

Tunneling Activity, Wood Consumption and Survivorship of *Coptotermes gestroi*, *Coptotermes curvignathus* and *Coptotermes kalshoveni* (Isoptera: Rhinotermitidae) in the Laboratory

by

Boon-Hoi Yeoh^{1,2} & Chow-Yang Lee^{1,3}

ABSTRACT

The tunneling activity, wood consumption and survivorship of three species of subterranean termites, *Coptotermes gestroi* (Wasmann), *C. curvignathus* Holmgren and *C. kalshoveni* Kemner were compared in the laboratory using two methods: petri dish and glass jar methods. The petri dish method consisted of a 15-cm petri dish containing 5% agar and 4 rubber wood blocks placed at positions of 0, 90, 180 and 270°. The second method consisted of a glass jar containing 200 g sand moistened with 40 ml distilled water and a piece each of rubber and pine wood on the top of the sand surface. Termites were released into the dish and jar, and allowed to tunnel freely for 28 days. At the end of the experiment, the tunneling activity was semi-quantitatively ranked, the set-up was disassembled, and termite survivorship and wood consumption rate were determined. Results suggested that *C. curvignathus* has the most active tunneling activity, followed by *C. gestroi*. On the other hand, *C. kalshoveni* has the least tunneling activity, but highest wood consumption rate, possibly showing a higher resource fidelity. The survivorship of the termites was >75% after the 28-day experimental period.

Key words: tunneling activity, *C. gestroi*, *C. curvignathus*, *C. kalshoveni*, wood consumption, survivorship.

INTRODUCTION

The genus *Coptotermes* consists of some of the most destructive subterranean termite species in the world. In Peninsular Malaysia, three *Coptotermes* species can be commonly found infesting buildings and structures, namely *C. gestroi*

¹Urban Entomology Laboratory, Vector Control Research Unit, School of Biological Sciences, Universiti Sains Malaysia, 11800 Penang, Malaysia.

²Present address: Rentokil Initial Singapore Pte Ltd, 16 & 18 Jalan Mesin, Singapore 368815.

³Corresponding author. Email: chowyang@usm.my.

(Wasmann), *C. curvignathus* Holmgren and *C. kalshoveni* Kemner. Among the three species, the former is the most economically important (Kirton & Brown 2003), while *C. curvignathus* has been reported as a forest pest that attacked tropical conifers (Kirton & Wong 2001) as well as an agricultural pest in rubber, oil palm, and coconut plantations.

Kirton & Azmi (2005) reported the varying prevalence of infestations of the three *Coptotermes* species in buildings and structures in Malaysia. We speculated that this could be due to differences in their tunneling activities and wood consumption rate. In this study, we compared the tunneling activity and wood consumption rate of these three species in the laboratory by using two methods, namely the petri dish and the jar methods.

MATERIALS AND METHODS

C. gestroi, *C. curvignathus* and *C. kalshoveni* were each collected from in-ground monitoring stations that were established earlier in Minden campus, Universiti Sains Malaysia, Penang Island. The collected termites were brought back to the laboratory and were separated from the soil debris according to the method described by Tamashiro *et al.* (1973).

Two methods were used to study the tunneling activity of the termites: the petri dish and glass jar methods. In the first method, 5% agar was poured into a petri dish (inner diameter: 15 cm) making a layer of tunneling medium (~ 1 - 1.3 cm). Four rubber wood blocks (*Hevea brasiliensis*) (Wild. ex A. Juss.) Muell. Arg. measuring 2 x 1 x 1 cm each were inserted into the agar layer before it became solid (partially buried) at positions of 0°, 90°, 180° and 270°. Two hundred workers (undifferentiated larvae of at least 3rd instar) and 10 soldiers were then introduced into the tunneling arena to allow free tunneling activities. For each termite species, experiment was replicated 10 times. Bioassay units were kept in a chamber (25.2 ± 0.2 °C, 56.3 ± 0.7 % RH) and left undisturbed for 28 days in complete darkness. Results were recorded using a scanner (Canon CanoScan LiDE20) on day 28. Tunnel formation was compared between termite species.

The numbers of tunnels entering each wood block were counted. Differences among the numbers of tunnels in 4 directions for each termite species were compared using one-way ANOVA and means were separated using Tukey's HSD. Intensity of tunnels formed was also ranked: 0 = no tunneling activ-

ity, 1 = tunneling activity throughout >0 - 25 % of the arena, 2 = tunneling activity throughout 26 – 50 %, 3 = tunneling activity throughout 51 – 75 % and 4 = tunneling activities throughout \geq 75 %. These ranks were statistically analyzed using the Kruskal-Wallis analysis of variance.

The second method, similar to that described by Grace *et al.* (2004), consisted of a glass jar (inner diameter: 5 cm x height: 10 cm) that was filled with 200 g of sieved (mesh 40) sterile sand moistened with 40 ml distilled water. A piece each of pre-weighted oven-dried rubber wood and pine wood (2 x 2 x 1 cm), layered with aluminum foil (3.5 x 4.0 cm), were placed side by side on the sand surface. Two hundred workers and 10 soldiers were then introduced into the test jar. Experiments were replicated 10 times for each termite species. All test jars were kept in a dark chamber (25.2 ± 0.2 °C, 56.3 ± 0.7 % RH) for 28 days. Tunnel formation facing the glass jar surface was observed at the end of the experiment. Total wood consumed for both wood types and termite survivorship were determined. Differences in total amounts of wood consumed by each termite species were determined using one-way analysis of variance (ANOVA) and means were separated by using Tukey's HSD. Differences in consumption rates of rubber wood and pine wood were compared using a paired-t test at $\alpha = 0.05$. Termite survivorship (in percentages) was transformed into arc-sine values and analyzed using one-way ANOVA and means were separated using Tukey's HSD. All statistical analyses were performed using Statistix 7.0 Analytical Software.

RESULTS AND DISCUSSION

There were distinct differences in the tunneling patterns of the three *Coptotermes* species. Based on visual observation, it was found that *C. curvignathus*'s tunnels were wide and highly branched, similar to those made by *C. gestroi*. On the other hand, *C. kalshoveni* showed the least tunneling activity, where most of its tunnels were straight and hardly branched (Fig.1). *C. curvignathus* was the most aggressive species, with significantly higher tunneling activity when compared to the other species (Table 1). However, both *C. gestroi* and *C. curvignathus* made similar numbers of entries into each wood block, compared to *C. kalshoveni*, which made the fewest tunnels. This suggested that *C. kalshoveni* may have higher resource fidelity. *C. gestroi* and *C. curvignathus* were observed to continue searching for other food sources even

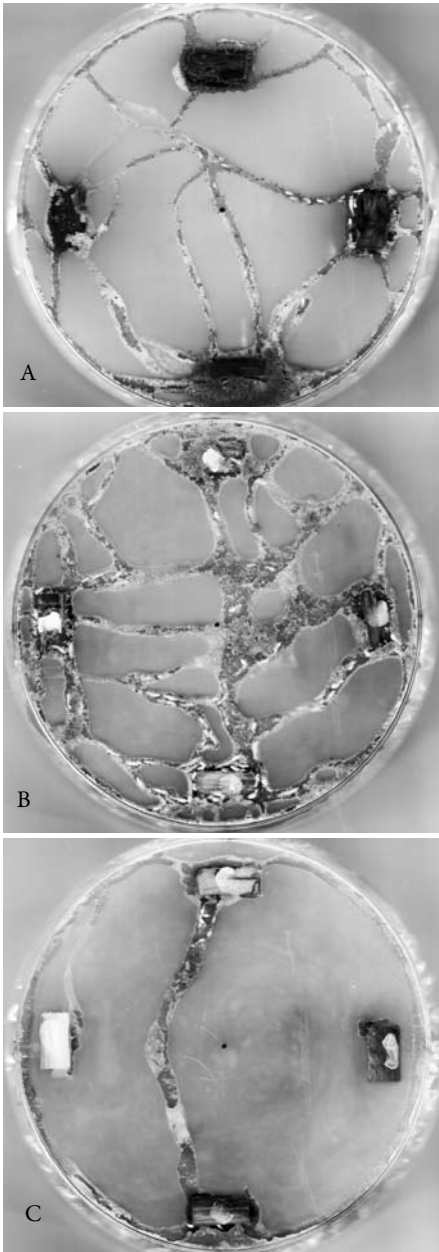


Fig.1. Tunneling activity of (A) *C. gestroi*, (B) *C. curvignathus*, and (C) *C. kalshoveni* in agar-filled petri dishes.

after they had encountered their first food source, and never abandoned any of the feeding sites during the experimental period. The aggressive searching pattern in these two *Coptotermes* may indirectly explain why these two species were more frequently found to be infesting buildings in Peninsular Malaysia, as reported by Kirton & Azmi (2005).

Despite the differences in the tunneling patterns, all three termite species foraged along the perimeter upon reaching the edge of the petri dish. This corresponded well with reports by Campora & Grace (2001) and Su (2005) that termites foraged in a relatively linear fashion along the edges of the test arena when no food attractant was available.

Termite survivorships were high (>75%) in all replicates. Agar served as a good tunneling medium in this study as it enabled clear observation of the tunneling activity, while at the same time providing sufficient moisture for the termites (Su *et al.* 1987). The problem of termites backfilling the tunnels (Tucker *et al.* 2004) can also be overcome with this medium.

The jar method was adopted in this study because it has been reported that *C. gestroi* survived well in a glass bottle with moistened sand (Sornnuwat *et al.* 1995). Haverty (1979) also suggested

Table 1. Tunneling activity and numbers of tunnels entering each wood block of *Coptotermes curvignathus*, *Coptotermes gestroi* and *Coptotermes kalshoveni*, after a 28 day evaluation period in the petri dish method (Mean ± SE, n=10)

Termite species	Mean (± SE) ranking for tunneling activity ¹	Mean (± SE) tunnels entering each wood block ²
<i>Coptotermes gestroi</i>	2.1 ± 0.2 b	5.5 ± 0.3 a
<i>Coptotermes curvignathus</i>	4.0 ± 0.0 a	5.5 ± 0.2 a
<i>Coptotermes kalshoveni</i>	1.0 ± 0.0 c	1.8 ± 0.2 b

¹Tunneling activity: 0 % area = 0, 1-25 % area = 1, 26-50 % area = 2, 51-75 % area = 3, 76-100 % area = 4. Means followed by same letters were not significantly different at α = 0.05, Kruskal-Wallis Test.

²Means followed by same letters were not significantly different, p > 0.05 (Tukey HSD).

the use of sand and water mixture for short-term testing of termites. In this method, all three termite species did not have any systematic tunnel formation pattern (Fig. 2). This method, however, confirmed our findings with the petri-dish method that *C. gestroi* and *C. curvignathus* were more aggressive species than *C. kalshoveni*. *C. gestroi* and *C. curvignathus* tunneled ~90 % of total area, making numerous branches, with most of the tunnels interconnected to one another.

C. curvignathus showed equal affinity towards rubber and pine woods (Table 2). The result was consistent with that reported by How (2004). However, other researchers have reported that *C. curvignathus* preferred pine over rubber wood (Ngee *et al.* 2004). The variation may due to the differences in termite sample size, medium and colony used in these studies. In contrast, *C. gestroi* and *C. kalshoveni* significantly preferred to feed on rubber wood (wood consumption = 0.1902 ± 0.0190 g and 0.3086 ± 0.0302 g, respectively) than pine wood (wood consumption = 0.0979 ± 0.0075 g and 0.1203 ± 0.0275 g, respectively). Ngee *et al.* (2004) also found 2 out of 3 colonies of *C. gestroi* that they evaluated to prefer rubber over pine woods.

C. kalshoveni had a significantly higher wood consumption rate in the glass jar method, which supports our suggestion that this species has higher resource fidelity. Hedlund & Henderson (1999) had reported that termite wood consumption rate was inversely related to tunneling volume. This observation corresponded well with that observed in this study where *C. kalshoveni* had the highest total wood consumption among the three *Coptotermes* species,

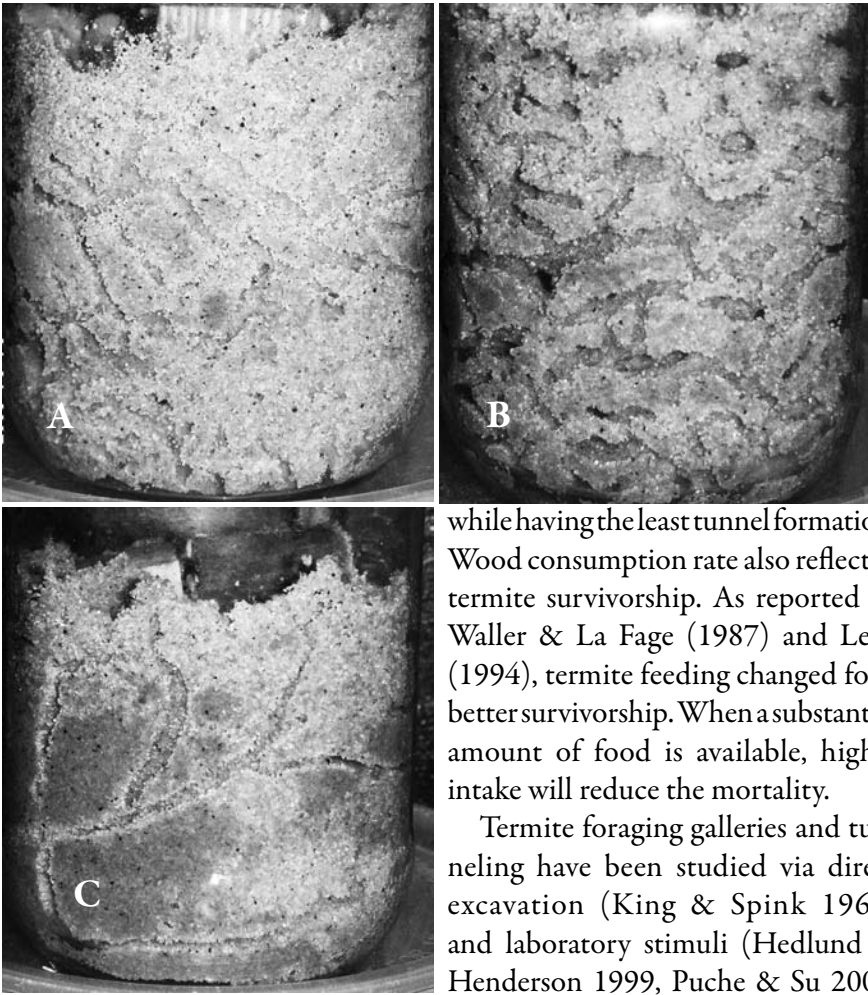


Figure 2 Tunneling activities of (A) *C. gestroi*, (B) *C. curvignathus*, and (C) *C. kalshoveni* in sand-filled glass jars.

while having the least tunnel formation. Wood consumption rate also reflected termite survivorship. As reported by Waller & La Fage (1987) and Lenz (1994), termite feeding changed for a better survivorship. When a substantial amount of food is available, higher intake will reduce the mortality.

Termite foraging galleries and tunneling have been studied via direct excavation (King & Spink 1969) and laboratory stimuli (Hedlund & Henderson 1999, Puche & Su 2001, Su 2005). These studies showed that termites formed intensive searching or branching patterns at the beginning of food-searching activity (Robson *et al.* 1995, Hedlund & Henderson 1999, Campora & Grace 2001, Cornelius & Osbrink 2001, Puche & Su 2001, Grace *et al.* 2004). This branching strategy is energy-saving, while increasing the probability of encountering possible food sources (Hedlund and Henderson 1999). Often, termites followed the direct route once they had located the food source (Hedlund & Henderson 1999) and enlarged the gallery (King

Table 2: Wood consumption and survivorship of *C. gestroi*, *C. curvignathus* and *C. kalshoveni* and their survivorship (Mean ± SE) after a 28 day experimental period by using the jar method (n=10).

Termite species	Wood consumption [g] (Mean ± SE) ¹		Mean (± SE) ranking for tunneling activity ²	Survivorship, % (Mean ± SE) ³
	pine	rubber		
<i>Coptotermes gestroi</i>	0.0979 ± 0.0075 a (a)	0.1902 ± 0.0190 b (a)	3.8 ± 0.1 a	76.35 ± 2.84 b
<i>Coptotermes curvignathus</i>	0.1787 ± 0.0268 a (a)	0.2357 ± 0.0163 a (ab)	3.9 ± 0.1 a	81.15 ± 2.02 ab
<i>Coptotermes kalshoveni</i>	0.1203 ± 0.0275 a (a)	0.3068 ± 0.0302 b (b)	1.3 ± 0.2 b	83.55 ± 1.24 a

¹ Means in the same row followed by same letters were not significantly different at $\alpha = 0.05$ (Paired-t test). Means in the same column (in parentheses) followed by same letters were not significantly different, $p > 0.05$ (Tukey HSD).

² Tunneling activity: 0 % area = 0, 1-25 % area = 1, 26-50 % area = 2, 51-75 % area = 3, 76-100 % area = 4. Means followed by same letters were not significantly different ($p > 0.05$, Kruskal-Wallis multiple range test).

³ Means in % were transformed to arcsine values before analysis of variance, and were later separated with Tukey HSD. Means followed by same letters were not significantly different, $p > 0.05$.

& Spink 1969, Robson *et al.* 1995, Reinhard *et al.* 1997, Campora & Grace 2001, Cornelius & Osbrink 2001, Puche & Su 2001, Swoboda & Miller 2004) by further recruitments (Ettershank *et al.* 1980).

In summary, this laboratory study found: (1) The tunneling activity for the three *Coptotermes* species: *C. curvignathus* > *C. gestroi* > *C. kalshoveni* (most to least active), (2) *C. kalshoveni* showed the fewest tunneling branches, (3) Wood consumption rate [rubber wood]: *C. gestroi* ≥ *C. curvignathus* ≥ *C. kalshoveni*, (4) Survivorship of >75% after a 28 day experimental period.

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