

Physical and anatomical variations in culm characteristics of *Pseudoxytenanthera bourdillonii* (Gamble) H.B.Naithani – a lesser studied endemic bamboo in the Western Ghats, Kerala

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Abstract: *Pseudoxytenanthera bourdillonii* (Gamble) H.B.Naithani, locally known as “Arayambu”, represents one of the lesser-known bamboo species in the Western Ghats region of Kerala (restricted to Palakkad, Thrissur, Idukki and Malappuram districts). The present study evaluated physical and anatomical properties of this species including fibre characters across various culm height. Experimental methods outlined in IS 6874 (2008) were used for moisture content and basic density studies. Franklin method was adopted for fibre maceration. It was observed that the moisture content consistently showed a decreasing trend from bottom to top whereas the basic density found a reverse trend. The plant volumetric shrinkage reaches its peak in the middle of the culm. The highest fibre length (2.65 ± 0.16 mm) was noticed in the middle slivers taken from the bottom portion of culm. The fibre width and lumen width of the bottom part showed a significant difference compared to middle and top culm ($P < 0.05$). There was no significant difference in wall thickness between height position and radial position. The average Runkel ratio found to be greater than one. Vascular bundle numbers showed an increasing trend from

bottom to top. Additionally, the vessel diameter was higher in inner compared to middle and outer part. There was a significant difference in parenchyma diameter between outer and inner portion. Despite having a Runkel ratio greater than one, other fibre parameters of *P. bourdillonii* make it suitable for pulping and remains a favorable option for handicraft sector owing to its non-thorny nature substantial intermodal length, and attractive golden yellow color of both the culm and culm sheath. However, *P. bourdillonii* culms of two-year age group is less preferred for high strength demanding purpose due to high moisture content and lower basic density.

Keywords: *P. bourdillonii*, fibre characteristics, Runkel ratio, culm physical properties

Introduction

Non-Timber Forest Products (NTFPs) play a crucial role in sustaining livelihood for 80% of the world's population, meeting essential needs like livestock support, fuelwood, furniture etc. (Charlotte *et al.*, 2021). For centuries, bamboo has served as a socially and economically significant non-timber forest product, meeting the daily needs of millions of people across widely distributed tropical and subtropical regions (Ahmad *et al.*, 2021; Lobovikov *et al.*, 2004). With higher potential productivity, rapid growth and ease of propagation, bamboo emerges as a promising alternative to traditional wood resources, positioning itself as a significant non wood forest asset. (Zhan *et al.*, 2015). The continuous progress in technology has significantly amplified the potential application of bamboo, with nearly 4000 commercial products crafted from this versatile material now being utilized on a global scale (Singh, 2008). Globally, bamboo forest

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span a vast expanse of 36 million hectares, with India accounting for 16 million hectares, followed by China with 6.5 million hectares, and Brazil with 4 million hectares. The international trade value of bamboo was pegged at 68.8 billion USD in 2018, significantly influencing the lives of 2.5 billion people directly or indirectly worldwide (FSI, 2019; INBAR, 2019).

Bamboo stands out as a viable alternative to the diminishing resources of forest timber. Non-woody plants, including bamboo, offer various advantages, such as rapid growth cycles, quick maturation, modest requirements for irrigation and fertilization, and a low lignin content. The latter characteristic is particularly significant as it contributes to a reduction in the energy and chemicals needed during the pulping process, making bamboo a sustainable and environmentally friendly option for various industrial applications (Navaee-Ardeh *et al.*, 2004). Bamboo species, with their distinct properties, emerge as valuable alternatives in the realm of pulp and paper industry. The increasing demand for wood and fibre, and the low availability of traditional wood supplies, have prompted researchers to actively seek non wood plants possessing fibre properties comparable to those of wood (Worku *et al.*, 2023).

However, there are still some underexploited and lesser known endemic bamboo species, which if studied and properly utilized could bring about a significant change in productivity and end use patterns. *P. bourdillonii* (Fig.1). stand out as underutilized and least studied bamboo resources in Kerala, holding immense potential for exploration and development. *P. bourdillonii* is endemic to Kerala

regions of western Ghats locally referred to as “arayambu”. This species thrives in the challenging terrain of steep precipices and wet rocks within moist deciduous forests flourishing at altitudes ranging from 750 -1300 meters. This species is prized by local indigenous tribes for its easy workability and attractive shiny nature of end products in traditional handicrafts. The other versatile applications of this bamboo include using its culms for crafting tooth-picks, creation of honey bottles, and basketry, while the culm sheath finds its purpose in the production of various handicrafts (Seethalakshmi *et al.*, 1998; Kumar, 2009). The mature culms are also favoured in farm and house construction by indigenous tribes in Nelliampathy, Palakkad in Kerala.

Understanding the properties of Bamboo is crucial, as they vary significantly between species and even within the culm of the same plant. Comprehensive studies of different bamboo species are essential to ascertain their suitability for specific end uses. Morphological characters play a pivotal role in this assessment. Moreover, physical properties such as moisture content, basic density, and hollowness are key determinants of bamboo strength and its appropriateness for diverse applications. These factors collectively contribute to the overall performance and durability of bamboo in various contexts, ranging from construction to crafts and other industrial uses. Therefore, a thorough investigation into these morphological and physical characteristics is indispensable for informed decision-making in utilizing bamboo effectively and sustainably (Selvan *et al.*, 2017).



Fig 1. Natural distribution of *P. bourdillonii* in Nelliampathy (A), Culm of *P. bourdillonii* (B)

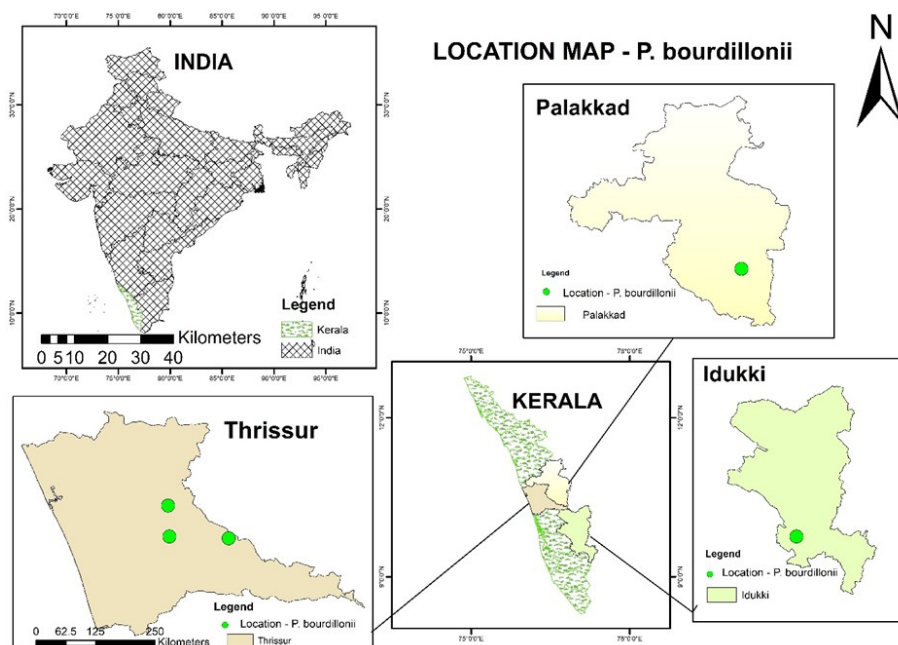


Fig 2. Study sites of *P. bourdillonii*

The literature review revealed comprehensive information on the morphology, occurrence and revisionary studies of the selected species. However, there is a significant lack of research addressing fundamental aspects like physical properties and anatomical properties of, *P. bourdillonii* to date. Thus, this study aims to evaluate the anatomical and physical properties of *P. bourdillonii* across different culm heights and locations, aiming to contribute valuable information that enhances potential applications of these species.

Materials and methods

Culm samples of *P. bourdillonii* were methodically collected from five distinct locations (Fig. 2) which includes two different locations in Chimmini Cheriya Ponmudi Thrissur (N 10° 31' 12.6" E 076° 21' 24.0"), (N10°25'53.3" E 076° 31'19.0"), vicinity of Valanjanganam waterfall in Kuttikkanam (N 09° 33' 45.15" E 076° 58' 40.20") Seetharkunde areas in Nelliampathy (N 10° 33' 05.17" E 076° 42' 20.01") and from KFRI-FRC Palapilly bambusetum (N 10° 26' 10.6" E 076° 21' 36.6").

The two-year-old culms were randomly collected from these sites ensuring a cutting height above 30 cm to prevent hindrance to the growth of remaining

culms. The harvested culms are subsequently divided into bottom, middle, and top portions.

Physical properties

P. bourdillonii is a moderate sized, straggling bamboo forming open clumps. Culms reach a height of 15-20 m height with a diameter of 8-10 cm at the base, tapering to 2-4cm. The average internodal length at the base of harvested culms is 50 – 60 cm.

Moisture content

Three centimetres long round culms representing five sites and three culm heights (base, middle, and top) were cut from fresh culms to determine its initial moisture content. The green weight of each specimen was measured using an analytical balance with a 0.01g accuracy (IS 6874, 2008). The specimens were then oven-dried at a temperature of (103±2) °C until attaining constant weight. The moisture content was calculated using Equation

$$\text{Moisture content} = \frac{\text{GW} - \text{ODW}}{\text{ODW}} \times 100\%$$

where GW is the green weight (the weight of the sample before drying), OD W is the weight of the specimen after drying in oven up to constant weight (Oven dried weight).

Basic density

Three centimeters long specimens, (representing 5 sites and three culm heights), were cut from fresh culms of *P. bourdillonii* used for the determination of basic density. The green weights of all specimens were measured using an analytical balance with an accuracy of 0.01 g. The water displacement method was used to determine the volume of each specimen. The specimens were then oven-dried at a temperature of $(103 \pm 2)^\circ\text{C}$. The repeated measurement of weight was recorded until the constant weight was reached. Basic density was determined based on IS 6874 (2008). Basic density was calculated using the equation

$$\text{Basic density} = \frac{\text{OD weight (g)}}{\text{Green volume (cm}^3\text{)}}$$

Volumetric shrinkage

Specimens each 3-cm in length and representing five distinct site of the species at three different culm height were initially measured for green volume using water displacement method. Subsequently, the specimen underwent oven drying at a controlled temperature of $(103 \pm 2)^\circ\text{C}$ until a constant weight was achieved with the final measurement recorded as oven dried volume. The determination of shrinkage followed by the standards outlined in ASTM D 143-94 (1990) and Razak, 2012. The shrinkage was calculated using the equation

$$\text{Volumetric shrinkage} = \frac{\text{GV} - \text{ODV}}{\text{GV}} \times 100\%$$

Where GV is the green volume (the volume of the sample before drying), ODV is the volume of the specimen after drying in oven (Oven dried volume)

Anatomy assessment

The bamboo culms were divided into bottom, middle and top portions. The fixation process involves Formalin -acetic acid (FAA) composed of 90% ethanol (conc. 70%), 4% glacial acetic acid, and 6% Formaldehyde (conc. 37-48%) was applied immediately after felling the specimen blocks. These were subsequently stored in sealed containers, preserving them for further anatomical observations (Razak, 1998).

Observation on anatomical features

The observations focus primarily on key anatomical features including number of vascular bundles, meta

xylem vessel size, fibres, parenchyma diameter and parenchyma lumen width. These investigations adhered to the methodologies established by Jane, 1933; Abdul Latif and Mohd Tamizi, 1992. Bamboo sample blocks were cut into sections of $10 \text{ mm} \times 10 \text{ mm} \times \text{culms wall thickness}$ sliced into $25 \mu\text{m}$ thick transverse sections by sledge microtome with a 15-degree knife angle. Each section was stained with 4 drops of aqueous safranin. They were washed with 50% ethanol then dehydrated through alcohol series of 70, 80, 90 and 95%, and 3 changes of absolute ethanol for 1 minute. Observations were based on three replicate slides. Cell measurements were done using Leica image analyser.

Number of vascular bundles, meta xylem vessel size, parenchyma diameter and parenchyma lumen width were measured in outer middle and inner regions of bottom middle and top sections. The distributions of vascular bundles were determined by counting the number of vascular bundle on a cross-section per mm^2 .

Maceration process

Match size Samples prepared separately from outer, middle inner slivers of bottom, middle and top portions using a sharp knife. The splints were subsequently placed in a marked vial, and an equal volume (1:1) of 10% glacial acetic acid and 30% Hydrogen Peroxide (H_2O_2) was added. The mixture subjected to boiling at $100 \pm 2^\circ\text{C}$ until the splints achieved a softened and bleached white state following the maceration protocol adopted by Franklin, 1945. At the end of the maceration period, the softened splints will be carefully washed with distilled water. Subsequently the vials were half filled with distilled water and securely capped. Drops of safranin were introduced to contrast the fibre's images.

Fibre dimension Measurements

The fibre dimensions measured include fibre length, fibre diameter, and lumen diameter. 2.5x magnification used to measure fibre length while other dimensions determined using 40x magnification. Fibre double wall thickness obtained from the difference between fibre diameter and lumen diameter. Twenty-five complete and reasonably straight fibres was measured.

Derived morphological fibre characteristics:

$$\text{i Runkel Ratio/ Rigidity Coefficient} = \frac{2 \times \text{cell wall thickness}}{\text{Lumen width}}$$

$$\text{ii} \quad \frac{\text{Slenderness ratio / Felting Coefficient / Aspect ratio}}{\text{Fibre length}} = \frac{\text{Fibre width}}{\text{Fibre width}}$$

$$\text{iii} \quad \frac{\text{Flexibility Ratio/ Elasticity Coefficient}}{\text{Lumen width}} = \frac{\text{Fibre width}}{\text{Fibre width}}$$

Statistical analysis

For each sample, 25 readings were taken from inner, outer, and middle slivers of the bottom, middle and top portions to measure fibre morphology. More than three replications from each location were used to measure physical properties such as moisture content, density, and volumetric shrinkage. The average results with SD are reported in the manuscript. A 95% confidence limit of P value less than or equal to 0.05 was used to determine significance. Two-way ANOVA followed by the least significant difference test as post hoc analysis was done for comparing each parameter between height position and between radial position.

Result and Discussions

Moisture content

The present study examined moisture content of *P. bourdillonii* by analysing samples from 5 different locations and assessing variation in moisture level at bottom, middle and top positions of 2-year-old culms. The mean moisture content obtained from five different sites was $160.60 \pm 84.72\%$. The moisture content decreases progressively from the base to the top of the culm as its height increases. The bottom portion exhibits highest moisture content

$175.79 \pm 73.32\%$ followed by the middle $171.05 \pm 98.43\%$ and $135.55 \pm 79.15\%$ in the top. Statistical analysis reveals no significant variation ($p > 0.05$) indicating that the position of the culm does not significantly influence the moisture content. This moisture content is higher than 133% recorded at the bottom of *B. balcooa* (Kamruzzaman et al., 2008). The decrease in moisture content aligned with prior observation in other bamboo species indicating a consistent trend as the culm height increased (Wahab et al., 2009). Conversely, the decline in the initial moisture content could be attributed to a lower proportion of vascular bundles at the base compared to the upper section of the culm (Anokye et al., 2014).

Basic density

The average basic density was $0.45 \pm 0.14 \text{ g/cm}^3$. The average basic density of *P. bourdillonii* is lower than *B. balcooa* (0.595 g/cm^3) (Kamruzzaman et al., 2008). Contrary to moisture content, the basic density exhibited an upward trend with values of $0.424 \pm 0.117 \text{ g/cm}^3$, $0.447 \pm 0.154 \text{ g/cm}^3$, $0.485 \pm 0.153 \text{ g/cm}^3$ observed from the base to top (Fig. 3). Several researchers have documented trends consistent with this study, observing rise in basic density as the height of the culm increases from base to top (Wahab et al., 2009; Vetter et al., 2015). No significant variation was observed along the culm height ($p > 0.05$). Typically, density in bamboo falls within the range of approximately $0.4 - 0.9 \text{ g/cm}^3$, a variation influenced by its anatomical structure, as indicated by the quantity and distribution of fibres around the vascular bundles (Mussa et al., 2023).

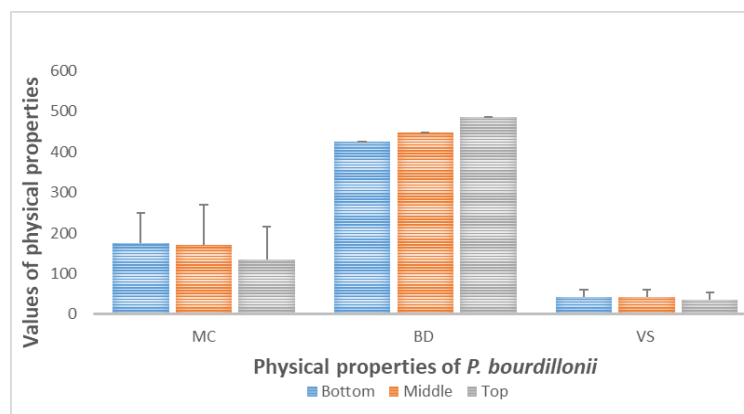


Fig 3. Average physical property values of *P. bourdillonii*; error bars represent mean standard deviation; MC-moisture content (%), BD –basic density (kg/m^3), VS- volumetric shrinkage (%).

Table 1. Comparison of different physical parameters in *P. bourdillonii* between bottom, middle and top portion of culm

Variable	Bottom	Middle	Top	F-value(P-value)
Green weight	79.60 ^a + 24.31	33.70 ^b + 9.5	9.12 ^c + 4.13	142.6**(<0.001)
Oven- dried weight	29.12 ^a + 7.49	13.27 ^b + 4.09	3.92 ^c + 1.53	167.2**(<0.001)
Green volume	73.67 ^a + 24.27	31.58 ^b + 9.58	8.91 ^c + 4.31	120.2**(<0.001)
Oven-dried volume	39.17 ^a + 11.55	17.08 ^b + 5.12	5.23 ^c + 1.94	141.1**(<0.001)
Moisture content	175.79 + 73.32	171.05 + 98.43	135.55 + 79.15	1.747 ^{ns} (0.181)
Basic density	0.424 + 0.117	0.447 + 0.154	0.485 + 0.153	1.207 ^{ns} (0.305)
Volumetric Shrinkage	43.05 + 16.81	43.09 + 17.18	35.71 + 17.28	1.598 ^{ns} (0.209)

** Significant at 0.01 level; ns non-significant Means having different small letter as superscript differ significantly within a row

Volumetric shrinkage

The mean value of volumetric shrinkage of *P. bourdillonii* is 40.59 ± 17.11 %. with the highest peak value observed in the middle of the culm having average value 43.09 ± 17.18 %. The bottom and middle part showed relatively similar values with the bottom recorded 43.05 ± 16.81 %. Lowest volumetric shrinkage at the top 35.71 ± 17.28 %. The identified pattern previously reported in Iron bamboo (*Guadua angustifolia*), where a lower volumetric shrinkage consistently observed in the top of culm (Villareal et al., 2020). There was no significant variation in volumetric shrinkage along culm height.

The variation in shrinkage could be attributed to the anatomical structure of the culm, where the inner portion comprise more parenchyma cells but fewer fibres and conducting cells compare to the outer portion of the culm (Wahab et al., 2010). The higher moisture content in the inner tissue induces increased stress and collapse, resulting in the greater shrinkage

as observed (Anokye et al., 2014). Table 1 represents physical parameters of *P. bourdillonii* across various culm heights.

Anatomical properties of *P. bourdillonii*

The culm tissue comprised of two cell types parenchyma and vascular bundles. In the bamboo culm approximately 50% composed of parenchyma, 40% fibres, and 10% vessels and sieve tubes (Maye et al., 2013; Liese 1998). Vascular bundle type IV with combination of Type III is found in *P. bourdillonii* (Fig. 4 & 5). Basal internodes show type IV, middle - combination of type IV and III and top parts showing Type III bundles. Vascular bundle type IV consist of two isolated fibre strand at the phloem and protoxylem side. Besides the central strand Type III has a separate fibre strand. Type IV predominantly occurs in basal internodes where additional support is often required and is provided by two fibre bundles (Liese, 1998).

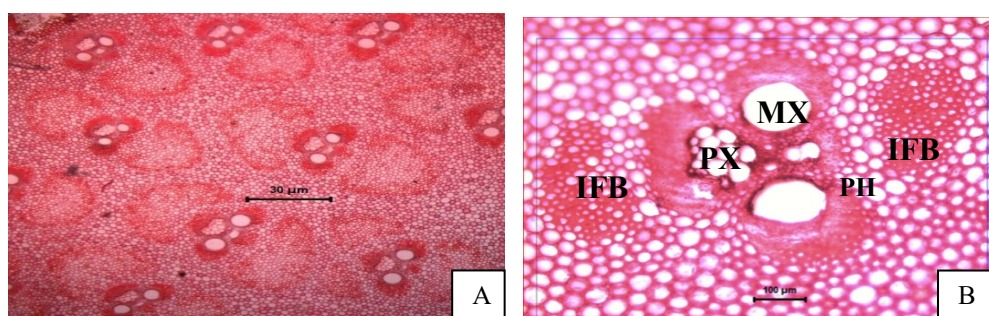


Fig 4. Type IV vascular bundle in the Bottom of *P. bourdillonii* - 2.5x magnification (A), Single vascular bundle having two isolated fibre strands –10x magnification (B)

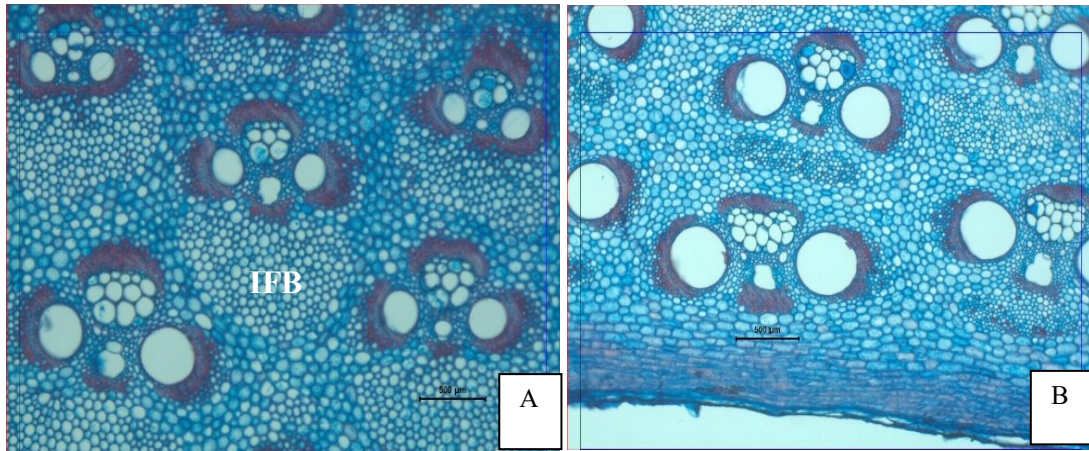


Fig 5. Type III vascular bundles in the top of *P. bourdillonii* - 6.3x magnification (A) & (B).



Fig 6. Fibre length of *P. bourdillonii* – 2.5x magnification

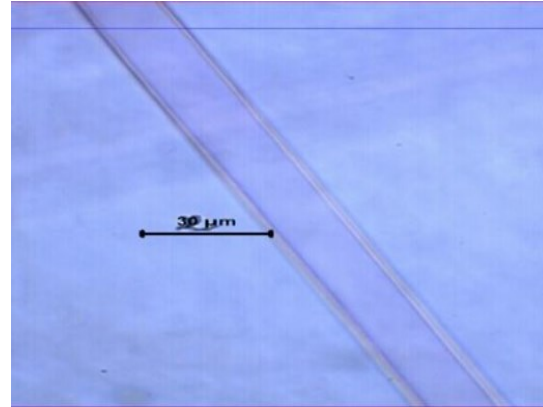


Fig 7. Fibre width of *P. bourdillonii* - 40x magnification

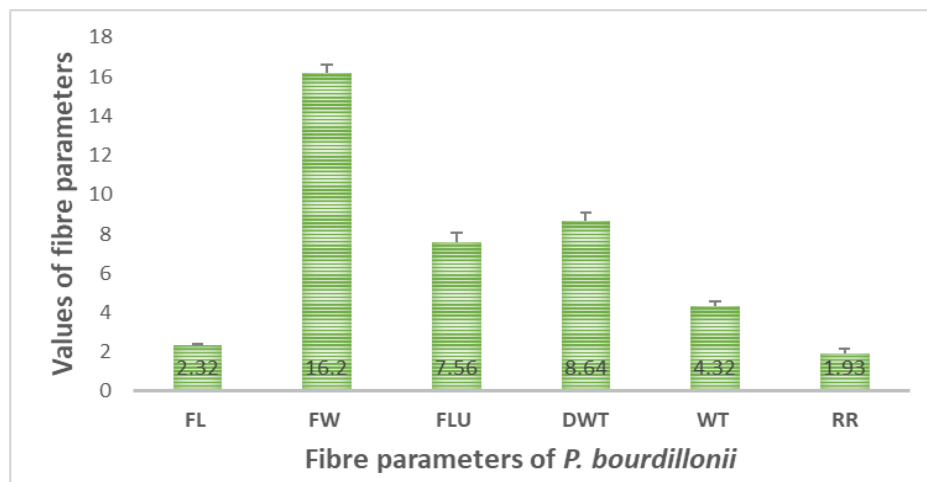


Fig 8. Average fibre parameter values of *P. bourdillonii*; error bars represent mean standard deviation; FL-fibre length,(mm) FW–fibre width(µm), FLU- fibre lumen (µm), DWT- double wall thickness(µm), WT- wall thickness(µm), RR-runkel ratio.

Fibre morphology

Characterized by a slender form, the fibres exhibit a length that is elongated and tapered at both ends, occasionally featured forked structures. The overall strength of the culm and its pulping properties are directly influenced by the length of the fibres (Liese, 1998). The characteristics of fibre stand out as a pivotal variable in assessing appropriateness as raw material for pulp and paper production (Kayama, 1979). Fig 6 & 7 illustrates fibre morphology of *P. bourdillonii*. The mean fibre parameter values are shown in Fig 8.

Fibre length

International Association of Wood Anatomists (IAWA) classifies fibres into three groups: medium-length (0.91-1.60 mm), moderately long (1.61-2.20 mm) and very long (2.21-3.00 mm) (Adi et al., 2014).

The mean fibre length of *P. bourdillonii* is 2.32 ± 0.08 mm categorises the plant as long fibre plant. Longer fibre length enhances the tearing resistance of paper (Sharma et al., 2011). This fibre length is higher than *Ochlandra travancorica* (2.20mm) (Azzini and Zalgado, 1982), and *Bambusa vulgaris* (2.10mm) (Sadiku et al., 2016).

The largest mean fibre length is found at the middle slivers of bottom part, and shortest fibres at the outer slivers of middle part. There is no significant difference in fibre length of bottom, middle and top part. However, a significant variation seen in fibre length of outer and middle parts, with the middle part exhibiting the highest average value 2.55 ± 0.10 mm (Table 2). There is no significant difference in the fibre length of middle and inner parts. The interaction between height position and radial position found to be statistically non-significant.

Table 2. Results of comparison of fibre length between height position and between radial position in *P. bourdillonii*

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	1.99 ± 0.32	2.65 ± 0.16	2.55 ± 0.25	2.40 ± 0.16
Middle	1.65 ± 0.13	2.62 ± 0.14	2.62 ± 0.21	2.30 ± 0.15
Top	2.11 ± 0.19	2.38 ± 0.21	2.35 ± 0.20	2.28 ± 0.11
Overall	$1.92^b \pm 0.13$	$2.55^a \pm 0.10$	$2.51^a \pm 0.12$	2.32 ± 0.08

Between F-value Height position = 0.267ns; P-value = 0.767 Between F-value radial position = 8.64**;
P-value = 0.001 Interaction between height position and radial position = 1.007ns; P-value = 0.417

** Significant at 0.01 level; ns non-significant Means having different small letter as superscript differ significantly within a row

Table 3. Results of comparison of fibre width between height position and between radial position

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	17.53 ± 2.03	18.09 ± 0.79	18.01 ± 0.66	$17.88^A \pm 0.71$
Middle	13.89 ± 1.35	16.08 ± 0.95	16.68 ± 1.21	$15.55^B \pm 0.71$
Top	14.82 ± 1.01	16.44 ± 0.28	14.30 ± 0.53	$15.19^B \pm 0.44$
Overall	15.41 ± 0.91	16.87 ± 0.46	16.33 ± 0.61	16.20 ± 0.40

Between F-value Height position = 5.334**; P-value = 0.009 Between F-value radial position = 1.355ns; P-value = 0.271 Interaction between height position and radial position = 0.777ns; P-value = 0.548

** Significant at 0.01 level; ns non-significant Means having different capital letter as superscript differ significantly within a column.

Table 4. Results of comparison of fibre lumen between height position and between radial position

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	9.14 ± 1.67	10.61 ± 1.27	10.68 ± 1.52	10.14 ^A ± 0.82
Middle	6.26 ± 1.44	7.70 ± 1.35	7.57 ± 1.83	7.17 ^B ± 0.85
Top	4.89 ± 1.20	6.49 ± 1.35	4.71 ± 0.51	5.36 ^B ± 0.62
Overall	6.76 ± 0.91	8.27 ± 0.85	7.65 ± 0.99	7.56 ± 0.53

Between F-value Height position = 8.989**; P-value = 0.001 Between F-value radial position = 0.884ns; P-value = 0.422 Interaction between height position and radial position = 0.165ns; P-value = 0.955

** Significant at 0.01 level; ns non-significant; Means having different capital letter as superscript differ significantly within a column.

Fibre width

The average fibre width of *P. bourdillonii* is found to be $16.20 \pm 0.40 \mu\text{m}$. The highest fibre width was found in the middle slivers of bottom position with an average value $18.09 \pm 0.79 \mu\text{m}$. The shortest fibre width found in the inner position of the top part $14.30 \pm 0.53 \mu\text{m}$. The fibre width of the bottom part vary significantly with the fibre width of middle and top positions ($p < 0.05$), there is no significant difference in fibre width at the outer middle and inner parts ($p \geq 0.05$), (Table 3). This overall fibre width of *P. bourdillonii* surpasses that of various bamboo species such as *Bambusa vulgaris* ($14.8 \mu\text{m}$) as reported by Egbewole et al. (2015), as well as *Gigantochloa apus* ($14.5 \mu\text{m}$) reported by Sugesty et al. (2015), and even exceeds the measurements observed in the hardwood of *Melia azedarach* ($13.45 \mu\text{m}$) as reported by Megra et al. (2022).

Fibre lumen Diameter

The highest lumen diameter of *P. bourdillonii* was obtained from the inner slivers of the bottom part with an average value $10.68 \pm 1.52 \mu\text{m}$. The shortest lumen found at the inner slivers of top position is $4.71 \pm 0.51 \mu\text{m}$. The average value of fibre lumen diameter of *P. bourdillonii* is $7.56 \pm 0.53 \mu\text{m}$. *P. bourdillonii* exhibits higher fibre lumen width compared to several other bamboo species *Dendrocalamus strictus* ($4.33 \mu\text{m}$), *D. latiflorus* ($3.44 \mu\text{m}$), *D. giganteus* ($5.66 \mu\text{m}$), *D. asper* ($3.97 \mu\text{m}$), *B. vulgaris*

($3.81 \mu\text{m}$), and *B. beecheyana* ($3.55 \mu\text{m}$) (Rusch et al., 2021). The width of the fibre lumen plays a crucial role in the pulp beating process, with a larger lumen contributing to improved beating efficiency. This is due to the enhanced penetration of the liquids in to the fibre lumen, facilitating more effective pulp treatment (Sharma et al., 2011). The fibre lumen width of bottom $10.14 \pm 0.82 \mu\text{m}$ varies significantly with that of middle $7.17 \pm 0.85 \mu\text{m}$ and top $5.36 \pm 0.62 \mu\text{m}$ ($p < 0.05$)- Table 4. But the fibre lumen diameter of outer, middle and inner slivers not differ statistically.

Double wall thickness

Double wall thickness determined by subtracting fibre diameter with lumen diameter. The average double wall thickness of *P. bourdillonii* is found to be $8.64 \pm 0.47 \mu\text{m}$. The highest double wall thickness found in the middle part of the top of culm $9.95 \pm 1.45 \mu\text{m}$. There is no significant difference of double wall thickness between height positions and radial positions (Table 5).

Wall thickness

The average fibre wall thickness of *P. bourdillonii* is $4.32 \pm 0.23 \mu\text{m}$. The thickest wall found in the outer slivers of top of the culm $4.96 \pm 0.46 \mu\text{m}$ while the thinnest at the inner slivers of bottom position $3.67 \pm 0.82 \mu\text{m}$. Wall thickness increases from bottom to top. The difference in wall thickness between the height position and radial position is not statistically

significant (Table 6). The wall thickness of *P. bourdillonii* is lower than *B. vulgaris* (5.06µm), *Ochlandra travancorica* (6.00µm), and *D. asper* (5.69µm). *A. donax*, which has been utilized as an excellent raw material for producing handmade paper, has a cell wall thickness of 5.36µm. Plants having thinner cell walls contribute to the production of high-quality paper (Boadu et al 2020). The wall thickness of *P. bourdillonii* is slightly higher than *Eucalytus* sp. with 3.29-3.86 µm (Viane et al., 2009), and is comparable to *G. scortechinii* 4.3 µm (Razak et al., 2012)

Fibre derived values of *P. bourdillonii*

Runkel ratio

The runkel ratio of different height position is $1.19 \pm$

0.25 in bottom 1.96 ± 0.41 in the middle and top having 2.66 ± 0.34 . The average runkel ratio of *P. bourdillonii* is 1.93 ± 0.21 . There is no significant difference in the outer middle and inner slivers. Runkel ratio of top and bottom vary significantly ($p < 0.05$) (Table 7). If the runkel ratio is less than 1 it is more suitable for paper making Xu et al., 2006. Bottom part of *P. boudillonii* get almost similar range 1.19 ± 0.25 . The fibres with runkel ratio below 1 are considered to be thin-walled and have good mechanical properties (Neto et al., 1996 ; Oluwadare and Sotannde, 2007). The average runkel ratio of *P. bourdillonii* is lower than that of *Ochlandra travancorica* (2.44) (Azzini and Zalgado, 1982), *G. angustifolia* (6.57), *G. superba* (5.64) (Azzini et al., 1977) *D. asper* (3.51), *D. giganteus* (2.37) and *B. vulgaris* (3.13) (Ciaramello and Azinni, 1971).

Table 5. Results of comparison of double wall thickness between height position and between radial position

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	8.39 ± 1.08	7.48 ± 1.34	7.33 ± 1.64	7.73 ± 0.74
Middle	7.63 ± 1.05	8.38 ± 1.81	9.11 ± 2.39	8.37 ± 1.00
Top	9.93 ± 0.91	9.95 ± 1.45	9.60 ± 0.93	9.82 ± 0.60
Overall	8.65 ± 0.6	8.60 ± 0.87	8.68 ± 0.97	8.64 ± 0.47
Between F-value Height position = 1.581 ^{ns} ; P-value = 0.220 Between F-value radial position = 0.002 ^{ns} ; P-value = 0.998 Interaction between height position and radial position = 0.209 ^{ns} ; P-value = 0.932				

Table 6. Results of comparison of single wall thickness between height position and between radial position

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	4.20 ± 0.54	3.74 ± 0.67	3.67 ± 0.82	3.87 ± 0.37
Middle	3.82 ± 0.53	4.19 ± 0.91	4.56 ± 1.20	4.19 ± 0.50
Top	4.96 ± 0.46	4.97 ± 0.72	4.80 ± 0.46	4.91 ± 0.30
Overall	4.33 ± 0.30	4.30 ± 0.44	4.34 ± 0.49	4.32 ± 0.23
Between F-value Height position = 1.581 ^{ns} ; P-value = 0.220 Between F-value radial position = 0.002 ^{ns} ; P-value = 0.998 Interaction between height position and radial position = 0.209 ^{ns} ; P-value = 0.932				

* Significant at 0.05 level; ns non-significant

Slenderness ratio

The mean slenderness ratio of *P. bourdillonii* is 152.76 ± 4.82 (Table 8). This value is higher than the slenderness ratio of other bamboo species such as *G. angustifolia*(120), *G. superba*(136) (Azzini et al, 1977) *D.asper*(136), *D. strictus*(122)(Ciaramello and Azinni, 1971) and *Ochlandra travancorica*(131) (Azzini and Zalgado, 1982). There is an increase in slenderness ratio from bottom to top. There is no significant difference of slenderness ratio between height position but the slenderness ratio of outer slivers is significantly lower than that of middle and

inner slivers. The obtained slenderness ratio of *P. bourdillonii* is higher than *O. abyssinica* 109.98 ± 0.21 (Worku et al., 2023). The fibres extracted from all parts of *P. bourdillonii* have a slenderness ratio greater than 70, indicating their potential to yield high quality sheets suitable for various applications such as packing. The mean slenderness ratio of *P. bourdillonii* exceeds the values obtained for conventional paper making sources *Eucalyptus grandis* (55.18), *Eucalyptus tereticornis* (52.66) and *Eucalyptus camadulensis* (53.33) (Dutt and Tyagi, 2011; Pillai et al., 2013).

Table 7. Results of comparison of runkel ratio between height position and between radial position

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	1.38 ± 0.45	1.05 ± 0.38	1.12 ± 0.53	$1.19^B \pm 0.25$
Middle	2.00 ± 0.64	1.80 ± 0.79	2.08 ± 0.87	$1.96^{AB} \pm 0.41$
Top	2.87 ± 0.59	2.49 ± 0.77	2.62 ± 0.47	$2.66^A \pm 0.34$
Overall	2.08 ± 0.34	1.78 ± 0.39	1.94 ± 0.38	1.93 ± 0.21

Between F-value Height position = 4.094*; P-value = 0.025 Between F-value radial position = 0.174^{ns}; P-value = 0.841 Interaction between height position and radial position = 0.025^{ns}; P-value = 0.999

* Significant at 0.05 level; ns non-significant Means having different capital letter as superscript differ significantly within a column

Table 8. Results of comparison of slenderness ratio between height position and between radial position

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	123.19 ± 17.20	158.64 ± 13.87	151.03 ± 13.93	144.29 ± 9.03
Middle	126.36 ± 10.98	171.91 ± 3.89	163.39 ± 6.44	153.89 ± 6.69
Top	155.76 ± 22.61	152.45 ± 12.18	172.13 ± 12.70	160.11 ± 9.14
Overall	$135.11^b \pm 10.18$	$161.00^a \pm 6.21$	$162.18^a \pm 6.57$	152.76 ± 4.82

Between F-value Height position = 1.022^{ns}; P-value = 0.370 Between F-value radial position = 3.764*; P-value = 0.033 Interaction between height position and radial position = 0.919^{ns}; P-value = 0.464

* Significant at 0.01 level; ns non-significant, Means having different small letter as superscript differ significantly within a raw

Table 9. Results of comparison of flexibility ratio between height position and between radial position.

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	0.49 ± 0.07	0.57 ± 0.06	0.58 ± 0.08	0.55 ^A ± 0.04
Middle	0.43 ± 0.08	0.47 ± 0.08	0.46 ± 0.11	0.45 ^{AB} ± 0.05
Top	0.32 ± 0.06	0.39 ± 0.08	0.39 ± 0.08	0.36 ^B ± 0.04
Overall	0.41 ± 0.04	0.48 ± 0.04	0.47 ± 0.05	0.45 ± 0.03

Between F-value Height position = 4.153*; P-value = 0.024 Between F-value radial position = 0.604ns; P-value = 0.552 Interaction between height position and radial position = 0.037ns; P-value = 0.997

Significant at 0.05 level; ns non-significant Means having different capital letter as superscript differ significantly within a column

Table 10. Results of comparison of vessel diameter between height position and between radial position

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	75.58 ± 4.21	122.21 ± 6.94	151.95 ± 4.44	116.58 ± 8.87
Middle	80.11 ± 6.20	126.71 ± 3.25	175.65 ± 4.10	127.49 ± 10.72
Top	83.50 ± 5.52	122.77 ± 8.96	163.10 ± 10.16	123.12 ± 9.79
Overall	79.73 ^c ± 3.00	123.90 ^b ± 3.68	163.56 ^a ± 4.47	122.40 ± 5.58

Between F-value Height position = 2.228^{ns}; P-value = 0.122 Between F-value radial position = 129.93^{**}; P-value < 0.001 Interaction between height position and radial position = 0.886^{ns}; P-value = 0.482

^{**} Significant at 0.01 level; ns non-significant, Means having different small letter as superscript differ significantly within a row

Flexibility ratio

The flexibility ratio signifies the ease with which fibre interconnects influencing the resultant tensile and bursting strength (David *et al.*, 2016). The observed average flexibility ratio of *P. bourdillonii* is found to be 0.45 ± 0.03 (Table 9). This was higher than that of *Ochlandra travancorica* (.29) (Azzini and Zalgado, 1982), *D. asper* (.22), *D. giganteus* (.29) and *B. vulgaris* (.24) (Ciaramello and Azinni, 1971). There is a significant difference in the flexibility ratio of bottom (0.55 ± 0.04) and top fibres (0.36 ± 0.04). The flexibility ratio of outer, middle and inner slivers not differ statistically.

Meta xylem vessel diameter

The mean values of bottom, middle and top meta

xylem vessel diameters are 116.58 ± 8.87 µm, 127.49 ± 10.72 µm, and 123.12 ± 9.79 µm respectively. The average vessel diameter of *P. bourdillonii* is 122.40 ± 5.58 µm. There is no statistical difference in vessel diameter of the bottom, middle and top positions (Table 10). The size difference of the outer, middle and inner slivers is statistically significant. The outer metaxylem vessels exhibit a lower diameter 79.73 ± 3.00 µm followed by the middle vessels with a diameter of 123.90 ± 3.68 µm, while the inner vessels show a higher diameter 163.56 ± 4.47 µm. This observation aligns with findings of Wahab *et al.* (2010) indicating a progressive increase in vessel diameter from the outer to the inner part of the bamboo culm.

Parenchyma diameter

The parenchyma diameter varies from $39.37 \pm 1.47 \mu\text{m}$ highest at the bottom, $35.58 \pm 1.68 \mu\text{m}$ at the middle and lowest value $34.08 \pm 1.59 \mu\text{m}$ in the top (Table 11). While there is no statistically significant difference between parenchyma diameter among different height positions, a notable contrast observed between outer and inner positions with the outer position has the lowest diameter $32.92 \pm 1.78 \mu\text{m}$, and inner position displays largest parenchyma diameter $38.86 \pm 1.37 \mu\text{m}$ (P-value < 0.05). Parenchyma diameter of *P. bourdillonii* is higher than $26.6 \mu\text{m}$ of *Bambusa vulgaris* (Wahab et al., 2009). Fig 9 illustrates Parenchyma of *P. bourdillonii*.

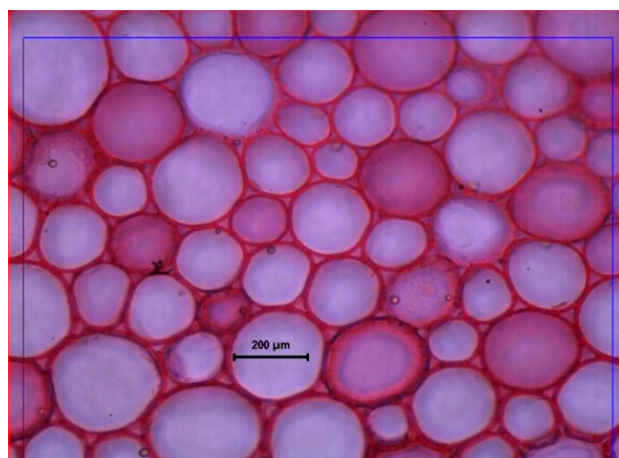


Fig 9. Parenchyma of *P. bourdillonii*

Table 11. Results of comparison of parenchyma diameter between height position and between radial position

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	36.61 ± 3.50	40.11 ± 2.43	41.38 ± 1.14	39.37 ± 1.47
Middle	31.52 ± 2.11	37.22 ± 2.88	38.00 ± 3.24	35.58 ± 1.68
Top	30.64 ± 3.39	34.41 ± 2.00	37.20 ± 2.35	34.08 ± 1.59
Overall	$32.92^b \pm 1.78$	$37.25^{ab} \pm 1.46$	$38.86^a \pm 1.37$	36.34 ± 0.95

Between F-value Height position = 3.145ns; P-value = 0.055 Between F-value radial position = 3.992*; P-value = 0.027 Interaction between height position and radial position = 0.084ns; P-value = 0.987

Means having different small letter as superscript differ significantly within a row

Table 12. Results of comparison of parenchyma lumen diameter between height position and between radial position

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	31.67 ± 2.72	35.40 ± 1.78	35.83 ± 0.97	$34.30^A \pm 1.16$
Middle	27.16 ± 2.16	32.60 ± 2.43	33.37 ± 2.47	$31.04^{AB} \pm 1.46$
Top	25.93 ± 2.72	29.55 ± 1.40	31.87 ± 1.67	$29.12^B \pm 1.26$
Overall	$28.25^b \pm 1.51$	$32.52^a \pm 1.21$	$33.69^a \pm 1.06$	31.49 ± 0.80

Between F-value Height position = 4.592*; P-value = 0.017 Between F-value radial position = 5.486**; P-value = 0.008 Interaction between height position and radial position = 0.122ns; P-value = 0.974

** Significant at 0.01 level; ns non-significant Means having different small letter as superscript differ significantly within a row, Means having different capital letter as superscript differ significantly within a column

Parenchyma lumen diameter

Parenchyma lumen diameter exhibits a descending trend from the bottom ($34.30 \pm 1.16 \mu\text{m}$), to the middle ($31.04 \pm 1.46 \mu\text{m}$) reaching the lowest value at the top ($29.12 \pm 1.26 \mu\text{m}$). significantly, there is a notable statistical significance in parenchyma lumen diameter between the bottom and top positions (Table 12). Moreover, the outer position ($28.25 \pm 1.51 \mu\text{m}$) differs statistically from the middle ($32.52 \pm 1.21 \mu\text{m}$) and inner position ($33.69 \pm 1.06 \mu\text{m}$).

Vascular bundle number / mm^2

The results reveal a gradual increase in the mean vascular bundle number per square millimetre with values of, 1.96 ± 0.24 at the bottom, 2.35 ± 0.22 in the middle and 2.75 ± 0.39 at the top. The average vascular bundle / mm^2 of *P. bourdillonii* is 2.35 ± 0.17 .

Notably there is a statistically significant difference in vascular bundle frequency between bottom and top parts, evidenced by a P-value < 0.05 . The difference in vascular bundle frequency of outer, middle, and inner part is statistically significant at 1% level ie, P-value < 0.001 with outer measurements 3.55 ± 0.29 , middle 1.98 ± 0.14 and top values 1.52 ± 0.13 bundles/ mm^2 (Table 13). This vascular bundle count is higher than the reported values of *Bambusa vulgaris* by Wahab et al., 2009 *G. scortechinii* (1.76 bundles/ mm^2), *G. wrayi* (1.44 bundles/ mm^2) and *G. brang* (1.37 bundles/ mm^2) (Mustafa et al., 2011). A high vascular bundle count can serve as an indicator of elevated lignin content which implies stronger material (Mulyaningsih et al., 2022). Average anatomical values are provided in Fig 10.

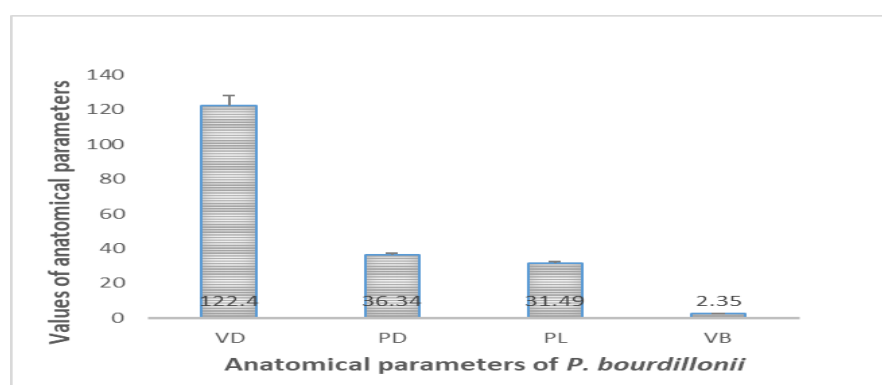


Fig 10. Average anatomical parametes of *P. bourdillonii*; error bars represent mean standard deviation; VD-metaxylem vessel diameter(μm), PD-parenchyma diameter (μm), PL-parenchyma lumen diameter(μm), VB- vascular bundles/ mm^2 .

Table 13. Results of comparison of vascular bundle number / mm^2 between height position and between radial position

Height position	Radial position			Overall
	Outer	Middle	Inner	
Bottom	2.99 ± 0.27	1.64 ± 0.29	1.25 ± 0.24	$1.96^B \pm 0.24$
Middle	3.41 ± 0.23	2.05 ± 0.09	1.58 ± 0.11	$2.35^{AB} \pm 0.22$
Top	4.26 ± 0.76	2.26 ± 0.26	1.73 ± 0.30	$2.75^A \pm 0.39$
Overall	$3.55^a \pm 0.29$	$1.98^b \pm 0.14$	$1.52^b \pm 0.13$	2.35 ± 0.17

Between F-value Height position = 4.132*; P-value = 0.024 Between F-value radial position = 30.236**; P-value < 0.001 Interaction between height position and radial position = 0.489ns; P-value = 0.744

** Significant at 0.01 level; ns non-significant Means having different small letter as superscript differ significantly within a row, means having different capital letter as superscript differ significantly within a column

Table 14. Correlation of anatomical and fibre parameters with physical properties

Anatomical and fibre parameters	Physical parameters		
	Moisture content	Basic density	Volumetric shrinkage
Fibre length	-0.132	0.155	-0.146
Fibre width	-0.103	0.101	-0.107
Fibre lumen	0.738**	-0.706**	0.706**
Double wall thickness	-0.897**	0.859**	-0.863**
Single wall thickness	-0.897**	0.859**	-0.863**
Runkel	-0.853**	0.845**	-0.825**
Slenderness ratio	-0.029	0.037	-0.042
Flexibility ratio	0.844**	-0.807**	0.812**
Vessel diameter	-0.335	0.509	-0.417
Parenchyma diameter	-0.574*	0.615*	-.664**
Parenchyma lumen	-0.407	0.431	-0.486
Vascular bundles/mm ²	-0.179	0.083	-0.056

** Significant at 0.01 level; * Significant at 0.05 level

Correlation between physical and anatomical properties

The correlation between the physical and anatomical properties of *P. bourdillonii* is provided in the Table 14 which indicate a significant inverse correlation between moisture content and key anatomical parameters including fibre wall thickness, runkel ratio and parenchyma diameter. Conversely, there is a noteworthy significant positive correlation between fibre lumen and flexibility ratio with moisture content. All other parameters exhibit non-significant correlation with moisture content. A study by Mohmod *et al.* (1993) on *B. blumeana* fibre length and vessel diameter was reported to have a non-significant correlation with moisture content. In *P. bourdillonii*, fibre lumen and flexibility ratio show a significant negative correlation with basic density, while, wall thickness, parenchyma diameter, and runkel ratio show a significant positive correlation to density. The volumetric shrinkage demonstrates a positive and significant correlation with fibre lumen and flexibility ratio and a negative significant correlation with wall thickness, runkel ratio and parenchyma diameter.

The positive significant correlation of basic density with that of fibre wall thickness matches with the findings of Rusch *et al.*, 2019 indicating a direct influence of basic density on fibre wall thickness. This suggests that species with lower density tend to have fibres with thinner walls.

Conclusion

The research focused on examining the physical and anatomical properties of 2-year-old *P. bourdillonii* along various culm heights. The recorded values of moisture content exhibit a decreasing trend from the bottom to the top of the culm. In contrast, the average basic density of *P. bourdillonii* shows a reverse trend compared to that of moisture content. The volumetric shrinkage reaches its peak in the middle of the culm. Statistical analysis reveals no significant difference in physical properties across different culm heights. A notable variation is observed between fibre length of the outer and middle parts, with middle slivers showing the highest average value. Fibre width and lumen width of the bottom part exhibit a significant difference compared to middle and top ($P < 0.05$). However, no significant

difference was observed in wall thickness between height position and radial position. The results indicate that the runkel ratio exceeding 1 and the slenderness ratio is higher than the standard value 70. Anatomical parameters like vessel size, vascular bundle number, parenchyma diameter and lumen were also quantified. Moisture content and shrinkage showed a negative significant correlation with wall thickness, runkel ratio, and parenchyma diameter while showing a positive correlation with fibre lumen and flexibility ratio. Basic density shows a significant positive correlation with wall thickness and parenchyma diameter. Despite the runkel ratio of *P. bourdillonii* exceeding the recommended threshold, its other parameters such as long fibre length $2.32 \pm 0.08\text{mm}$ and a slenderness ratio greater than 70 (152.76 ± 4.82) make *P. bourdillonii* suitable for pulping. The two-year age group of *P. bourdillonii* is less preferred for high-strength demanding purposes due to its higher moisture content and lower basic density when compared to other major bamboo species such as *B. bambos* and *B. balcooa*. However, with its non-thorny nature, long internodes, thin culm wall, and aesthetically pleasing golden yellow colour of the culm and culm sheath *P. bourdillonii* is well suited for handicraft sector.

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