

Research paper

The changes of extractive contents of young *Swietenia mahagoni* (L.) Jacq trees during heartwood formation

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Abstract. We investigated the distribution of extractive, phenolic, and polysaccharides content of young *S. mahagoni* trees aged 1 to 5 years. The wood powder materials in two vertical positions (bottom and top) were successively extracted using n-hexane, methanol, and hot water solutions. The results showed that there was a significant interaction between the tree age and axial position in all parameters. The methanol and total extractive content decreased significantly from 1 to 3-year-old at the top part. In contrast, the amount of hot water and total extractive increased significantly from 3 to 4-year-old trees at the bottom part. Furthermore, a significant increase was also found at the bottom heartwood for hexane, methanol, and the total extractive from 4 to 5-year-old trees. The reverse trend was observed in the polar fractions (methanol and hot water) at the top of the trees. Generally, the extractive pattern tends to decrease from bottom to top with the highest proportion of methanol extract (70% based on the total extractive weight), and it increased from sapwood to heartwood. The total phenolic content (TPC) and total soluble polysaccharides (TSP) generally increase with tree age (except for TSP from 2 to 3-year-old trees at the top part). The high content of TPC in the heartwood and TSP in the sapwood indicated the process of heartwood formation. In addition, a drastic decrease in the methanol extract and TSP at the age of 3 years at the top indicated that the TSP was transferred from top to bottom for the heartwood formation at the bottom part of 4-year-old trees.

Key words: extractive content, phenolics, polysaccharides, heartwood formation, *S. mahagoni*

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Introduction

Swietenia mahagoni (L.) Jacq (*S. mahagoni*) is one of the most commercially valuable woods in tropical countries including Indonesia. This species belongs to the *Meliaceae* family and it is a native plant in Flori-

da in the southeastern region of the United States (Nahed *et al.*, 2017). This species is used for making furniture and musical instruments (He *et al.*, 2020). It is well known as raw material for veneer decoration. In addition, *S. mahagoni* has durability and resistance classes III and II–III against dry

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wood termites of *Cryptotermes cynocephalus* (Martawijaya *et al.*, 2005). It has a wood density of 0.56 to 0.72 g. cm⁻³, with a total tree height of more than 18 m, diameter at breast height (dbh) of 30 to 105 cm. The heartwood part of mahogany is very important, and the international market is expected to have a premium level (Haslett *et al.*, 1991; Wadsworth & Gonzales, 2008). Furthermore, this part has a reddish or pinkish color, and it becomes darker with increasing age to dark red or brown, while the sapwood is predominantly yellowish (Lemmens, 2005).

The part of the wood cell (parenchyma cells) that is physiologically inactive and filled with a non-organic structure (extractives) which contributes to its colorations and natural durability is the heartwood. This natural durability is related to the extractive content in the heartwood, in particular phenolic extractives, which are dominant and contain antioxidant activities (Rababah *et al.*, 2011). Rastogi & Mehrotra (1993) reported that there was biological activity in the heartwood part of *S. mahagoni*. They reported that *S. mahagoni* heartwood had the content of cyclo swietenol, lupleol, benzoate hedergenin, cycloartenol, and β -sitosterol. Additionally, Falah *et al.* (2008) found (+)-catechin, (-)-epicatechin and a new amorphous compound, namely swietem acrophyllanin in the hot water extractive of mahogany bark. These compounds showed a strong red color, and usually are used as tannin for tanning skin. Sukardiman & Ervina (2020) also reported that *S. mahagoni* contains some phytochemicals, such as phenols, flavonoids, phospholipids, alkaloids, anthraquinones, cardiac glycosides, saponins, terpenoids, volatile oils, and long-chain unsaturated acids.

The extractives are responsible for the odor, wood color, and protection from microbe and insect attacks (Umezawa, 2000). Furthermore, they are chemical constituents located in the lignocellulosic tissue that contains a higher diversity of organic compounds, such as triglycerides, neutral

compounds, and phenolic compounds (Nascimento *et al.*, 2013). The composition and distribution depend on species, growth location, position within the tree, and genetic factors (Hillis, 1987). Additionally, the content varies in axial and radial positions within the tree. Santana *et al.* (2012) reported that in the juvenile phase the extractive content is increased with increasing tree age. Yeh *et al.* (2006) mentioned that the extractive content of benzene alcohol in the juvenile phase increased from bottom to top. Meanwhile, Dünisch *et al.* (2010) stated that the content is increased from the juvenile heartwood phase to mature heartwood.

Studies related to extractives in mahogany were extensively conducted on the bark (Falah *et al.*, 2008; Arisandi *et al.*, 2019b; Masendra *et al.*, 2020), leaves (Rastogi & Mehrotra, 1993; Abdelgaleil *et al.*, 2006), seeds (Ekimoto *et al.*, 1991; Chen *et al.*, 2007) and twigs (Lin *et al.*, 2011). However, variations in phenolic, and polysaccharide contents in wood or heartwood parts of mahogany are still limited. Only a small number of studies on the extractive and phenolic contents of *Swietenia* sp. and *S. macrophylla* King wood were reported (Batubara *et al.*, 2012; da Silva *et al.*, 2013). In addition, a previous study showed that heartwood formation in *S. mahagoni* begins after a tree reaches 4 years of age (Arisandi *et al.*, 2021). However, the change of the chemical properties such as the extractive, phenolic, and polysaccharide content, which is related to the durability of mahogany wood, was not discussed.

Because the presence of heartwood is very important as it is related to natural durability and its quality, research is necessary to investigate the extractive changes that occur during the heartwood formation process. The formation process transforms sapwood into heartwood (Niamké *et al.*, 2011). The transition of metabolic activities from sapwood to heartwood extractives form (phenolic compounds) from reserve materials (non-structural carbohydrates/

NSC) was also reported (Datta & Kumar, 1987; Nobuchi *et al.*, 1996). Niamké *et al.* (2011) found that NSC (starch, glucose, fructose, and sucrose) decreased drastically from sapwood to heartwood in teak wood. In contrast, it was found in the phenolic components (H1, P1, 1,4 naphthoquinone, 2-(hydroxymethyl) anthraquinone, anthraquinone-2-carboxylic acid, lapachol, and tectoquinone. In the temperate species, heartwood formation was well documented but poorly understood in tropical species (Magel *et al.*, 2001; Niamké *et al.*, 2011). Understanding heartwood formation is important due to its economic significance (Mishra *et al.*, 2018). Therefore, the aim of this study was to investigate the distribution of extractive, phenolic and polysaccharide contents in the process of heartwood formation. The variation of tree age and axial position in all parameters are also discussed. We hypothesized that tree age and stem height had an influence on the distribution of extractives, phenolics, and soluble polysaccharides content. In addition, the content of phenolics and soluble polysaccharides assumed plays an important role in the process of heartwood formation.

Material and Methods

Five species of *S. mahagoni* from each tree ages were felled in Perum Perhutani plantation located in Temanggung Regency, Central Java, Indonesia (7°14'S, 110°2'E). The mean rainfall is more than 2000 mm in a year. The mean temperature is 20°C, with mean humidity of 74.4% and altitude of 500 to 1450 m above sea level (Setyawan *et al.*, 2019) with a soil type of clay and landscape of moderate slope. Tree spacing is 3 m x 2 m that had been generated by seedlings in the plantation where no thinning and pruning had been carried out. For the preliminary study, 4-year-old trees were collected in December 2018, while the 2- and 3-year-old trees were observed in October 2019. Subsequently, tree ages of 1 and 5

years were obtained in October 2020. Sampling of trees was carried out in the rainy season. The selected trees were straight trunks with low defects. Thirty trees were selected and felled, with each tree age represented by 5 trees. Each tree was cut into disc samples (4 cm in thickness) at the bottom (5% of total tree height) and top part (50% of total tree height) containing heartwood (Figure 1). After felling, the discs were immediately wrapped in plastic to avoid moisture loss and transported to Universitas Gadjah Mada for testing. After that, the samples were brought to the laboratory, and stored in a freezer before heartwood area was measured.

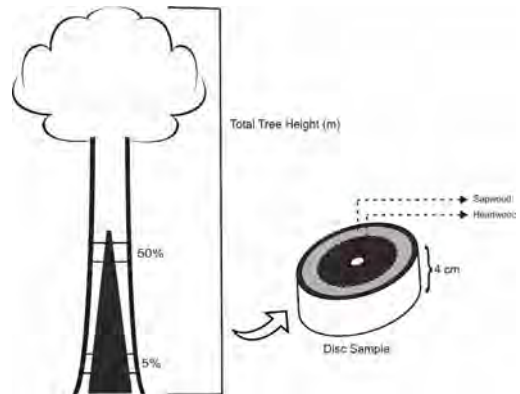


Figure 1. Wood sampling scheme for extractive analysis.

By visual inspection, trees within the age of 1 and 2 years contained sapwood only. Meanwhile, in the case of 4–5-year-old tree samples which possessed a heartwood area, each disc was divided into sapwood and heartwood. The heartwood had a distinctive salmon pink to red color compared with the light sapwood color. Lemmens (2005) stated that the *S. mahagoni* heartwood is reddish or pink in color, and darker with increasing age to dark red or brown, which is clearly different from the part of sapwood that is usually yellow in color. Based on the area in cross-section, the range of heartwood proportion at the age of 4 years from the bottom to top part

was 51.6% to 25.8%, while at the age of 5 years the range was from 49.0% to 19.8%.

Furthermore, the disc samples were stored at room temperature and air-dried after measuring the area of heartwood. Afterwards, air-dried wood specimens were collected by drilling in a disc from sapwood (0.5 cm from bark-sapwood border) and heartwood (1.0 cm from the sapwood-heartwood border). The drilled wood was then separately milled to a powder and sieve-screened (to pass a 1 mm sieve) for chemical analysis. Table 1 shows more detailed information on wood specimens.

Table 1. Description of tree samples from young *S. mahagoni* trees.

No	Age (year)	Average of DBH (cm)	Average of total merchantable height (m)
1	1	1.08 ± 0.28 a	0.54 ± 0.10
2	2	1.99 ± 0.32	1.72 ± 0.16
3	3	4.46 ± 0.10	4.25 ± 0.10
4	4	6.13 ± 1.12	4.80 ± 0.77
5	5	8.05 ± 1.17	6.60 ± 1.20

Note: a = Diameter is taken at the level height 10%

Chemicals

Sodium carbonate, sodium hydroxide, aluminum chloride, and Folin-Ciocalteu's phenol reagent were purchased from Merck (Darmstadt, Germany), while standard components of gallic acid and D-glucose were purchased from Sigma-Aldrich (Chemie GmbH, USA).

Extractives content determination

The powder (0.8 to 1.5 g) was successively extracted by *n*-hexane and methanol solutions for 6 hours in a Soxhlet apparatus, while hot water refluxing in separating the extraction for 3 hours. The number of percolations during the extraction was 8 cycles per hour. Furthermore, the solvent was evaporated by a rotary evaporator, then the extract was dried in an oven (103±2°C) and the extractive content was quantified.

Total phenolic content determination

The total phenolic content (TPC) was determined by the Folin-Ciocalteu method (Singleton *et al.*, 1999). 2.5 ml of diluted Folin Ciocalteu phenol reagent and distilled water (1:9, v/v) were mixed with 0.5 ml of the methanolic extract (0.25 mg/ml) in a 9 ml glass. After an interval of 2 min, 7.5% aqueous Na₂CO₃ (2 ml) was added, and the mixture was allowed to stand for 30 min at ambient temperature. Then, the absorbance for testing of standard solutions was performed against the blank at 765 nm with a Visible spectrophotometer (model VIS-WPA S800⁺). In addition, TPC was performed as the mean ± standard deviation of five tree replication measurements and was expressed as milligrams gallic acid equivalents (mg GAE/g dried extract).

Total soluble polysaccharides determination

Total soluble polysaccharides (TSP) were measured according to DuBois *et al.* (1956). Meanwhile, the hot water extract was dissolved in distilled water (0.25 mg/ml). 1 ml of the solution was mixed with 1 ml phenol reagent solution 5% (5 g in 100 ml in distilled water) before adding 5 ml of sulfuric acid (98%). Furthermore, the mixture was allowed to stand for 20 min at room temperature, and the absorbance for testing of standard solutions was performed against the blank with the same procedure (without extract) at 490 nm, using a Visible spectrophotometer (model VIS-WPA S800⁺). The TSP was performed as the mean ± standard deviation of five tree replication measurements and was expressed as milligrams glucose equivalents (mg GE/g dried extract).

Statistical analysis

The data were statistically handled using the SPSS program (version 16 IBM, New York, USA). Analysis of variance (ANOVA) was conducted and the significant differences were set at a 95% confidence level. Also, two-way ANOVA was applied

to determine the effect of tree age and axial position (bottom and top) on the extractive content, TPC, and TSP. All the obtained data were normally distributed after analysis, and Duncan's test was performed to evaluate groups that differ.

Results

Extractive content, total phenolic content, total soluble polysaccharides

The average value of n-hexane, methanol,

and hot water extractive content from bottom to top parts were 2.39 to 0.71%, 7.81 to 1.61%, and 5.21 to 1.01%, respectively (Table 3). In addition, Figure 2a showed that the total extractive content ranged from 3.93 to 13.7% of dried wood. Methanol solution has the highest contribution of total extractives, it ranged from 25 to 70.2% based on extractive weight (Figure 2b). Meanwhile, the value for hot water and n-hexane ranged from 13.6 to 50.3% and 10 to 29%, respectively.

Table 3. The interaction of tree age with an axial position on the extractive content of *S. mahagoni* wood.

Solvent	1 year		2 years		3 years	
	Bottom	Top	Bottom	Top	Bottom	Top
	Sapwood	Sapwood	Sapwood	Sapwood	Sapwood	Sapwood
<i>n</i> -Hexane	0.71 ± 0.14 a	1.09 ± 0.24 bc	1.26 ± 0.08 bc	1.28 ± 0.20 bcd	1.41 ± 0.41 cd	0.96 ± 0.13 ab
Methanol	4.30 ± 0.85 bcde	7.48 ± 2.34 f	4.48 ± 0.48 abcde	4.36 ± 1.04 cde	3.72 ± 1.09 abcd	2.57 ± 0.98 abc
Hot water	2.09 ± 0.70 bc	2.09 ± 0.50 bc	3.57 ± 1.19 e	2.22 ± 0.36 bc	5.21 ± 0.65 f	1.01 ± 0.20 a
Total	7.10 ± 1.23 bc	10.7 ± 2.27 de	9.31 ± 1.25 cde	7.86 ± 0.69 cd	10.3 ± 1.59 de	4.54 ± 0.95 ab
Solvent	4 years					
	Bottom		Top			
	Sapwood	Heartwood	Sapwood	Heartwood		
<i>n</i> -Hexane	1.64 ± 0.28 de	1.90 ± 0.42 e	1.14 ± 0.10 bc	1.22 ± 0.06 bc		
Metanol	4.16 ± 1.34 bcde	4.74 ± 1.32 cde	1.66 ± 0.33 a	1.93 ± 0.27 ab		
Hot water	3.19 ± 0.47 de	3.40 ± 0.54 e	1.13 ± 0.18 a	1.50 ± 0.43 ab		
Total	8.98 ± 1.57 cde	10.0 ± 1.16 de	3.93 ± 0.52 a	4.65 ± 0.56 ab		
Solvent	5 years					
	Bottom		Top			
	Sapwood	Heartwood	Sapwood	Heartwood		
<i>n</i> -Hexane	1.31 ± 0.47 bcd	2.39 ± 0.16 f	1.29 ± 0.11 bcd	1.42 ± 0.13 cd		
Metanol	6.42 ± 0.51 ef	7.81 ± 1.33 f	5.19 ± 2.38 cde	7.55 ± 3.96 f		
Hot water	3.20 ± 0.35 de	3.52 ± 0.62 e	2.42 ± 0.86 bcd	2.69 ± 1.26 cde		
Total	10.9 ± 0.41 e	13.7 ± 1.80 f	8.90 ± 2.29 cde	11.7 ± 4.16 ef		

Note: Average of five trees ± the standard deviation; The same letters in the same row are not significantly different at $p < 5\%$ by Duncan's test.

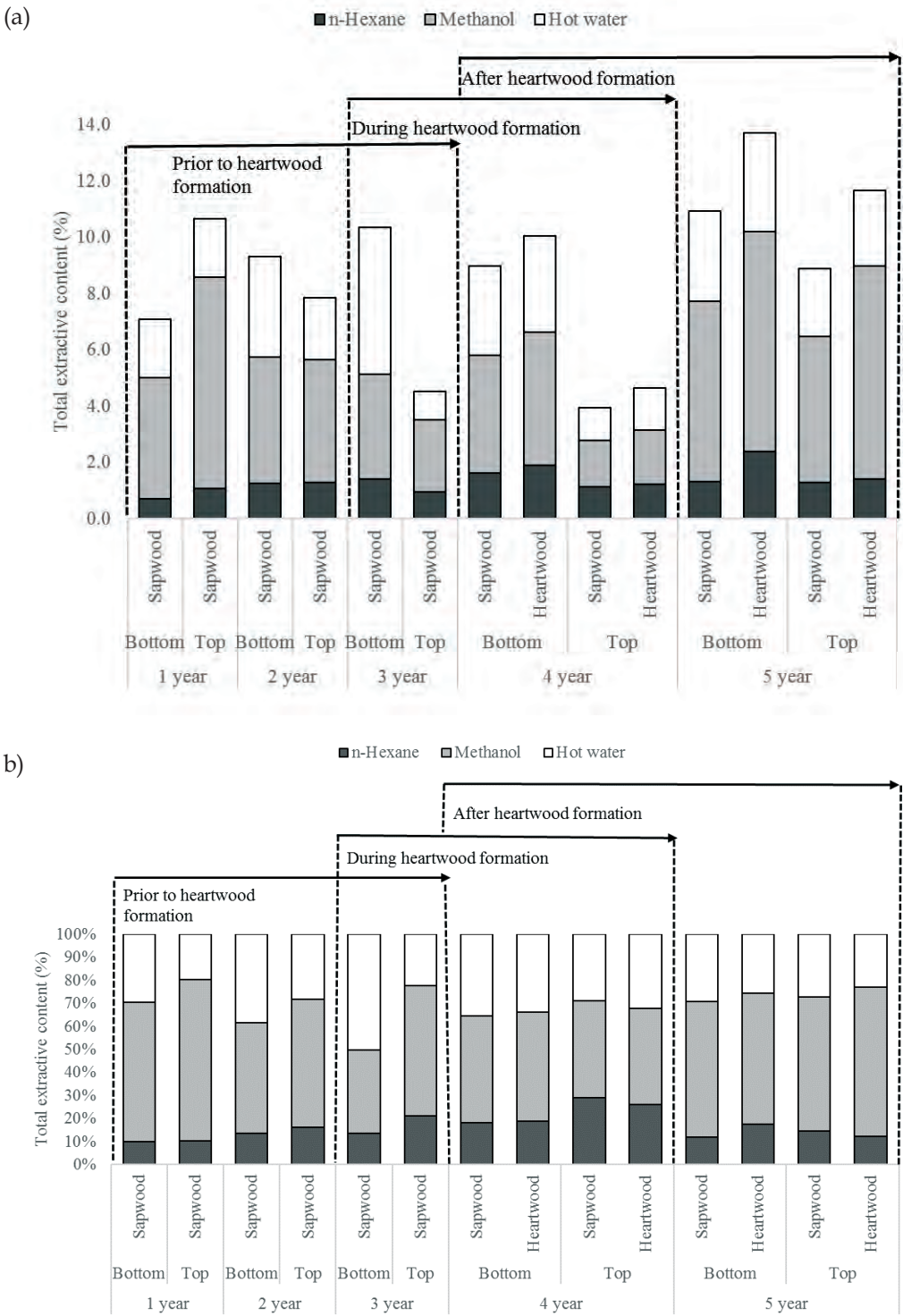


Figure 2. Total extractive content of *S. mahagoni* based on oven-dry wood mean (a) and extractive weight (b).

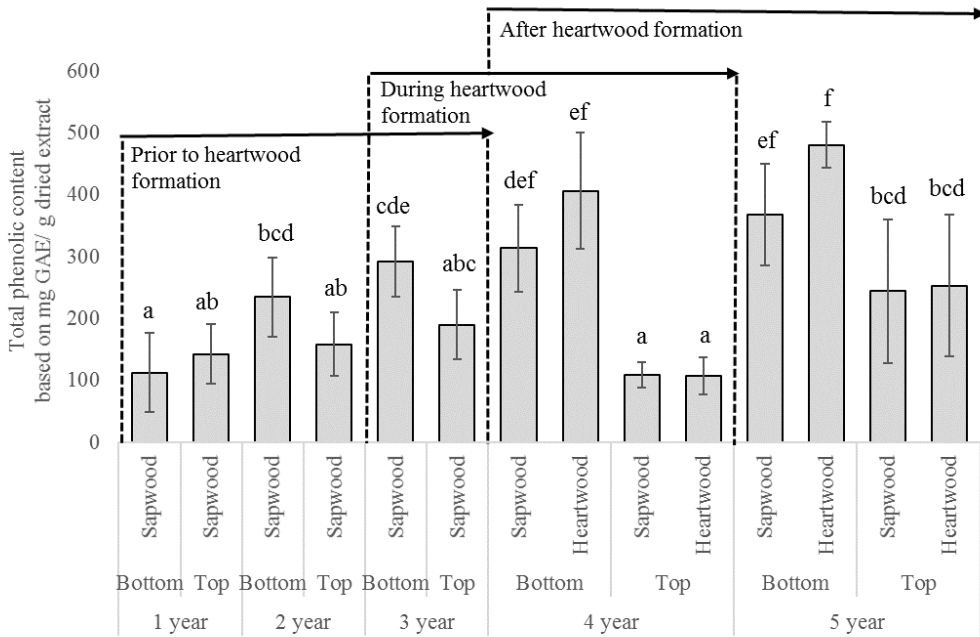


Figure 3. Total phenolic content based on mg GAE/g dried extract from *S.mahagoni* (means of five trees) on the interaction of tree age and axial position with error bar as standard deviation. The same letters on the histograms implied no significant differences ($p < 0.05$ by Duncan's test).

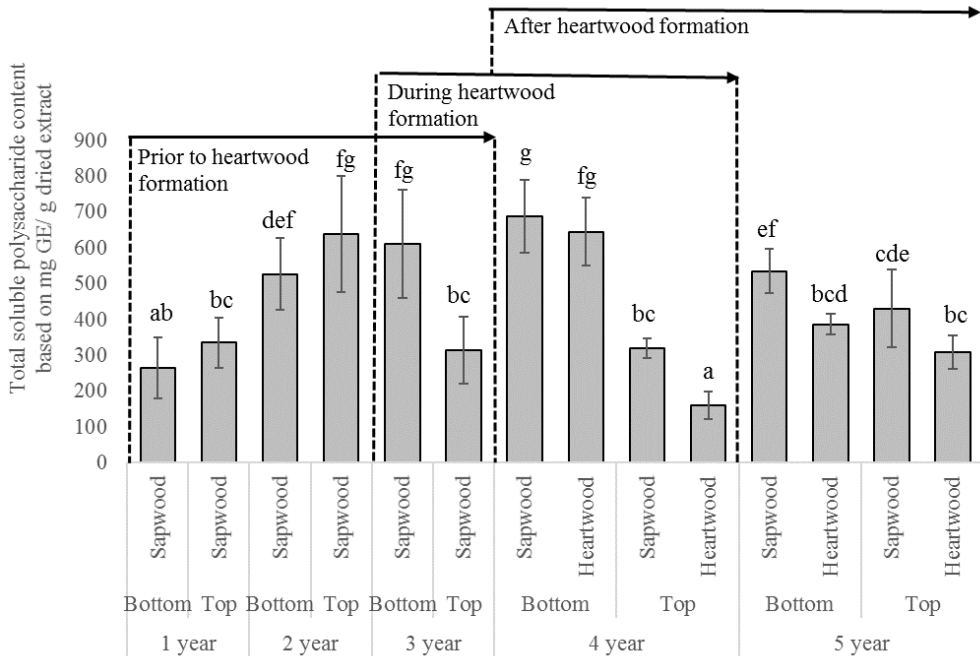


Figure 4. Total soluble polysaccharide content based on mg GE/g dried extract from *S.mahagoni* (means of five trees) on the interaction of tree age and axial position with error bar as standard deviation. The same letters on the histograms implied no significant differences ($p < 0.05$ by Duncan's test).

Figure 3 showed that the TPC in the methanol extracts ranged from 107.3 to 480.2 mg GAE/g dried extract. Furthermore, the amount of TSP ranged from 160 to 688 mg GE/ g dried extract with the mean values of 440.3 mg GE/ g dried extract (Figure 4).

Among and within tree variation

Extractive content

The ANOVA of extractive contents showed significant tree age and axial position interactions (Table 2). Prior to heartwood formation (1 to 3 years) (Table 3) generally the extractive content increased with the increasing tree age at the bottom of the tree, where the content of hexane and hot water extract increased significantly from the age of 1 to 2 and 1 to 3 years. Meanwhile, the methanol extract slightly decreased from the age of 2 to 3 years, after slightly increasing from the age of 1 to 2 years. Furthermore, the total extractive content increased significantly from the age of 1 to 3 years. At the top of the tree, hexane and hot water extract decreased significantly (hot water extract) at the age of 3 years, although slightly increased previously at the age of 1 to 2 years. Meanwhile, a significant decrease was found in the methanol extract (from tree age of 1 to 2 and 1 to 3 years) and in the total extractive content (from tree age of 1 to 3 years and 2 to 3 years).

From the age of 3 to 4 years in sapwood, the levels of hexane and methanol increased

with tree age at the base of the tree, while the reverse pattern was found in the amount of hot water extract and total extractives, where the hot water content decreased significantly. At the top part, a slight increase in the extractive content was found in the amount of hexane and hot water extract, while the content of methanol and total extractives were found to decrease. After the formation of heartwood (4 to 5 years), the content of methanol and hot water extract increased (except for hexane) with tree age in the bottom sapwood. The same pattern was shown in heartwood, except for the hot water extract which slightly increased. Further at the top of the tree, both in sapwood and heartwood, the extractive content of methanol and hot water increased significantly from the age of 4 to 5 years. A similar trend was shown by the total extractive content, where its level drastically increased with increasing tree age.

In the axial position, the extractive content generally descends from the bottom to the top of the tree before heartwood formation (Table 3, Figure 2). A significant reduction was found in the amount of hexane extract at the age of 3 years and hot water at the age of 2 and 3 years. The reverse trend was found at the age of 1 year, where the extractive content increased significantly in hexane and methanol extracts. Furthermore, a similar pattern was also shown in the total extractive content, where the levels decreased significantly at the age of 3 years and increased significantly at the age of 1 year.

Table 2. Two-way ANOVA analysis for extractive, TPC, and TSP contents of *S. mahagoni* wood.

Source of variation	df	Parameters					
		<i>n</i> -Hexane	Methanol	Hot water	Total Extractive	TPC	TSP
Tree age (T)	10	***	***	**	***	***	**
Axial (A)	1	***	n.s	***	***	***	***
T x A	10	***	**	***	***	**	***
Error	46						
Total	67						

df degrees of freedom;

n.s. not significant at 5% level, * P<0.05, **P<0.01, ***P<0.001.

In addition, from the age of 3 to 4 years, the content of extractive patterns was similar to before heartwood formation (except for methanol extract at the age of 3 years and hexane extract at the age of 4 years, which slightly decreased). After the formation of heartwood, at the age of 4 years both in sapwood and heartwood, the extractive content decreased significantly from the bottom to the top of the tree. The same trend was also shown at the age of 5 years, although the decrease was not significant (except for methanol extract). Furthermore, in the radial profiles, extractive content in the heartwood both at the base and at the upper part of the tree was higher than in the sapwood at the age of 4 years. The same pattern was also shown at the age of 5 years, where the content of hexane extract in heartwood was significantly larger compared to sapwood at the bottom and a similar trend was shown by the methanol extract at the top of the tree.

Total phenolic content (TPC)

There was a significant interaction between age and axial position in TPC in the ANOVA test (Table 2). Before heartwood formation (Figure 3), TPC increased with tree age. It increased significantly from the age of 1 to 2 years at the bottom of the tree. A similar trend was also found in the TPC at the age of 3 to 4 years at the bottom sapwood part, although TPC decreased at the top of the tree. However, a reverse pattern was found from the age of 4 to 5 years after heartwood formation, where the TPC increased significantly at the top sapwood part.

Axially, before heartwood formation (1 to 3 years), the TPC content decreased from the bottom to top part except for 1-year-old trees, which slightly increased from the base to the upper part of the trees. The same pattern was shown during heartwood formation. A significant decrease was found at the age of 4 years in sapwood. In addition, a similar trend was also shown after the formation of heart-

wood, where both in sapwood and heartwood (4 and 5 years old), the TPC content significantly decreased from the bottom to the top of the tree. Furthermore, in the radial variation, both at the age of 4 and 5 years, the difference in TPC between sapwood and heartwood was more visible at the bottom than at the top, where the TPC of heartwood was higher than that in the sapwood samples.

Total soluble polysaccharide (TSP)

The ANOVA of TSP showed significant tree age and axial position interactions (Table 2). Generally, the TSP level increased with the increasing tree age before the formation of heartwood (Figure 4), where both at the bottom and top of the tree, a significant increase was found from the age of 1 to 2 years. Meanwhile, at the top part, the amount of TSP from the age of 2 to 3 years decreased significantly. Furthermore, the TSP content slightly increased again from the age of 3 to 4 years both at the bottom and top of the tree in sapwood. Then, TSP at the bottom both in sapwood and heartwood decreased significantly from the age of 4 to 5 years after the formation of heartwood. In contrast, the amount of TSP increased significantly at the top heartwood of the tree.

In axial position, the TSP increased from the bottom to the top of the tree (except for the age of 3 years) before heartwood formation. TSP from the age of 3 to 4 years, with the axial pattern being the same as that of TPC, decreased drastically from the base sapwood to the upper sapwood. Furthermore, the TSP trend at the age of 4 and 5 years was the same as that of TPC at the same age where the TSP level dropped from the bottom to the top of the tree. Meanwhile in the radial variation, TSP in sapwood was higher than in heartwood. A significant difference was found at the age of 4 years at the top, and at 5 years at the base of the tree.

Discussion

Extractive content, total phenolic content, and total soluble polysaccharides

Theoretically, the non-polar solution (*n*-hexane) removed fats, waxes, resins, and sterols (Lukmandaru, 2011). Meanwhile, cold and hot water-soluble extractives dissolve mineral salts, sugars, tannins, phenolic compounds, dyes, pectins, free acids, and others (Fengel & Wegener, 1989; Han & Rowell, 1997). The extractive contents of this result were slightly lower compared to that of Batubara *et al.* (2009, 2010, 2012) and da Silva *et al.* (2013) who observed the values of methanol and hot water extractive in other *Swietenia*. The value obtained for *S. macrophylla* in Moto Grosso, Brazil was 5.93% to 7.97%. Meanwhile, 4.01% to 14.83% was obtained for *Swietenia* sp. wood in Samarinda, Indonesia. Taylor *et al.* (2008) obtained that the yield of toluene/ethanol (2:1), 95% ethanol, and hot water extract ranged from 11.8% to 28.1% of mahogany wood from Central and South America. Additionally, Jankowsky & Galvão (1979) reported that the amounts of alcohol-benzene and water-soluble extract in *S. macrophylla* wood were 7.03% and 8.64%, respectively. The observed yield of extractive of *S. mahagoni* was lower compared to the reported values because the samples were used from a young age.

In contrast, extractive levels of this study were higher compared to the content of petrol ether (0.8%), acetone (3.2%), methanol (3.4%), hot water (1.1%), and cold water (2.1%) (Rutiaga Quiñones *et al.*, 1998a). Gala *et al.* (2020) found that the extractive content of ethanol and hot water solution of *S. mahagoni* wood from Malang, Indonesia was 4.85% and 4.95%, respectively. In addition, the contribution of the three solvents in the total of extractive contents was similar to the results of Morais & Pereira (2012), where methanol contributed greatly to dissolve the extract. The proportions of particular components in wood

depend on the species, the climate, the time of year, the site conditions, the part of the tree, and the health condition (Bikovens *et al.*, 2013; Miranda *et al.*, 2017; Szczepkowski *et al.*, 2007; Zobel & Sprague, 1998). In addition, the composition and amount of extractives depend on the solvent used (e.g., ethanol-benzene mixture, ethanol, acetone, dichloromethane, and ethyl ether) (Lachowicz *et al.*, 2019).

TPC of this study was lower compared to previous research. Batubara *et al.* (2012) reported that TPC in *Swietenia* sp. was 673.8 mg GAE/ g. In *Swietenia* species, such as *S. macrophylla*, it was 6.81% (da Silva *et al.*, 2013). Furthermore, the same pattern was also shown by TSP, the levels of which were lower compared to the value reported by Rutiaga Quiñones *et al.* (1998b). They reported the content in *S. macrophylla* as 63.3%, dominated by the glucose component.

Among and within tree variation

In general, the extractive content increased with increasing tree age. The same pattern has been reported in previous studies in *Eucalyptus globulus* Labill., *Eucalyptus grandis* W. Hill, *Eucalyptus urophylla* S.T. Blake, and *Tectona grandis* L. f. (Miranda & Pereira, 2002; Lukmandaru, 2009; Santana *et al.*, 2012; Lestari *et al.*, 2016). The vascular cambium of older trees is more likely to divide and multiply into parenchyma cells, so that the formation of parenchyma cells increases. Therefore, the extractive content tends to rise (Hamidah *et al.*, 2009). In addition, a significant increase in the extractive content was also found after the formation of heartwood (4 to 5 years) at the top of the tree. This content increased due to the largest contribution from the extractive content of methanol and hot water which increased significantly after heartwood formation. This may be related to non-structural carbohydrate (NSC) and phenolic compounds, considering that methanol and hot water solutions are the best solvents to dissolve sugar and pheno-

lic components (Fengel & Wegener, 1989; van Putten *et al.*, 2014). This argument was supported by the significantly increased TSP and TPC content at the top of the tree. Piispanen & Saranpää (2001) reported that some TSP (sucrose and glucose content) are more abundant in the top parts of the crown, such as the stems, which are closer to photosynthesizing leaves, where the assimilation of CO₂ takes place. In addition, Magel *et al.* (1994) reported that the high amount of the main monosaccharide components in sapwood could be a high demand of NSC which were degraded for energy and the carbon skeleton needed for metabolic activities, such as cell respiration, photosynthesis and the formation of secondary metabolites, such as phenolics. In addition, the significant decrease in the TSP content after the formation of heartwood (4 to 5 years) at the base of the tree may be related to the formation of heartwood, where sugar is converted into phenolic components. This is indicated by the increased TPC content at the base both in sapwood and heartwood after the formation of heartwood.

However, in this study it was also found that the extractive content decreased from a certain tree age, namely in the hot water extract at the bottom and in the methanol extract at the top part from the age of 3 to 4 years. In addition, especially at the top of the tree from 1 to 3 years, the extractive content decreased significantly. This may be influenced by the certain proportion of sugar components which decreases with age at the top of the tree. Berrocal *et al.* (2004) reported that the sugar component, such as xylose, galactose, and arabinose in *Pinus radiata* D. Don decreased with tree age (1 to 15 years old). In addition, Rencoret *et al.* (2011) reported that the hot water extractive of *E. globulus* clone wood from Pontevedra, Spain, decreased with tree ages of 1 month, 18 months, and 9 years. Stolarski *et al.* (2011) observed in willow shoots that the hot water extract decreased from the age of 1, 2, and 3-year-old trees in

the species obtained from North of Poland. Miranda & Pereira (2002) reported that the total extractive content of *E. globulus* from Portugal decreased from 3 to 6 years old. In addition, a drastic decrease in methanol and TSP levels at the top of 3-year-old trees suggests that a lot of TSP was transferred from the top to bottom part in the process of heartwood formation in 4-year-old trees at the base. This finding was supported by the high extractive content of hot water at the bottom part of 3-year-old trees, where the hot water solvent is known as a good solvent of sugar components (van Putten *et al.*, 2014). Prayitno (1992) reported that the axial position in the bottom part has the highest extractive content.

In axial variation, the extractive content at the bottom part was greater compared to the top of the tree (except for 1-year-old trees). A similar trend was reported in previous work in other species, such as superior teakwood, *Styrax sumatrana* J.J. Sm., and *Eucalyptus globulus* (Iswanto *et al.*, 2019; Gominho *et al.*, 2015; Zulkahfi *et al.*, 2020). Gominho *et al.* (2001) and Caron *et al.* (2013) also found the high concentrations of extractives at the base of hardwood. The high extractive content at the base part may be due to the intense polymerization reaction during the aging process of the tree. Meanwhile, the high content at the top part is caused by the vicinity to the canopy as a place of photosynthesis (Lukmandaru *et al.*, 2021). Yeh *et al.* (2006) found that benzene alcohol extractives in the top part were higher compared to the bottom. This is because the living moiety on the top of a tree was higher. Meanwhile, TPC and TSP generally decreased from the bottom to the top of the tree (after 2 years of age). A similar pattern was also found by Neverova *et al.* (2013) in *Larix sibirica* Ledeb. However, a reverse trend was found in previous studies that TPC increased from the base to the top of the tree in other tropical species, such as superior teakwood aged 11 years and *Eucalyptus pellita* F. Muell. from natural forest (Arisandi *et al.*, 2019a; Zu-

Ikahfi *et al.*, 2020). This might be one of the characteristics of the species *S. mahagoni* compared to other tropical wood species in the period until the age of 5 years. Other reasons might be due to differences in tree age, species, and growth location (Hillis, 1987; Freire *et al.*, 2005).

In the radial profiles, the extractive content in heartwood was higher than in the sapwood part. A similar trend has been reported in other species, such as *Tectona grandis*, *Acacia melanoxylon* R. Br., *Eucalyptus pellita*, and *Eucalyptus globulus* (Miranda *et al.*, 2006; Lourenço *et al.*, 2008; Lukmandaru, 2011; Morais & Pereira, 2012; Arisandi *et al.*, 2019a, 2020). Studies on the pigments of the heartwood parts were almost never carried out in mahogany (Yazaki, 2015). Rastogi & Mehrota (1993) reported that the heartwood of *S. mahagoni* contained cycloswietenol, lupleol, hederagenin benzoate, cycloartenol, and -sitossterol. In addition, the hot water extractive from mahogany bark exhibits a strong red color, and is usually used as a tannin for leather tanning. Bark extracts found in hot water solution were (+)-catechin, (-)-epicatechin and a new amorphous compound, switemacrophyllanin (Falah *et al.*, 2008).

The high levels of extractives in heartwood are related to the heartwood formation process, where the accumulation of extractives is a key feature in the formation of heartwood (Hillis, 1971). Umezawa (2000) reported that heartwood is richer in polyphenols and resin acids (diterpenes). In heartwood (with a lower pH condition), the most abundant of the soluble sugars (xylose, mannose, and arabinose) were derived from hydrolyses. This is supported by the decreasing TSP content from sapwood to heartwood as well as a reverse pattern found in TPC. Thus, the decline in TSP from sapwood to heartwood indicates that the content of certain TSP compounds is metabolized at the sapwood-heartwood boundary (Niamké *et al.*, 2011). Hillis & Hasegawa (1963) postulated that NSC becomes the main source of carbon frame-

work for synthesizing extractives of heartwood (phenolic compounds).

In the transition zone, the amount of starch can be hydrolyzed to glucose by hydrolyzing enzymes (amylglycosidases) (Magel *et al.*, 1997, 2001). In other species, like teak, key NSC catabolism enzymes, such as succinate dehydrogenase and glucose-6-phosphate are involved in reducing NSC and accumulate in the inner sapwood and in the transition zone (Data & Kumar 1987). Niamké *et al.* (2010) reported that the depletion of NSC from sapwood to heartwood suggests that they may be precursors for the synthesis of wood extractives (phenolic compounds) during heartwood formation. Burtin *et al.* (1998) reported that hydrojuglone glucoside was the precursor of juglone, and their polymers in heartwood *Juglans nigra* L., whereas dihydrorobinetin was the precursor of robinetin synthesis in *Robinia pseudoacacia* L. (Magel *et al.*, 1994). However, this study was limited in the general composition of TSP and TPC. Therefore, further research is needed to investigate the changes in sugar and phenolic compositions in the component level, in order to investigate what compounds play an important role in the heartwood formation of *S. mahagoni*.

Conclusions

Based on the ANOVA analysis, there was a significant interaction between tree age and axial position in all parameters. The distribution of the extractive content, TPC, and TSP increased with increasing tree age, except for a certain age before heartwood formation (3 years) at the top of the tree (for methanol and TSP content). The highest increase was found after heartwood formation (4 to 5 years) in hexane, methanol, and the total extractive fractions. On the other hand, the three parameters generally have a similar pattern both in axial and radial variation (except TSP and TPC), where in axial position, its distribution

decreased from the bottom to top of the tree, particularly after the age of 2 years. This might be one of the characteristics of *S. mahagoni* species until the age of 5 years compared to other tropical wood species. Meanwhile, in radial profiles, extractive and TPC levels were higher in heartwood, while a reverse trend was found in TSP. This finding indicated that this occurs due to the process of heartwood formation in *S. mahagoni*. In addition, a drastic decrease in the methanol extract and TSP content at the top of the tree at 3 years, indicated that a lot of the TSP content was transferred from the upper to the lower part of the tree for the process of heartwood formation at the beginning of the base at 4 years of age.

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