Effect of vessel diameter on variation of fiber morphology in Acacia mangium

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Effect of vessel diameter on variation of fiber morphology in *Acacia mangium*

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ABSTRACT

Paper quality depends on fiber diameter and wall thickness, and their derivatives. Fiber deformation occurs due to pressure from the vessel during development. The diameter and wall thickness of the fibers were measured following the direction of pressure exerted by the vessel on the face of the fiber cells. Fiber cell diameter measured perpendicular to and parallel with vessel enlargement was referred to as radial and tangential diameter, respectively, and likewise for fiber wall thickness. Differences in radial and tangential diameter and wall thickness of fiber cells in relation to their distance from vessels were analyzed. The radial diameter of fibers adjacent to large vessels decreased from the first to the fifth fiber, and from the first to the second fiber adjacent to small vessels. Conversely, tangential fiber diameter increased from the first to the fifth fiber for fibers adjacent to large vessels, and from the first to the second fiber adjacent to small vessels. The fibers adjacent to the vessel seem to have thicker walls in both the tangential than radial directions up to 2 and 5 fibers for small and large vessels, respectively. The first two fibers adjacent to small diameter vessels may produce higher strength paper than those up to five fibers from large diameter vessels, because the Runkel ratio, Coefficient of rigidity and Muhlsteph ratio values of fibers adjacent to small vessels are lower than fibers adjacent to large vessels. The opposite occurs for flexibility coefficient values.

Keywords: *A. mangium*; fiber diameter; fiber wall thickness; vessel size.

INTRODUCTION

Fast-growing *Acacia mangium* Willd. is a major forest plantation timber species in Indonesia (Jasmani & Adnan 2017; Permadi *et al.* 2017). Wood from this species is used for solid wood and paper products (Yong *et al.* 2011; Griffin *et al.* 2014).

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It is commonly known that the diameter and wall thickness of fibers and their derived values affect the quality of paper (Dutt & Tyagi 2011). Thin-walled fibers, due to their wide lumens, will easily be bonded to other fibers, which consequently increases the strength of the paper produced (Tofanica *et al.* 2011). Thin-walled fibers will also improve the smoothness of the paper surface. The Runkel ratio, Coefficient of rigidity, Muhlstep Ratio and flexibility coefficient determine the suitability of fiber for paper production (Yahya *et al.* 2010). Therefore, knowing the factors that influence variation in diameter and wall thickness of fibers as well as their derived values is needed in order to increase paper quality.

The diversity of fiber properties is generally driven by specific environmental factors (Briggs 2010; Sisi *et al.* 2012; Meyer *et al.* 2013; Zumaya-Mendoza & Terrazas 2016) or internally controlled genetic factors (Yahya *et al.* 2010; Dutt & Tyagi 2011; Elissetche *et al.* 2011; El Moussaouiti *et al.* 2012; Gomes *et al.* 2015). The effect of these two factors on paper quality has been reported in many studies. However, biomechanical factors may also be important. During tree growth, internal stress due to the development and dimension of fibers always occurs. Processes during maturation determine fiber dimensions (Ridoutt & Sands 1993; Ohshima *et al.* 2011; Rao *et al.* 2011; Aref *et al.* 2014). The fusiform initial is different for fibers in radial and tangential rows. One factor that determines the length of the fiber is the length of the fusiform initial (Ridoutt & Sands 1993; Ohshima *et al.* 2011; Rao *et al.* 2011). The final size of the fiber is also influenced by the state of maturity of other xylem elements in the vicinity (Honjo *et al.* 2006; Yahya *et al.* 2011; Yahya *et al.* 2015). Hence, better knowledge of factors controlling fiber properties will allow more effective use of fiber materials (Pirralho *et al.* 2014).

The effect of other cells on fiber dimensions, such as diameter and thickness, and their derived values, should be considered. Many cells undergo dimensional enlargement, including elongation and increase in diameter, before they reach maturation (Panshin & de Zeeuw 1980). Among all types of cells in hardwood, vessels experience the most significant increase in diameter. In the wood of *A. mangium*, vessels occupy 12.1% (Yahya *et al.* 2010).

However, it is very difficult to measure fiber dimensions in relation to distance from a vessel. Maceration is the quickest and most direct method to measure fiber dimensions (Yahya *et al.* 2010). However, as maceration affects the position between cells the wood cell position, including that of fibers and vessels cannot be measured. Consequently, this method cannot be used to study the relationships between vessels and fiber dimensions (Yahya *et al.* 2017).

The three-dimensional (3D) serial cross-section method was introduced by Honjo *et al.* (2006) and other previous researchers to measure fiber length in relation to distance from vessels. However, this method is complicated, tedious, and time-consuming, and not practical for where large sample sizes are needed, such as tree breeding programs.

The 3D method was refined by Yahya *et al.* (2011) to speed up the measurement of fiber properties in relation to distance from vessels. Through the newly developed 3D method, fiber length up to five and two fibers away from a vessel in the radial and tangential directions, respectively, was found to be significantly shorter than fibers at greater distances from the vessels (Yahya *et al.* 2011). The diameter and wall thickness of fibers in *A. mangium* have also been found to vary based on distance from a vessel (Yahya *et al.* 2015). The size of the vessel diameter used in that study ranged from 180–220 µm. Nugroho *et al*. (2012) found that vessel diameter in *A. mangium* was varied. Because the vessels cause deformation as mentioned above, it is suspected that variation in vessel diameter will affect the diameter and wall thickness of fibers and their derived values based on their distance from the vessel. The objective of the present study was to observe the influence of vessel lumen diameter on fiber diameter and wall thickness and their derived values, in *A. mangium*.

MATERIALS AND METHODS

A wood block of 10 mm × 5 mm × 20 mm (in radial, longitudinal and tangential directions) was cut from near the bark of a seven-year-old *A. mangium* tree. The tree was randomly selected from the trial area of a private forest plantation, Musi Hutan Persada (MHP) Company, in South Sumatra, Indonesia. The sample was prepared as described below.

The wood block was first softened by heating it at 160°C for 15 min in a small autoclave with a mixture of alcohol and glycerol (1:1 ratio). The softened wood block was then serially sectioned resulting in 200 slices of 25 µm thick cross-sections. The cross-sections were sequentially mounted on glass slides. A confocal laser scanning microscope (LSM 5 Pascal Ver. 2.5; Carl Zeiss, Heidelberg, Germany) was used to observe the samples. Photographs were taken of the sections with an image size of 2048 × 2048 pixels and a pixel resolution of 0.45 µm. The position at the middle of the optical slice was specifically used for further analysis to avoid the distortion of the structure caused by sectioning (Yahya *et al.* 2011).

The software 'Reconstruct' (Fiala 2005) was applied to align the series of images of samples. The 3D Viewer function of ImageJ software was used to further examine the aligned set of sequential images. An image containing the largest diameter along the fiber in these serial sections was used for measurement of fiber cell diameter and lumen diameter. Before the measurement, we differentiated between fibers and paratracheal parenchyma which has thinner cell walls than the fibers (Fig. 1a; Sahri *et al.* 1993). Wall thickness of fibers was calculated according to the following formula:

$$\text{Fiber wall thickness} = (\text{fiber diameter} - \text{fiber lumen})/2 \tag{1}$$

Fiber cells that are distant from a vessel have a shape sharply defined by the radial and tangential directions in the tissue. In contrast, fiber cells adjoining a vessel deviate from this well-defined shape as a result of vessel enlargement. Fiber/lumen diameter and fiber wall thickness were measured in radial and tangential directions to the vessel (Fig. 1; Yahya *et al.* 2015). The following formulas were applied to calculate the average diameter and the cell wall thickness of fibers:

$$\text{Mean fiber diameter} = (\text{radial diameter} + \text{tangential diameter})/2 \tag{2}$$

$$\text{Mean fiber wall thickness} = (\text{radial thickness} + \text{tangential thickness})/2 \tag{3}$$

Changes in fiber diameter and wall thickness were observed in relation to distance from the vessel. Tangential lumen diameter of the vessel was used as the basis for measuring the changes in fiber diameter and cell wall thickness. Based on preliminary measurements of

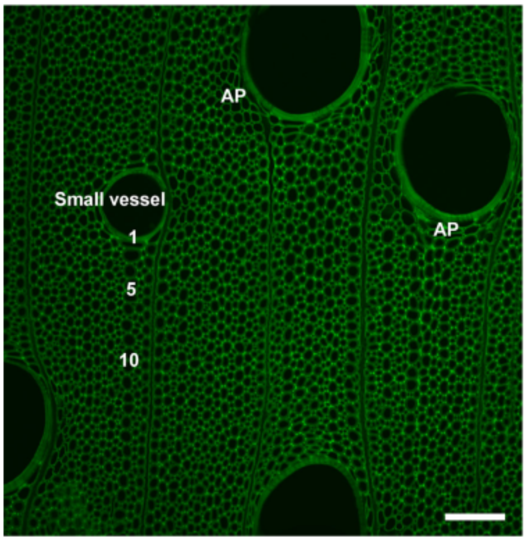

Figure 1. Transverse section of *Acacia mangium* showing fiber cells radiating outwards from the vessel and fiber cell shape changes parallel with and perpendicular to vessel enlargement. AP = axial parenchyma. Scale bar = 100 µm.

lumen diameter, the lumens in the present study were categorized as large (171–212 µm) and small (85–109 µm). The radial distance from a vessel was marked by cell number, with the vessel-adjoining fiber as the 1st cell. The radial and tangential diameter and wall thickness of the fibers were compared using the *t*-test.

Average values of fiber diameter and wall thickness (radial and tangential) were calculated based on their distance from the vessel, then fiber derivative values were calculated as follows:

$$\text{Runkel ratio,} \quad RR = 2 \times FWT/FLD \quad \text{(Pirralho et al. 2014)} \tag{4}$$

$$\text{Coefficient of rigidity,} \quad CR = FWT/FD \quad \text{(Tamolang \& Wangaard 1961)} \tag{5}$$

$$\text{Flexibility coefficient,} \quad FC = FLD/FD \quad \text{(Pirralho et al. 2014)} \tag{6}$$

$$\text{Muhlsteph ratio,} \quad MR = (FD)^2 - (FLD)^2/(FD)^2 \quad \text{(Tamolang \& Wangaard 1961)} \tag{7}$$

where

FWT = Fiber wall thickness
FLD = Fiber lumen diameter
FD = Fiber diameter

RESULTS AND DISCUSSION

Comparison of fiber diameter and wall thickness in relation to distance from small and large vessels

Fiber cells adjacent to small vessels were larger and more elongated than those adjacent to large vessels (Fig. 2). Cells adjoining small vessels (1st cells) measured 35 µm and 19 µm, in radial and tangential directions respectively, whereas cells adjoining large vessels (1st cells) measured 25 µm and 15 µm in radial and tangential directions, respectively. The pressure exerted by large vessels appeared evident until the 5th cell as radial and tangential diameter values converged at the 6th cell (19 µm for both). Small vessels appeared to exert less influence on cell radial and tangential diameters with distance, radial diameter dropped sharply from the 1st to 2nd cell (35 to 24 µm, respectively) while tangential diameter increased (19 to 21 µm, respectively), such that radial and tangential diameters had converged before the 3rd cell. Following convergence, fiber radial diameters up to the 13th fiber were relatively constant, ranging from 16–20 µm and 18–19 µm, in proximity to small and large vessels, respectively. Likewise, tangential diameters ranged from 20–22 µm and 19–21 µm, in proximity to small and large vessels, respectively.

The average tangential diameter of fibers adjoining small vessels (35 µm) was much higher than that of fibers adjoining large vessels (25 µm). This difference could be due to two things. First, small vessels may exert less pressure on fibers than larger vessels resulting in a smaller degree of deformation (flattening) in fibers adjoining the small vessels. Second, fibers adjacent to small vessels may have grown larger than those adjacent to large vessels because more space was available next to the smaller than expected vessels for fiber development. The final dimensions of wood fibers will also be influenced by the maturity level of other surrounding cells (Honjo *et al.* 2006; Yahya *et al.* 2011).

Further observation showed that the number of flattened fibers in relation to their distances from small or large vessels differed. The number of flattened fibers adjacent to large

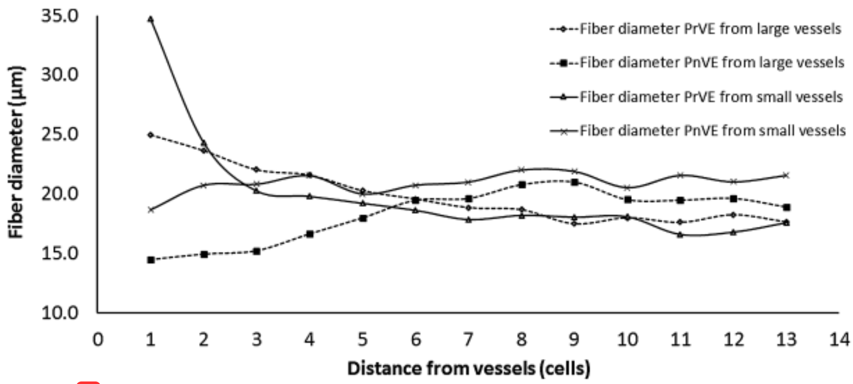

Figure 2. Variation in fiber radial and tangential diameter in relation to distance from large and small vessels. PrVE = parallel to vessel enlargement, PnVE = perpendicular to vessel enlargement.

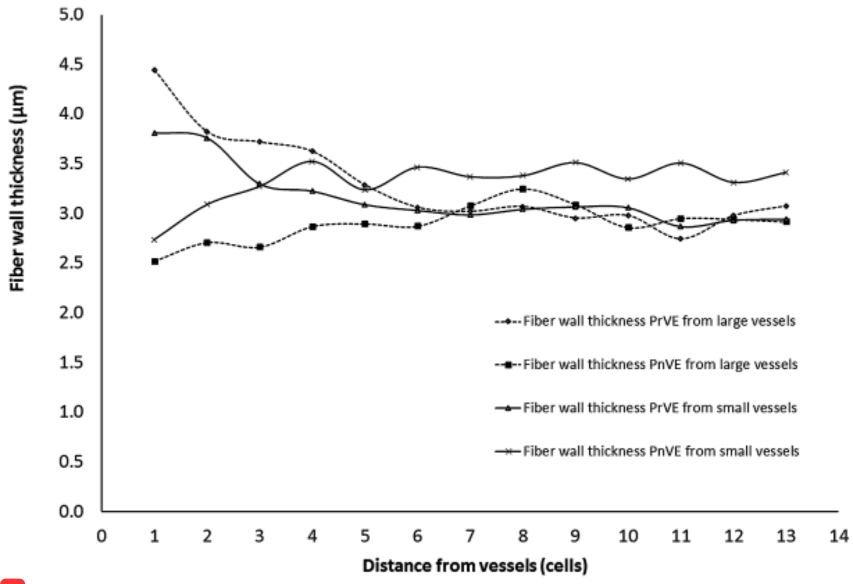

Figure 3. Variation in fiber wall thickness in relation to distance from large and small vessels. PrVE = parallel to vessel enlargement, PnVE = perpendicular to vessel enlargement.

vessels was five. In contrast, only two fibers adjacent to small vessels were flattened. This phenomenon was likely due to the higher pressure exerted on the fibers by large vessels as compared with small vessels.

The fibers adjacent to the vessel seem to have thicker walls in the tangential compared to the radial directions (Fig. 1a). For large vessels, fiber tangential wall thickness gradually decreased with distance from the vessel. Wall thickness of fibers adjacent to large vessels decreased from 4.5 µm for the first fiber to 3.3 µm for the fifth fiber (Fig. 3). For fiber radial wall thickness there was a gradual increase from vessel-adjoining fibers outwards. The increment for fibers adjacent to small vessels was from 2.7 µm for the first fiber to 3.1 µm for the second fiber. For others, there was no detectable difference in the cell wall thickness.

Comparison of fiber quality adjacent to large and small vessels as raw material for paper production

Runkel ratio and coefficient of rigidity values of fibers adjacent to small vessels (up to two fibers away from the vessel) were lower than those for fibers adjacent to large vessels (up to five fibers away from the vessel; Fig. 4). The suitability of fiber for paper production is determined by its Runkel ratio value. A decrease in the Runkel ratio value tends to indicate increased paper strength (Istikowati *et al.* 2016; Takeuchi *et al.* 2016). The Runkel ratio value is negatively correlated with burst (Elmas *et al.* 2018) and tensile strength of paper (Dutt & Tyagi 2011; El Moussaouiti *et al.* 2012). A Runkel ratio lower than 1 is preferred for producing good paper characteristics (Sharma *et al.* 2011; Kiaei *et al.* 2014).

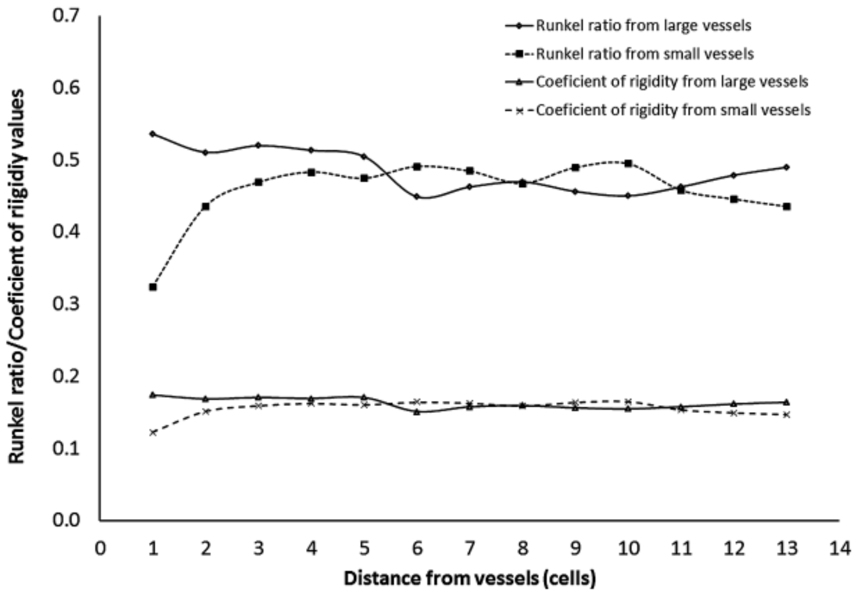

Figure 4. Variation in Runkel ratio and Coefficient of rigidity values in relation to distance from large and small vessels.

Paper strength will decrease if there is an increase in the coefficient of rigidity of the fiber. The coefficient of rigidity value is inversely proportional to the tensile, tear, burst and double fold resistance of paper (Elmas *et al.* 2018). To produce writing and printing paper, a low coefficient of rigidity is needed (Akgul & Tozluoglu 2009). Based on the results of the present study, we anticipate that *A. mangium* wood fibers adjacent to small vessels will produce higher strength paper than that utilizing fibers adjacent to large vessels.

Flexibility coefficient values of fibers adjacent to small vessels were higher than those adjacent to large vessels, while the opposite was recorded for Muhlstep ratio values (Fig. 5). Fibers with high flexibility coefficient values are flexible, collapse readily and produce good surface contact and fiber to fiber bonding (Kiaei *et al.* 2014). Fibers with a flexibility coefficient greater than 60 are preferred in paper production (Rydholm 1965) because paper strength is directly proportional to the flexibility coefficient of fibers. Fibers that have a high flexibility coefficient will produce paper that is high in burst, tensile and fold strength (Ogbonnaya *et al.* 1997).

Muhlsteph ratio is directly proportional to cell wall thickness. Thin-walled fibers will produce paper with high burst, tensile strength and folding endurance (Sharma *et al.* 2011; Tofanica *et al.* 2011; Kiaei *et al.* 2014).

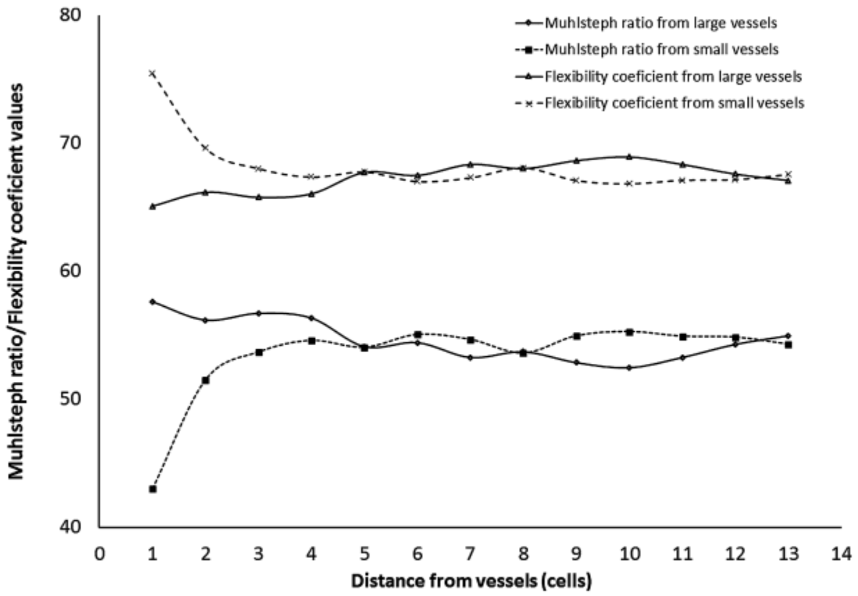

Figure 5. Variation in Muhlsteph ratio and flexibility coefficient values in relation to distance from large and small vessels.

Table 1.
Results of *t* tests on the differences in mean radial and tangential diameter and wall thickness of fibers distant from large and small vessels.

Parameter	Radial		Tangential		*p*
	Mean	SD	Mean	SD	
Fiber diameter distant from small vessel	21.2	0.62	18.3	1.14	0.000
Fiber diameter distant from large vessel	19.9	0.73	18.3	0.74	0.004
Fiber wall thickness distant from small vessel	3.4	0.99	3.1	0.12	0.000
Fiber wall thickness distant from large vessel	3.0	0.13	3.0	0.11	0.864

Note: Distant fibers for small and large vessels mean starting from the 3rd and 6th fibers respectively, until the 13th fiber from a vessel.

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