

Faculty of Forestry, University of Toronto, Toronto, Ontario, Canada

Termiticidal activity of boron dusts (Isoptera, Rhinotermitidae)

By J. K. GRACE and A. ABDALLAY

Abstract

Mortality of *Reticulitermes flavipes* (Kollar) workers was evaluated 1, 2, 4, and 8 days after exposure to boron dusts. Mortality was most rapid after exposure to barium metaborate (Busan 11-M1), followed by boric acid. Exposure to either of these two dusts resulted in 100% mortality within 8 days. Neither sodium borate (borax) nor zinc borate elicited significant mortality, and increasing the exposure period from 1 to 10 minutes had no effect on termite mortality. In assays simulating field conditions, treatment of 10% of the termite population with boric acid or barium metaborate dusts elicited high mortality. Mortality was reduced, although still significant, when 5% of the population was treated. Field collection of subterranean termites in traps, treatment of captured termites with an insecticidal dust, and subsequent release back into the colony may be an effective method of control, although these results indicate that a relatively large proportion of the colony would have to be exposed to boric acid or barium metaborate dusts.

1 Introduction

Prior to the development in the 1940's of organic insecticides and modern fumigants for termite control, inorganic insecticidal dusts were commonly used in the control of both drywood termites (Kalotermitidae) living in structural timbers, and soil-dwelling termites (Rhinotermitidae and Termitidae) occupying subterranean gallery systems (KOFOID and WILLIAMS 1934; RANDALL and DOODY 1934). In this technique, the dust is injected into a portion of the gallery system, spread to other portions of the system on the bodies of termites coming in contact with it, and passed among colony members by grooming behaviour. The highly toxic compound arsenic trioxide is still used for this purpose in several countries (EDWARDS and MILL 1986; FRENCH 1988; LIN 1987).

In the past decade, concerns over possible adverse effects on human health from the extensive application of persistent organic insecticides and organic fumigants have stimulated interest in the development of alternative termite control techniques, such as the use of toxic baits. Bait development has proceeded slowly, largely due to the difficulty of identifying an appropriate toxicant (FRENCH 1988; LA FAGE 1984).

This study was initiated to determine whether insecticidal dusts might be applicable to a bait method of subterranean termite control. Small traps designed for collecting subterranean termites at urban sites (GRACE 1989a; SU and SCHEFFRAHN 1986) are suitable for collecting large numbers of insects. Theoretically, captured termites could be coated with a dust and released back into the colony through these same traps, in a manner analogous to the release of dyed termites in mark-release-recapture studies of termite populations (GRACE et al. 1989). Several boron compounds were chosen for evaluation because of their low mammalian toxicity and demonstrated toxicity to termites (GRACE 1989b; RANDALL et al. 1934; WILLIAMS and AMBURGEY 1987). Such relatively safe compounds might also be injected into the channels within infested wood in efforts to control above-ground termite infestations.

2 Materials and methods

2.1 Collection and bioassay conditions

Eastern subterranean termites, *Reticulitermes flavipes* (Kollar) (Rhinotermitidae), were collected in traps consisting of short lengths of plastic pipe containing rolls of corrugated paper (GRACE 1989a), buried just below the soil surface at a site in Scarborough, Ontario. Termites collected while feeding on the corrugated paper (ca 1,000–10,000 per trap) were kept in plastic boxes in an unlighted incubator at $27 \pm 0.5^\circ\text{C}$ and $90 \pm 5\%$ relative humidity. Bioassays employed termite workers, undifferentiated individuals older than the third instar as determined by size, and were also performed in this incubator.

Borates were evaluated in the form of undiluted commercial powders: *ortho*-boric acid (Fisher Scientific, Ontario), sodium borate (disodium tetraborate decahydrate) (Borax, Drug Trading Co. Ltd., Ontario), zinc borate (ZB-325, Humphrey Chemical Corp., Maryland), and barium metaborate (90+ % active barium metaborate monohydrate) (Busan 11-M1, Buckman Laboratories of Canada Ltd., Quebec). Termite workers were placed in a 5 cm diameter glass petri dish containing a thin layer of powder (permitting walking), and the dish shaken once to coat the insect with the powder. The insects were then allowed to remain in the dish for either a 1 or 10 min interval, and the dish shaken once more before transferring the worker with a small brush to an assay vial. Termites used in the control assays were treated similarly, but with no powder in the petri dish.

Mortality of termites in the treatment and control groups in each experiment were subjected to analysis of variance with a one-way layout, and means significantly different at the 0.05 level were separated by the Ryan-Einot-Gabriel-Welsch multiple F test (SAS Institute 1987).

2.2 Comparative toxicity of borate compounds

Three groups of 20 *R. flavipes* workers per treatment were exposed to either boric acid, sodium borate, zinc borate, or barium metaborate powder for a one minute interval. Three control groups of workers were handled similarly in clean petri dishes. Each group of 20 termites was then placed in a 44.8 ml polystyrene vial (60 × 36 mm diameter) containing 7 ml (ca 10 g) sand, a ca 2 × 6 cm strip of Whatman No. 1 filter paper as food, and 1.5 ml deionized water. These vials were capped with polyurethane foam plugs and placed in the incubator.

Cumulative mortality in each group of workers was evaluated after 1 day (24 h), 2 days (48 h), and 8 days. To minimize boron accumulation in the sand in the assay vials, surviving workers in each group were transferred to newly-prepared vials after each evaluation.

2.3 Effect of exposure time on toxicity

Using the methods described above, 3 groups of 20 workers per treatment were exposed to boric acid, sodium borate, zinc borate, or barium metaborate powders for either 1 min or 10 min. Following this exposure, each group was transferred to an assay vial and mortality evaluated after 1 day (24 h) and 4 days. Again, termites surviving 1 day were transferred to newly-prepared vials to minimize boron accumulation in the sand.

2.4 Simulation of field treatments

Two bioassays were designed to assess the effect of introducing *R. flavipes* workers coated with borate dusts into a larger, untreated, termite population. In the first assay, 6 groups of 18 workers per treatment were placed in assay vials containing sand, water, and filter paper, prepared as described above. These untreated termites were allowed to acclimate to the vials in the incubator for 24 h, at which time tunnelling was clearly visible in all vials. After the 24 h acclimation period, 2 workers (10 % of each test group) exposed for 1 min to either boric acid or barium metaborate (or handled similarly as a control) were added to each vial. Total group mortality was independently evaluated (3 vials per time per treatment) 8 and 16 days after introduction of the dust-treated workers.

The second field-simulation assay employed an apparatus originally designed to evaluate termite tunnelling behaviour (GRACE, in prep.) to provide greater physical separation of the test population from the site of introduction of the dust-treated workers. This apparatus consists of 3 serially connected compartments: two assay vials are connected by 1 cm lengths of tygon tubing inserted through holes drilled in the side of each vial (below the surface of sand in the vial) to either end of a 7.5 cm long glass microscope slide "sandwich". This centre sandwich separating the two vials is composed of two standard microscope slides, spaced 3–4 mm apart, with the ends sealed with plastic spacers and silicone caulking. The sandwich is placed horizontally in an upright position, with the bottom long edge secured by silicone caulking to a third flat glass slide, then filled with sand (ca 8 g), and the top long edge sealed with plaster of paris. A strip of Whatman No. 1 filter paper is placed in

the first assay vial as a feeding substrate, and a small block of red pine (*Pinus resinosa* Ait.) wood decayed by the fungus *Gloeophyllum trabeum* (Pers. ex Fr.) Murr. is placed on the surface of the sand in the second vial. Termites placed in the first vial thus have food (filter paper) available, but must pass through the first tygon tube, tunnel through the 7.5 cm sandwich, and then pass through the second tygon tube to reach the vial containing the preferred decayed wood.

In this second assay, 38 *R. flavipes* workers were placed in the first vial, and allowed a 24 h acclimation period to explore the apparatus. Only groups successful in tunnelling through the sandwich and reaching the decayed wood in the second vial were used in the subsequent assay. Two workers (5% of the total test group) exposed for 1 min to either boric acid or barium metaborate were then added to the vial containing the decayed wood, simulating the introduction of dusted termites at one location in the large gallery system characteristic of *R. flavipes* field colonies (GRACE et al. 1989). As in the first field-simulation assay, mortality was independently evaluated (3 groups per time per treatment) 8 and 16 days after introduction of the dust-treated insects.

3 Results and discussion

Twenty-four hours after exposure to the boron dusts, independent of the length of exposure, only those *R. flavipes* workers exposed to barium metaborate suffered significant mortality (table 1, 2). However, two days after dust exposure, mortality among those exposed to boric acid, although less than that of the barium metaborate group, was also significant. Four days after exposure, the same relationship was apparent, although mortality continued to increase in both the barium metaborate and boric acid groups (table 2). Complete mortality was recorded eight days after exposure to these two dusts (table 1).

Table 1. Cumulative percent mortality (mean \pm SD) of *R. flavipes* workers following a 1 min exposure to boron dusts¹

Chemical	Cumulative % mortality		
	1 day	2 days	8 days
Boric acid	5.0 \pm 5.0b	15.0 \pm 5.0b	100.0 \pm 0a
Sodium borate	0 \pm 0b	3.3 \pm 2.9bc	56.7 \pm 23.6b
Zinc borate	3.3 \pm 5.8b	5.0 \pm 8.7bc	50.0 \pm 32.8b
Barium metaborate	33.3 \pm 16.1a	56.7 \pm 2.9a	100.0 \pm 0a
Control	0 \pm 0b	1.7 \pm 2.9c	13.3 \pm 7.6b

¹ Each mean represents 3 groups of 20 workers. Means in each column followed by the same letter are not significantly different at the 0.05 level.

Neither sodium borate (borax) nor zinc borate elicited significant mortality within eight days (table 1), independent of the length of the exposure period (table 2). The reduced solubility in aqueous solution of these two borates, in comparison to that of boric acid, may result in less boron being made biologically available to the insects. Although barium metaborate is only 0.3% soluble in water, the acute toxicity of barium (BUCKMAN Laboratories 1983) may have contributed to the high mortality observed in these assays. Although no information is available on the toxicity of barium alone to *R. flavipes*, KOFOID and WILLIAMS (1934) reported barium chloride dust to be more toxic than either boric acid or sodium borate to *Zootermopsis angusticollis* (Hagen). These authors also reported sodium borate dust to be more toxic than boric acid, but this was likely due to differences in assay procedures. In their assay, the insects were placed directly on damp filter paper, which could leach the more soluble boric acid from the surface of the cuticle and thereby reduce the actual boron exposure through dilution.

In assays simulating field treatments, introduction of 2 termites treated with either boric acid or barium metaborate into established groups of 18 workers, representing treatment of

Table 2. Cumulative percent mortality (mean \pm SD) of *R. flavipes* workers following a 1 min or 10 min exposure to boron dusts¹

Exposure period	Chemical	Cumulative % mortality	
		1 day	4 days
1 min	Boric acid	8.3 \pm 5.8b	31.7 \pm 12.6b
	Sodium borate	0 \pm 0b	15.0 \pm 8.7bc
	Zinc borate	1.7 \pm 2.9b	16.7 \pm 2.9bc
	Barium metaborate	41.7 \pm 10.4a	70.0 \pm 10.0a
	Control	0 \pm 0b	3.3 \pm 2.9c
10 min	Boric acid	13.3 \pm 7.6b	45.0 \pm 10.0b
	Sodium borate	5.0 \pm 8.7b	13.3 \pm 23.1c
	Zinc borate	0 \pm 0b	15.0 \pm 8.7c
	Barium metaborate	36.7 \pm 5.8a	86.7 \pm 2.9a
	Control	0 \pm 0b	1.7 \pm 2.9c

¹ Each mean represents 3 groups of 20 workers. Means in the same column within each exposure period followed by the same letter are not significantly different at the 0.05 level.

10% of the total group, resulted in complete mortality within 16 days (table 3). Since these simulation assays were conducted in single vials, and the sand within the vials was not changed during the assay, boron accumulation in the sand cannot be excluded as a contributing mortality factor, although the amount vectored in the vial on the cuticle of two workers would be rather small. When the proportion of treated termites in the group was reduced to 5% and the termites were allowed to move throughout three compartments, the same trend was apparent although mortality was less, with only boric acid differing significantly from the controls after 16 days (table 3).

Of the boron dusts examined in this study, boric acid and barium metaborate were effective termiticides. The field simulation assays with these compounds demonstrate their transfer to other colony members through grooming behaviour, and possibly trophallaxis. However, reduction of the proportion of dust-treated termites in the test groups from 10% to 5% led to a concomitant decrease in termite mortality, indicating that a large proportion of the termite colony would have to be exposed to the dust to achieve acceptable control. Injection of insecticidal dusts into galleries within infested timbers may

Table 3. Percent mortality (mean \pm SD) of *R. flavipes* workers in assays simulating field treatments where a small number of termites are treated with boron dusts and re-introduced to the general population¹

Type of assay	Proportion of dust-treated termites	Chemical	Percent mortality	
			8 days	16 days
1 vial	10 %	Boric acid	31.7 \pm 15.3ab	100.0 \pm 0a
		Barium metaborate	48.3 \pm 10.4a	100.0 \pm 0a
		Control	11.7 \pm 2.9b	23.3 \pm 5.8b
3 compartments	5 %	Boric acid	25.8 \pm 7.6a	65.8 \pm 30.2a
		Barium metaborate	27.5 \pm 10.0a	27.5 \pm 13.0b
		Control	1.7 \pm 1.4b	14.2 \pm 3.8b

¹ See text for description of assays. Each mean represents 3 groups of 20 (1 vial assay) or 40 (3 compartment assay) workers. Means in each column within each assay type followed by the same letter are not significantly different at the 0.05 level.

be an appropriate strategy with termites nesting above ground (Kalotermitidae), but is unlikely to result in contamination of a sufficient number of individuals to control ground-nesting subterranean species. However, the collection traps described by GRACE (1989a) and SU and SCHEFFRAHN (1986) are suitable for collecting large numbers of subterranean termites, which could then be dusted with a toxicant and released into the field colony by replacement in these same traps.

In a single collection from traps at one site, GRACE (1989a) collected 56,737 *R. flavipes*, and GRACE et al. (1989) removed over 200,000 termites from the same site in a two-week period. Although multiple trapping cycles would be necessary to affect the *R. flavipes* population of 3.2 million termites estimated at this site (GRACE et al. 1989), treatment and release of termites from a single collection might be sufficient to control smaller colonies. HOWARD et al. (1982), for example, estimated the mean size of *R. flavipes* colonies in the southern United States to be 244,445 termites. Under these conditions, dust-treatment and release of ca 25,000 termites would, theoretically, achieve control. The results of our laboratory study provide justification for the development of field studies of this methodology, and indicate that certain boron dusts could be effective toxic agents.

Acknowledgements

This is part of a study supported by the Ontario Ministry of the Environment, Ontario Ministry of Housing, Canada Mortgage and Housing Corporation, Ontario Real Estate Association Foundation, Toronto Real Estate Board, and the municipalities of East York, Etobicoke, Guelph, Hamilton, Kincardine, Leamington, North York, Oakville, Scarborough, and Toronto.

Zusammenfassung

Termitizide Aktivität von Borpulver (Isoptera, Rhinotermitidae)

Die Sterblichkeit von Arbeitern der Termitenart *Reticulitermes flavipes* wurde 1, 2, 4 und 8 Tage nach Behandlung mit Borpulver gemessen. Die besten Ergebnisse wurden mit Bariummetaborat (Busan 11-M1) und Borsäure erzielt. Behandlung mit beiden Produkten führte nach 8 Tagen zum Tod der Insekten. Weder Natriumborat (Borax) noch Zinkborat riefen signifikante Sterblichkeit hervor. Eine 1 bis 10 Minuten dauernde Behandlung mit diesen beiden Substanzen führte nicht zum Tod der Insekten. Versuche, die Freilandbedingungen simulierten, ergaben eine hohe Sterblichkeit, wenn 10 % einer Termitenpopulation mit Borsäure oder Bariummetaborat-Pulver behandelt wurden. Wurden nur 5 % der Population behandelt, so war die Mortalität zwar geringer, aber immer noch signifikant. Die Behandlung von Termiten, welche im Freiland mit Fallen gefangen wurden, mit insektizidem Pulver könnte eine wirksame Kontrolle, besonders nach Freilassung der Insekten, darstellen. Andererseits zeigen diese Versuche aber auch, daß ein relativ großer Anteil der Kolonie mit Borsäure oder Bariummetaborat behandelt werden müßte.

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Authors' addresses: Dr. J. K. GRACE (for correspondence), Department of Entomology, University of Hawaii, 3050 Maile Way, Rm. 310, Honolulu, HI 96822, USA; and A. ABDALLAY, Faculty of Forestry, University of Toronto, Toronto, Ontario M5S 1A1, Canada