The effect of bait design on bait consumption in

termites (Isoptera: Rhinotermitidae).

Theodore A. Evans and Patrick V. Gleeson

CSIRO Entomology, Canberra ACT 2601 Australia

Abstract.

The efficacy of baiting as a pest control method relies on the bait appealing to the pest species. In the case of wood-eating termites, bait stations should be designed to encourage termite presence and to maximize their consumption of bait matrix in order to expedite control in minimal time. A field experiment examined the effect of bait size (one large bait or four small baits of equivalent total size, with commensurate inspection and replacement schedules), compaction (tightly rolled or loosely folded) and composition (paper only or paper plus wood) on termite presence and on untreated bait paper removal rates over four months in a field experiment. All three factors were significant, with bait size the most important factor, followed by compaction and then composition. The least effective baits were small, compacted (rolled) paper-only baits with monthly inspections; these had the highest abandonment rate (70%) and had the least paper removed (mean of 24g). The most effective baits were large, folded paperplus-wood baits with inspections at two months; these had the lowest abandonment rate (20%) and had the highest paper removal (mean of 112g). The more than four-fold difference between these baits types demonstrates that bait efficacy can be altered considerably merely by changing bait design without adding new ingredients to the bait matrix.

1 Introduction

2 Baiting has been promoted as a desirable method of termite pest control. It is lauded as 3 environmentally sound as it uses very small amounts of insect specific toxicants that are 4 administered in localized baits that are targeted at the pest species (i.e. not large amounts of 5 toxicants spread over large areas around a house). However, in order for baiting to work 6 successfully termites must find and consume the bait matrix and for the toxicant contained 7 therein to be transferred back to the nest. These requirements are not inconsequential: a 8 successful baiting program can take up to nine months (e.g. 3-9 months Su, 1994; 7 months 9 Tsunoda et al., 1998; 3-7+ months Su & Scheffrahn, 2000), which is much slower control than 10 that provided by other methods.

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12 How can control using baiting be accelerated? A good understanding of termite foraging 13 behaviour, and in particular food preferences, may contribute to the design of a bait station 14 'optimized' for termite foraging. 'Optimized' in this context means that the preferred termite 15 food is presented in the most appealing fashion so that termites collect the bait matrix and return 16 it to their nest faster, which in turn should increase the speed at which control occurs. It is 17 important to put such activity in the context of total foraging activity of a termite colony as the 18 bait stations will be in competition with other food sources for attention from foragers 19 (Forschler, 1996; Perrott et al., 2004).

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Work has been conducted on termite food and forging preferences (e.g. Howard & Haverty,
1979; Rust *et al.*, 1996; Suoja *et al.*, 1999), yet little of this appears to have been included into
bait station design. This is because most economically important, wood-eating termite species

1 evolved to eat relatively large pieces of timber (which is the reason why these species are pests), 2 including whole trees (e.g. Coptotermes species; Hill, 1942; Greaves, 1962, 1967; Lenz, 1994), 3 however most bait systems are much smaller than natural food resources and typically do not 4 include wood. Instead, either paper, cardboard or purified cellulose powder, are offered as a bait 5 matrix inside various plastic bait stations, both those manufactured commercially (e.g. Su, 1994; 6 Tsunoda et al., 1998; Su & Scheffrahn, 2000) and otherwise (e.g. see French et al., 1995). It is 7 of interest to note that such materials are not accepted uniformly (e.g. toilet paper LaFage et al., 8 1973; French & Robinson, 1980).

9

10 From the perspective of the termites, inspection and replacement of baits creates another, 11 possibly important, deviation from foraging on natural foods. In natural habitats, termites 12 feeding inside of large pieces of timber would not be disturbed frequently, whereas relatively 13 small bait stations require frequent, typically monthly, inspections to maintain supply of the bait 14 matrix. There are no published data that demonstrate how bait station design and the inspection 15 regime, commercial or otherwise, affect termite foraging and therefore whether bait station 16 design and the inspection regime has been 'optimized' for any bait (see Lenz & Evans, 2002 for 17 a discussion).

18

Therefore, given the differences between natural foods and bait systems, the possibility of altering bait station design so as to increase bait matrix removal (consumption) and so accelerate control may be great. This study examined the effects of three bait station factors: bait size (and related inspection frequency), compaction and composition, on termite foraging at artificial bait stations. The study aimed to determine the combination of these factors that would have the

least negative effect on termite presence in baits and the most positive effect on matrix removal,
 in order to determine the optimized design of termite bait stations.

3

4 Methods

5 The field site was in the Brindabella Mountains of the Australian Capital Territory (35°17' S, 6 149°13' E, elevation c. 800m, c. 30 km W of Canberra). The topography varies from mild to 7 steep slopes, the native *Eucalyptus* forest of *E. dalrympleana* Maiden, *E. delegatensis* R.Baker 8 and *E. fastigata* Deane and Maiden (Boland *et al.*, 1992) was cleared originally for grazing but 9 now has been replaced with the introduced *Pinus radiata* D. Don (Pinaceae) plantations for 10 wood production. The endemic *Coptotermes lacteus* (Froggatt) has adapted to the pine 11 plantation and mound-colonies are common.

12

13 This experiment was designed to test whether bait station design affected bait matrix removal, 14 not whether bait design affected bait discovery. Therefore it was decided that bait stations 15 should be placed onto active foraging sites and thereby be encountered immediately. This bait 16 placement on active feeding sites (as suggested by French 1991) is similar to commercial "above 17 ground bait stations" (Su et al., 1997). The foraging sites were created along trenches that 18 encircled 10 termite mounds. These were 10 cm wide x 25 cm deep x 10 m \emptyset , lined with pieces 19 of *P. radiata* and *E. regnans* F. Muell. (preferred timbers; Gay *et al.*, 1957). The foraging sites 20 were milk cartons (opened one litre waxed paper cartons, 24 x 7 x 7 cm, Southcorp Packing 21 Australia), holed eight times on four sides (2 cm \emptyset), filled with corrugated cardboard and 22 wooden slats (all 24 x 5 x 0.7 cm) (after Evans, 2002). These 'carton-bases' were buried (top 23 flush with soil surface, wood wetted, covered with a transparent plastic lid (7 x 8 cm) and opaque plastic sheet (40 x 40 cm) and soil) and spaced evenly around the circular trench (ca. 1.6 m
 intervals).

3

The carton-bases were interred in March 1997 and left undisturbed over winter (when foraging
activity is low) and inspected (lifting the opaque plastic sheet but not the transparent plastic lid)
in November 1997. Termites had contacted between 16 and 20 milk cartons at each moundcolony and were foraging actively in them; this was seen through the transparent plastic lids that
covered the milk cartons. This contact rate was sufficient for the experiment to commence and
coincided with summer, the height of foraging activity (Evans & Gleeson, 2001).

10

11 Three different bait attributes were tested. The first was size and commensurate inspection 12 frequency. Large baits had 144 g of paper (14 sheets of 490 x 405 mm Versatowel (a 13 multipurpose medical towel; Kimberley Clark Australia). These were placed once, at the 14 beginning of the experiment, and left as undisturbed as possible for the four months of the 15 experiment. Small baits were one quarter the size of large baits, 36 g of paper (3.5 sheets of 16 Versatowel). One small bait each was placed on the appropriate carton bases at the beginning of 17 the experiment, and these were inspected approximately monthly for four months, with a new 18 small bait placed each month. Thus carton bases with small baits received a total of 144 g of 19 paper, the same amount as for large baits, over the four month period.

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21 The second attribute was compaction, so termed because the paper was either rolled tightly or
22 folded loosely (concertinaed). Paper rolled tightly has a minimal volume and surface area,
23 which may be useful for reasons to do with manufacturing, storage and transport, but minimizing

1 the volume and surface area means that fewer termites can gain access to the paper, and this may 2 reduce consumption rate. Paper folded loosely will maximize edges and surfaces onto and into 3 which greater numbers of termites could crawl, and so may increase consumption rate. 4 5 Composition was the third attribute: paper-only or paper-plus-wood (E. regnans). The wood 6 was not considered to be part of the bait matrix and only paper removal (consumption) was 7 measured and analysed. Wood was present to determine simply whether an additional natural 8 food source increased termite activity. Large baits had two large slats (24 x 5 x 0.7 cm) and 9 small baits had one short slat $(12 \times 5 \times 0.7 \text{ cm})$ (one quarter the wood) in the plus-wood 10 treatment. 11 12 Three treatments created eight treatment combinations: small, rolled, paper-only; small, folded, 13 paper-only; small, rolled, paper-plus-wood; small, folded, paper-plus-wood; large, rolled, paper-14 only; large, folded, paper-only; large, rolled, paper-plus-wood; and large, folded, paper-plus-15 wood. Large bait treatment combinations were housed in whole milk cartons (as for carton-16 bases, above), holed eight times on two sides (all holes $2 \text{ cm } \emptyset$). The small bait treatment 17 combinations were housed in half milk-cartons (12 cm long), holed four times on two sides (see 18 Figure 1). 19 20 Two replicates of each treatment combination were placed at random on the contacted carton-

bases around each mound (Figure 1); a total of 20 replicates of each treatment combination on

21

22 (20 January 1998). The clear plastic covers were removed and the baits placed on top of the

23 carton base, and the plastic sheets were replaced on top. Termite activity and the estimated

1	amount of paper removed from the small baits were recorded at each inspection (25 February =
2	36 days post placement, 27 March = 66 days, 24 April = 94 days, 16 June = 147 days). Small
3	baits were inspected by lifting the opaque plastic cover, lifting the baits, and examining the
4	paper. Once the large baits were placed they were not disturbed, except for a brief inspection
5	(slightly lifting the opaque plastic cover) at two months (27 March = 36 days) and for the final
6	inspection at four months (16 June = 147 days). At this time all baits were collected and dry-
7	weighed in order to calculate total weight of bait removed. Twenty carton-bases were left
8	completely undisturbed to measure abandonment caused by bait placement and inspection.
9	
10	Termite activity was monitored for carton-bases and baits; abandonment of bases and baits was
11	analysed using chi-squared tests (Sokal & Rohlf, 1995). The total amount of paper removed (i.e.
12	cumulative paper removed at four months) was compared by three-way ANOVA with bait size,
13	compaction and composition as factors. All posthoc comparisons were done with Bonferroni
14	adjustments. Possible differences in paper removal between mound-colonies were examined
15	with one-way ANOVA. (n.b. 'Removal' is used instead of 'consumption' throughout the text as
16	bait matrix that has been removed from the bait station is not necessarily consumed (i.e. eaten by
17	the termites) – it can be used for construction and it might be stored for long periods prior to
18	consumption). All analyses were performed with Systat 9 (SPSS Inc. 1996).
19	

Results

The number of undisturbed bases with termite activity by the end of the experiment is an
important measure against which all bait treatments should be compared – it indicated the level
of abandonment without disturbance from baiting. Only one base of 20 was abandoned and then

only by the final inspection; a rate of 5% after four months (Table 1), and this was due to the wood in the carton base being eaten completely by this time. Therefore, it seems appropriate to consider that the termites would continue to forage in the bases while wood was available and therefore any abandonment of bases with wood was likely to be due to disturbance from baiting.

5

Bases used for baiting were abandoned over the course of inspections: 9 of 160 bases (5.6%) had been abandoned by 27 March and this doubled to 20 bases (12.5%) abandoned by 16 June (Table 1). Therefore, bait placement caused ~ 8.5% abandonment of established feeding sites. Bait placement, not bait type, appeared to be the cause, as there were no significant differences found between treatments (16 June data): small baits compared with large (12 : 8, $\chi^2_1 = 0.80$, P =0.37), rolled baits versus folded (13 : 7, $\chi^2_1 = 1.80$, P = 0.18); paper-only versus plus-wood baits (10 : 10, $\chi^2_1 = 0.0$, P = 1.0).

13

14 Baits were abandoned more frequently than bases over the course of inspections: on the second inspection (27 March) 65 of 160 baits (41%) had been abandoned, whereas by the fourth and 15 16 final inspection (16 June) this had increased to 75 baits (47%). In general, the termites sealed off 17 the bases from the baits, and the baits were then ignored. However, baits could be re-attacked, and consequently the pattern differed slightly between bait-treatments from March to June. In 18 March, significantly more small than large baits were abandoned (41 : 24, $\chi^2_1 = 4.45$, P = 0.035), 19 but other bait variables were not different (rolled : folded, 37 : 28, $(\chi^2_1 = 1.25, P = 0.264, \text{ and})$ 20 paper-only : plus-wood, $38 : 27\chi^2_1 = 1.86$, P = 0.172). By June these patterns were more marked 21 and were significant for small and large baits (57 : 18, $\chi^2_1 = 20.28$, P < 0.001) and rolled versus 22 folded also (49 : 26, $\chi^2_1 = 7.05$, P = 0.008), but there was no significant difference between 23

1 paper-only and plus-wood (34 : 41, $\chi^2_1 = 0.65$, P = 0.419), due to the increased abandonment on 2 small baits (Table 1, Figure 2).

3

4 The total (cumulative) amount of paper removed after four months (at the end of the experiment) 5 follows the patterns found for bait abandonment (Figure 3). The three-way ANOVA of total 6 paper removed found that no interaction terms were significant, therefore the main treatments 7 could be considered separately. All three were significant, with size being the most important (large vs small $F_{1,152} = 38.3$, P < 0.001), followed by compaction (folded vs rolled $F_{1,152} =$ 8 6.78, P = 0.010) and then composition (paper-only vs plus-wood $F_{1,152} = 4.26$, P = 0.041). The 9 analysis explained 25% of the variability observed ($r^2 = 0.25$), therefore other factors were 10 11 important. 12 13 Colony differences were important also: colonies removed significantly different amounts of paper ($F_{9,150} = 8.81$, P < 0.001), and colony differences in paper removal explains 35% of the 14 variability observed ($r^2 = 0.35$). In general, colonies 4, 8 and 10 removed significantly less paper 15

16 (means of 20.6g, 16.6g and 24.5g respectively), which is explained by the reluctance of termites

17 from these colonies to come out of the bases and attack the baits (Figure 4). Colonies 3, 6 and 9

18 removed significantly more paper (means of 98.8g, 108.5g and 104.9g respectively), which

19 corresponded with a greater number of baits attacked (Figure 3).

20

21 **Discussion**

22 Of the three attributes tested in this study, bait size (with commensurate inspection frequency)

and compaction were consistently important in affecting bait consumption. More of the paper in

the large baits was removed than that in the small baits, more paper that was folded loosely was removed compared with that which was tightly rolled. The composition attribute, i.e. the presence of wood, had a small but discernable effect, with more paper being removed in baits with wood compared with those without. These effects are relatively simple to explain.

5

6 Large baits allowed termites to chew the bait at their own rate; and several studies have shown
7 that termites (in the laboratory) will vary their consumption of food inversely with the size of the
8 food – i.e. they eat small food more slowly (Lenz, 1994; Hedland & Henderson, 1999). Given
9 that the results of this study have a consistent pattern with those of the laboratory studies,
10 perhaps this effect is found in the field also? Alternatively, and perhaps more plausibly, the
11 termites were not delayed by the monthly inspection regime, as they would have been when they
12 were fed small baits.

13

14 Another advantage of the large baits is that they were placed only once, at the beginning of the 15 study. Therefore they did not need to be inspected and have more bait placed each month, thus 16 decreasing the disturbance to the foraging termites, compared with the small baits. It is well 17 known that termites dislike disturbance and will avoid it if the levels are high (Lai, 1977; Jones, 18 1991; French et al., 1995), but unfortunately disturbance levels have not been quantified. Of 19 course the present study measured the combined effects of disturbance (from inspections) and 20 bait size, and therefore cannot quantify the disturbance effect separately. Nevertheless, the 21 results from this experiment suggest that disturbance is an important effect for baiting and that 22 minimizing it is likely to improve bait removal.

1 The success of loosely folded paper has one simple explanation; the greater surface area of paper 2 immediately available for gnawing. The small increase in paper removal rates with added wood 3 has two possible explanations. The first is that wood, and especially *E. regnans*, is highly 4 palatable for C. lacteus and so might attract more termites into the bait. The second is that the 5 wood was not eaten as quickly, and therefore if the termites had removed all the paper in a small bait before the next inspection, then the termites would remain in the bait eating the wood. The 6 7 latter reason may be the correct one as termites generally abandoned baits when all the paper had 8 been removed, typically sealing the carton-bases with mudding.

9

10 Termites abandoned many baits but few bases as a consequence of the disturbance suffered 11 during bait placement and inspection, especially for the small baits (Figure 2). This phenomenon 12 would be of great concern in a real baiting program in a house, following the analogy that the 13 baits of the current study were functionally similar to "above ground" baits (French, 1991; Su et 14 al., 1997). Not only would this suggest that termites would not be consuming the bait matrix, 15 they might continue to forage on wood in the house. It is possible that some people might 16 interpret a reduction in termite activity in baits to mean colony reduction or elimination when in 17 fact the termites have simply abandoned baits that are too small or inspected too frequently.

18

The results of this study show that a change in the presentation of materials in the bait station can have large effects on bait matrix removal. No new ingredients were necessary to increase paper removal four fold after only four months, if small, paper-only, rolled baits are compared with large, plus-wood, folded baits. This is a small step towards bait station optimization. There are many termite pest species in many different countries against which baits are deployed. The

results for *C. lacteus* in this study may not be applicable to other species in other situations. That said, further consideration of bait station design from the perspective of the termite, especially designs that increase bait size and minimize disturbance, is likely to produce increases in bait matrix removal, and so further advances in optimization and efficacy of bait stations. Evidently, much more work on food and forging preferences is required to design more efficacious baiting protocols.

Acknowledgments

We thank Michael Lenz for his help throughout this study and for his helpful comments that improved the original manuscript. We also thank ACT Forests for allowing us to have access to their properties.

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Table 1. The number of bases and baits (X, Y) with *Coptotermes lacteus* activity at each inspection. N.b. initially there were 20 bases per bait treatment combination (20/1); large baits were not inspected in February or April. Undisturbed bases did not receive baits and are indicated with a dash.

Bait:			Inspection date:				
Size	Composition	Compaction	25/2	27/3	24/4	16/6	
Large	Paper only	Folded	-	20, 13	-	18, 16	
		Rolled	-	17, 12	-	16, 13	
	Plus wood	Folded	-	19, 17	-	20, 18	
		Rolled	-	18, 14	-	18, 15	
Small	Paper only	Folded	19, 12	20, 10	20, 12	19, 8	
		Rolled	18, 9	18, 7	18, 3	17, 2	
	Plus wood	Folded	20, 14	20, 12	19, 11	16, 12	
		Rolled	19, 15	19, 10	16, 7	16, 1	
Undisturbed bases			20, -	20, -	20, -	19, -	

Figure legends

Figure 1. Schematic of experimental setup and apparatus: (a) *Coptotermes lacteus* mound colony and circular trench with (b) enlargement of 'carton-bases' viewed looking along the trench; two examples of baits (c) large, folded, paper-plus-wood in a whole milk carton and (d) small, rolled, paper-only in a half milk carton. See text for quantities and measurements.

Figure 2. The number of bases and baits with *C. lacteus* activity at the final inspection (fourth inspection, June) separated according to bait treatment combination. Open columns indicate bases, filled columns indicate baits.

Figure 3. The average $(\pm SE)$ cumulative amount of paper removed (g) from the bait treatment combinations, at the end of the experiment (fourth inspection, June).

Figure 4. Average (± SE) total quantity of paper (i.e. from all baits of all treatment combinations) removed (g) from each colony of *C. lacteus* at the end of the experiment. The top number is the number of baits attacked during the experiment (maximum of 44: 8 large baits and 32 small baits), the bottom number is the number of infested bases used for baits (maximum of 16).















