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AMBROSIA BEETLE'S (COLEOPTERA: CURCULIONIDAE) OCCURRENCE AND DIVERSITY IN FOREST PLANTATIONS IN WESTERN SARAWAK, MALAYSIA

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ABSTRACT

Ambrosia beetles (Coleoptera: Curculionidae) are wood-boring beetles that play an important role in temperate forests. They live symbiotically with microorganisms such as fungi that can cause plant wilt and death. In Sarawak, Malaysia, the ambrosia beetles have been found attacking exotic tree species such as *Acacia mangium*, *Acacia crassicarpa* and *Eucalyptus pellita* in some forest plantations. This research aimed to investigate the occurrence of the ambrosia beetles and determined their diversity in forest plantations located in western Sarawak, Malaysia. This research was conducted in exotic plantations at Licence Planted Forest 42 (LPF42), Sampadi, Lundu, and Sabal Model Forest, Simunjan from January 2022 until October 2022. A total of 20 units of modified intercept panel traps baited with 70% ethanol were used in each study site to trap the adult ambrosia beetles. The occurrence and diversity of ambrosia beetles were later analysed using Palentological Statistics Software (PAST). As a result, a total of 170 ambrosia beetles representing 12 species of Scolytinae and two species of Platypodinae were collected. There are seven species comprising 129 individuals of ambrosia beetles collected at LPF42 and 11 species comprising 41 individuals at Sabal Model Forest, respectively. The Shannon-Wiener and Simpson's indices showed the greatest values in Sabal Model Forest (1.45 and 0.58 respectively), meanwhile, in LPF42 (0.50; 0.28) indicated a range of low to medium species diversity but low species evenness in both study sites. In general, *Xylosandrus crassiusculus* was dominant in both sites with a total capture of 141 individuals (82.94%). Overall, this study provided baseline information on ambrosia beetle occurrence and their diversity in forest plantations in western Sarawak.

Keywords: Abundance, fungus-farming beetle, pest, weevil, *Xylosandrus crassiusculus*

ABSTRAK

Kumbang Ambrosia (Coleoptera: Curculionidae) adalah Kumbang Pengorek Kayu yang memainkan peranan penting dalam hutan temperat. Kumbang ini hidup secara simbiosis dengan mikroorganisma seperti kulat yang boleh menyebabkan tumbuhan layu dan mati. Kumbang ambrosia di Sarawak telah ditemui menyerang spesies pokok eksotik seperti *Acacia mangium, Acacia crassicarpa* dan *Eucalyptus pellita* di beberapa ladang. Kajian ini bertujuan untuk mengkaji taburan kumbang ambrosia dan menentukan kepelbagaian kumbang ambrosia di ladang yang terletak di bahagian barat Sarawak, Malaysia. Kajian ini telah dijalankan di ladang iaitu di Licence Planted Forest 42 (LPF42), Sampadi, Lundu, dan Sabal Model Forest, Simunjan dari Januari 2022 sehingga Oktober 2022. Perangkap panel pintasan yang diubah suai dan menggunakan etanol 70% sebagai umpan digunakan untuk menangkap kumbang ambrosia dewasa. Taburan dan kepelbagaian kumbang ambrosia selanjutnya dianalisis menggunakan perisian *Palentological Statistics* (PAST). Hasilnya, sejumlah 170 kumbang ambrosia mewakili 12 spesies Scolytinae dan dua spesies Platypodinae telah dikumpulkan. Terdapat tujuh spesies yang terdiri daripada 129 individu kumbang ambrosia masing-masing dikumpulkan dari LPF42, manakala 11 spesies daripada 41 individu dari Hutan Model Sabal. Indeks Shannon-Wiener dan Simpson menunjukkan nilai tertinggi di Hutan Model Sabal, (masing-masing 1.45 dan 0.58), sementara itu di LPF42 (0.50; 0.28) menunjukkan julat kepelbagaian spesies rendah hingga sederhana tetapi kesamarataan spesies rendah di kedua-dua lokasi kajian. Secara amnya, *Xylosandrus crassiusculus* mendominasi di kedua-dua lokasi kajian dengan jumlah tangkapan 141 individu (82.94%). Secara keseluruhannya, kajian ini memberikan maklumat asas mengenai kehadiran kumbang ambrosia dan kepelbagaian mereka dalam ladang di barat Sarawak.

Kata kunci: Kelimpahan, kumbang penternak kulat, serangga perosak, kekabuh, *Xylosandrus crassiusculus*

INTRODUCTION

The ambrosia beetle belongs to the Curculionidae family in the Coleoptera, found in either the Scolytinae or Platypodinae subfamilies of weevils, and exhibits an obligatory association with nutritional fungal symbionts (Hulcr et al. 2015). While most ambrosia beetles are benign and of little economic significance as wood-boring insects, there are also members of this group that possess invasive tendencies and can cause substantial damage to trees (Hulcr et al. 2007). The majority of ambrosia species target dying or deceased trees, with a select few posing a threat to healthy trees (Sittichaya et al. 2019). These beetles create tunnels within the xylem, where they introduce symbiotic fungi. Consequently, they are commonly referred to as fungus-farming beetles, engaging in a symbiotic relationship with microorganisms such as fungi, bacteria, and yeast. This symbiosis can result in plant diseases that lead to wilting and eventual demise of the affected plants (Tarno et al. 2016). The ambrosia beetle is linked to a diverse range of fungi (Harrington 2005).

Scolytinae represents the more prevalent group, while Platypodinae is less common, as identified by Kirkendall et al. (2015). There are over 6,000 species of Scolytinae and around 1,400 species of Platypodinae documented, with the majority of these species being native to tropical or subtropical regions (Kirkendall et al. 2015; Sitompul et al. 2023). These ambrosia beetles play a pivotal ecological role and primarily serve as decomposers of wood in forest ecosystems. Their adaptive nature allows them to thrive in various climates and environments. In Southeast Asia, the initial checklist by Beaver and Browne in 1975 documented 33 species of Xyleborini, mainly concentrated in the northern regions of Thailand. Subsequent studies, including the comprehensive work by Sittichaya et al. (2019), have expanded the known diversity to a remarkable 156 xyleborine species in Thailand. The intricate biodiversity of Scolytinae is not limited to Thailand, as studies in Indonesia have revealed nine distinct species of ambrosia beetles in teak plantations (Setiawan et al. 2018) employed ethanol-baited traps in monoculture and polyculture systems, identifying *Xylosandrus crassiusculus* as the predominant species in both scenarios. Besides that, Sitompul et al. (2023) has highlighted several Coffee Berries from Indonesia under the subfamily Scolytinae.

Ambrosia beetles play a crucial role in maintaining the ecological balance of temperate forest ecosystems, yet their presence is accompanied by significant economic losses (Lindgren & Raffa 2013). According to Nair (2007), these beetles have been documented attacking a wide array of forest plants contributing to widespread mortality of deciduous and coniferous trees in forested and urban areas (Kühnholz et al. 2001. Reports highlight instances of ambrosia beetles, particularly the *Euplatypus paralellus* Fabricius species, infesting Angsana trees (*Pterocarpus indicus* Willd.) in southern Thailand (Bumrungsri et al. 2008) and Malang, Indonesia (Tarno et al. 2014). Notably, in the middle of Tennessee, the chestnut trees faced significant threats from *Xylosandrus crassiusculus* and *Xylosandrus germanus* (Oliver & Mannion 2001) and (Gugliuzzo et al. 2021). The impact of these invasive beetles extends beyond their ecological role, underscoring the broader implications for forestry and horticulture.

In Sarawak, based on our surveillance, the ambrosia beetles were found attacking *Acacia crassicarpa, Acacia mangium* and *Eucalyptus pellita* plantation. However, there is a lack of ecological studies on ambrosia beetles in the Sarawak Forest plantation. Such studies are vital for informing sustainable forestry practices and mitigating economic and environmental challenges associated with bark beetle infestations. Appropriate research should be initiated, to enhance detection, monitoring, and management programs. Thus, this study aimed to investigate the occurrence and diversity of the ambrosia beetle in an exotic forest plantation located in western Sarawak, Malaysia.

MATERIALS AND METHODS

Study Sites

Trappings of ambrosia beetles were carried out at two study sites in western Sarawak, Malaysia (Figure 1). Site 1 is a forest plantation called the Licensed Planted Forest (LPF) 42 located at Sampadi, Lundu, a licensed planted forest owned by the company Polima Forest Bintulu. It is located at 1^0 30' 34.4 N, 109^0 55' 01.4 E. Second site is the Sabal Model Forest located within Sabal Forest Reserve, $01^0$05'10.5 N, $110^0$56'12.4 E, within the wildlife conservation area and forest reserve area. Surrounding faunas are indigenous tree species such as Engkabang and Meranti trees. There are four trapping transect lines (100 metres each transect line) established in each study site (Table 1).

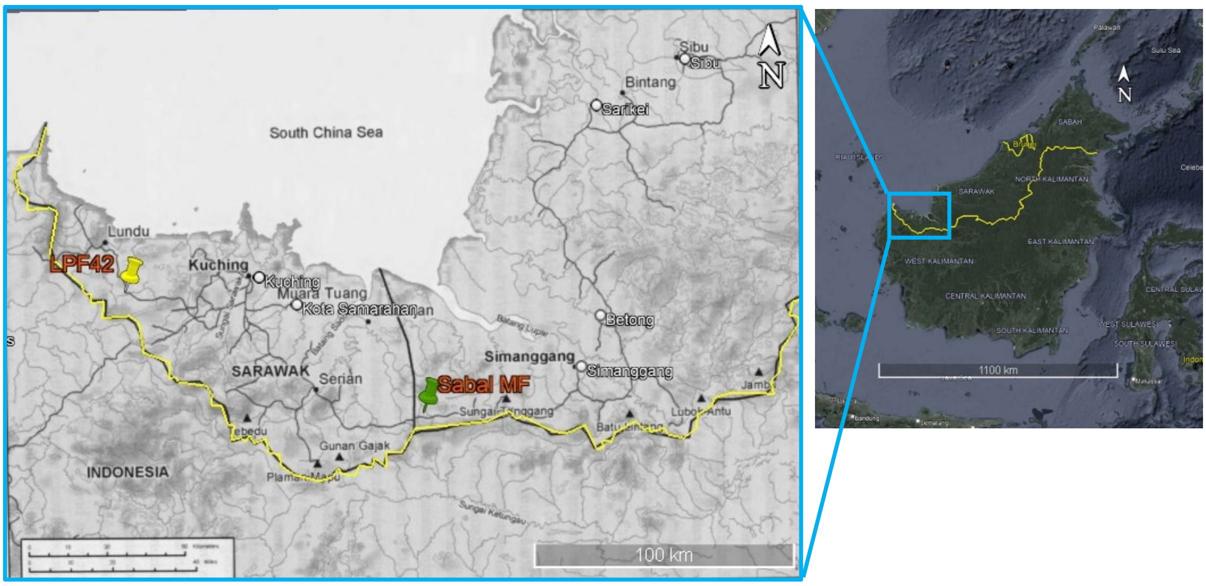

Figure 1. Study sites for ambrosia beetles' trapping in western Sarawak (Source: Google Earth ProSoftware)

Table 1. Plant species and planting date of each trapping plot in Site A and Site B (Source: Forest Department Sarawak)

Study Site	Trapping Transect Line	Host Plant	Year of Planting
LPF42	TP1	*Acacia mangium*	2015
LPF42	TP2	*Acacia crassicarpa*	2015
LPF42	TP3	*Eucalyptus pellita*	2015
LPF42	TP4	*Acacia mangium, Acacia crassicarpa, Eucalyptus pellita, Falcataria molluccana* & oil palm trees	2017
Sabal MF	TP1	*Acacia* spp. (Hybrid)	2011
Sabal MF	TP2	*Acacia mangium*	2011
Sabal MF	TP3	*Eucalyptus pellita* (Adjacent to wooded forest)	2011
Sabal MF	TP4	*Eucalyptus pellita* 2 (Adjacent to *Acacia mangium* plot)	2011

Sampling Procedure

The ambrosia beetles were collected through the utilisation of a modified Intercept panel trap that was attractively baited with 95% ethanol (Figure 2). Ethanol-lured traps have proven to be effective in the surveillance of ambrosia beetle populations (Reding & Ranger 2020; Reding et al. 2013, 2011; Ranger et al. 2021). Ethanol, which is the identical volatile compound released by debilitated plants in response to stressors of a biotic or abiotic nature, has the potential to stimulate the attack by ambrosia beetles (Kelsey et al. 2014; Ranger et al. 2016, 2010, 2020). The baited panel traps were tied up in the tree stands approximately 1 metre above the ground (Steininger et al. 2015; Setiawan et al. 2018. Six-panel traps were positioned at each trapping site, along a transect line spanning 100 metres, with a separation of 20 metres between each trap. The trappings were carried out for five continuous days in each of the subsequent months: January 2022, April 2022, July 2022, and October 2022. The trapped ambrosia beetles were collected every day during these periods. The collected ambrosia beetles were then preserved in 70% ethanol in vials and were labelled with date and sampling locality and brought to the laboratory for identification.

Beetle Identification

The identification of the ambrosia beetle was carried out based on its morphological characteristics, such as body size, elytra, and body color. This identification process was conducted using a stereo microscope, model Olympus SZ61 installed with an industrial digital camera (18MP ½.3" Color USB3.0 APTINA CMOS Sensor) and images of the specimen captured using Top View 3.7 software. The identification keys used specifically for ambrosia beetles were mostly from Rabaglia et al. (2006), Gomez et al. (2018), and Pérez-De la Cruz et al. (2016). The identification of the ambrosia beetle took place at the Entomology Laboratory, Industrial Forest Research Centre, Forest Department of Sarawak, Malaysia. To further confirm and verify the species, high-quality images of the beetles were emailed to the UF Forest Entomology Laboratory at the University of Florida, USA.

Statistical Analysis

The collected data was then analysed using *Palentological Statistics* (PAST) software version 4.03 (Hammer et al. 2001). The diversity of the ambrosia beetle was assessed using several indices (Table 1), including the Shannon-Wiener diversity index (H'), Species Evenness index (E), and Simpson's dominance index (C) (Tarno et al. 2016).

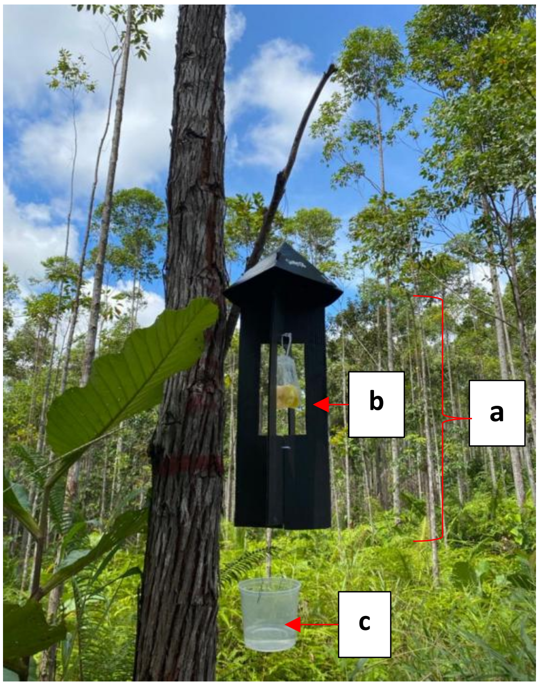

Figure 2. Modified Intercept panel trap baited with ethanol used in this study. a). Body, made from PP corrugated sheet, b) Ziplock plastic bag, containing a small sponge and filled with 95% of ethanol, and c) Collecting container, filled with water for trapping the beetle

Table 2. Value and description of index for the diversity indices (Tarno et al. 2016)

Indices	Index Value	Description
Shannon-Weiner (H')	<1	Low level of diversity, low individual distribution of each species
	1-3	Middle level of diversity, middle level of individual distribution for each species
	>3	High level of diversity, high level of individual distribution for each species
Simpson Dominance (C)	0.00<C<0.50	Low dominance of species
	0.50<C0.75	Middle dominance of species
	0.75<C<1	High dominance of specie
Species Evenness (E)	0.00<E<0.50	Evenness is low, community is under pressure
	0.50<E<0.75	Evenness is medium level, community is unstable
	0.75<E<1.00	Evenness is high, community is stable

RESULTS AND DISCUSSION

The Occurrence of Ambrosia Beetle

Table 3 presents data on the prevalence of beetle species in the Scolytinae and Platypodinae subfamilies. Overall, the Scolytinae subfamily contributes significantly to the total beetle count with 141 individuals, while the Platypodinae subfamily has 29 individuals. The large quantity of Scolytinae beetles captured in the forest plantations may be attributed to their ability to adapt to the specific conditions of these plantations. The plantations undergo extensive management practices, such as regular planting, pruning, and harvesting if compared to natural and secondary forests. Thinning and logging operations frequently remove trees (3 to 4 years old), resulting in the formation of debris piles that are left in the stands (Stilwell et al. 2014). These tree remains serve as attractive habitats for a variety of scolytines (Perez-De la Cruz et al. 2016). Many ambrosia beetle species are primarily attracted to the ethanol emitted from the vascular tissues of newly dead or decaying trees, which serve as suitable hosts for numerous species of Scolytinae (Hulcr et al. 2008; Rudinsky 1962). Both study sites are particularly vulnerable to environmental stress factors, including attacks from other insect pests such as termites.

Within the subfamily Scolytinae, *Xylosandrus crassiusculus* (Figure 3a-b) was the most common species, making up 82.94% of the total individuals. Other species in this subfamily exhibit varying abundance. *Xylosandrus crassiusculus* is known for its wide range of host plants and polyphagous nature, colonising numerous species across various plant genera (Horn & Horn 2006; Pennacchio et al. 2003). It occurs in a wide variety of host plants, such as clove (*Syzygium aromaticum*), mahagony (*Swietenia mahagoni*), and albizia (*Paraserianthes falcataria*) in East Java (Tarno et al. 2021). The distribution of *X. crassiusculus* in temperate forests is primarily influenced by climatic factors rather than the types of forests, as noted by Ranger et al. (2016). It is a polyphagous species, adapting well to warm and humid regions (Kirkendall & Faccoli 2010). The high number of *X. crassiusculus* individuals in both study sites may be attributed to their attraction to traps baited with ethanol, as demonstrated by Reding et al. (2011) study using ethanol-baited bottle-style traps. *Xyleborus affinis* was also captured in both sites but with only four individuals in total. Despite being recognised as highly polyphagous on 248 host plant species (Wood 1982), the present study recorded a low number of *X. affinis* individuals. This may be attributed to the trapping method used, as suggested by

Steininger et al. (2015) who found that *X. affinis* had weak attraction to ethanol. *Xylosandrus crassiusculus* is also captured in greater numbers every sampling month which may indicate to have several overlapping generations per year. In the tropics, breeding is continuous throughout the year, with overlapping generations, so that the species is present at all times and in all stages of development (Browne 1969).

In the subfamily Platypodinae subfamily, *Dinoplatypus pseudocupulatus* (Figure 3c-d) dominated, constituting 4.12% of the total beetles. *Dinoplatypus pseudocupulatus* and *Euplatypus parallelus* has been found to infest the *Acacia crassicarpa* trees during the study. The symptoms observed were the presence of numerous small holes (1mm in diameter) and black-stained sap out of the holes. Recently, *E. parallelus* expanded its presence in East Java and started infesting *Pterocarpus indicus* Willd., as reported by Gomez et al. (2018). This species became prevalent in attacking *P. indicus* in Malang and Batu, East Java. Tarno et al. (2022) noted that *E. parallelus* also caused a significant economic impact by targeting large-diameter stems and living trees. The factors influencing the distribution of Platypodinae beetles remain under-researched. The distribution of their fungal symbionts might also affect the beetles' spread. Thus, studying their fungal symbioses, as suggested by Vanderpool et al. (2018), is key to understanding possible distribution constraints.

Table 3. Total number of ambrosia beetlesl collected in the intercept panel traps baited with ethanol in exotic forest plantations from two study sites in western Sarawak

Subfamily	Species	LPF42	Sabal MF	Total no. of beetle (N)	(% of N total)
Scolytinae	*Xylosandrus crassiusculus*	115	26	141	82.94
	Dryocoetes himalayensis	2	0	2	1.18
	Xyleborus affinis	1	3	4	2.35
	Xyleborus artestriatus	0	3	3	1.76
-	*Eccoptopterus spinosus*	0	1	1	0.59
	Coccotrypes sp.	0	2	2	1.18
	Xyleborus perforans	0	1	1	0.59
	Xyleborus ferrugineus	2	1	3	1.76
	Arixyleborus setosus	0	1	1	0.59
	Scolytinae sp.	0	1	1	0.59
	Xyleborus pfeillii	1	1	2	1.18
Platypodinae	*Dinoplatypus pseudocupulatus*	7	0	7	4.12
	Platypodinae sp.	0	1	1	0.59
	Euplatypus paralellus	1	0	1	0.59
	Total	129	41	170	100

The Diversity of Ambrosia Beetle

The data from the two study sites based on Table 4, LPF42 and Sabal MF, show differences in ambrosia beetle diversity. At LPF42, seven species of ambrosia beetle were collected and the Shannon-Wiener diversity index is low at 0.50, indicating low species diversity. In contrast, Sabal MF displays a moderate level of diversity with a Shannon-Wiener index of 1.45 and 11 species of ambrosia beetle collected. According to Tarno et al. (2016), an index value between 1 and 3 is categorised as intermediate diversity.

The evenness and Simpson indices in LPF42 are also low (0.24 and 0.20), respectively, suggesting uneven species distribution and low dominance of species (Tarno et al. 2016). The evenness index in Sabal MF, while still low at 0.39, is slightly higher than LPF42, indicating a more balanced species distribution. The Simpson index at 0.58 is in the middle range, signifying a more diverse and moderate dominant ecosystem. Tarno et al. (2016) stated that the value of Simpson index between 0.50 and 0.75, is categorised at the medium level.

Sabal MF was located within a forest reserve and wild conservation area with a dense hardwood forest. Forest composition and higher forest richness influenced the occurrence of ambrosia beetle (Rassati et al. 2016). Plant abundance impacts insect diversity (Dinnage et al. 2012, and ambrosia beetle abundance correlates with host abundance and size (Reed & Muzika 2010. Low captures of some species were attributed to ethanol trap inefficiencies (Atkinson et al. 1988; Lindgren 1983). Increased ethanol lures enhanced beetle capture (Ranger et al. 2011; Reding et al. 2011), and an ultra-high ethanol release rate proved more effective (Viloria et al. 2021).

Table 4. Species diversity indices in two study sites at western of Sarawak, Malaysia

Studt Site	**Shannon-Wiener (H')**	**Evenness (e^H/S)**	**Simpson (1-D)**	**Species Richness**
LPF42	0.50 (Low)	0.24 (Low)	0.20 (Low)	7
Sabal MF	1.45 (Middle)	0.39 (Low)	0.58 (Middle)	11

*Licenced Planted Forest (LPF), Model Forest (MF)

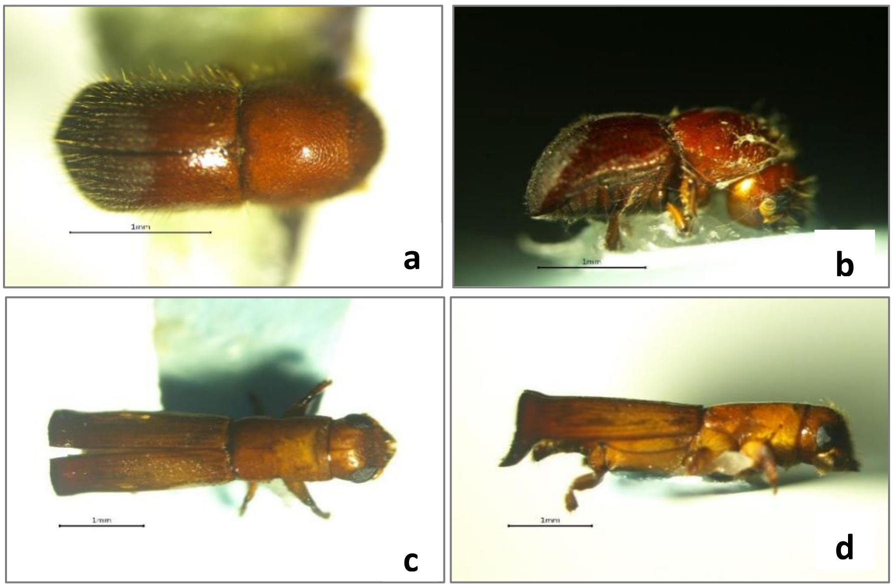

Figure 3. Images of the dominant species recorded for subfamily Scolytinae (a & b) and Platypodinae (c & d) captured using internal digital camera installed on the stereo microscope; a) Dorsal view of *X. crassiuculus*, magnification of 2×; b) Lateral view of *X. crassiusculus*, magnification of 1.5×; c) Dorsal view of *D. pseudocupulatus*, magnification of 1.2×; and d) Lateral view of *D. pseudocupulatus*, magnification of 1.2×

CONCLUSION

In conclusion, a total of 129 ambrosia beetles representing seven species and 41 ambrosia beetles from 11 species were collected in LPF42 and Sabal MF respectively. The abundance of ambrosia beetles was highest in LPF42, meanwhile, across the trapping sites, ambrosia beetles were abundant in the *A. mangium* plot in both study sites. *X. crassiussculus* showed a strong attraction to the ethanol-baited trap and was the dominant species. The Shannon-wiener and Simpson's index showed the greatest values in Sabal MF, categorised in the medium diversity category and low species evenness in both study sites. Overall, this study had provided baseline information on ambrosia beetle occurrence and their diversity in forest plantations in western Sarawak.

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AUTHORS DECLARATIONS

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Conflict of Interest
The authors declare that they have no conflict of interest.

Ethics Declarations
No ethical issue required for this research.

Data Availability Statement
My manuscript has no associated data.

Authors' Contributions
ITD and WA conceived this research, ILIP designed the experiments; ILIP, ITD and WA do the research and collect the samples from the field; ILIP, ITD and WA participated in the interpretation of the data; ITD and WA performed the plant identification; ILIP, ITD and WA wrote the paper and participated in the revisions of it. All authors read and approved the final manuscript.

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