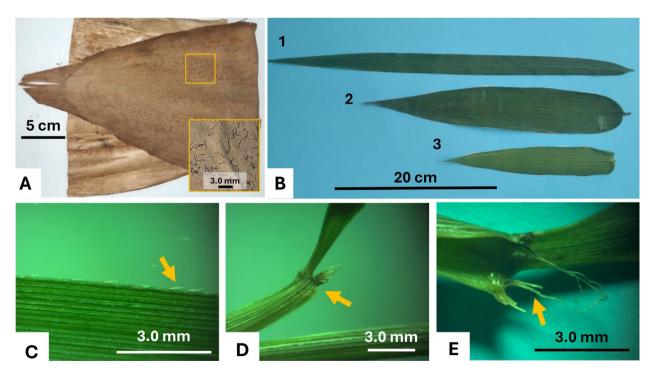
In a recent study, Portal-Cahuana et al. (2023) observed that the culm consists of vascular bundles and parenchymatous tissue, showing a gradual distribution of vascular bundles from the outer to the inner region. Three distinct regions can be recognized within the bamboo wall: outer, central, and inner regions (Figures 5A-C). The outer region shows small, dense vascular bundles surrounded by thick sclerenchymatous sheaths. In the central region, the vascular bundles are larger, and the amount of parenchymatous tissue increases (Figure 5B). The inner region is characterized by even larger vascular bundles, with thinner sclerenchymatous sheaths and a higher proportion of parenchymatous cells. When examining the vascular bundles in isolation, Figure 5D reveals a fiber cord positioned at the lower section of the sclerenchyma sheath. Additionally, Figure 5E illustrates a fibrovascular bundle that features a fiber cord located at the upper part of the sclerenchyma sheath of the phloem, and along with intercellular spaces originating from the protoxylem. Finally, Figure 5F depicts a fibrovascular bundle found in the inner zone, comprising a central vascular strand accompanied by a fiber cord situated above the sclerenchyma sheath of the phloem.

Anatomical variations in vascular bundles enable the classification of the genus into five distinct types: I, II, III, IV, and V. According to Liese (1985), four to five major vascular bundle types can be distinguished:

**Type I**: Comprising a single central vascular strand, with supporting tissue exclusively in the form of sclerenchyma sheaths. This type is characteristic of genera such as *Arundinaria*, *Phyllostachys*, *Fargesia*, and *Sinobambusa*.

**Type II**: Also consisting of a single central vascular strand with supporting tissue restricted to sclerenchyma sheaths. A distinguishing feature is the presence of a significantly larger sheath surrounding the intercellular space (protoxylem) compared to the other three sheaths. This type is observed in genera such as *Cephalostachyum*, *Pleioblastus*, *Melocanna*, and *Schizostachyum*.

**Type III**: Composed of two structural components: a central vascular strand with sclerenchyma sheaths and a single isolated fiber cord. This type is found in species belonging to *Melocanna*, *Schizostachyum*, *Bambusa*, *Dendrocalamus*, *Gigantochloa*, *Sinocalamus*, and *Oxytenanthera* (for which only Type III has been reported).



**Figure 4:** (A) Culm leaf sheath of *Guadua acreana* reddish-brown in color. Highlighted, conspicuous trichomes on the surfaces of the sheath blade that resemble small spines. (B) Coloration of the photosynthetic leaves of three bamboo species: 1 - *Guadua acreana*; 2 - *Guadua weberbaueri*; 3 - *Bambusa vulgaris*. (C) Pubescence on the photosynthetic leaf of *Guadua acreana*. Trichomes along the blade margin resembling eyelash-like structures at 40x magnification. (D) Discrete auricles observed at 20x magnification. (E) Liqules visible at 40x magnification.

Table 1: Key characteristics that distinguish Guadua acreana from other giant Guadua species.

Characteristics	G. acreana	G. lynnclarkiae	G. chaparensis	G. angustifolia
Photosynthetic leaves	8-10 per complement	10-12 per complement	5-10 per complement	2-12 per complement
Leaves shape	Lanceolate, Elliptic- lanceolate or linear- lanceolate	Lanceolate to linear- lanceolate	Lanceolate with fine margins	Lanceolate or oblong
Leaves length	21-40 cm	15-23 cm	4-15 cm	6-25.6 cm
Leaves wide	1.5-2.8 cm	1.6-4.2 cm	0.6-2 cm	0.6-5 cm
Trichomes	Present, pubescence on both surfaces and ciliate margins	Present, adaxial surface pubescent, abaxial surface glabrescent, ciliate margins	Present, glabrescent toward the margins	Present, usually glabrous to sparsely pubescent, margins scabrous
Ligules	Present (up to 3 mm)	Present (2-3 mm)	Present	Present (0.1-1 mm)
Auricles	Discrete	Present	Not specified	Absent, if present mainly on juvenile leaves
Culm Diameter	Up to 22 cm	6-17 cm	3.5-12 cm	7 to 23 cm
Culm Height	Can exceed 30 m	Up to 27 m	Up to 25 m	Up to 30 m
Internode length	28-30 cm	15-49 cm	22-70 cm	14-34 cm
Wall thickness	1.9-2.3 cm	2-5 cm	1-1.5 cm	1-3 cm
Thorns	Present (2-5 per node)	Present (1-5 per node)	Curved thorns on the secondary branches (1-4 per node)	Present (1-5 per node)
Culm leaf at the base		Coffee-colored, measuring 68 to 91 cm in length and 68 to 72 cm in width	Whitish-green with red markings when young, transitioning to cream-brownish as it matures; measuring 29 to 57 cm in length and 35 to 54 cm in width	Coffee-colored, measuring 18 to 78 cm in length and 9 to 55 cm in width
References	Afonso (2024); these authors	Londoño (2013)	Londoño and Zurita (2008)	Young and Judd (1992); Londoño (2013)

**Type IV**: Characterized by three structural components: a central vascular strand with small sclerenchyma sheaths, accompanied by two isolated fiber cords located externally and internally to the central strand. This type includes species of *Bambusa*, *Dendrocalamus*, *Gigantochloa*, and *Sinocalamus*.

**Type V**: A semi-open vascular bundle type, representing a further step in the evolutionary continuum. Although the authors did not specify any genera exhibiting this type, they indicated its existence.

A controversial classification for *Guadua angustifolia* Kunth has been proposed, designating it as type IV vascular bundle, as noted by Prado Gárate et al. (2021). In our analysis, which involved a qualitative anatomical characterization of the vascular bundles across the three zones of the culm wall in *G. acreana*, we did not observe the three distinct components characteristic of type IV, namely a central vascular strand and two separate fiber cords. In the central zone of *G. acreana*, we identified two fiber cords; however, these were situated together with the central vascular strand (Figure 6).

Then, the vascular bundles were classified as type V according to the system proposed by Liese (1985),

characterized by a central vascular strand surrounded by sclerenchymatous sheaths and condensed fiber cords (Figure 6A). Each vascular bundle consists of a metaxylem vessel, a protoxylem vessel, and a phloem portion associated with sclerenchymatous cells (Figure 6B). The presence of a fiber cord located at the lower part of the sclerenchyma sheath surrounding the metaxylem and protoxylem is evident, while Figure 6C depicts a fibrovascular bundle comprised of a fiber cord situated at the upper portion of the sclerenchyma sheath of the phloem and presence of intercellular space derived from protoxylem. The supporting tissue contains sclerenchyma sheaths within the central vascular strand, prominently featuring a protoxylem sclerenchyma sheath that is significantly larger than the others. Additionally, the fiber cords surrounding the protoxylem and metaxylem are densely clustered, contributing to the structural support around the central vascular strand.

The phloem tissue is located adjacent to the xylem and associated with fibers and the ground tissue is mainly composed of parenchymatous cells, which are polygonal and thin-walled. Figures 6D and 6E depict fibrovascular bundles in the central zone,

consisting of a central vascular strand and two fiber cords. One fiber cord is positioned above the phloem sclerenchyma sheath, while the other is located near the protoxylem sclerenchyma sheath. The supporting tissue also contains sclerenchyma sheaths within the central vascular strand, with fiber cords adjacent to the protoxylem and metaxylem condensed along the central vascular strand. It is important to note that the protoxylem sclerenchyma sheath decreases in size as it approaches the inner zone.

Sclerenchymatous fibers are long and thick-walled, forming a continuous sheath around the vascular bundles. Figure 6F illustrates a fibrovascular bundle in the inner zone, comprising a central vascular strand and a fiber cord located above the phloem sclerenchyma sheath. The supporting tissue shows sclerenchyma sheaths within the central vascular strand, with the protoxylem sclerenchyma sheath significantly smaller compared to others.

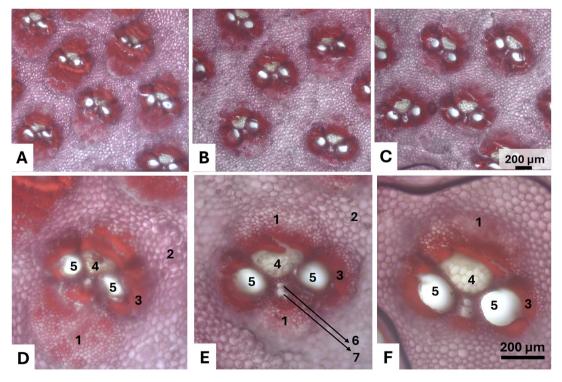
Recognizing the significance of comprehending the anatomical classification of vascular bundles, we propose an illustration of type V vascular bundles, showcasing their distribution in the outer (Figure 7A, 7B and 7C), central (Figures 7D and 7E), and inner (Figure 7F) zones of the culm wall in *G. acreana*. Grosser and Liese (1971) differentiated the zones of bamboo culms into four categories: peripheral,

transitional, central, and interior. In the present study, we did not employ this differentiation for the species analyzed; however, it is important to note that both the peripheral-to-central and central-to-interior transitions occur, as evidenced by the anatomy of the vascular bundles detailed in Figure 7.

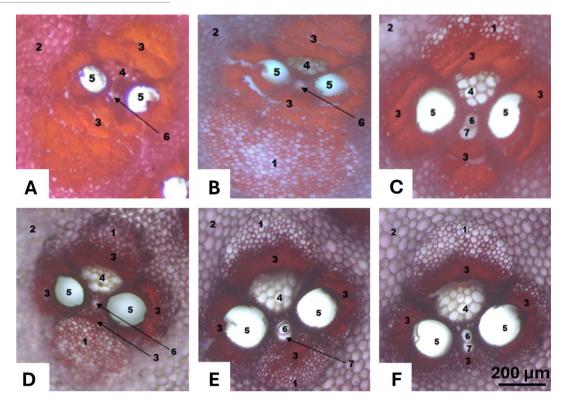
According to Grosser and Liese (1971) and Liese (1998), the vascular system of bamboo culms is characterized by the presence of one or two protoxylem elements. In our study, we demonstrate that the species *G. acreana* exhibits up to three protoxylem elements in its outer zone (Figures 8A and 8B). Additionally, illustrations of these bundles are provided (Figures 8C and 8D).

#### **DISCUSSION**

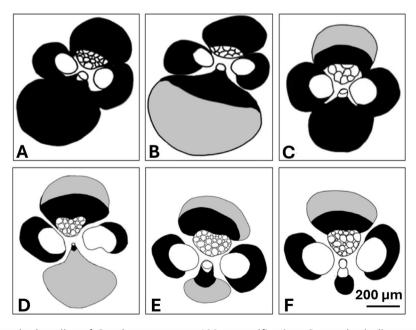
The morphological and anatomical features of *Guadua acreana* described in this study contribute to understanding the taxonomic and functional diversity of Amazonian bamboos. The morphological parameters of *G. acreana* fall within the general characteristics observed for the genus; however, certain traits, such as culm color and the shape of the sheath blade, are distinct. These traits are important for accurate species identification, that distinguish *G. acreana* from other *Guadua* species.



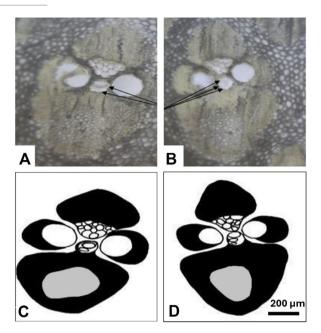
**Figure 5:** (A, B, C) Vascular bundles in the three zones of the culm wall of *Guadua acreana* at 40x magnification: (A) Outer zone. (B) Central zone. (C) Inner zone. (D, E, F) Vascular bundles of *Guadua acreana* at 100x magnification. (D) Vascular bundles from the outer zone. (E) Vascular bundle from the central zone. (F) Vascular bundle from the inner zone. Legend: 1 - fiber cord; 2 - parenchyma; 3 - sclerenchyma sheath; 4 - phloem; 5 - metaxylem; 6 - protoxylem; 7 - intercellular space derived from protoxylem.



**Figure 6:** Type V vascular bundles of *Guadua acreana* at 100x magnification. These figures served as models for creating the schematic material presented in Figure 7. (A, B, and C) Vascular bundles from the outer zone. (D and E) Vascular bundle from the central zone. (F) Vascular bundle from the inner zone. Legend: 1 - fiber cord; 2 - parenchyma; 3 - sclerenchyma sheath; 4 - phloem; 5 - metaxylem; 6 - protoxylem; 7 - intercellular space derived from protoxylem.



**Figure 7:** Type V vascular bundles of *Guadua acreana* at 100x magnification. Gray color indicates the fiber cord of the vascular bundle, black designates sclerenchyma sheath, and white highlights the metaxylem, protoxylem, and phloem. The fibers in the fiber cord are less dense than those in the sclerenchyma sheath, represented by lighter coloration. (A, B, and C) Vascular bundles located in the outer zone; (D and E) Vascular bundles located in the central zone; (F) Vascular bundles located in the inner zone.



**Figure 8:** Vascular bundles from the outer zone of *Guadua acreana* at 100x magnification. (A) Vascular bundle with two protoxylem elements. (B) Vascular bundle with three protoxylem elements. (C) Schematic illustration of a vascular bundle with two protoxylem elements. (D) Schematic illustration of a vascular bundle with three protoxylem elements. Legend: arrows indicate protoxylem elements.

Quantitative morphological characteristics play a crucial role in selecting the most suitable bamboo species for various applications (Wahab et al., 2010). Taller bamboos with greater wall thickness and culm diameter are often utilized in construction, charcoal production, and furniture manufacturing (Generoso et al., 2016), as well as for building materials (Vasata et al., 2021). In contrast, species with shorter culms and smaller diameters are recommended for ornamental and landscaping purposes (Generoso, 2016).

The analyzed culms of *G. acreana* are upright and straight, with 12-year-old specimens averaging a height of 12 to 14 m. In our study, mature culms exhibited an average diameter of 9 cm and a wall thickness of 1.9 cm, whereas young bamboos showed a larger average diameter of 11 cm and a wall thickness of 2.3 cm. The observed differences in diameter and wall thickness between young and mature culms can be attributed to the higher turgor pressure in the cells of young bamboo, which is associated with their increased water content. In contrast, mature bamboos often display lower water content, which may influence these morphological measurements. As a bamboo clump matures, new culms emerge with progressively larger diameters, while older culms typically exhibit smaller diameters compared to the newest shoots. It is important to note that each new culm develops with a defined diameter and does not increase in diameter after its initial emergence, as bamboo lacks secondary growth (Long et al., 2023).

The species *G. acreana* is distinguished from *G. lynnclarkiae* by the presence of velutinous cataphylls. Our analysis revealed that unique characteristics were

identified, including small thorns on the branches and a reddish-brown sheath blade adorned with small dark brown trichomes resembling spines. In the green leaves, the species exhibits pubescence on both the adaxial and abaxial surfaces, as well as along the margins of the leaf blades. Additionally, discrete auricles were identified, and an external liqule was present at the junction between the blade and sheath on the abaxial surface. We observed branches with 8 green leaves, liqules reaching lengths of up to 3 mm, and auricles lengths of up to 1.5 mm. According to Afonso (2024), the culm leaf sheath is either coriaceous or chartaceous, measuring between 45 and 86.9 cm in length and 39 to 76 cm in width, with an inner liqule measuring 1.2 to 2 mm long. The foliage leaves number between 5 and 8 per complement, featuring an inner ligule of 0.2 to 0.3 mm in length, with margins that have trichomes. The outer ligule ranges from 0.3 to 0.4 mm, is chartaceous, and has smooth margins. Depending on the stage of development, the botanical structures may vary.

In contrast, *G. lynnclarkiae* is characterized by pubescent cataphylls and a coriaceous culm leaf sheath measuring between 68 and 91 cm in length and 68 to 72 cm in width, featuring an inner ligule that is 2 to 3 mm long. The foliage leaves number between 10 and 12 per complement, with an inner ligule approximately 0.5 mm long and margins that may be smooth or have trichomes (Londoño, 2013). *G. lynnclarkiae* differs from *G. angustifolia* in that it has cauline leaves with oral setae. The leaf blade measures 15 to 23 cm in length and 1.6 to 4.2 cm in width, exhibiting falcate-lanceolate auricles and fimbriae on the upper part of the leaf sheath. The leaf blade of *G. angustifolia* ranges

from 6 to 20 cm in length and 0.5 to 2 cm in width and does not have auricles (Londoño, 2013).

*G. acreana* can be distinguished from *G. chaparensis* by the latter's leaf blades that are one-fourth to one-fifth the length of the sheath, varying from 7 to 12 cm. The adaxial surface is densely pubescent between the nerves, featuring two types of hairs: firm, transparent hairs up to 2.5 mm long and short, soft hairs up to 0.5 mm. The margins are papery, shiny, and smooth in the middle and upper portions, with ciliation at the base and conspicuous fimbriae up to 2.5 mm long. Additionally, the leaf blades of *G. chaparensis* are glabrous on the abaxial surface (Londoño and Zurita, 2008).

Observed changes in color between young and mature culms may serve as a practical guide in the field. These distinguishing characteristics can aid in the selection of bamboo culms within a clump, facilitating their allocation to various applications, particularly those that demand specific qualities. We observed that mature culms of G. acreana are characterized by a dull brownish-green color, marked by spots caused by lichen growth, and prominent horizontal nodal lines. Mature bamboo culms can host fungi and lichens on their surfaces. This phenomenon is attributed to the increased resistance of bamboo as it ages, due to reduced moisture and sap circulation, which lowers the risk of insect attacks. Furthermore, the position of the bamboo within the clump influences the colonization of these organisms. Culms that are more exposed to direct sunlight and wind tend to have a lower incidence of fungi and lichens, while those in shadier, more sheltered positions are more prone to colonization (Greco and Zannin, 2017).

Leaf coloration is a fundamental characteristic for identifying bamboo species in their natural environment and can serve as a visual parameter for distinguishing between individuals. Advances in image analysis technology have enabled the use of drones equipped with RGB sensors to estimate plant biomass through artificial intelligence. Moscovini et al. (2024) demonstrated that the Normalized Difference Vegetation Index (NDVI) can be accurately predicted from calibrated RGB images, eliminating the need for costly hyperspectral or multispectral sensors. This methodology enables the application of affordable remote sensing techniques for small-scale farmers and researchers seeking to monitor vegetation and estimate bamboo biomass in tropical forests, providing valuable data for the sustainable management of these species.

Furthermore, variations in leaf coloration can significantly influence ecophysiological processes, such as light absorption and photosynthesis. Generally, darker green leaves tend to exhibit higher rates of photosynthesis compared to lighter green leaves (Zhao et al. 2020; Su et al. 2023). Consequently, the leaf coloration of native species *G. acreana* and *G. weberbaueri* can indicate their greater efficiency in carbon sequestration compared to the exotic species *B. vulgaris*, reflecting their superior adaptability to their respective environments.

In addition to the darker green coloration of its leaves, *Guadua* can be distinguished from *Bambusa* by the presence of thorns on its culms and branches. Furthermore, *Guadua* features a triangular culm leaf where the margins of

the sheath and blade are contiguous or nearly so, and it has a distinctive band of hairs on the nodal lines (Soderstrom and Londoño, 1987). Additionally, *Guadua* differs from *Bambusa* in its geographic distribution; *Guadua* is known as American bamboo, while *Bambusa* has its center of origin in Asia (Young and Judd, 1992). Specifically, the mature culms of *B. vulgaris* are characterized by their green or yellow coloration, often adorned with green stripes, further distinguishing this species within the genus (Wahidah et al., 2021).

The anatomical composition of culms, particularly in terms of parenchyma cells, can vary significantly based on species, seasonal changes, and the age of the culms (Nunes et al., 2021). In *G. acreana*, the culm consists of a diverse array of tissue types, including parenchyma cells, vascular bundles, and fibers. Examination of the three zones of the culm wall revealed that the outer zone is characterized by denser fiber bundles and smaller vascular elements. As one moves inward toward the central and inner zones, there is a gradual increase in the number of parenchyma cells. In these inner zones, the vascular bundles become more widely spaced and are accompanied by a comparatively lower density of fibers.

Grosser and Liese (1971) reported that the vascular bundles in the outer zone of bamboo culms are smaller and more numerous, resulting in a reduced presence of parenchyma cells interspersed among them. Li and Shen (2011) demonstrated that both the longitudinal modulus of elasticity and the strength of the vascular bundles increase linearly from the inner to the outer regions of the culms.

According to Rusch et al. (2018), the anatomical differences in vascular bundles are influenced by internode height and wall thickness, resulting in distinct structures in the internodes at the base compared to those in the middle and upper portions of the culm. While there is a general pattern of fibrovascular bundles within the internodes, variations in their shape, size, and quantity are observed across the outer, central, and inner zones (Liese, 1985). This indicates a correlation between morphological and histological characteristics, particularly as anatomical differences become more pronounced with increasing wall thickness

Grosser and Liese (1971) and Liese (1985) reported that all leptomorph bamboo genera exhibit a similar vascular bundle structure, characterized by the simple Type I. Furthermore, their study demonstrated that pachymorph bamboo genera could be classified into three distinct vascular bundle types (II, III, and IV). The anatomical characterization showed that the vascular bundles of *G. acreana* correspond to the type V pattern, similar to that observed in other species of the genus, such as *G. angustifolia* and *G. weberbaueri* (Brea and Zucol, 2007; Rodrigues et al., 2020), suggesting a broader structural evolutionary continuum within the genus.

According to Liese (1985), type V bundles are characterized by a central vascular strand surrounded by a continuous sclerenchymatous sheath and fibers, which confer mechanical strength to the culm. These findings highlight the need for further anatomical investigations

to refine the classification of vascular bundle types in *Guadua* bamboos.

This anatomical pattern is consistent with the findings of Amada et al. (1996), who observed a correlation between the density of vascular bundles and the mechanical strength of bamboo species. Janssen (1981) found that the outer region of the culm is more resistant to tension and compression due to the higher concentration of fibers.

Portal-Cahuana et al. (2023) observed that *B. vulgaris* possesses both types III and IV vascular bundles, while *Gigantochloa apus* (Schultes) Kurz. contains only type III bundles. They also classified *G. weberbaueri* as having solely type III. However, when comparing their findings with others, Rodrigues et al. (2020) noted that in *G. weberbaueri*, the fiber cords are condensed along the central vascular strand, a characteristic also observed in *G. acreana*.

A significant finding of this research is the reclassification of the type of vascular bundle. Previous studies categorized other species of *Guadua* as type III or as type IV, however the vascular bundles of *G. acreana* are classified as type V because, although the fiber cord is present, it is not separate from the central vascular strand; rather, it is condensed alongside it. Thus, this study not only revises the classification proposed in earlier research but also enriches the literature by identifying a fifth type of vascular bundle within the genus *Guadua*. This new classification may enhance the identification and understanding of the anatomical characteristics of this genus.

The distribution of vascular bundles across the culm wall—denser in the outer region and larger toward the inner portion—directly influences the mechanical performance of the bamboo.

### CONCLUSION

The morphological and anatomical characterization of *Guadua acreana* provides new information about this bamboo species, which was recently reclassified and is endemic to the state of Acre.

The morphological traits, such as darker green leaves, smaller ligules, and shorter auricles, distinguish this species from other *Guadua* species and from *Bambusa vulgaris*.

This study provides a comprehensive analysis of *G. acreana*, highlighting an average height of 12 to 14 m, diameters ranging from 9 to 11 cm, and wall thickness of 1.9 to 2.3 cm at 12 years of age. Notable characteristics include small thorns on the branches and a reddish-brown sheath blade with dark brown trichomes resembling spines.

Anatomically, the culm of *G. acreana* presents type V vascular bundles distributed from the outer to the inner region of the wall, with an increase in metaxylem and phloem size toward the center, indicating a distinct anatomical structure that enriches the literature and revises previous classifications of other *Guadua* species as types III or IV.

The anatomical organization of *G. acreana* suggests that this species has favorable mechanical properties for

construction and structural applications. The findings support the integration of this species into agroforestry and reforestation initiatives in the Amazon region.

The results obtained in this study contribute to knowledge of Amazonian bamboo diversity and highlight the importance of *G. acreana* as a species of economic and ecological value for the region. Future studies should investigate the mechanical, chemical, and technological properties of this species to expand its potential applications in sustainable development.

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## **DATA AVAILABILITY**

The datasets supporting the conclusions are included in the article

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