












Diversity and Species Composition of Bark and Ambrosia Beetles Captured Using Ethanol Baited Traps on Different Hosts in East Java, Indonesia

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Bark and ambrosia beetles are a diverse group that causes widespread mortality of deciduous and coniferous trees. The present study aimed to investigate the species compositions and richnesses of bark and ambrosia beetles in six species of plant hosts in East Java, Indonesia. Bark and ambrosia beetles were sampled using bottle traps baited with ethanol. Studies were conducted at two sites of monoculture and polyculture systems for each host plant species. At each site, 20 ethanol-baited traps were deployed on a linear transect along the forest. Six host tree species examined were used, namely *Tectona grandis* (Teak), *Syzygium aromaticum* (Clove), *Swietenia mahagoni* (Mahogany), *Pinus merkusii* (Sumatran Pine), *Paraserianthes falcataria* (Moluccan Albizia), and *Mangifera indica* (Mango). The data were analyzed using R software. A total of 4823 beetles were collected, representing 26 ambrosia beetle and eight bark beetle species. The abundance of bark and ambrosia beetles was significantly highest at the sites of *T. grandis* ($F = 13.88$, $P < 0.01$). *Xylosandrus crassiusculus* showed a strong attraction to the ethanol lure and was the dominant beetle species (50.65% of the total number of individuals). The Shannon-Wiener diversity index of all beetles captured in this study was the highest in the *S. mahogany* polyculture (2.28) and the lowest in the *T. grandis* polyculture (0.47). According to Bray-Curtis analysis, the *T. grandis* monoculture and *T. grandis* polyculture had a high similarity value of bark and ambrosia beetle species compositions (91% similar). There were no significant differences between two cultural systems of host plants in the compositions of bark and ambrosia beetle species (ANOSIM, $R = -0.1537$, $P = 0.961$).

Key words: Ambrosia beetles, Cultural system, Ethanol-baited, Species richness, NMDS.

BACKGROUND

Bark and ambrosia beetles are a diverse group that cause widespread mortality of deciduous and coniferous trees in forested and urban areas (Kühnholz et al. 2001; Oliver and Mannion 2001). The ambrosia beetle guild shows the lowest host specificity among whole herbivore guilds, and the bark beetle guild shows relatively high host specificity (Novotny et al. 2010). In general, ambrosia beetle females bore into the xylem and feed on symbiotic fungi, whereas bark beetles feed on the phloem of their host trees (Rabaglia et al. 2006).

Herbivorous insects have a degree of host specificity, from monophagous to polyphagous, and defensive capability, such as resistance to physical host defenses and its chemical compounds (Ødegaard et al. 2005; Agrawal 2007). The host specificity of herbivorous insects is one of the key predictors of patterns of biodiversity and has been widely used in the calculation of local species richness (Hamilton et al. 2010; Novotny et al. 2012). A recent study showed, in a tropical rainforest, that the model using host specificity is the best one for estimating species richness in herbivorous and nonherbivorous insect taxa (Basset et al. 2012).

In this study, six different plant species were used to estimate the species richness of bark and ambrosia beetles namely *Tectona grandis* (Teak), *Syzygium*

aromaticum (Clove), *Swietenia mahagoni* (Mahogany), *Pinus merkusii* (Sumatran Pine), *Paraserianthes falcataria* (Moluccan Albizia), and *Mangifera indica* (Mango) in East Java, Indonesia. The present study also used bottle traps baited with ethanol. Traps with chemical attractants are commonly an effective control used for studying population dynamics, estimating species richness, predicting outbreaks, and mass trapping to control pests (Burbano et al. 2012). Some studies have shown that a bottle trap baited with ethanol can be used to collect various species of bark and ambrosia beetle (Reding et al. 2011; Galko et al. 2014). The collected information is needed to optimize detection, monitoring, and management programs for pest species in different plant hosts. The research objectives were to investigate the species compositions and richnesses of bark and ambrosia beetles in six different plant hosts in East Java, Indonesia.

MATERIALS AND METHODS

Sampling Protocol

Bark and ambrosia beetles were sampled using bottle traps baited with ethanol (Fig. 1). Six plant species were used in this study: *Tectona grandis* (Teak), *Syzygium aromaticum* (Clove), *Swietenia mahagoni*

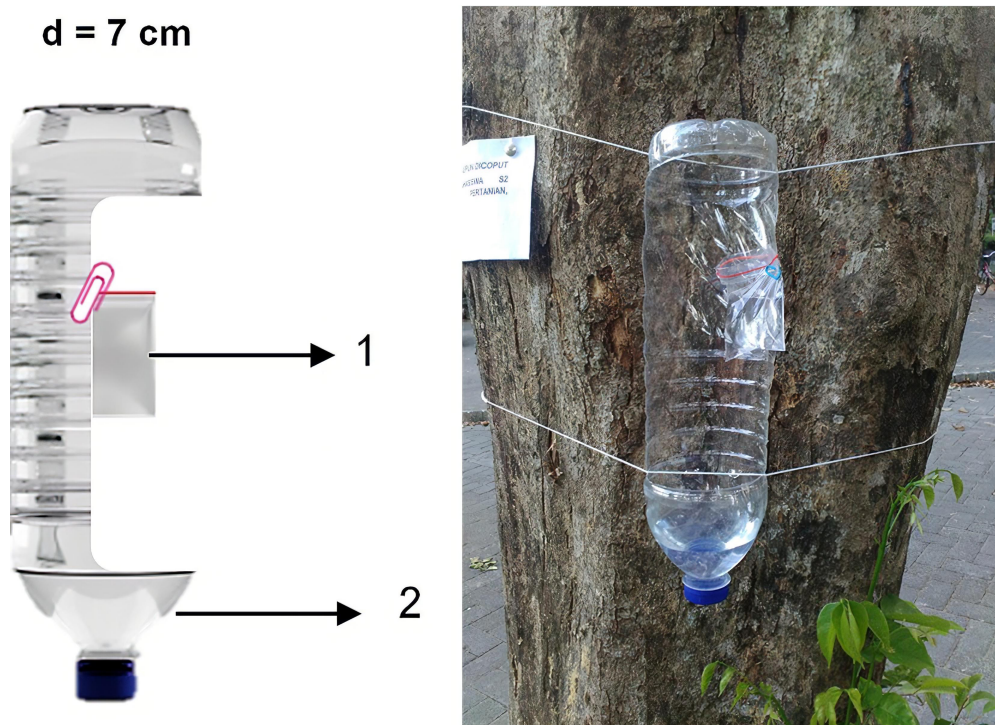


Fig. 1. Bottle trap baited with ethanol used in this study. 1: Plastic zipper bag containing 95% ethanol bait, and 2: Soap solution.

(Mahogany), *Pinus merkusii* (Sumatran Pine), *Paraserianthes falcata* (Moluccan Albizia), and *Mangifera indica* (Mango). Diameter at breast height of the selected sample trees ranged from 20 to 40 cm, except teak (> 40 cm). For each species, the study was divided into two sites based on cultural systems *i.e.*, monoculture and polyculture. At each site, 20 ethanol-baited traps were deployed on a linear transect along forest edges, separated by 20 m to reduce inter-trap interactions. The trap was made using a transparent bottle (volume = 1.5 L) with one window cut on the side and specimen container (containing soap solution) in the bellow part. It was baited with 95% ethanol. Each trap was ca. 7 cm in diameter (21.5 cm × 15 cm in size), attached to each trunk of sampled tree at ca. 1.5 m above the ground (Fig. 1). Bark and ambrosia beetles were collected eight times at 3-day intervals. This research was conducted in Malang, Blitar, Mojokerto, Batu, Kediri, and Pasuruan from March to April 2017

and 2018, and from December 2018 to January 2019 (Table 1 and Fig. 2). Based on figure 2, the selected location of each site depended on the suitability of the cultural system of each plant host species.

Preservation and Identification of Beetles

Bark and ambrosia beetles were preserved in 95% ethanol in small tubes. The bark and ambrosia beetle specimens were placed in the specimen bottle and labeled (date and site of observation). The identification of bark and ambrosia beetles was performed on the basis of morphological characters using an Olympus SZ51 stereo microscope. The identification of bark and ambrosia beetles was conducted at the Plant Pest Laboratory, Department of Plant Pests and Diseases, Faculty of Agriculture, Universitas Brawijaya. Bark and ambrosia beetles were identified using ambrosia beetle identification keys (Rabaglia et al. 2006; Wood 2007;

Table 1. Plant species, culture technique (monoculture or polyculture system), longitude, latitude, and altitude at each of the study sites

Host plant species	Family	Order	Cultural system	Study site
<i>Tectona grandis</i>	Lamiaceae	Lamiales	M	Malang District
			P	Malang District
<i>Syzygium aromaticum</i>	Myrtaceae	Myrtales	M	Malang District
			P	Blitar District
<i>Swietenia mahagoni</i>	Meliaceae	Sapindales	M	Mojokerto District
			P	Mojokerto District
<i>Pinus merkusii</i>	Pinaceae	Pinales	M	Mojokerto District
			P	Pasuruan District
<i>Paraserianthes falcata</i>	Fabaceae	Fabales	M	Kediri District
			P	Batu City
<i>Mangifera indica</i>	Anacardiaceae	Sapindales	M	Pasuruan District
			P	Pasuruan District

Host plant species	Beetle collection (Year)	Coordinates		Altitude (m.a.s.l.)
		Longitude	Latitude	
<i>Tectona grandis</i>	March to April 2018	90°90'92.8"	6°87'79.0"	396
	March to April 2018	90°91'10.6"	6°87'55.3"	392
<i>Syzygium aromaticum</i>	March to April 2017	112°42'50.9"	8°19'27.2"	360
	March to April 2017	112°24'43.25"	8°1'53.49"S	611
<i>Swietenia mahagoni</i>	March to April 2017	112°35'09.61"	7°37'19.17"	435
	March to April 2017	112°35'08.42"	7°37'16.87"	422
<i>Pinus merkusii</i>	March to April 2017	112°34'52.17"	7°39'38.40"	738
	March to April 2017	112°44'38"	7°49'19"	449
<i>Paraserianthes falcata</i>	March to April 2017	112°12'28.36"	70°47'10.66"	730
	March to April 2018	112°31'53"	7°53'25"	469
<i>Mangifera indica</i>	December 2018 to January 2019	112°52'51"	7°2'9"	37
	December 2018 to January 2019	112°45'59"	7°39'8"	36

Note: M = Monoculture and P = Polyculture.

Hulcr and Smith 2010).

Statistical Analysis

The populations of bark and ambrosia beetles in each host plant species were analyzed by using analysis of variance (ANOVA) ($P < 0.05$). Following significant results from ANOVA, the means were separated by Duncan's Multiple Range Test (DMRT) ($\alpha = 0.05\%$) using the R program version 3.3.3 with the vegan package Agricolae. The data were analyzed by using the Shannon-Wiener diversity index (H'), Evenness index (E), and Simpson Dominance Index ($1-D$) (Krebs 1999; Tarno et al. 2016). Bark and ambrosia beetle compositions were compared among the different host species based on the Bray-Curtis dissimilarity index and further analyzed using nonmetric multidimensional scaling (NMDS). All the data were analyzed by using the R program version 3.3.3 with the vegan package (Oksanen 2015; R Core Development Team 2019).

RESULTS

During the sampling period, a total of 4823 beetles, representing 26 species of ambrosia beetle and eight species of bark beetle, were collected (Table 2). Among the six host trees examined, the abundance of bark and ambrosia beetles was significantly higher in *T. grandis* ($F = 13.88$, $P < 0.01$) (Fig. 3). In all six different host plants investigated, the number of ambrosia beetles exceeded the number of bark beetles. The former accounted for 95% (4586 individuals) of the total number of collected individuals, and the

latter accounted for only 5% (237 individuals) of the total number of individuals. The ambrosia beetle *X. crassiuscullus* was the dominant species, with a total of 2443 individuals (50.65% of the total number of individuals collected) (Table 2).

The Shannon-Wiener diversity index of all beetles captured in this study was the greatest in the polyculture site of *S. mahogany* (2.28) and the lowest in the polyculture site of *T. grandis* (0.45) (Table 3). The species evenness index value for the polyculture site of *P. falcata* (0.89) was higher than that for other hosts, and the lowest value was for the polyculture site of *T. grandis* (0.23) (Table 3). The Simpson's dominant index value was the highest (0.83) in the polyculture site of *T. grandis*, and the lowest (0.13) in the polyculture site of *S. mahogany* (Table 3).

NMDS ordination analysis showed the compositions of bark and ambrosia beetle species in different host plants and culture systems (Fig. 3). NMDS ordination analysis showed no significant differences in the composition of bark and ambrosia beetle species between monoculture and polyculture (ANOSIM, $R = -0.1537$, $P = 0.961$) (Fig. 4). Based on Bray-Curtis analysis, the species compositions of bark and ambrosia beetles on *T. grandis* trees were 91% similar between the polyculture and monoculture system sites (Fig. 5).

DISCUSSION

Ambrosia and bark beetles in different plant host species in East Java were dominated by ambrosia beetle species, which constituted 95% of the total number of individuals and 5% of the total number of bark

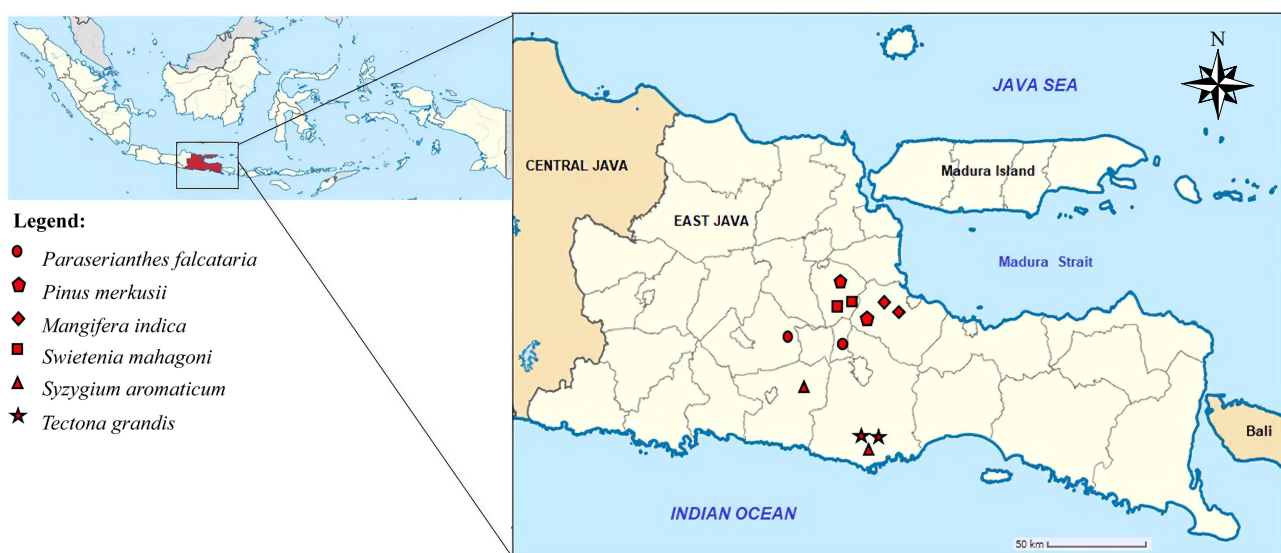


Fig. 2. Distribution of collection sites of each host plant species for bark and ambrosia beetles in East Java, Indonesia.

beetles in this study. Ambrosia beetles (Platypodidae and Scolytinae) also dominated in the teak forest and *Pterocarpus indicus* plant in the Malang District and Batu City, East Java (Tarno et al. 2014 2015; Setiawan et al. 2018). Five species of Platypodidae—namely

Crossotarsus sp., *Dinoplatypus* sp., *Dinoplatypus pallidus*, *Platypus solidus*, and *E. parallelus*—were identified in this research. One of the species, *E. parallelus*, had previously been reported in Malang and Batu City, East Java (Tarno et al. 2014). *Xylosandrus*

Table 2. Total number of bark and ambrosia beetles collected in bottle traps baited with ethanol in different host species in East Java, Indonesia

Guild and Subfamily	Species	<i>Tectona</i>		<i>Syzygium</i>		<i>Swietenia</i>		<i>Paraserianthes</i>		<i>Pinus</i>		<i>Mangifera</i>		Total no. of beetle (N)	(% of N total)
		<i>grandis</i>		<i>aromaticum</i>		<i>mahagoni.</i>		<i>falcataria</i>		<i>merkusii</i>		<i>indica</i>			
		TGm	TGp	SAm	SAp	SMm	SMp	PFm	PFp	PMm	PMp	MI m	MIp		
Ambrosia beetles (Fungal chewers)															
Platypodidae	<i>Euplatypus parallelus</i>	5	-	10	6	-	-	1	-	-	-	19	12	53	1.10
	<i>Dinoplatypus</i> sp.	-	-	10	-	-	-	-	-	-	-	-	-	10	0.21
	<i>Crossotarsus</i> sp.	-	-	-	7	-	-	-	-	-	-	-	-	7	0.15
Scolytinae	<i>Platypus solidus</i>	-	-	-	-	-	-	-	4	-	1	-	-	5	0.10
	<i>Dinoplatypus pallidus</i>	-	-	-	-	-	1	-	-	-	-	-	-	1	0.02
	<i>Xylosandrus crassiusculus</i>	916	1,045	8	190	72	19	53	121	10	9	-	-	2,443	50.65
	<i>Xyleborus affinis</i>	10	14	1	567	21	21	113	36	14	7	330	44	1,178	24.42
	<i>Cryptoxyleborus</i> sp.	-	-	332	-	-	-	-	-	-	-	-	-	332	6.88
	<i>Xylosandrus morigerus</i>	36	32	-	8	25	5	-	32	-	5	-	-	143	2.96
	<i>Premnobius cavipennis</i>	17	-	7	3	2	3	-	34	-	3	11	-	80	1.66
	<i>Arixyleborus</i> sp.	30	30	-	-	-	1	-	-	-	1	-	-	62	1.29
	<i>Xyleborus ferrugineus</i>	23	-	5	-	-	17	-	-	15	2	-	-	62	1.29
	<i>Premnobius adjunctus</i>	-	-	52	-	-	-	-	-	-	-	-	-	52	1.08
	<i>Amasa resectus</i>	-	-	25	2	-	-	-	-	-	-	-	-	27	0.56
	<i>Euwallacea fornicates</i>	4	-	-	-	1	-	-	4	-	-	10	3	22	0.46
	<i>Diuncus haberkorni</i>	1	1	-	1	-	-	-	16	-	-	-	-	19	0.39
	<i>Xyleborinus saxesenii</i>	-	19	-	-	-	-	-	-	-	-	-	-	19	0.39
	<i>Xylosandrus compactus</i>	7	-	-	-	-	4	-	-	4	-	-	-	15	0.31
	<i>Ambrosiodmus pseudocitri</i>	-	-	-	4	4	3	-	-	-	-	-	-	11	0.23
	<i>Scolytoplatypus</i> sp.	-	-	-	-	-	-	-	10	-	-	-	-	10	0.21
	<i>Xyleborus celsus</i>	-	-	-	-	-	-	-	8	-	-	-	-	8	0.17
	<i>Ambrosiodmus minor</i>	-	-	-	7	-	-	-	-	-	-	-	-	7	0.15
	<i>Euwallacea similis</i>	-	-	-	-	3	2	-	-	-	2	-	-	7	0.15
	<i>Beaverium</i> sp.	-	-	-	-	-	-	-	6	-	-	-	-	6	0.12
<i>Xyleborinus andrewesi</i>	-	-	-	-	-	-	-	4	-	-	-	-	4	0.08	
<i>Eccoptopterus spinosus</i>	-	-	-	-	-	-	1	2	-	-	-	-	3	0.06	
Bark beetles (Phloem chewers)															
Scolytinae	<i>Ambrosiodmus</i> sp.	-	-	-	-	-	-	-	74	-	-	-	-	74	1.53
	<i>Hypothenemus</i> sp.	12	11	-	8	-	2	1	-	2	-	29	6	71	1.47
	<i>Monarthrum</i> sp.	-	-	-	-	-	21	-	-	21	-	-	-	42	0.87
	<i>Dendrocranulus</i> sp.	-	-	-	-	-	16	-	-	16	-	-	-	32	0.66
	<i>Cryphalus</i> sp.	-	-	-	2	-	3	-	-	-	3	-	-	8	0.17
	<i>Cocotrypes</i> sp.	-	-	-	-	1	-	3	-	-	-	-	-	4	0.08
	<i>Cryptocarenus</i> sp.	-	-	-	-	-	2	-	-	-	2	-	-	4	0.08
	<i>Hypocryphalus</i> sp.	-	-	-	-	-	1	-	-	1	-	-	-	2	0.04
	Total no. of ambrosia beetles	1,049	1,141	450	795	128	76	168	277	43	30	370	59	4,586	95
Total no. of bark beetles	12	11	0	10	1	45	4	74	40	5	29	6	237	5	
Grand total no. of beetles	1,061	1,152	450	805	129	121	172	351	83	35	399	65	4,823	100	

Note: The first & second letters indicate the host plant (TG: *Tectona grandis*, SA: *Syzygium aromaticum*, SM: *Swietenia mahagoni*, PF: *Paraserianthes falcata*, PM: *Pinus merkusii*, MI: *Mangifera indica*), and the third letter indicates the cultural types (m: Monoculture, p: Polyculture).

crassiusculus were the dominant species in this study, accounting for 50.65% of the total catch. Setiawan et al. (2018) reported that *X. crassiusculus* was the dominant species in polyculture and monoculture teak plant systems in the Malang District. Pennacchio et al. (2003) also reported that *X. crassiusculus* is a polyphagous species with various genera of plant hosts including forest trees, shrubs, and vines. It is also a pest in a

variety of hosts, including 124 hosts and 48 families that occur mostly in tropical regions, including pine, cocoa, coffee, mahogany, rubber, tea, and teak (Horn and Horn 2006). Reding et al. (2011) also reported that bottle traps baited with ethanol lure captured *X. crassiusculus* effectively.

The present study showed that *T. grandis* had the highest total number of individuals. The species

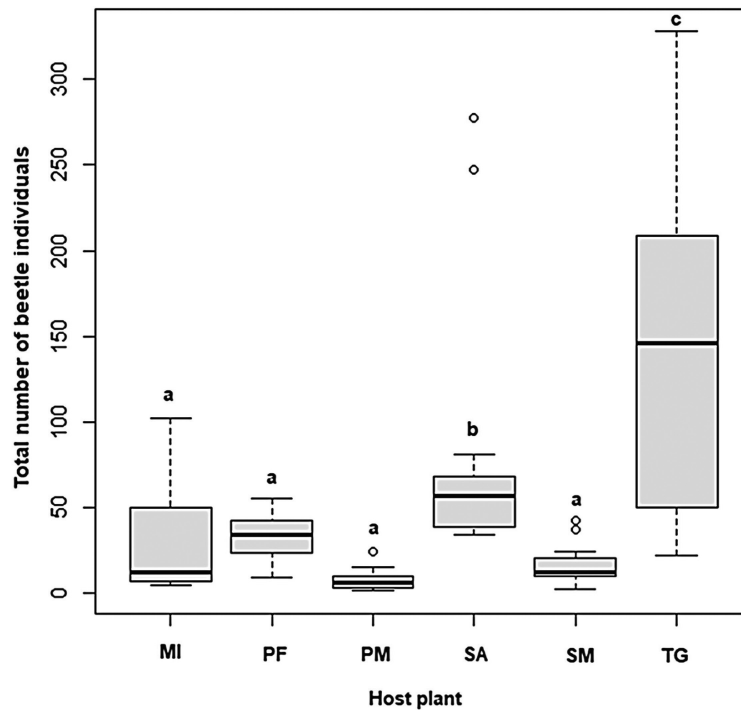


Fig. 3. The total number of bark and ambrosia beetles collected in six host plant species, including *Mangifera indica* (MI), *Paraserianthes falcata* (PF), *Pinus merkusii* (PM), *Syzygium aromaticum* (SA), *Swietenia mahagoni* (SM), and *Tectona grandis* (TG).

Table 3. Species diversity indices in different host plant species

Study site	Indices			Species richness
	Shannon-Wiener Diversity	Evenness	Simpsons	
TGm	0.67 (Low)	0.28 (Low)	0.75 (High)	11
TGp	0.47 (Low)	0.23 (Low)	0.83 (High)	7
SAm	1.00 (Middle)	0.45 (Low)	0.57 (Middle)	9
SAp	0.88 (Low)	0.35 (Low)	0.56 (Middle)	12
SMm	1.27 (Middle)	0.61 (Middle)	0.38 (Low)	8
SMp	2.28 (Middle)	0.82 (Middle)	0.13 (Low)	16
PFm	0.79 (Low)	0.44 (Low)	0.53 (Middle)	6
PFp	1.95 (Middle)	0.76 (High)	0.20 (Low)	13
PMm	1.81 (Middle)	0.87 (High)	0.18 (Low)	8
PMp	2.06 (Middle)	0.89 (High)	0.16 (Low)	10
MIIm	0.68 (Low)	0.42 (Low)	0.70 (Middle)	5
MIp	0.93 (Low)	0.67 (Middle)	0.51 (Middle)	4

Note: The first & second letters indicate the host plant (TG: *Tectona grandis*, SA: *Syzygium aromaticum*, SM: *Swietenia mahagoni*, PM: *Pinus merkusii*, PF: *Paraserianthes falcata*, MI: *Mangifera indica*), and the third letter indicates the cultural types (m: Monoculture, p: Polyculture).

diversity of bark and ambrosia beetles was the greatest in *S. mahogany* baited with ethanol. According to Tarno et al. (2016), an index value between 1 and 3 is categorized as intermediate diversity, and the distribution of each species are also moderate. The species evenness index value for *P. falcata* indicated

the highest species evenness. According to Tarno et al. (2016), an index value between 0.75 and 1 is categorized as high species evenness and a stable species community. The Simpson's dominant index value of *P. falcata* indicated low species dominance. According to Krebs (1999), when the index value is

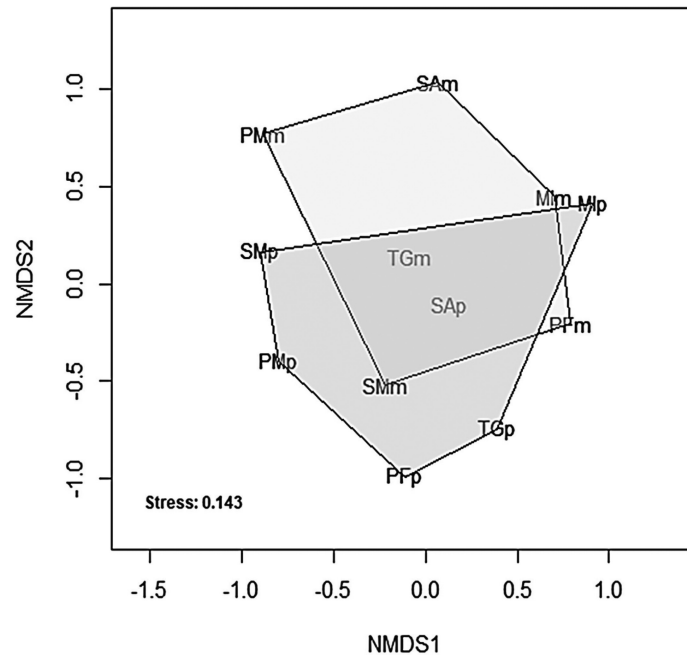


Fig. 4. Variation in bark and ambrosia beetle compositions between study sites, in non-metric multidimensional scaling (NMDS) ordination (based on abundance data and a Bray-Curtis distance metric). The first & second letters indicate the host plant (TG: *Tectona grandis*, SA: *Syzygium aromaticum*, SM: *Swietenia mahagoni*, PM: *Pinus merkusii*, PF: *Paraserianthes falcata*, MI: *Mangifera indica*), and the third letter indicates the cultural types (m: Monoculture, p: Polyculture).

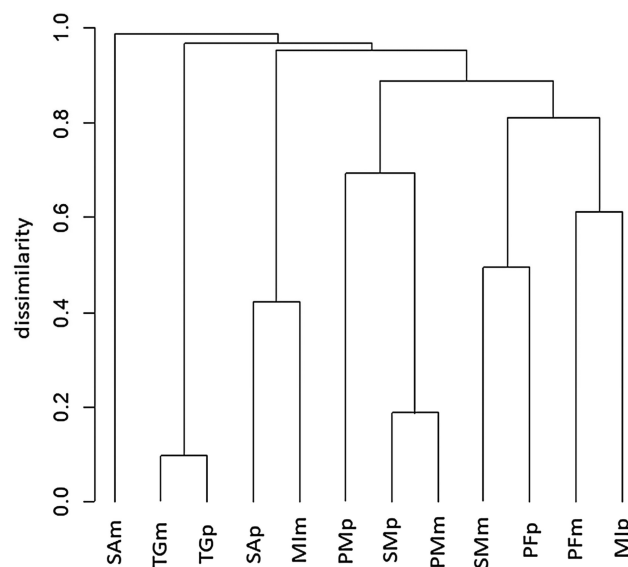


Fig. 5. Dissimilarity cluster between the host and the culture system in bark and ambrosia beetle species compositions. The first & second letters indicate the host plant (TG: *Tectona grandis*, SA: *Syzygium aromaticum*, SM: *Swietenia mahagoni*, PM: *Pinus merkusii*, PF: *Paraserianthes falcata*, MI: *Mangifera indica*), and the third letter indicates the cultural types (m: Monoculture, p: Polyculture).

> 0.5 , there are no dominant species in the community. Species diversity can be influenced by several factors, including host specificity and mechanisms of plant resistance to herbivorous insects. In this study, the diversity of ambrosia beetles in various host species differed depending on the resistance mechanism of each plant species. The host specificity of herbivorous insects is one of the key predictors of patterns of biodiversity (Novotny et al. 2002). Host plants varied in defensive capabilities, *i.e.*, chemical compounds and physical defenses, such as lignin or wood toughness (Agrawal 2007). The lowest value of diversity was *T. grandis*, because this plant has natural resistance properties to insects. Ngee et al. (2004) reported that teak is a less preferred species against the native pests *Coptotermes*, *Microceratotermes*, *Globitermes*, and *Macrotermes* termites in a preference test. The chemical components provide a good indication of the natural durability of teak, and the composition of extractives of teak wood was reported to be quite complex (Yamamoto et al. 1998). According to Lukmandaru and Takahashi (2008), tectoquinone (2-methylanthraquinone), and the n-hexane-extractible content has insecticidal properties, conferring insect resistance.

In this study, the *T. grandis* monoculture and polyculture had a high similarity value. Differences in species compositions between plant host species and *X. affinis* were found at all sites. In this study, each host plant species had different locations, which differed in available resources, and altitude. At the study sites, *S. aromaticum* and *M. indica* had the lowest altitude. Warmer temperatures and climate changes at lower elevations may lengthen the flight activity period and increase the number of generations produced per year, thus inducing beetles to migrate to higher elevations (Bale et al. 2002). *Xyleborus affinis* was found at all sites, thus it had the broadest host range. *Xyleborus affinis* is extremely polyphagous and has a known host range of 248 species, angiosperms as well as gymnosperms (Wood 1982). Although it is among the most widespread and common ambrosia beetles in forested areas around the world (Sobel et al. 2015). Steininger et al. (2015) also reported that *X. affinis* was only weakly attracted to ethanol, the most commonly used lure for ambrosia beetle monitoring.

The NMDS ordination analysis showed that there was no significant difference in the compositions of bark and ambrosia beetle species between monoculture and polyculture, suggesting that the cultural systems are similar. Reed and Muzika (2010) reported that different forms of forest management may not modify the ambrosia beetle community. Setiawan et al. (2018) also mentioned that in teak forest, monoculture and polyculture systems had the same diversity categories

based on ambrosia beetle abundant. Previous studies provide evidence about the similarities between the two cultural systems of six different host plants related to the ambrosia and bark beetles abundant in East Java, Indonesia.

CONCLUSIONS

A total of 4823 beetles were collected, representing 26 ambrosia beetle and eight bark beetle species. The abundances of bark and ambrosia beetles were significantly highest in *T. grandis*. *X. crassiuscullus* showed a strong attraction to the ethanol lure and was the dominant species. The Shannon-Wiener diversity index of all beetles captured in this study was the greatest in the *S. mahogany* polyculture and the lowest in the *T. grandis* polyculture. According to Bray-Curtis analysis, the *T. grandis* monoculture and *T. grandis* polyculture had a higher similarity value of bark and ambrosia beetle species compositions than cultures collected at other sites. There were no significant differences between culture systems in the compositions of bark and ambrosia beetle species.

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