

Journal of Asian Scientific Research

Online ISSN 2223-1331

Vol 1 No 7

November, 2011



Asian Economic and Social Society
www.aessweb.com



Anatomical Properties and Microstructures Features Of Four Cultivated Bamboo *Gigantochloa* Species

Abstract

Anatomical properties and microstructure features of four (4) cultivated species of popular tropical bamboo genus *Gigantochloa* were studied. *Gigantochloa brang*, *G. levis*, *G. scotechinii* and *G. wrayi* of age-group 3 were selected, harvested and processed for the anatomical and microstructure studies. The studies focussed mainly on the vascular bundles and fiber cells located at the internodes and nodes 8 at the outer, middle and inner layers of the bamboo. The sizes of the vascular bundles length, vascular bundles width, fiber length, fiber diameter, fiber lumens diameter, fiber walls thickness and fiber Runkle's ratio were measured between each of the species in relation to the samples positions at the internodes, nodes, and positions in the cross-section of the bamboo culms. The results in the fibers morphology studies showed that the fibers for each species has different lengths, diameters, cell walls thickness and lumen sizes. The size of vascular bundle is smaller at outer position and become bigger at the inner position. All the four (4) bamboo species exhibited similar in characteristics but having different sizes in anatomy and microstructure features.

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Key words: Cultivated *Gigantochloa*
species, anatomy, vascular bundles, fibers,
microstructure features.

Introduction

Bamboo has been the focus of research and development in recent years. They are considered to be among the fastest growing plant on earth and the best possible alternative to replace timber in the future. Research and development which covers all aspects in silviculture, propagation, processing, properties and utilization of bamboo found naturally growing wild in the forest and cultivated has been intensified. However, study on cultivated bamboo stands has so far mostly confined to selected species in silviculture and fertilizers application to enhance growing (Azmy *et al.* 2007). Information on the properties such as anatomical and structural properties is rather limited.

The anatomy and physical properties of bamboo culms have been known to have significant effects

on their durability and strength (Latif & Tamizi, 1993; Liese, 1985; Razak, 1998). Studies on the anatomical and physical properties of cultivated *Bambusa vulgaris* conducted by Razak *et al.* (2010) support this statement. Information generated on the anatomical properties of bamboo can be used to determine their possible proper utilization. Currently, bamboo used for making traditional products such as handicraft, basketry, and high-value added products of panels, parquets, furniture and construction materials. *Gigantochloa* species of bamboo are among the most popular tropical bamboo species for plantation. These bamboo are easily cultivated and possess thick culms wall, and having uniform sizes between the nodes and internodes. This makes them perfect as materials for industrial usage.

The objectives of the study were to determine the differences in the anatomical and structural properties between four (4) *G. brang*, *G. levis*, *G. scortechinii* and *G. wrayi*. The anatomical structures were investigated due to their relationship with strength, preservative absorption, distribution and likely pathways for colonisation by micro-organisms (Razak *et al.* 2005, 2002).

Material and Methodology

Materials

Samples of the four bamboo species from genera *Gigantachloa* namely *G.brang*, *G.levis*, *G.scortechinii* and *G.wrayi* were harvested from The Bambusetum Plot, Forest Research Institute Malaysia (FRIM), Kepong, Selangor, Malaysia. Culms of 3-year-old were selected for the study as the culms of this age was found to be most suitable as material for industrial uses. The bamboos had their age verified from the tags and had been monitored since the sprouting stage. The plants were harvested in January 2010. The bamboo culms were cut at about 30 cm above ground level. These culms were taken from randomly selected clumps with diameter range from 8-17 cm diameter, depend on species. Each stem was marked and cut at nodes and internodes 8. An end-coating paint was applied to the cut surfaces before the samples were transported to the laboratory. This was done to minimize evaporation and prevent fungal and insect attacks on the bamboo. The number of specimens taken were 10 culms per species.

Sample Preparation

The bamboo culms were divided according to species, and further sub-divided into node and internodes, position in the bamboo culms wall (outer layer, middle layer and inner layer). Specimen blocks intended for anatomical investigations and were fixed in formalin-acetic acid (FAA) immediately after felling and kept in closed bottles. The mixture of FAA consists of 90% ethanol (conc. 70%), 4% glacial acetic acid and 6% formaldehyde (conc. 37-48%) (Razak, 1998). Each culm was consistently cross cut into position with and without nodes with the sampling preparation protocol for each respective investigation.

Anatomy Assessment

The technique used by Latif and Tamizi (1993) was used with some modification in measuring and counting in the distribution of the vascular bundles on the bamboo surface at the cross section. The anatomical characteristic of the four (4) bamboo species with two locations (node and internode) and three (3) positions (outer, middle and inner layers) of the bamboo culm were studied.

Vascular Bundles

Method of measuring the vascular bundles distribution and fibre dimensions was adopted from the technique used by Latif and Tamizi (1993).

Vascular Bundle size

The sizes of the vascular bundles were measured by the scanning electron microscope (SEM) images through it measuring tools.

Determination of Fiber Morphology

Bamboo Maceration

The bamboo splits of size 20 mm x 10 mm x thickness were cut tangentially and divided into 3 equal portions (inner, middle and outer layers). Each portion were splits radials into match stick sizes using a sharp knife.

Macerates were prepared from match-stick sizes bamboo by placing them in solution containing glacial acetic acid (M=60.05g/mol) and hydrogen peroxide (30% and M=34.01 g/mol) at ratio 1:1. The bamboo in the solution were heated over a water bath inside a fume chamber for 2-3 hrs until it become soft and white. One or two drops of sodium hydrogen carbonate crystals were added to neutralize the acid before the mixture was decanted and washed with distilled water. A through shaking of the mixture was done to separate the individual fibers. Safranin was used to colour the extracted fiber to red.

One hundred (100) undamaged or unbroken fibers were measured for their length (L), fiber widths (d), lumen diameter (l) and cell wall thickness(w). Quantimeter Image Analyzer equipped with Lecia Microscope and Hipad Digitizer (Quantimet 520, Cambridge Instruments) was used to observed and measured at computer images at 10 x (length), 100 x (diameter) and 100 x (lumen) magnifications. The calculations of Runkel's ratio (2w/l), coefficient of suppleness or flexibility ratio (l/d) and felting factor (L/d) were carried out using the equations (1) and (2) below:

$$\text{Cell wall thickness} = (\text{Fiber Diameter} - \text{Lumen diameter}) / 2 \quad \dots\dots\dots(1)$$

$$\text{Runkel's ratio} = (2 \times \text{fiber wall thickness} (\mu\text{m})) / (\text{lumen diameter} (\mu\text{m})) \quad \dots\dots\dots (2)$$

Cell Wall Structure in Electron Microcopy

Scanning electron microscope (SEM) and transmission electron microscope (TEM) were used to analyses the microstructure of the bamboo cell

walls. For SEM analysis, the samples were then selected and cut into a smaller size for shorter duration of pre vacuum process. The surface portion of samples was cut using high speed microtome blade to ensure the smooth surface. The samples went through pre vacuum process on thin plate before the Aurum coating process took place (about 20 nm) to ensure the efficient conductivity for the analysis process. The apparatus for the coating process is called 'sputters coater' Fison SC 515. Scanning analysis was performed using 'Leica Cambridge S - 360', with magnification up to 4000 times.

The samples for TEM analysis were dehydrated in an ethanol series and embedded in Spurr resin. For cell wall structure of bamboo fiber, they were chosen according to species and position in bamboo culm and cut into pieces of 2 x 3 blocks. Samples were then dehydrated in an ethanol series and embedded in Spurr resin (Epon), which polymerized for 24 hours at 60°C. Transverse sections (1µm) were cut from the embedded material, using the Sorvall ultra microtome (MT 5000) and stained with 1% Toluidine Blue for lignin distribution determination. This gives a high contrast to lignin rich structure such as middle lamellas and cell corners. The section was viewed under polarized microscope (Nikon YS2-H). Ultra-thin section (0.1µm) were obtained from embedded samples, stained with 2% uranyl acetate and lead citrate and finally viewed under TEM (energy filter - Zeiss Libra® 120).

Result and Discussion

Vascular bundle distribution

The result for the vascular bundles distribution on the four (4) selected *Gigantochloa* species are shown in Table 1. The mean number of vascular bundle for *G. scortechinii* was 6.38 bundle/4 mm² follow by *G. wrayi* at 6.84 bundle/4 mm² and *G. brang* at 6.38 bundle/4 mm² under one group and the lowest was *G. levis* at 4.33 bundle/4 mm². These were in agreement with Latif (1991) finding of the number of vascular bundles in *G. scortechinii*. The anatomical features within and between culm of different or even the same bamboo species may vary as the individual characteristic of the bamboo itself (Pattanath, 1972; Soeprayitno *et al.*, 1990).

Different number of vascular bundle in the node and the internode sections were observed in the bamboo culm. The distributions of vascular bundles in the internodes were higher than the nodes. The vascular bundles were also observed to be higher in

number and more compacted at the outer layers of the bamboo culm than those at the inner layers. This is acknowledged by other researchers (Liese, 1992, Latif & Tamizi 1993, Hisham *et al.*, 2006). Li (2004) in his studies on a monopodial bamboo *P. pubescens* found that the number of vascular bundles were higher compared to the simpodial bamboo species. Hisham *et al.* (2006) studied on the anatomical, physical and chemical properties the characterization of bamboo *G. scortechinii* at different ages found that the number of vascular bundles increases from the inner zone towards the outer zone. Similar trend was reported in sympodial bamboo *Phyllostachys pubescens* (Wenyue *et al.* 1981). This indicates that bamboo possesses long and small vascular bundle at the outer zone, but short and big towards inner the inner zone (Liese, 1985).

Vascular Bundle Length

The results on the the measurement of the vascular bundles length is shown in Table 2. The higher mean of vascular bundle length at internodes were *G. levis* (1171.14 µm) followed by *G. brang* (788.82 µm), *G. scortechinii* (787.19 µm), and *G. wrayi* (754.06 µm).

The mean average for vascular bundle length at the nodes were *G. levis* (1193.89 µm) followed by *G. scortechinii* (1078.20 µm), *G. brang* (1000.27 µm), and *G. wrayi* (963.41 µm). The vascular bundles length were longer at the node than the internodes. The mean average of vascular bundle length for outer layer position was 748.54 µm, middle layer 1013.25 µm and for inner layer was 1131.42 µm. The vascular bundles length were longer at the middle than at the outer and inner periphery.

Vascular Bundle Width

The results on the measurement of the vascular bundles width is showed in Table 3. The higher mean of vascular bundle width was *G. levis* (798.26 µm), *G. wrayi* (532.88 µm), *G. brang* (509.47 µm) and *G. scortechinii* (501.38 µm).

The mean average at different position showed significant difference between the internodes and nodes. The nodes vascular bundle width were 720.43 µm for *G. levis*, 685.82 µm for *G. wrayi*, 587.89 µm for *G. scorechinii* and 551.14 µm for *G. brang*. The vascular bundles width were observed to be higher at the node than the internodes. The vascular bundle width was widened at the inner and smaller toward the outer periphery position. This may be due to the smaller size of the vascular bundle at the outer layer, compared to the inner layer of bamboo culms.

Table 1: Mean number of vascular bundle (per 4 mm²) of various *Gigantochloa* species

Position	Position	<i>G. brang</i>	<i>G.levis</i>	<i>G.scortechinii</i>	<i>G.wrayi</i>
Internode	Outer	9.65 (±1.54)	7.46 (±1.72)	13.24 (±1.75)	8.91(± 1.73)
	Middle	6.04 (±1.15)	3.00 (±0.54)	6.44 (±1.12)	6.27 (±1.01)
	Inner	3.45 (±0.69)	2.54 (±0.40)	3.50 (±0.64)	5.35 (±1.35)
	Mean	6.38 (±1.13)	4.33 (±0.87)	7.73 (±1.17)	6.84 (±1.36)
Node	Outer	6.06 (±1.27)	5.94 (±3.40)	10.55(±1.77)	6.69 (±3.08)
	Middle	4.18 (±1.11)	3.56 (±1.03)	5.80 (±1.40)	3.81 (±1.20)
	Inner	3.46 (±1.00)	2.87(± 0.88)	2.75 (±1.13)	3.44 (±1.23)
	Mean	4.57 (±1.13)	4.12 (±1.77)	6.37 (±1.43)	4.65 (±1.84)

Values in bracket represent the standard deviation,

Table 2: Mean vascular bundle length (µm) of various *Gigantochloa* species

Position	Position	<i>G. brang</i>	<i>G.levis</i>	<i>G.scortechinii</i>	<i>G.wrayi</i>
Internode	Outer	706.34 (±141.07)	928.73 (±303.07)	625.77 (±232.89)	685.45 (± 56.25)
	Middle	828.67 (±71.07)	1176.49 (±144.23)	882.32 (±74.07)	692.74 (± 49.88)
	Inner	831.46 (±121.93)	1408.20(± 210.11)	853.60 (±110.02)	818.66 (±52.57)
	Mean	788.82 (±113.67)	1171.14 (±219.13)	787.19 (±138.99)	754.06 (±52.90)
Node	Outer	752.50 (±123.57)	769.09 (± 129.79)	785.40 (±193.88)	735.06 (±125.33)
	Middle	1102.92(±117.22)	1387.66 (±65.41)	999.55 (±157.05)	1035.68(±142.97)
	Inner	1145.39(±180.68)	1424.92 (±99.85)	1449.64(±172.69)	1119.49(±114.70)
	Mean	1000.27 (±140.49)	1193.89 (±98.35)	1078.20 (±174.54)	963.41 (±127.67)

Values in bracket represent the standard deviation,

Table 3: Mean vascular bundle width (µm) of various *Gigantochloa* species

Position	Position	<i>G. brang</i>	<i>G.levis</i>	<i>G.scortechinii</i>	<i>G.wrayi</i>
Internode	Outer	357.15 (±69.33)	610.61 (±129.41)	382.41 (±141.22)	383.27 (±43.91)
	Middle	438.81 (±94.14)	723.01 (±93.36)	494.11 (±73.87)	582.74 (±75.64)
	Inner	732.46 (±43.89)	1061.18 (±103.56)	627.62 (±101.20)	632.62 (±43.07)
	Mean	509.47 (±69.12)	798.26 (±108.78)	501.38 (±105.43)	532.88 (±54.21)
Node	Outer	430.60 (±48.21)	570.89 (± 89.88)	478.04 (±46.60)	524.83 (±94.20)
	Middle	594.08 (±60.97)	752.93 (±76.08)	593.74 (±72.24)	619.67 (±90.79)
	Inner	628.23 (±98.00)	837.48 (±73.18)	691.88 (±92.30)	846.01 (±68.39)
	Mean	551.14(±69.06)	720.43 (±79.71)	587.89 (±70.38)	685.82 (±84.46)

Values in bracket represent the standard deviation

Fiber morphology

Fiber Length

The results for the fiber lengths study of the various *Gigantochloa* species are showed in Table 5. The longest fiber length were obtained from the *G. levis* (2039.98 µm) follow by *G. brang* (1909.68 µm), *G. wrayi* (1798.79 µm) and *G. scortechinii* (1745.27 µm).

The fiber length at the internode were significantly longer than those found at the nodes. It shows there was significant different in the fiber length between position at internode and node. At the internode, the anatomy structure were quite consistant constant but at the node the structure were quite twisted (see Figures 1, 2, 3 and 4). These were observed in all the four bamboo species used in this study. The fiber length shows considerable differences between species as well as variations within one culm (Liese & Grosser 1972). The mean average

for fiber length in genera *Gigantochloa* from this study was 1600 - 2000 μm . The result obtained by Hisham (2006) studies on *G. scortechinii* was between 2350-2630 μm . While Ireana 2009, study on *B. blumeana* found the length of fiber was around 2900 μm . *B. vulgaris* fiber length is 3600-4700 μm (Razak, 2010), 1940 - 2430 μm (Latif, 1995).

The results from this study showed that bamboo fiber length from *Gigantochloa* genera was longer than the fiber from *P. Pubescens* which growth in large areas of China, Japan, Taiwan and Indochina. The fiber length for this species was about 1300 μm length (Liese, 1992) compared with the genera *Gigantochloa* (1750-2040 μm). Walter Liese (1992) studied the structure of bamboo in relation to its properties and utilization. They reported that the fibers contribute 60-70% by weight of the total culm tissue. Certain species generally have shorter fibers, such as *Phyllostachys edulis* (1.5 mm), *Ph. pubescens* (1300 μm), other longer ones like *Dendrocalamus giganteus* (3200 μm), *Oxytenanthera nigrociliata* (3600 μm), *D. membranaceus* (4300 μm).

The fiber length of *Gigantochloa* (1600-2000 μm) genera was clearly shorter than the Softwood fiber length (3600 μm), but still longer than hardwood (1200 μm). The bamboo fiber length were in fact it is longer than *Eucalytus spp* (960-1.0400 μm) a popular as a source of fiber pulp for paper industry (Horn & Setterholm, 1990; Ververis *et al.*, 2004). The fiber pulp using by paper mill in Malaysia are mostly imported from Brazil (*Eucalytus spp.*) and Canada (Softwood). This shows that the *Gigantochloa* fiber has a potential to be use as a pulp for in the future.

Fiber diameter

The results on the fiber diameter study in the *Gigantochloa* genera are showed in Table 5. The larger mean average of fiber diameter were *G. brang* (22.75 μm), followed by *G. levis* (22.67 μm), *G. wrayi* (17.86 μm) and *G. scortechinii* (17.26 μm). The fiber diameter at the node at greater than at the internodes. Significant different existed between the fiber diameter in position at the internodes and nodes. The fiber diameter at different position showed that the middle layer has greater in sizes followed by the inner and outer layer. The fiber diameter of the *Gigantochloa* genera in this study ranged between 17-22.8 μm . The previous studies on the fiber diameter for *G. scortechinii* were 26 μm (Hisham, 2006), 23-37 μm (Abd. Latif, 1995), while studies on the species of *bambusa* genera found that; fiber diameter for *B. blumeana* were 12.0 μm (Ireana, 2009), *B. vulgaris* was 16.9-18.0 μm (Razak, 2010), 20-42 μm (Latif, 1995). This study found that the fiber diameter

were smaller than the previous studies. The diameter of the fiber of this study was 17-22.8 μm and is smaller than the Softwood (35 μm) and hardwood (25 μm). The comparison between the fiber diameter on this study showed that the fiber diameter *Gigantochloa* genera (17-22.8 μm) was bigger than *Eucalytus spp* (15.5 - 16.3 μm).

Lumen diameter

The lumen diameter for *G. scortechinii*, *G. brang*, *G. levis* and *G. wrayi* were 8.60 μm , 4.75 μm , 4.75 μm and 4.75 μm respectively (see Table 5). The results obtained in *G. scortechinii* were almost the same as obtained by Hisham (2006), but smaller than those obtained by Latif (1995). The lumen diameter for *Bambusa* were 1.6 μm for *B. blumeana* (Ireana, 2009), 2.3-2.6 μm for *B. vulgaris* (Razak, 2010). The lumen diameter for *Eucalytus spp* was 8.5-9.5 μm . The lumen diameter at the nodes were larger than those at the internodes. The lumen diameter at the inner layer were larger than those at the middle and outer layers.

Wall thickness

The results on the measurement of the wall thickness of various *Gigantochloa* species are shown in Table 5. *G. levis* (9.34 μm) possess thicker wall compare to the *G. brang* (9.02 μm), *G. wrayi* (7.02 μm) and *G. scortechinii* (4.30 μm). The fiber wall thickness were thicker at the nodes than at the internodes. The wall thickness at the middle layer were thicker at the middle followed by the outer and inner layers respectively. From this study the fiber wall thickness for *Gigantochloa* genera ranged 4.3-9.34 μm . The fiber wall thickness for *G. scortechinii* obtained by Hisham *et al* (2006) was 8-10 μm and Latif (1995) was 12.5-30.1 μm . The fiber wall thickness of *G. wrayi* (9.02 μm), *G. brang* (9.34 μm) was thicker than *B. blumeana* which was 5.01 μm (Ireana, 2009), *B. vulgaris* which was 7.1-7.6 μm (Razak, 2010), 2.5-13.3 μm (Latif, 1995). As a comparison, the fiber wall thickness of *G. scortechinii* almost similar with fiber wall thickness of *Eucalytus spp* which was 4.3 μm and 3.29-3.86 μm (Viane *et al.*, 2009), respectively.

Analysis of Variance

The analysis of variance (ANOVA) for the above studies are shown in Tables 4 and 5. Table 4 shows the ANOVA on the vascular bundle distribution, vascular bundle length and vascular bundles width between the bamboo species at nodes and internodes. Table 5 shows the ANOVA on the length, fiber diameter, fiber lumen diameter, fiber wall thickness and Runkle's ratio between the bamboo species, position and position.

Fiber Runkle's ratio

The results on the fiber Runkle's ratio of various *Gigantochloa* species in Table 6. The higher mean of fiber Runkle's ratio was *G. levis* (5.32) followed by *G. brang* (4.90), *G. wrayi* (4.13) and the lowest was *G. scortechinii* (1.35). The mean average of fiber Runkle's ratio for internode was 4.17 and for node was 3.68. It shows there was significant different of the Fiber Runkle's ratio between position at node and internode of the bamboo.

The result showed the fiber Runkle's ratio was bigger at the node as compare to the internodes and it was a significantly difference between this two position. Table 6 showed the value of fiber Runkle's ratio at node and internode. The mean average for fiber Runkle's ratio at difference position showed that at the outer layer was 7.03, middle 8.43 and at the inner layer was 6.80. The result showed that the fiber Runkle's ratio is bigger at the middle and thinner toward the inner and outer layer. It was a significantly difference between this three position. Table 6 showed the fiber Runkle's ratio value for every species, position and position. Runkle's ratio of fiber in this study was higher than 1.0 for *G. brang*, *G. wrayi* and *G. levis*. *G. scortechinii* was the only species has the value of Runkle's ratio less than 1.0. The Runkle's ratio value more than one, this main the fiber properties was hard and difficult to felting during the paper production. The quality of the paper will be gross and poor bonding if Runkle's ratio value more than one. If the Runkle's ratio less than one, it indicates the fiber has a thin fiber wall and easily to felting. The quality of the paper will be better and bonding will be good. This indicates that *G. scortechinii* could be a source to replace short-fiber pulp that was imported from abroad. *Eucalytus spp*, the Runkle's ratio is less than 1.0, namely 0.7 and 0.8 (Viena *et. al.*, 2009) was even shorter fiber than *G. scortechinii*. The Runkle's for hardwood and Softwood was 0.4-0.7 and 0.35 respectively. Kenaf has Runkle's ratio of 0.5-0.7 to prove they are good fiber felting power. Runkle's ratio for *G. scortechinii* was lowest than value one, which was 0.97 to prove it can still be used.

Ultrastructure Study

The ultrastructure studies on the four (4) selected bamboo from *Gigantochloa* genera are shown in Figures 1 to 8. Figures 1 to 4 shows the ultrastructures of the internodes and nodes focusing at the vascular bundles of the *G. brang*, *G. levis*, *G. scortechinii* and *G. wrayi* taken using the SEM.

Figures 5 to 8 shows the images of the bamboo species taken using TEM focusing at the fibers cells. The cells wall of the fibers clearly shows that they possess more than two layers, which were S1, S2, S3 and Sn.. All the four bamboo species shows clear that they belong to bamboo of are bamboo in class type. They possesses of a vascular sheath fiber and one fiber strand. Eventhough all the cells are similar in shape but they are however different in sizes in position at internodes and nodes, and position in the bamboo at either the outer, middle and the inner layers. The distribution of the vascular bundles per mm², vascular length, mvascular bundle width are shown in Tables 1, 2, 3 and 4. The fibers length, diameter, lumen diameters, wall thickness and the Runkle's ratio are given in Tables 5 and 6.

Conclusion

The anatomical structure varies significantly with the species. The distribution and the size of vascular bundle are differences between species and even in the same genera. The vascular bundle of four species almost similar and were classified under namely Type III consisting of single vascular sheath fiber and one fiber strand. But, the vascular bundle size was significantly different between position (node and internode) and position (outer, middle and inner layers).

The fiber morphologi for each species has a different measure of size in terms of length, diameter, lumen diameter and wall thickness. The study identified that there was differences in fiber dimensions on the position (node and internode) and position (outer, middle and inner layer) in the same species. Fiber length was longer at the internode node. While the middle layer has the longest fiber length compare to the outer and inner layer.

Ultra structures of bamboo genera *Gigantochloa* have different properties than wood. Bamboo fiber has a small lumen and thick wall Thickness. Wall Thickness of wood only has two layers which was S1 and S2, while the bamboo fiber has more than two layer, which were S1, S2, S3 and Sn. *G. scortechinii* fiber has great potential to be a resource of fiber production. It will be a source to replace short fiber from of hardwood, to combine with soft wood pulp for paper production in the future.

Table 4: Analysis of variance for anatomical properties between bamboo species, position & position.

		Anatomical Properties		
		No. Vascular bundle	Vascular bundle length	Vascular bundle Width
Species	<i>G. brang</i>	5.47b	894.55c	530.22c
	<i>G. levis</i>	4.23c	1182.51a	759.35a
	<i>G. scortechinii</i>	7.05a	932.71b	544.63c
	<i>G. wrayi</i>	5.75b	847.84d	598.02b
Position	Internode	6.32a	869.87b	585.42b
	Node	4.93b	1058.94a	630.70a
Position	Outer layer	8.56a	748.54c	467.23c
	Middle layer	4.89b	1013.25b	599.76b
	Inner layer	3.42c	1131.42a	757.19a

Table 5: Analysis of variance for fibre morphology between species, position & position.

		Fibre Morphology				
		Fibre Length	Fiber Diameter	Lumen Diameter	Wall Thickness	Runkle's Ration
Species	<i>G. brang</i>	1909.68b	22.75a	4.75b	9.02a	4.90b
	<i>G. levis</i>	2039.98a	22.67a	4.00c	9.34a	5.32a
	<i>G. scortechinii</i>	1745.27c	17.26b	8.66a	4.30c	1.35d
	<i>G. wrayi</i>	1798.79c	17.86b	3.83c	7.02b	4.13c
Position	Internode	2074.24a	18.23b	4.43b	6.90b	4.17a
	Node	1672.62b	22.04a	6.18a	7.02a	3.68b
Position	Outer layer	1698.52c	18.49c	5.44c	7.03b	4.04b
	Middle layer	2060.41a	22.36a	5.51b	8.43a	4.29a
	Inner layer	1861.35b	19.56b	5.96a	6.80c	3.45c

Values followed by the same letter in a column is not significant different at 95% probability level.

Table 6: Runkle's ratio of various *Gigantochloa* species

Sample Position	Position	<i>G. brang</i>	<i>G. levis</i>	<i>G. scortechinii</i>	<i>G. wrayi</i>
Internode	Outer	5.64 (±3.05)	4.71 (±2.80)	2.16 (±1.50)	4.12 (±1.34)
	Middle	5.67 (±1.95)	7.19 (±3.09)	1.42 (±0.95)	4.90 (±1.84)
	Inner	4.51 (±2.27)	5.44 (±2.09)	0.97 (±0.78)	3.28 (±1.05)
Node	Outer	4.85 (±1.84)	4.62 (±2.09)	1.71 (±0.96)	4.52 (±2.49)
	Middle	4.45 (±1.91)	5.29 (±2.64)	1.06 (±0.64)	4.33 (±1.64)
	Inner	4.30 (±7.63)	4.64 (±2.55)	0.79 (±0.57)	3.63 (±1.59)

Values in bracket represent the standard deviation,

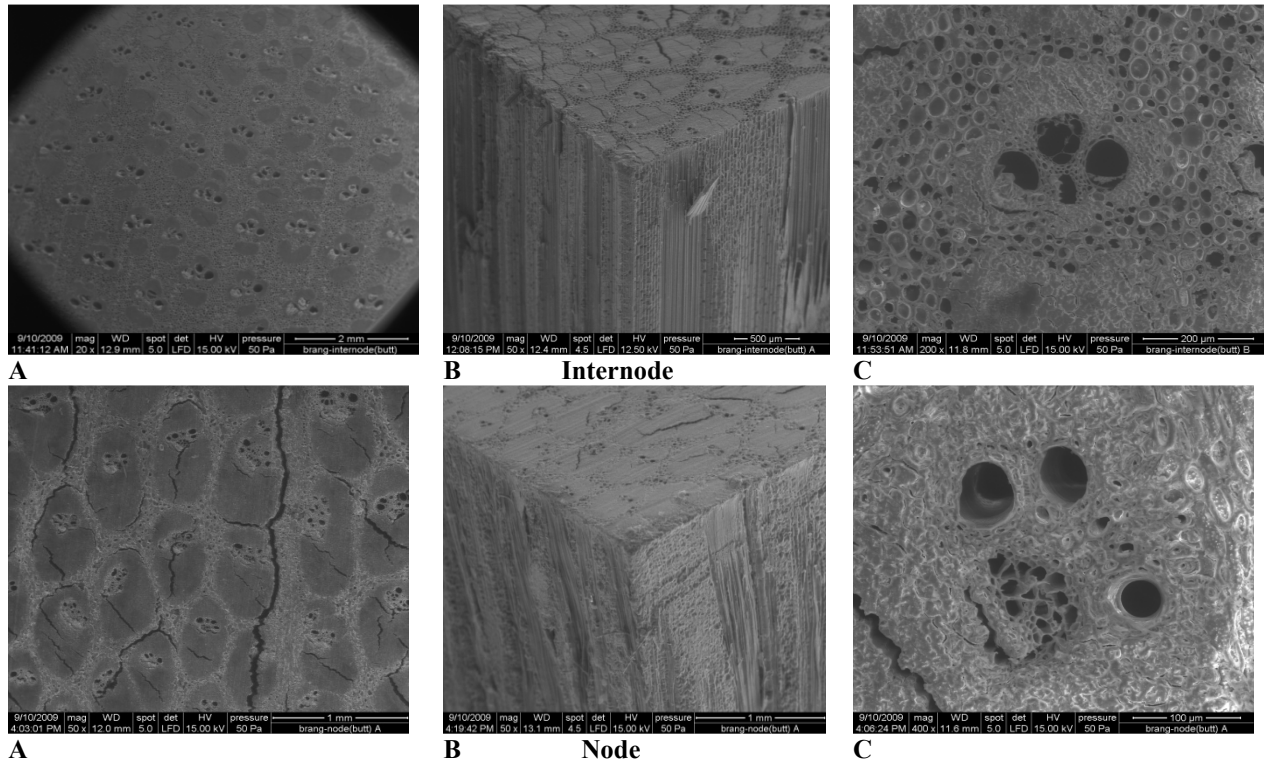


Figure 1: SEM images of the vascular bundles at internodes and nodes of the *G. brang*

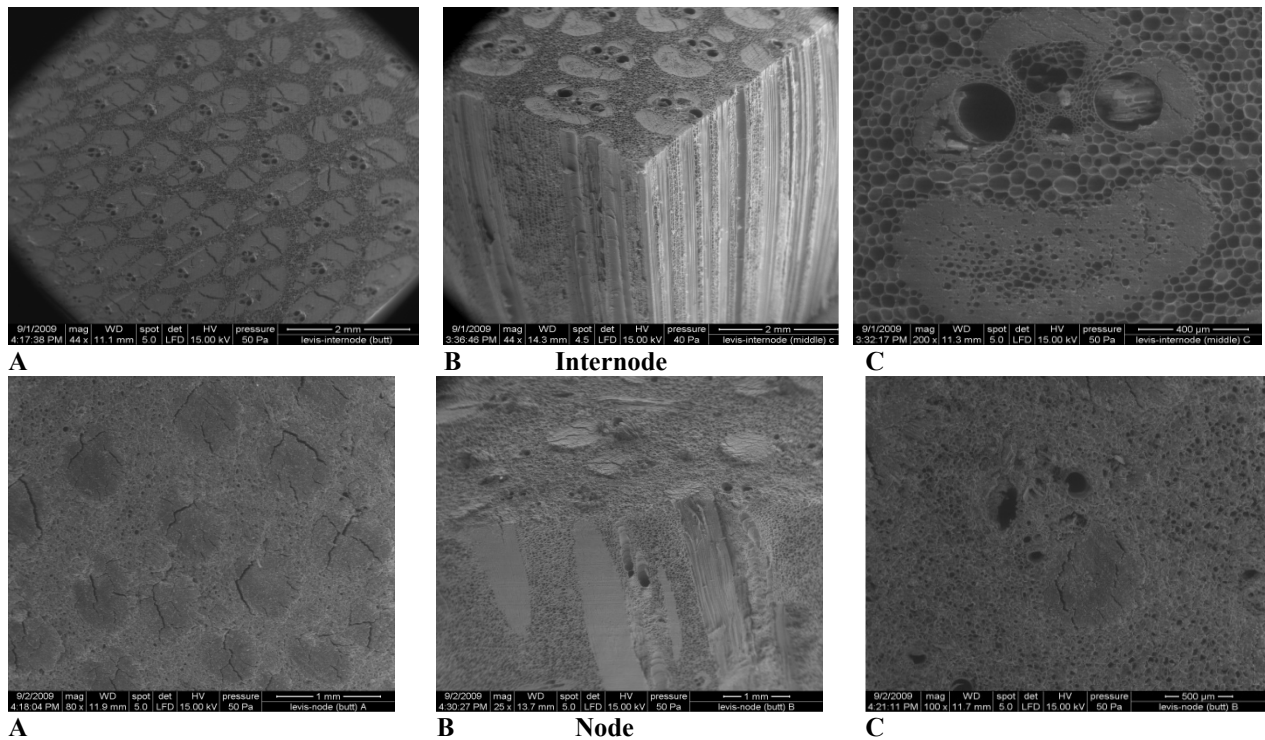


Figure 2: SEM images of the vascular bundles at internodes and nodes of the *G. levis*

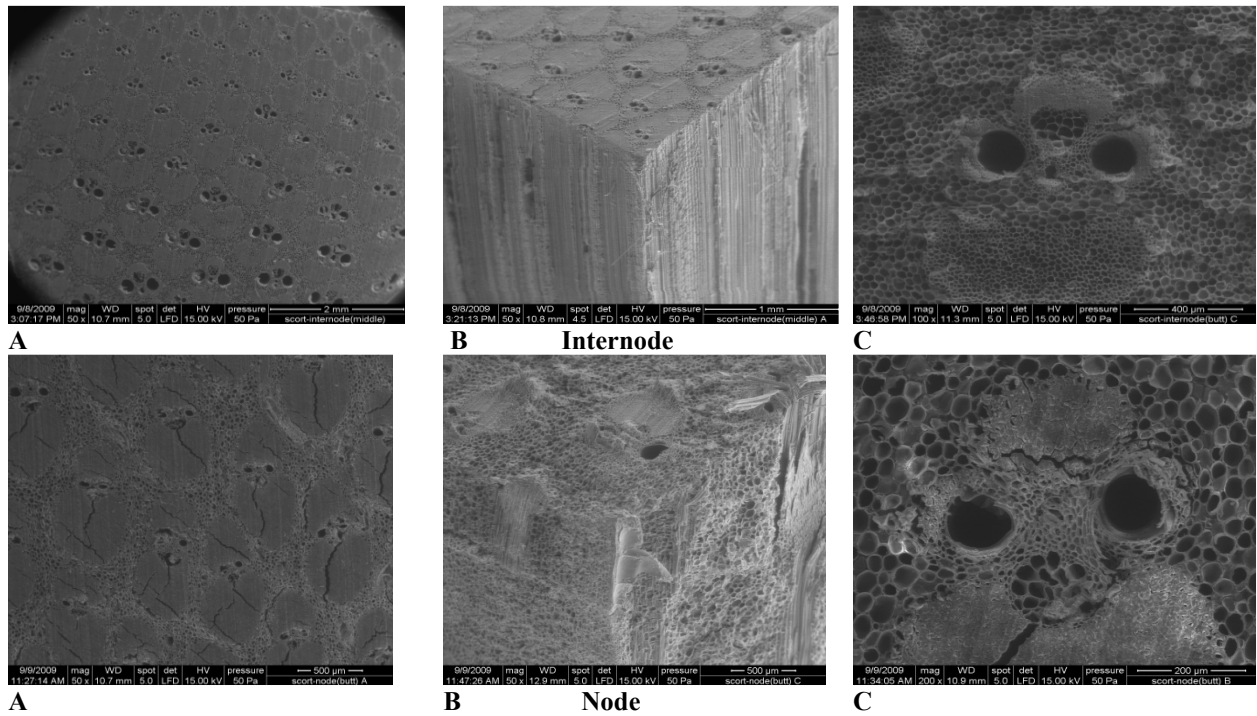


Figure 3: SEM images of the vascular bundles at internodes and nodes of the *G. scortechinii*

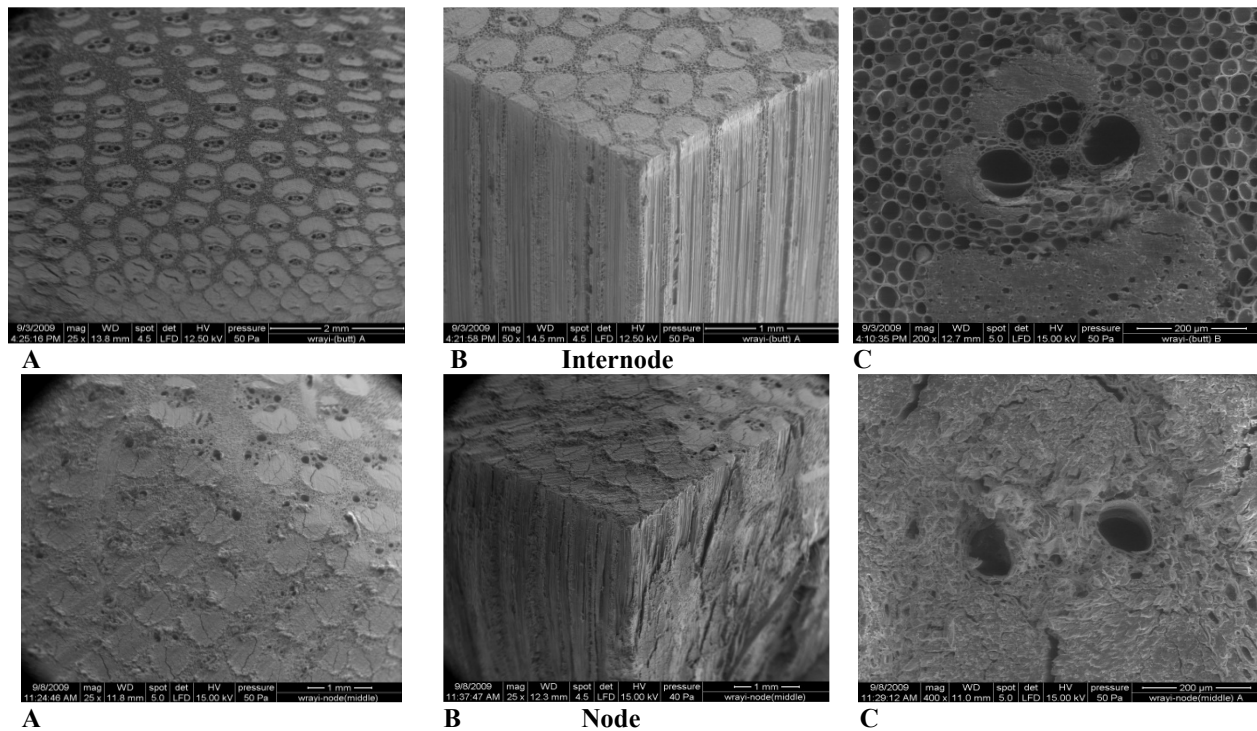


Figure 4: SEM images of the vascular bundles at internodes and nodes of the *G. wrayi*

Note: SEM image at the internodes and nodes of *G. brang*, *G. levis*, *G. scortechinii* and *G. wrayi*. Distribution of vascular bundle (left), Isometric view (central) and vascular bundle image (right).

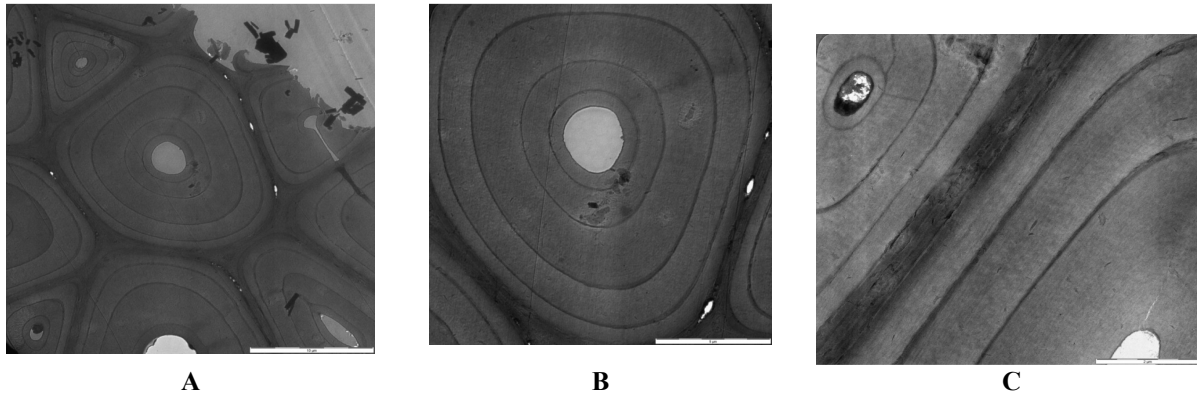


Figure 5: Fibre cells (A, B) and middle lamella (C) at internodes of the *G. brang*

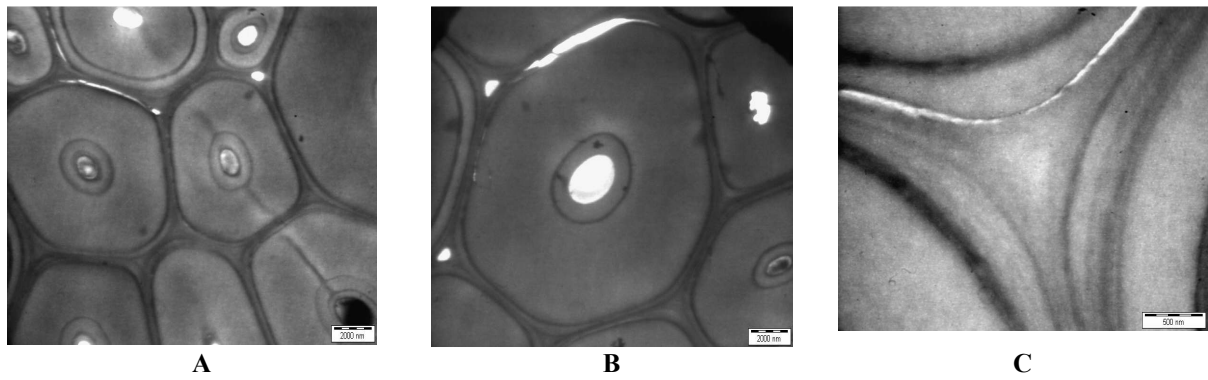


Figure 6: Fibre cells (A, B) and middle lamella (C) at internodes of the *G. levis*

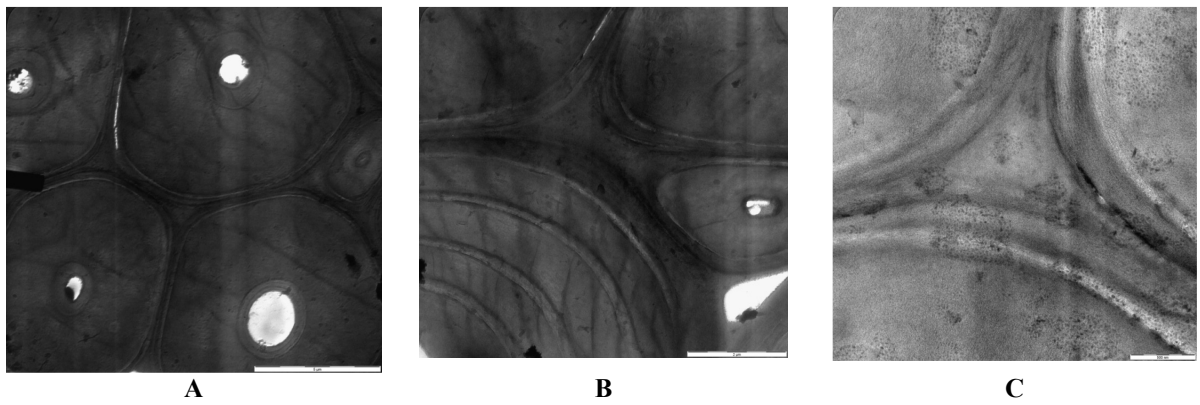


Figure 7: Fibre cells (A, B) and middle lamella (C) at internodes of the *G. scortechinii*

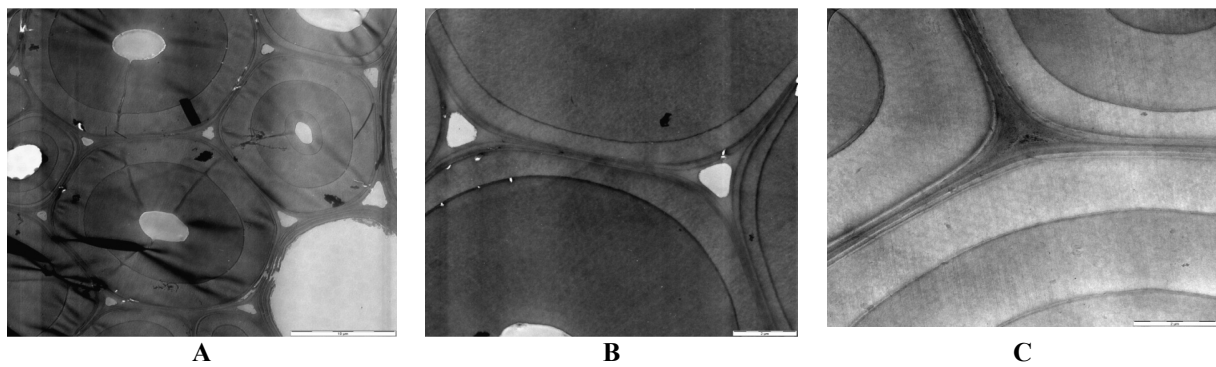


Figure 8: Fibre cells (A, B) and middle lamella (C) internodes of the *G. wrayi*

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