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Define Colony Number of Subterranean Termites *Coptotermes gestroi* (Isoptera: Rhinotermitidae) in Selected Infested Structures

(Penetapan Jumlah Koloni Anai-Anai Bawah Tanah *Coptotermes gestroi* (Isoptera:Rhinotermitidae) yang Menyerang Struktur Bangunan Terpilih)

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ABSTRACT

Termites are one of the social insects living in large colonies that can cause economic loss. The objective of this study was to estimate foraging territory of infested subterranean termites on building structure. A mark-recapture study was conducted on eight Coptotermes gestroi *colonies located at selected infested building structures in Penang, Malaysia. From the foraging study, the population of* C. gestroi *was estimated to be within the range of 106,592±6,968 to 4,185,000±2,127,328. Additionally, the foraging territory was from 13 to 300 m² of the infested building structures. Meanwhile the maximum foraging distance was from 4 to 30 m of the infested structures. The results indicated that each of the building structures was infested by a single colony. This study also showed that the triple mark recapture technique used to estimate the population size of the termite colony was capable of providing rough estimates of foraging population of* C. gestroi.

Keywords: Coptotermes gestroi*; foraging distance; foraging population; foraging territories; triple mark recapture*

ABSTRAK

Anai-anai adalah salah suatu serangga sosial yang hidup di dalam koloni besar yang boleh menyebabkan kerugian ekonomi. Objektif kajian ini adalah untuk menganggarkan wilayah keluasan mencari makanan anai-anai bawah tanah pada struktur bangunan. Suatu kajian tanda tangkap dan lepas semula telah dijalankan ke atas lapan koloni Coptotermes gestroi *yang menyerang bangunan terpilih di Pulau Pinang, Malaysia. Daripada kajian mencari makanan, populasi* C. gestroi *telah dianggarkan antara 106,592±6,968 dan 4,185,000±2,127,328. Selain itu, wilayah mencari makanan adalah dari 13 hingga 300 m². Sementara itu, jarak maksimum linear mencari makanan adalah dari 4 hingga 30 m. Keputusan kajian menunjukkan bahawa setiap struktur bangunan telah diserang oleh satu populasi anai-anai. Kajian ini juga menunjukkan bahawa tangkap-tanda dan lepas sebanyak tiga kali yang digunakan untuk membuat anggaran saiz koloni anai-anai mampu menyediakan anggaran kasar populasi mencari makanan oleh* C. gestroi.

Kata kunci: Coptotermes gestroi*; jarak mencari makanan; keluasan mencari makanan; saiz populasi; tangkap tiga kali bertanda*

INTRODUCTION

Foraging and population territory studies are important to determine the termite population and foraging size in order to achieve effective termite control. Foraging population of subterranean termite can be estimated using two methods, mark recapture and direct counting. However, none of these methods have been absolutely accurate, although mark recapture is generally more acceptable because of its practicality for studying termite populations in the field (Nutting & Jones 1990).

Two mark- recapture methods that have been used intensively to estimate the foraging populations of subterranean termites are single mark recapture and triple-mark recapture techniques. The single mark recapture technique with Lincoln Index (Begon 1979) was used to estimate the foraging populations of *Coptotermes formosanus* Shiraki (Lai 1977; Su et al. 1984), *Reticulitermes* spp. (Forschler & Townsend 1996a), *Globitermes sulphureus* Haviland (Abdul Hafiz et al. 2008; Abdul Hafiz & Abu Hassan 2007; Lee et al. 2003a; Ngee & Lee 2002). The triple mark recapture technique using weighted mean method (Begon 1979) was used to estimate the populations of *C. formosanus* (Su & Scheffrahn 1988), *Reticulitermes flavipes* Kollar (Forschler & Townsend 1996b; Su et al. 1993), *Reticulitermes speratus* Kolbe (Tsunoda et al. 1998), *Coptotermes gestroi* Wasmann (Sornnuwat et al. 1996), *Coptotermes curvignathus* Wasmann (Abdul Hafiz & Abu Hassan 2006; Sajap 1999) and *Microtermes pakistanicus* Ahmad (Lee et al. 2003b). In this study, triple mark recapture technique was used because it increases the ratio of recaptured termites in the population as compared to single mark recapture with Lincoln Index on (Begon 1979).

Mark-recapture techniques have been used to estimate the foraging territories of the subterranean termites. Several different markers have been used in previous works. Lai

(1977) pioneered the use of histological dyes that have been adopted by other termite researchers (Evans et al. 1998; Haagasma & Rust 1993; Lai et al. 1983; Su et al. 1983). Spragg and Paton (1980) used radio isotope to trace the foraging territories of subterranean termites. Forschler (1994) and Miller (1993) used fluorescent paint. Other methods to estimate foraging territory include using direct counts of fumigated mounts (Darlington 1984), agonistic behavior of subterranean termites (Jones 1990; Pearce 1997) and direct excavation (Howard et al. 1982). In this study, Nile Blue A was used as markers. Nile Blue A is a type of stain used in the mark-recapture study to determine the efficiency of recapturing. It was chosen for marking the termites because the stain could last longer in the field (Haagasma & Rust 1993). Ngee and Lee (2002) found that the blue stained *Globitermes sulphureus* could remain blue as long as 60 days in the laboratory. At present, in Malaysia, Abdul Hafiz et al. (2007), Lee (2001), Lee et al. (2003b) and Sajap et al. (2000) had used Nile Blue A to estimate the termite foraging population of *C. gestroi*, *C. curvignathus* and *G. sulphureus*. Abdul Hafiz et al. (2007) discovered that the blue stain could last for 90 days in the field studies for *G. sulphureus*. The objectives of this paper were to identify the foraging distances and population territories of subterranean termites (*C. gestroi*) in selected structures of Penang, Malaysia.

MATERIALS AND METHODS

STUDY SITES AND TERMITE MONITORING STATION SETUP

Eight *C. gestroi* sites in Penang, Malaysia, were chosen for this study (Table 1). There were two types of termite monitoring stations - the above ground and the underground monitoring stations. Pre-installations of stations and pine wood billets similar to those described by Su and Scheffrahn (1988) were installed in the ground within the range of 1.5 to 3 m apart around buildings/structures. 100-300 pine stakes were used for every study site. Termite feeding consumption was higher using those two species of pine wood, *Pinus caribaea* and *Araucaria cunninghamii* (Abdul Hafiz & Abu Hassan 2007). Pine billets that were infested with termites were replaced with the in-ground monitoring stations. An in-ground monitoring station (IG) consisted of a hollow plastic container (20 cm in diameter by 19 cm in height) and a bundle of nine pine stakes, oven-dried for 48 h at 80°C, cooled on a tray and placed in the bucket. The buckets were buried at a depth of approximately 2.5 cm below the level of the ground. The in-ground monitoring stations were inspected biweekly to observe termite feeding activities. The above ground monitoring stations (AG) were black plastic boxes (6×11×6 cm). The box was painted black to minimize evaporation of moisture inside the box (Mohd Yusri et al. 2005). Two sets of rolled toilet tissue paper weighing between 40-80 g and oven-dried for 48 h at 50°C, were placed inside the box. The boxes were installed with overactive termite mud tubes. 10-20 boxes were used per-site. The above ground monitoring stations were examined every two weeks to observe termite feeding activities.

ESTIMATION OF TERMITE POPULATION AND FORAGING RANGE

After the establishment of ≥2 monitoring stations per site, a mark recapture program was carried out to estimate the foraging territory and the population size of the termites. The infested blocks from the in-ground monitoring stations were taken to the laboratory and carefully disassembled. The termites were removed by gently tapping the stakes over a plastic tray. They were separated from debris by allowing them access to a stack of five pine blocks (20×10 cm) that had been soaked in water for 24 h. After a 4-6 h aggregation on pine blocks, they were removed. The mean body weight of the termite workers was determined by weighing five groups of 10 individuals. The numbers of collected workers were determined by the total weight of collected workers and the mean worker weight to estimate the total of termites that were collected from the monitoring stations (Tamashiro et al. 1973). The termite workers collected from a station with a heavy feeding activity were stained with 0.1% (wt/wt) Nile Blue A by a no choice feeding of stained filter paper (Whatman No. 1, 9.0 cm in diameter) for 5 days (Su et al. 1991). The stained termites were released into the same station where they were collected. The total number of stained termites released depended on the total number of termites recaptured from the active monitoring station and it varied

TABLE 1. Locations and type of buildings infested by *C. gestroi*

Site	Location	Type of building	Number of in-ground station	Number of above-ground station
1	Bayan Baru	Double story terrace	0	6
2	Mengkuang	Single story bungalow	3	0
3	Gelugor	Double story bungalow	4	2
4	Guar Perahu	Wooden double story Malaysia traditional house	3	3
5	Bayan Lepas	Single story bungalow	0	3
6	Taman Rupawan	Single story bungalow	3	3
7	Islamic Centre, USM	A mosque	13	0
8	Plant house, USM	Greenhouse	7	0

for every study site. The monitoring stations at each site were checked 7 days after the release to record the number stained termites recaptured from the monitoring stations for that cycle (Su et al. 1991). The mark-released-recapture cycle was repeated for three rounds. Each round was run for two weeks. The foraging territory of a colony was defined as the area encompassed by the stations containing termites during the triple mark recapture cycles (TMR) (Su & Scheffrahn 1988). Meanwhile, the linear foraging distance was measured from one of the active monitoring stations containing blue termites to the furthest away monitoring station that contain blue termites at a site.

Data Analysis

The number of marked and unmarked workers was recorded for each cycle. A mean weight model (Begon 1979; Su et al. 1993) was used to estimate the foraging population (N) and the associated standard error (SE);

$$\mathrm{N} = \left(\sum M\mathrm{n}i \,/\, \left[\sum \mathrm{m}i\right) + 1\right]$$

$$\mathrm{SE} = N\sqrt{\left[1/\left(\sum mi+1\right)\right] + \left[2/\left(\sum mi+1\right)^2\right] + \left[6/\left(\sum mi+1\right)^3\right]},$$

where N is the mean foraging population; SE is the associated standard error for the *i*th cycle; m*i* is the total number of marked individuals up to the *i*th cycle; *Mi* is the number of marked individuals among the captured termites and n*i* is the number captured.

Results and Discussion

The triple mark recapture (TMR) method is widely used by termite researchers to estimate the foraging population of *C. formasanus* and *R. flavipes* (DeMark et al. 1995; Su et al. 1991). In Malaysia, triple mark recapture was used by Abdul Hafiz and Abu Hassan (2006) and Sajap et al. (2000) to estimate the foraging population of *C. curvignathus* and by Lee (2001) to estimate the foraging population *of C. gestroi*.

Our results showed that *C. gestroi* population in Penang, Malaysia was in the range of 56,127±11,925 and 4,185,000±2,127,328 at the eight sites (Table 2). Meanwhile in Thailand, *C. gestroi* population was smaller ranging from 1.13 to 2.75 × 10^6 (Sornnuwat et al. 1996). Sajap et al. (2000) reported that the foraging population of *C. curvignathus* was in the range from 1.6 to 7.1 × 10^5 with the foraging territory between 15 and 50 m^2 (Table 3).

The Malaysian population colonies of *Coptotermes* are smaller compared with the other *Coptotermes* species found outside Malaysia, such as *C. formasanus*. The population of *C. formasanus* was estimated in the range of 1.8 to 4.4 × 10^6 (Su et al. 1984), from 1.05 to 2.24 × 10^6 with the foraging territory was between 143 and 2189 m^2 (Su et al. 1984). The population estimation for *Reticulitermes flavipes* was even larger ranging from 4 to 19.1 × 10^6 (Grace et al. 1989; Haverty et al. 1999) in the United States.

The termite foraging population varies according to species. According to Lee et al. (2003a), the population estimate for *M. pakistanicus* was from 1.07 to 5.59 × 10^5 and consisted of five different colonies, with a foraging territory of 30.5-54.2 m^2. As for *G. sulphureus,* the foraging population was between 5.14 and 8.69 × 10^5 and consisted of three different colonies with a foraging territory of 5.9-42.5 m^2 (Lee et al. 2003b). In addition, by using Lincoln Index, Abdul Hafiz et al. (2007) estimated that the foraging population of *G. sulphureus* was 3.38 × 10^5 with the foraging territory of 96 m^2.

Environmental factors of soil characteristics such as pH, moisture, temperature, the presence of microorganism, organic materials (Chapman et al. 1982; Felsot 1989; Harris 1972; Macalady & Wolfe 1983; Tashiro & Kuhr 1978) and soil type (Campbell et al. 1971; Forschler & Townsend 1996a, 1996b; Harris 1972; Lange & Carlson 1956; Smith & Rust 1993) could also affect the foraging territory and estimated population of termites. Therefore, further study need to be carried to determine if soil types influence the foraging behavior of subterranean termites.

Site 8 (Plant House) had a larger foraging population size maybe due to the site being an open site where the colony was undisturbed by human activities. Site 5 (Bayan Lepas) had the smallest foraging population size, probably due to the size of the house and the colony was just below the house where human activities could have disturbed the foraging activities of the termites (Vasconcellos et al. 2010). Furthermore, it is possible that the age of termite colonies might be the reason of finding larger or smaller colonies in some places than others. Maybe, the smaller colonies are newly established colonies, while the larger ones have been in the area for a long time (Husseneder 2005; Vasconcellos et al. 2010).

The termite foraging behavior still requires more intensive and detail research. Owing to their epigeic foraging habits, ecologically prominent and abundant termites such as *Macrotermes, Schedorhinotermes, Hospitalitermes* and *Nasutitermes* would be excellent models for the study of the social organization and the divisions of labor that appear to enhance the efficiency of food collection. According to Reinhard et al. (1997), the efficiency of foraging and the addictiveness of the caste distributions that participate in different aspects of foraging have rarely been measured. In addition, knowing the population size and foraging territory of subterranean termites will help us determine how much termiticides is required to suppress the population. Therefore, less chemical usage could be applied to manage subterranean termite infestation inside the building structure.

Conclusion

This study showed the foraging population for *C. gestroi* was estimated to be between 106,592 ± 6,968 and 4,185,000 ± 2,127,328 individual number of foraging termite per colony with the foraging territory between 13 and 300 m^2 and the linear foraging distance between 5 and

TABLE 2. Triple mark recapture cycles, underground monitoring station, above-ground monitoring station [ ]. The number in the bracket indicates the monitoring station number where the marked termites were found in each cycle

Site	Recapture mark until *i*	*Mi*	*ni*	*mi*
1	1	8,521	1,1991	33
		[4]	[3,4,5,6]	
	2	11,828	7939	134
		[3,4,5,6]	[1,3,4,5,6]	
	3	7,514	1,833	33
		[1,3,4,5,6]	[1,2,6]	
2	1	12524	8924	155
		(3)	(1,2,3)	
	2	8924	4716	127
		(1,2,3)	(1,2,3)	
	3	4716	17260	130
		(1,2,3,)	(1,2,3)	
3	1	9,230	21,054	83
		(2)	(1,2,3,4) [1,2]	
	2	21,054	10,934	153
		(1,2,3,4) [1,2]	(1,2,3,4)	
	3	10,934	3,585	105
		(1,2,3,4)	(1,2,3,4)[1]	
4	1	3786	1213	65
		(2)	(2,3)[1,2,3]	
	2	1213	2481	142
		(2,3)[1,2,3]	[1,2,3]	
	3	2481	1091	28
		[1,2,3]	[1,2,3]	
5	1	1,873	2,718	5
		(3)	(3)	
	2	1,658	2,236	7
		(3)	(2,3)	
	3	2,236	11,359	3
		(1,2,3)	(1,2,3)	
6	1	3,351	12,223	2
		(2)	(1,2,3) [1]	
		6156	4528	19
	2	(1,2,3) [1]	(2,3,4)[1]	
		3948	9677	80
	3	(2,3,4)[1]	(1,2,3,4)[1]	
7	1	4980	30855	85
		(6)	(1,2,3,4,5,6,7,	
	2	30,855	8,9,10,11,12,13)	
		(1,2,3,4,5,6,7,	9383	
		8,9,10,11,12,13)	(1,2,3,4,5,6,7,8,9,	246
	3	9383	10,11,12,13)	
		(1,2,3,4,5,6,7,8,	329	37
		9,10,11,12,13)	(8,3)	
8	1	3,130	31,723	47
		(2)	(1,2,3,4,5,6,7)	
	2	31,723	16512	43
		(1,2,3,4,5,6,7)	(1,2,3,4,5,6,7)	
	3	16512	18,557	298
		(1,2,3,4,5,6,7)	(1,2,3,4,5,6,7)	

for each *i*th cycle, *Mi* = total number of marked individuals up to the *i*th cycle; m*i* = number of marked individuals among captured termites; and n*i*= the number of termites captured

TABLE 3. Foraging population, foraging territory and linear foraging distance of *C. gestroi*

Site	Location	Population ± standard error (N ± SE)	Foraging territory (m^2)	Linear foraging distance (m)
1	Bayan Baru	1,566,164 ± 111,017	40	5
2	Mengkuang	1,608,967 ± 79364	40	15
3	Gelugor	1,974,252 ± 107,224	257	18
4	Kg.Guar Perahu	106,592 ± 6,968	20	4
5	Bayan Lepas	1,559,181 ± 412,641	13	6
6	Taman Rupawan	2,108,986 ± 210,858	257	28
7	Islamic Centre USM	1,367,952 ± 71,405	300	30
8	Plant House, USM	4,185,0026 ± 2127328	216	24

30 m. Each of the building structures was only infested with single termite colony. This study also showed that the triple mark recapture technique used to estimate the population size of the termite colony was capable of providing rough estimates of foraging population of *C. gestroi*.

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