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Tannins in mangrove plants in Segara Anakan Lagoon, Central Java, Indonesia

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Abstract. *Hilmi E, Sari LK, Siregar AS, Sulistyo I, Mahdiana A, Junaedi T, Muslih, Pertiwi RPC, Samudra SR, Prayogo NA. 2021. Tannins in mangrove plants in Segara Anakan Lagoon, Central Java, Indonesia. Biodiversitas 22: 3508-3516.* Mangrove tannin is polyphenol compound and extractive matter in mangrove vegetation. Mangrove tannins have the potentials to support paint industry, animal feed, tanners and wood adhesives industry. This research is aimed to investigate the mangrove species in Segara Anakan, Central Java, Indonesia that produce tannins, and to analyze the distribution and contents of tannin in the part of mangrove plant. A total of 342 samples from 19 mangrove species was collected from 37 sampling points in Segara Anakan. UV-Visible Spectrophotometry was used to analyze the tannin content in the samples. The results showed that the tannins contained in bark and stems were higher (66.6%) than those in leaves (33.4%). Mangrove vegetation can be classified into five classes in terms of tannin percentage with *Heritiera littoralis, Nypa fruticans,* and *Rhizophora mucronata* had highest tannin percentage. The clustering analysis of mangrove tannin showed that *Ceriops decandra-Ceriops tagal, Acacia auriculiformis-Sonneratia alba, Sonneratia caseolaris-Xylocarpus granatum,* and *Avicennia marina-Rhizophora apiculata* had high similarity of tannin percentage. The distribution of tannins in mangrove species is as follow: 0.59-10.14 kg trees^{-1} (bark and stem of mangrove diameter > 10 cm) and 0.20-3.74 kg trees^{-1} (leaves of mangrove diameter > 10 cm) until 8.84-158.96 kg trees^{-1} (bark and stem of mangrove diameter > 40 cm) and 4.60-91.65 kg trees^{-1} (leaves of mangrove diameter > 40 cm). *R. mucronata* and *R. apiculata* had the highest total tannin content, ranged between 386.60-460.38 kg trees^{-1}.

Keywords: Clustering, distribution, mangrove tannin

INTRODUCTION

Mangrove is vegetation of intertidal zones between marine and terrestrial ecosystems (Bellotti et al. 2012; Rahim et al. 2008; Wang et al. 2018; Yang et al. 2018) has many ecosystem services such as carbon sequestration (Hilmi et al. 2017; Hilmi et al. 2019b), reducing coastal disasters (Hilmi 2018; Suhendra et al. 2018), water conservation, pollution reduction (Hilmi, Sari, and Setijanto, 2019; Hilmi et al. 2017; Sari et al. 2016; Su et al. 2012; Syakti et al. 2013a,b), bioremediation (Hidayati et al. 2011; Syakti et al. 2013a) and provide tannin as non-timber forest product. As the non-timber services, tannin has an important value similar with function to reduce coastal disaster (Hilmi 2018), heavy metal pollution (Syakti et al. 2013a; Hilmi et al. 2017), support REDD program (Hilmi et al. 2017) and activity of conservation and economic carbons (Canu et al. 2015; Avelar et al. 2017; Hilmi et al. 2017; Hilmi et al. 2019). Tannin is a water-soluble substance and extractive matter which is arranged by an aggregation of complex organic compounds (Tahir et al. 2019) and secondary metabolites, such as hydroxyl groups, which are made up of benzene rings, acid classes, and organic sugars (Martins et al. 2020). Tannin also has strong aqueous solution properties (Tahir et al. 2019), which are possibly degraded by the Tannase enzyme. Because of its toxic substances, tannin may act as a pathogen for other plants (Sharma 2019). Tannin also has correlation with the ability of plant protection, production of plants' secondary metabolites and defensive molecules, barriers against the phytopathogen invasions (El-Ashmawy et al. 2016; Sharma 2019; Martins et al. 2020), to protect plants against insects, pathogens, and herbivore animals regarding their defense activities (Sharma 2019). The other functions, tannins are also used to produce adsorbents of heavy metal ions (Martins et al. 2020; Matsutani and Nagai 2013; Oo et al. 2009), to produce tannase enzymes used to adsorb copper, lead, and other industrial products (Rahim et al. 2008; Oo et al. 2009; Zhang and Li 2017).

Basically, tannin produced by mangroves is classified into hydrolyzed and condensed tannins. Diluted acid concentrations heat hydrolyzed tannins to produce gallic and ellagic acids and more mixtures (Negri and Tabach 2013), while the condensed tannins are polymerized to deliver water-insoluble phlobaphene (Martins et al. 2020; Tahir et al. 2019). However, condensed tannin is also defined as flavonoid polymers and oligomers (Pan et al. 2019). Tannin is also divided into polyphenols, non-tannins, and flavonoids (El-Ashmawy et al. 2016; Jinhui et al. 2019; Pan et al. 2019). Also, it is used to assess antioxidant and antimicrobial activities (Medini et al. 2014; Zhang and Li 2017). Tannin extracted from mangrove is used to produce phloroglucinol nucleophiles, polar solvents, aqueous acetone and ethanol, water (Tan and Kassim 2011), phenolic resin (Hernes et al. 2001; Antoine et al. 2004; Lei et al. 2019; Pan et al. 2019; Liu et al. 2020), and phytopathogen (Erickson et al. 2012; Sharma 2019).

In mangroves, tannins are produced by many species, such as *Caesalpinia spinosa* (Bellotti et al. 2012), *Rhizophora apiculata* (Oo et al. 2009; Rahim et al. 2008a), *Rhizophora mucronata* (Tahir et al. 2019), *Hibiscus rosa-sinensis* and *Senna bicapsularis* (Mak et al. 2013), and *Kandelia obovat*a (Zhou et al. 2012). The tannins in mangrove are extracted from wasted barks (Oo et al. 2009), bark extracts (Tahir et al. 2019), leaves (Hernes et al. 2001), root and skin (Rahim et al. 2008a; Rahim et al. 2008), and other parts of plant (Mostafa et al. 2018), with concentrations between 107.39 mg/g-157.57 mg/g, (*Anadenanthera colubrina)* and 68.96 mg/g-76.55 mg/g (*Lafoensia replicata*) (Monteiro et al. 2014).

This research is aimed to investigate the mangrove species in Segara Anakan, Central Java, Indonesia that produce tannins, and to analyze the distribution and contents of tannin in the part of mangrove plant. While there are several studies on a similar topic, this research adds a new context of study area, i.e., Segara Anakan which is recognized as an important mangrove ecosystem remaining in Java Island.

MATERIALS AND METHODS

Study area

This research was conducted in Segara Anakan Lagoon, Cilacap District, Central Java, Indonesia (Figure 1). The Lagoon is influenced by the existence of several rivers including Donan, Sapuregel, Kembang Kuning, Citanduy, Cimeneng, and Cikonde (Hilmi et al. 2017; Sari et al. 2016).

The data were collected from 37 sampling site points (Hilmi et al. 2019) (Table 1, Figure 1). The research stations in West Segara Anakan were Sungai Ujung Gagak, Sungai Lorogan, Sungai Majingklak, Sungai Mauara Cawitali, Sungai Kebuyutan, Sungai Batu Macan, Sungai Jongor, Sungai Muara Legok, Sungai Kayu Mati, Sungai Langkap, Sungai Karang Braja, Sungai Klaces, Sungai Inti Ujung Gagak, Sungai Muara Bagian, Sungai Muara Masigitsela, Sungai Pertigaan Ujung Alang, Sungai Ujung Alang, Sungai Dermaga Ujung Alang, Sungai Kali Semak, Sungai Pertigaan Sudiro. While the research stations in East Segara Anakan were Kali Panas 1, Kali Panas 2, Kali Panas 3, Kali Panas 4, Kali Panas 5, Donan 1, Donan 2, Donan 3, Donan 4, Donan 5, Donan 6 (Sleko), Pelawangan Timur, Sapuregel 1, Sapuregel 2, Sapuregel 3, Kembang Kuning 1, and Kembang Kuning 2.

Samples collection procedure

Data collection was conducted through two stages: (i) *Vegetation samples.* Vegetation samples from mangrove stems, barks, and leaves were collected and stored in plastic materials used to extract and analyze the percent and weight of tannins (Rahim et al. 2008; Abdullah and Lee 2017; Hilmi et al. 2017). After the collection, vegetation samples were sliced in tiny proportions to ease the process of extraction. In the Segara Anakan Lagoon, the mangrove samples were collected from 19 dominant species with three replicates of each species and three replicates of stems, barks, and leaves (totaling 342 samples). (ii) *Sample extraction.* Sample extraction was conducted by the maceration method. As many as 700 g of mangrove powder in the macerator was added with 2 L of 70% ethanol for 24 hours and then evaporated using a rotary evaporator. While 10 g of leaves were blended with 25 mL of 70% ethanol, filtered with 25 ml of measuring liquid, and further added with 70% C_2H_5OH (ethanol). After 10 mins, 5 mL of samples were obtained and diluted with 25 mL of 70% ethanol (Antoine et al. 2004; Katwa et al. 1981).

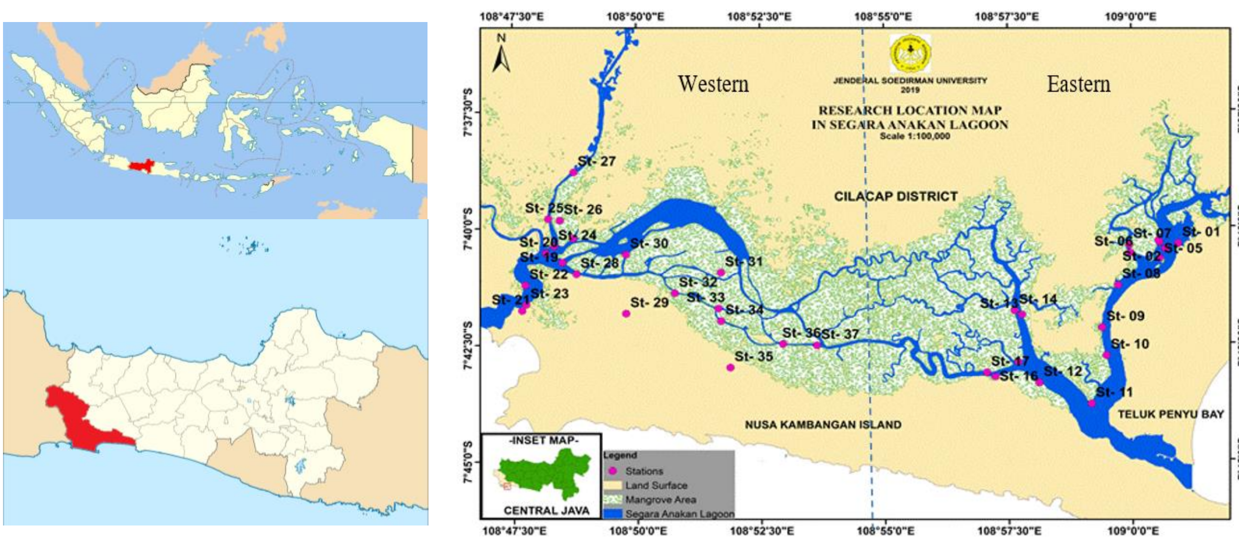

Figure 1. Map of the study area in Segara Anakan Lagoon, Cilacap District, Central Java, Indonesia

Table 1. List of research stations to collect mangrove samples in Segara Anakan Lagoon, Cilacap District, Central Java, Indonesia

Stations	Coordinate	
	Latitude (S)	Longitude (E)
West Segara Anakan		
Sungai Ujung Gagak	07◦40'13"	108◦48'43"
Sungai Lorogan	07◦40'44"	108◦48'30"
Sungai Majingklak	07◦40'32"	108◦48'1"
Sungai Mauara Cawitali	07◦41'46"	108◦47'41"
Sungai Kebuyutan	07◦41'13"	108◦47'45"
Sungai Batu Macan	07◦41'38"	108◦47'46"
Sungai Jongor	07◦40'23"	108◦48'20"
Sungai Muara Legok	07◦39'48"	108◦48'13"
Sungai Kayu Mati	07◦39'5"	108◦48'27"
Sungai Langkap	07◦38'48"	108◦48'44"
Sungai Karang Braja	07◦40'59"	108◦48'47"
Sungai Klaces	07◦41'5"	108◦49'47"
Sungai Inti Ujung Gagak	07◦40'34"	108◦49'47"
Sungai Muara Bagian	07◦40'58"	108◦51'42"
Sungai Muara Masigitsela	07◦41'24"	108◦50'46"
Sungai Pertigaan Ujung Alang	07◦41'44	108◦51'39"
Sungai Ujung Alang	07◦42'0"	108◦51'42"
Sungai Dermaga Ujung Alang	07◦42'6"	108◦51'53"
Sungai Kali Semak	07◦42'30"	108◦52'57"
Sungai Pertigaan Sudiro	07◦42'32"	108◦53'38"
East Segara Anakan		
Kali Panas 1	07^0 40’ 22,17”	109^0 0’ 56,36”
Kali Panas 2	07^0 40’ 28,91”	109^0 0’ 40,57”
Kali Panas 3	07^0 40’ 20,60”	109^0 0’ 33,62”
Kali Panas 4	07^0 40’ 18,26”	109^0 0’ 32,52”
Kali Panas 5	07^0 40’ 41,12”	109^0 0’ 33,98”
Donan 1	07^0 40’ 33,98”	108^0 59’ 58,10”
Donan 2	07^0 40’ 23,79”	108^0 59’ 56,90”
Donan 3	07^0 41’ 15,49”	108^0 59’ 43,22”
Donan 4	07^0 42’ 10,17”	108^0 59’ 23,75”
Donan 5	07^0 42’ 46,06”	108^0 59’ 29,10”
Donan 6 (Sleko)	07^0 43’ 48,07”	108^0 59’ 10,78”
Pelawangan Timur	07^0 43’ 20,95”	108^0 58’ 07,45”
Sapuregel 1	07^0 41’ 53,33”	108^0 57’ 46,71”
Sapuregel 2	07^0 41’ 47,97”	108^0 57’ 37,81”
Sapuregel 3	07^0 42’ 54,20”	108^0 57’ 42,07”
Kembang Kuning1	07^0 43’ 12,88”	108^0 57’ 14,24”
Kembang Kuning2	07^0 43’ 07,52”	108^0 57’ 03,97”

Analysis of tannin content procedure

The percent and weight of mangrove tannin were measured and calculated using UV-Visible Spectrophotometry with low-grade elements to determine the compound level percentage as a chromosphere structure. The percentage of mangrove tannin was measured by the absorbance method with the largest wavelength analysis of UV spectrophotometer. The UV-visible spectrophotometer was also used to calculate absorbed and reflected lights by calibrating the relative curve formula among the concentration series for the low-grade element analysis. Also, the UV method used the optical density principles of benzene chromosphere groups, which were related to the phenolic and tannin solutions at 280 nm (Antoine et al. 2004; Katwa et al. 1981).

Data analysis

Clustering tannin content

Clustering tannin content across mangrove species used euclidian distance analysis (Ludwig and Renold 1988) using following methods:

Stage 1. Calculation of Euclidian distance (ED) between species-i and species-j

$$ED_{jk} = \sqrt{\sum_{i=1}^{s}(xij - xik)^2}$$

Stage 2. Calculation distance between Euclidian among mangrove species

$$D\ (j,k)h = \propto_1 D(j,h) + \propto_2 D(k,h) + \ \beta\ D(j,k)$$

Stations	2	3	4	...	n
1	ED12	ED13	ED$_{14}$		
2		ED23	ED$_{24}$		
3			ED$_{34}$		
			...		
N			ED$_{n4}$		

Note: Ed*jk* is Euclidean Distance, i is species, X*ij* is density in station-*j* , X*ik* is density in station-*k* , D is Distance, α1 is 0.625, α2 is 0.625 and β is-0.25

Distribution of tannin across plant parts

The mangrove tannin distribution was analyzed used table stock analysis (initial stock, minimum stock, maximum stock and average stock). This analysis was used to describe distribution of mangrove tannins across plant parts for each mangrove species.

Distribution of tannin across landscape

The distribution of tanning across landscape was constructed by the distribution of its percent and weight from mangrove species. The landscaping of tannin showed the distribution of mangrove species in this ecosystem, following the characteristic environment, site, and potentials, as the main variable of this landscape. This landscaping aimed to develop the species zonation using tannin as an ecosystem service of mangrove ecosystem.

RESULTS AND DISCUSSION

Tannins contained in mangrove samples

Mangrove tannins were contained in the leaves, barks, and stems (samples total are 342 samples from 19 mangrove species with three tree replicates/species). The content of the tannins is shown in Table 2. The tannin in mangrove stems and barks was 253.6 mg g-1 or contributed to 67% total tannins in the samples while the tannin in mangrove leaves was 127.2 mg g-1 or 33% of the samples (Table 2).

As shown in Table 2, the barks and stems consisted a higher tannins than those in the leaves. Matsutani and Nagai (2013) found that tannin contents in the leaves of nine mangrove species ranged from 14.56 to 40.11% (dry wt). This result suggests that bark and stem of mangrove vegetation are main sources of tannin, which is similar to the results of research by Abdullah and Lee (2017), Rahim et al. (2008a), and Rahim et al. (2008). Basically, mangrove tannins were made up of chemical compounds of tannin-B and A-ring phenolic carbons (Hernes et al. 2001). Rahim et al. (2008) also discussed that the potential extractive matter of *Rhizophora* spp. stem had depolymerization activity of nucleophile phloroglucinol in order to develop the ethanol extractive agent. Furthermore, Tan and Kassim (2011) also analyzed stems and barks of *R. apiculata* to produce potential tannins, isopropyl proteins, and alcohol.

The higher tannin content in barks and stems than in the leaves of mangroves because bark and stem have cellulose, hemicellulose, lignin and extractive matters (Li et al. 2015; Patay et al. 2016; Sharma 2019; Martins et al. 2020). The potential tannin from barks and stems ranged 198.7 to 291.8 mg/g, while the leaves had tannin between 97.8 and 145.6 mg/g.

Composition of tannin in mangrove species

The composition of tannin in each mangrove species is shown in Table 3. The composition of tannin percentage shows the potential of extractive matters (including tannin) of mangrove species which correlates with the photosynthesis metabolism.

Table 2. Tannin contents in the samples of stems, barks and leaves of mangrove in Segara Anakan Lagoon, Cilacap, Indonesia

Part of mangrove	Tannin weight (mg/g)	Percentage of tannins (%)
Leaves (171 samples)	127.2	33.40
Barks and stems (171 samples)	253.6	66.60
Average	215.7	

Table 3. The composition of tannin in mangrove species in Segara Anakan Lagoon, Cilacap, Indonesia

Mangrove species	Tannin percentage (%)			
	Barks and stems		Leaves	
	Average	Sd	Average	Sd
Acacia auriculifromis	26.73	2.24	12.89	1.12
Acanthus ilicifolius	21.36	1.65	11.26	1.01
Aegiceras floridum	26.15	2.35	11.93	1.11
Avicennia alba	24.66	2.14	13.30	1.21
Bruguiera gymnorrhiza	19.87	1.15	13.45	1.2
Ceriops decandra	25.80	2.34	11.37	1.04
Ceriops tagal	25.73	2.23	11.27	1.01
Exoecaria agallocha	27.18	3.21	12.08	1.22
Heritiera littoralis	29.18	3.22	12.88	1.21
Hibiscus tiliaceus	26.17	2.05	9.78	0.98
Melaleuca leucadendron	23.37	2.11	11.26	1.12
Nypa fruticans	28.12	3.01	14.56	1.47
Rhizophora apiculata	25.30	2.05	13.76	1.38
Rhizophora mucronata	25.65	2.10	14.50	1.5
Rhizophora stylosa	23.40	1.54	12.67	1.28
Sonneratia alba	26.80	2.21	13.15	1.3
Sonneratia caseolaris	26.17	2.22	13.05	1.3
Xylocarpus granatum	24.36	2.01	12.08	1.21
Xylocarpus moluccensis	26.64	2.34	12.10	1.22

The data in Table 3 shows that *Xylocarpus moluccensis, Exoecaria agallocha, Nypa fruticans,* and *Heritiera littoralis* are the mangrove species with the highest tannin contents. The percentage of mangrove tannin ranged between 9.78-14.56% in leaves, and 19.87-29.18% in stem and barks. This result implies species' capability to produce tannin as extractive matter of mangrove vegetation (Rahim et al. 2008a; Hilmi et al. 2017).

Mangrove tannins are complex mixtures of biodegradable polyphenolic, non-toxic, and extracted compounds. The mangrove compound has two different classes of polyphenolic components including the condensed and hydrolyzed tannins (Nardeli et al. 2019). Also, the percentage of mangrove tannins has correlation with carbon content and organic compound, which are arranged by cellulose (Hilmi et al. 2017), hemicellulose, and extractive matter (Duncanson et al. 2017; Hilmi et al. 2019a).

Table 3 also show that the percentage of mangrove tannin is within the range of 7.2-14.3% (leaves) and 14.2-29.3% (stem and bark). The table also shows that many species had mangrove tannin percentage > 20% such as *H. littoralis*, *N. fruticans, Excoecaria agallocha, Aegiceras floridum, Ceriops decandra, Ceriops tagal, R. mucronata, R. apiculata, Sonneratia alba, S. caseolaris,* and *Xylocarpus moluccensis.* Matsutani and Nagai (2013) and Hilmi et al. (2015) state that the potential of phenolic compounds, such as mangrove tannin, was produced by plant’s leaves, roots, woods, barks, fruits, and buds. Further, the abundance of mangrove tannins is composed by 20% of dry weight.

Classification of mangrove tannin percentage

The classification of mangrove tannin percentage used the frequency class analysis (Willard 2020) and is shown in Table 4. The table shows that mangroves in our study area had five classes of tannin percentage which were arranged by potentials of stems and barks (19-32 %) and leaves (8-17 %). *H. littoralis* had the highest percentage of bark and stem tannin, between 29-33%, while *R. mucronata* and *N. fruticans* had high percentage of leaves tannin with 14-17%. The percentage of mangrove tannin has correlation with productivity of phytopathogen, antibiotics, tanners, and wood adhesives (Hernes et al. 2001; Rahim et al. 2008; Martins et al. 2020).

The classification of mangrove tannin percentage indicates the differential composition of both hydrolyzable tannins and hydrolysis of gallotannins. The percentage of the mangrove compound further explained the presence of C-C chains of tannin compounds (Negri and Tabach 2013), composition of lignin, with the potential of antioxidant and antimicrobial agents, in order to protect the plant’s self-defense (Medini et al. 2014; Rahim et al. 2008; Rahim et al. 2008). Tannins provide many benefits for plants, such as protection against insects, pathogens, and herbivore animals (Cohen et al. 2009; Subandriyo and Setianingsih 2016; Mostafa et al. 2018; Sharma 2019).

Table 4. The classification of mangrove species in Segara Anakan Lagoon, Cilacap, Indonesia based on the percentage of tannin contained in plant parts.

Class of percentage of mangrove tannin (%)	Species
Bark and stem	
15.0-20.0	*Bruguiera gymnorrhiza*
	Acanthus ilicifolius
20.5-25.0	*Melaleuca leucadendron*
	Rhizophora stylosa
	Xylocarpus granatum
	Avicennia alba
25.5-30.0	*Rhizophora apiculata*
	Rhizophora mucronata
	Ceriops tagal
	Ceriops decandra
	Aegiceras floridum
	Hibiscus tiliaceus
	Sonneratia caseolaris
	Xylocarpus moluccensis
	Acacia auriculifromis
	Sonneratia alba
	Exoecaria agallocha
	Nypa fruticans
30.5-35.0	*Heritiera littoralis*
Leaves	
08.0-10.0	*Hibiscus tiliaceus*
10.5-12.0	*Acanthus ilicifolius*
	Melaleuca leucadendron
	Ceriops tagal
	Ceriops decandra
	Aegiceras floridum
	Exoecaria agallocha
	Xylocarpus granatum
	Xylocarpus moluccensis
12.5-14.0	*Rhizophora stylosa*
	Heritiera littoralis
	Acacia auriculifromis
	Sonneratia caseolaris
	Sonneratia alba
	Avicennia alba
	Bruguiera gymnorrhiza
	Rhizophora apiculata
14.5-17.0	*Rhizophora mucronata*
	Nypa fruticans

Rahim et al. (2008a) also mentioned that *Rhizophora* spp (*R. mucronata, R. mangle, and R. apiculata)* had the highest tannin composition due to the fact that they contain antioxidants, precipitate pectinase, amylase, lipase, protease, β-galactosidase, cellulose, and other macromolecules (Sharma 2019). They also contain the highest complex macromolecular polyphenolic and natural polymers, such as cellulose, hemicellulose, and lignin (Pan et al. 2019; Ray et al. 2012; Verheyden et al. 2005)

Total tannin content in mangrove plant

The total tannin content in mangrove plants ranged between 0.59-158.96 kg/trees (bark and stem) and 0.20-91.65 kg/trees (leaves) (Tables 5 and 6). *R. apiculata and R. mucronata* had the highest total tannin (158.96 kg/trees). The total tannin has correlation with the biomass of barks, stems, and leaves (Hilmi et al. 2019a; Taillardat et al. 2018), carbon content, organic compound, and extractive matter. The results showed in Tables 5 and 6 suggest that *R. apiculata, R. mucronata, S. alba, Xylocarpus moluccensis, Exoecaria agallocha, N. fruticans,* and *Heritiera littoralis* have the potential to supply the raw materials of tannin industry (Bello et al. 2020) Our data also showed that the total tannins in mangrove species bigger than those from black acacia, *Acacia mearnsii*, and other breeds which ranged between 25.57-97.89 mg g^{-1} (Li et al. 2015; Nardeli et al. 2019). Accordingly, Salar et al. (2017), also mentioned that the potential of condensed tannins obtained from cultivars ranged between 40.96-99.91 mgCE/100 g, with coffee fruit at 0.402-1.46% (Patay et al. 2016), and medicinal plants at 17.15-44.02% (Moura et al. 2016).

Table 6 shows that total tannin content in mangrove plants can be classified into five classes: Class 1, which is the largest total mangrove tannins and consists of *R. apiculata* and *R. mucronata*; Class 2, which consists of *S. alba, S. caseolaris, Xylocarpus granatum, Avicennia alba/marina, Acacia auriculifromis, Aegiceras floridum,* and *Xylocarpus moluccensis*; Class 3, which includes *Ceriops* spp., *Bruguiera gymnorrhiza,* and *Rhizophora stylosa*; Class 4, which consists of *Melaleuca leucadendron, Exoecaria agallocha, H. littoralis, Hibiscus tiliaceus,* and *Nypa fruticans*; and Class 5, which includes *Acanthus ilicifolius.* The mangrove tannin content in classes 1 to 3 are still bigger than *Anadenanthera falcata, Lafoensia replicata,* and *Hymenaea stigonocarpa* as found by a study (Monteiro et al. 2014).

The regression analysis showed that potential tannin (in kg) can be estimated using the following formula

$$\text{Potential tannin (kg)} = -67.9 + 5.87 \text{ diameter (cm)}$$

in which the regression has strong correlation with 0.70 and variance analysis as presented in Table 7. This result suggests that the potential tannin of mangrove species has high significant correlation with trees diameter. This is not surprising since mangrove diameter will influence potential of cellulose, hemicellulose, lignin and extractive matter (Li et al. 2015; Martins et al. 2020; Patay et al. 2016; Sharma 2019).

Cluster of mangrove tannins

Cluster of mangrove tannin shows specific and similar distances based on the percentage and weight of the compound (Ludwig and Renold 1988; Rachmatin 2014; Rachmawati 2019) (Figure 2). The data explained that *Ceriops decandra-C. tagal, Acacia auriculiformis-S. alba, S. caseolaris-X. granatum,* and *Avicennia marina-R. apiculata,* are in cluster of species having the highest similarities.

Furthermore, Figure 3 shows boxplot of the clustering of mangrove tannins. Based on the tannins contained, mangrove vegetation in Segara Anakan Lagoon has four clusters: Cluster 1, consists of *((A. auriculiformis, S. alba), S. caseolaris), (Acanthus ilicifolius, Xylocarpus* *moluccensis), Excoecaria agallocha)*; Cluster 2, consists of *(((C. decandra, C. tagal),(A. marina, R. apiculata), R. mucronata)*; and Cluster 3, consists of *((Hibiscus tiliaceus, (H. littoralis, N. fruticans))* and *Cluster 4*, dominated by *((Acanthus ilicifolius, (Melaleuca Leucadendron, (Rhizophora stylosa, X. granatum), Bruguiera gymnorrhiza).*

Table 5. Total tannin content in mangrove species in Segara Anakan Lagoon, Cilacap, Indonesia

Mangrove species	Range of tannin total (kg/trees)											
	10				20				30			
	Barks and stems		Leaves		Barks and stems		Leaves		Barks and stems		Leaves	
	Max	Min	Max	Min	Max	Min	Max	Min	Max	Min	Max	Min
Acacia auriculifromis	5.77	4.88	1.86	1.56	31.99	27.04	10.31	8.66	87.09	73.62	42.12	35.38
Acanthus ilicifolius	0.68	0.59	0.24	0.20	3.79	3.25	1.35	1.13	10.32	8.84	5.51	4.60
Aegiceras floridum	7.07	5.90	2.16	1.79	35.80	29.90	10.92	9.06	92.45	77.21	42.30	35.10
Avicennia alba	6.68	5.62	2.41	2.01	34.12	28.67	12.32	10.26	88.55	74.41	47.94	39.95
Bruguiera gymnorrhiza	4.79	4.27	2.23	1.86	23.75	21.15	11.04	9.23	60.60	53.97	42.23	35.31
Ceriops decandra	6.98	5.82	2.05	1.71	35.35	29.47	10.39	8.65	91.29	76.10	40.26	33.51
Ceriops tagal	6.94	5.83	2.03	1.70	35.12	29.52	10.28	8.59	90.70	76.23	39.84	33.28
Exoecaria agallocha	1.86	1.47	0.54	0.44	7.44	5.87	2.17	1.77	16.74	13.20	7.33	5.98
Heritiera littoralis	1.98	1.59	0.57	0.48	7.93	6.36	2.30	1.90	17.85	14.30	7.76	6.43
Hibiscus tiliaceus	1.73	1.48	0.44	0.36	6.91	5.90	1.76	1.44	15.54	13.29	5.93	4.85
Melaleuca leucadendron	1.56	1.30	0.51	0.41	6.24	5.20	2.02	1.65	14.03	11.71	6.82	5.59
Nypa fruticans	1.91	1.54	0.65	0.53	7.62	6.15	2.62	2.14	17.15	13.83	8.83	7.21
Rhizophora apiculata	10.14	8.62	3.74	3.06	54.28	46.15	20.03	16.38	144.81	123.10	80.16	65.55
Rhizophora mucronata	8.37	7.10	3.22	2.61	53.62	45.51	20.61	16.75	158.96	134.90	91.65	74.47
Rhizophora stylosa	3.82	3.35	1.43	1.16	18.82	16.50	7.02	5.73	47.82	41.92	26.75	21.84
Sonneratia alba	7.23	6.13	2.40	1.97	36.93	31.31	12.26	10.06	95.85	81.25	47.74	39.15
Sonneratia caseolaris	7.08	5.97	2.39	1.95	36.14	30.49	12.18	9.97	93.80	79.13	47.41	38.82
Xylocarpus granatum	5.07	4.29	1.70	1.39	30.50	25.85	10.25	8.38	87.18	73.89	43.94	35.94
Xylocarpus moluccensis	5.57	4.67	1.71	1.39	33.52	28.10	10.27	8.39	95.79	80.32	44.04	35.97

Table 6. Classification of mangrove species in Segara Anakan Lagoon, Cilacap, Indonesia based on total tannin content

Class	Species	Max. of tannin total (kg/trees)
1	*Rhizophora apiculata*	386.60-460.38
	Rhizophora mucronata	
2	*Xylocarpus moluccensis*	230.22-258.55
	Acacia auriculifromis	
	Aegiceras floridum	
	Avicennia alba	
	Sonneratia alba	
	Sonneratia caseolaris	
	Xylocarpus granatum	
3	*Bruguiera gymnorrhiza*	167.57-227.58
	Ceriops decandra	
	Ceriops tagal	
	Rhizophora stylosa	
4	*Exoecaria agallocha*	32.21-39.64
	Heritiera littoralis	
	Hibiscus tiliaceus	
	Melaleuca leucadendron	
	Nypa fruticans	
5	*Acanthus ilicifolius*	25.23-27.90

Table 7. Analysis of variance potential of regression analysis on the correlation between mangrove tannin (kg) and diameter (sm)

Source	DF	SS	MS	F	P
Regression	1	327523	327523	65.23	0.000
Residual Error	74	371559	5021		
Total	75	699082			

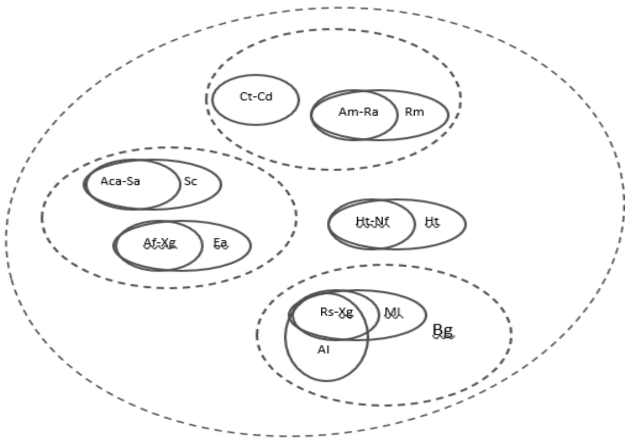

Figure 3. The boxplot of clustering analysis of mangrove species in Segara Anakan Lagoon, Cilacap, Indonesia based on tannin content

Basically, the clustering of mangrove tannins is influenced by nutrient absorption and environmental factors since one cluster has different characteristics from other groups (Rachmawati 2019; Widowati 2018). Using the density, Rachmawati (2019) and Widowati (2018) discussed that the mangrove ecosystem in Western and Eastern Segara Anakan had three clusters, respectively. Also, the clusterings of mangrove tannin and density are different because the compound among species is related to vegetation ability to produce extractive matter (i.e. tannin).

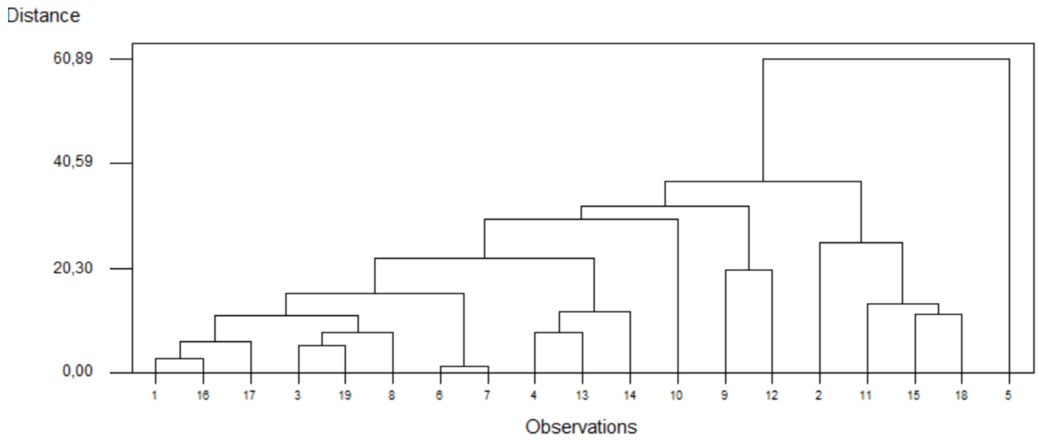

Figure 2. The dendrogram of clustering analysis of mangrove species in Segara Anakan Lagoon, Cilacap, Indonesia based on tannin content. Note: 1. *Aciacia auriculiformis,* 2. *Acanthus ilicifolius,* 3. *Aegiceras floridum,* 4. *Avicennia marina,* 5. *Bruguiera gymnorrhiza,* 6. *Ceriops decandra,* 7. *Ceriops tagal,* 8. *Excoecaria agallocha,* 9. *Heritiera littolaris,* 10. *Hibiscus tiliaceus,* 11. *Melaleuca leucadendron,* 12. *Nypa fruticans,* 13. *Rhizophora apiculata,* 14. *Rhizophora mucronata,* 15. *Rhizophora stylosa,* 16. *Sonneratia alba,* 17. *Sonneratia caseolaris,* 18. *Xylocarpus granatum,* 19. *Xylocarpus moluccensis*

In conclusion, tannins in mangrove vegetation in Segara Anakan Lagoon, Cilacap, Indonesia were contained in barks and stems (66.6%) and leaves (33.4%). The percentage of tannin composition ranged 21.36-29.18% in barks and stems and 9.78-14.50% in leaves. Mangrove species had varying tannin percentage with *H. littoralis, N. fruticans, Excoecaria agallocha, Xylocarpus moluccensis,* and *S. alba* having the highest tannin percentage. The total tannin content in mangrove plant were the largest in *R. apiculata* and *R. mucronata*. The clustering of mangrove tannin also showed that *C. decandra-C. tagal, A. auriculiformis-S. alba, S. caseolaris-X. granatum,* and *A. marina-R. apiculata* had high tannin percentage similarities with weight of the compound distributed at 0.59-10.14 kg/trees (bark and stem with diameter > 10 cm) and 0.20-3.74 kg/trees (leaves with diameter > 10 cm), till 8.84-158.96 kg/trees (bark and stem with diameter > 40 cm) and 4.60-91.65 kg/trees (leaves with diameter > 40 cm).

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