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The efficacy of flufenoxuron as potential bait toxicant against termites

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In this laboratory evaluation, *Coptotermes acinaciformis* (Froggatt) actively attacked in the first week of testing *Pinus radiata* (D. Don) wood blocks (50 x 25 x 15 mm) was treated with concentrations of Flufenoxuron (Flurox™) (that is, 25, 50, 100, 200 and 400 ppm); the blocks with highest concentration were the most attractive. Results were similar whether blocks were leached or unleached. The earliest mortality occurred in the 400 ppm treatments between the fifth and sixth week of testing. All levels of flurox treatments proved toxic to *C. acinaciformis*, within eight weeks. Termite mortality over the test period in the water treated and solvent treated controls remained at ten percent. Flurox stimulated active termite feeding and tended to override the termites' tendency to 'mud-up' their food source and surroundings. This suggests a strong 'attractancy-feeding response' induced by flurox. Since there was no significant difference between toxicity levels in the leached and unleached blocks, it may be concluded that flurox was firmly bound to the wood substrate of the timber specimens. These laboratory results strongly indicate that flurox has a role as a potential termite bait toxicant, particularly against *Coptotermes* species. Field trials were also conducted with parallel results.

Key words: Flurox, bait toxicant, *Coptotermes acinaciformis*, leached and unleached blocks, toxicity, feeding, mortality, laboratory evaluation, termite.

INTRODUCTION

Until recently in Australia, protection of timber-in-service from subterranean termites depended mainly on soil barriers, with the use of toxic, persistent chemicals such as the organochlorines and the organophosphate chlorpyrifos. Dusting termites in galleries and sheltertubes with arsenic trioxide was still permitted and was normally the first step taken to eradicate subterranean termites from existing buildings, followed by soil treatment. In 2007 (Archicentre, 2007), it was estimated that subterranean termites caused in excess of AUD $910 million damage to buildings each year in Australia.

In June 1995, the National Registration Authority (NRA) recommended that organochlorine insecticides be banned from use in termite control measures in all States and Territories of Australia, except the Northern Territory which was granted an extension (Anon, 1995). Prior to this time, protection of timber-in-service from subterranean termites depended mainly on soil barriers, with the use of toxic, persistent chemicals such as the organochlorines, the organophosphate chlorpyrifos (Australian Standard, 1995) and treated timber. Dusting termites in galleries and sheltertubes with arsenic trioxide was and is still permitted. Normally, the first step taken to eradicate subterranean termites from existing buildings

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was to dust using arsenic trioxide, followed by chemical soil treatment.

In 1986, it was estimated that subterranean termites caused in excess of $100 million damage to buildings each year in Australia, and over $5 – 6 million was spent annually on chemicals for termite control (French, 1986). Currently, the chemicals registered for use in subterranean termite control in Australia are arsenic trioxide, bifenthrin (Biflex), chlorpyrifos (Dursban), imidaclorprid (Premise), and fipronil (Termidor). Alternative termite control measures are actively being researched worldwide. Increased emphasis is being directed towards developing integrated termite pest management systems which will combine termite baiting techniques, bait and dust toxicants, chemical soil barriers and wood preservatives that have termiticidal properties (French, 1994, 1996; Forschler, 1996; Lewis et al., 1996; Gold et al., 1996; Grace et al., 1996; Myles, 1996; Pawson and Gold, 1996; Su and Scheffrahn, 1996).

More recently, termite bait system developments became an important step in termite control in Australia and around the world. Currently, some of the chitin synthesis inhibitors such as Requiem®, Nemesis® and Sentricon® AG, termite baits are registered for commercial use as bait toxicants against wood-feeding subterranean termites in Australia. Understand termite biology and foraging behaviour is critical for baiting system management. The *C. acinaciformis* termites are called subterranean termites and also are grouped as "lower termites". Their members are differentiated into various 'castes' a division of labour which allows the colony to undertake its complete range of essential activities (e.g. queen: egg-laying, population control; workers: foraging, food supply and construct galleries and nests; soldiers: defense; alates or winged reproductives: dispersal to establish new colonies and reproduction). Termite social life requires a complex chemical (hormonal) communication system within the nest, initiated by the queen.

This paper describes a laboratory evaluation of the compound flurox which is a member of the chitin synthesis inhibitor group as a potential bait toxicant when impregnated into timber specimens and exposed to subterranean termite attack in a no-choice situation.

MATERIALS AND METHODS

Formulation examined

Flurox technical grade (purity 96.6%) was supplied by Janssen Pharmaceutica N. V. Turnhoutseweg 30, B-2340 Beerse, Belgium.

Choice of test termite species

Coptotermes acinaciformis (Froggatt) was selected in our screening bioassays because it is the most economically important wood-feeding termite species in Australia. This termite is widely distributed throughout mainland Australia and is responsible for greater economic loss to timber-in-service than all other termite species combined.

Treated specimens

(i) Timber: Timber specimens (50 x 25 x 15 mm) with grain along the 50 mm direction were cut from the sapwood of several fast-grown *Pinus radiata* D. Don trees from a pruned and thinned forest which had been harvested in the Mount Gambier region of South-eastern Australia. The trees were 30 years old.
(ii) Randomisation: Timber specimens were pooled and randomly allocated treatment groups. Prior to timber treatment, specimens had a mean moisture content of 10%. Mean density of the specimens at 10% m.c. was 470 kg/m^3.

Timber treatment

(i) Solvent and water controls: Twenty timber specimens were impregnated in a vacuum desiccator. The schedule comprised 30 mins vacuum at -95 kPa, introduction of treatment solutions (acetone and deionised water) whilst under vacuum, and vacuum release. Specimens were left to soak in the solution at atmospheric pressure for 30 min and were weighed before and after treatment to determine their retentions.
(ii) Preservative treatment: Using the treatment method described for the controls, groups of 20 specimens were treated with flurox at five different concentrations. Flurox was dissolved in acetone to achieve concentrations of 400, 200, 100, 50 and 25 ppm. The average retentions of flurox achieved in wood were 0.187 kg/m^3, 0.0913 kg/m^3, 0.0465 kg/m^3, 0.0231 kg/m^3 and 0.0171 kg/m^3, respectively.

Artificial weathering

(i) Solvent release from treated specimens: After treatment and weighing, each replicate group of blocks was enclosed in a separate polyethylene bag and placed in an empty fish tank for two weeks. The fish tank lid was opened a few millimetres each day to allow specimens to slowly dry and for any chemical fixation/immobilisation to occur. Specimens were then transferred to drying racks in a fume cupboard for ten days. Water treated controls were allowed to air dry immediately after treatment. All specimens (including treated and solvent controls) were then subjected to a leaching/volatilisation schedule.
(ii) Leached specimens: From each group of twenty specimens, ten were placed in plastic jars filled with tap water (water three times the volume of wood). The jars were then transferred to a waterbath at 35ºC and shaken for five days. The water in each plastic jar was changed daily. After five days of agitation/leaching, the specimens were placed on drying racks for a further five days.
(iii) Unleached specimens: The remaining ten specimens were not leached but were allowed to condition in the fume cupboard until mass equilibrium was reached. This took up to four weeks.
(iv) Vacuum oven drying: Both leached and unleached specimens were vacuum oven-dried separately for five days at 40ºC and -95 kPa. After removal from the vacuum ovens, the specimens were cooled in a desiccator and weighed to obtain initial mass. At each stage, care was taken to ensure that specimens of different formulations and retentions were separate.

Termite source

Orphaned groups of workers and soldiers from two colonies of

C. acinaciformis were collected from active below-ground mound colonies in the semi-arid mallee country at Walpeup in the north-west of Victoria, and used within a week of collection.

Termite bioassay

After the conditioning periods, a single treated block from each conditioning regime (unleached and leached) was placed in glass jar (275 ml) containing sand (20%), *C. acinaciformis* mound material (80%), and 5 ml of deionised water. There were five replicate units per bioassay. To each jar 5 g of termites (approximately 1400 individuals) were added, and the glass jars sealed with a vented lid. All treatments were replicated five times using blocks nearest the mean retention. All the bioassay jars were placed in an insectary at 27ºC and 75% RH for eight weeks.

Termite mortality was visually estimated each week (1 to 7 weeks) and at the end of the test termites were removed from jars and counted. Observations of attractancy, repellency, and any morphological changes to the termites and wood blocks were routinely recorded. After eight weeks of laboratory bioassay the timber specimens were removed and oven dried to obtain final mass loss.

Wood consumption and toxicological data derived from these bioassays will provide a guide in selecting the economic lethal threshold levels (ELTL) for the treatment of wood blocks in the subsequent field test.

RESULTS AND DISCUSSION

Visual assessment of termite feeding/ foraging behaviour

The visual assessment of termites feeding on the untreated and flurox-treated wood blocks over the eight week test period is shown in Table 1. While this data is qualitative, rather than quantitative, it is important to record termite behaviour when they are presented with a potential bait toxicant or active ingredient. Normally, it is only after the bioassays have been completed that differences, some subtle, are discovered when taking a more 'holistic' view of the evaluation. So, when a visual assessment of termite feeding behaviour on the treated wood blocks is made and compared to the percentage mortality of *C. acinaciformis* (Figure 2), it is seen that there are indeed, important differences. For instance, there was an initial attractancy to the high loadings of flurox. Termites were actively attacking the high loadings (that is, 200 and 400 ppm) compared to the low loadings in the first week of the bioassay (Figure 3). However, termites became physiologically affected, and hardly moved off the treated blocks even when the light was turned on. This trend continued until the end of the third week, when the number of termites recorded on the 400 ppm blocks was significantly less than for the other loadings. Yet, 100% mortality was not recorded until the fifth week (Figure 2). There was a similar 'lag phase' with termites feeding on the 200 ppm flurox treated blocks. By way of contrast, the termites feeding on the water and solvent controls were initially slow to aggregate and feed on the blocks (Figures 2 and 3).

Also, termite feeding between some groups was markedly different, in ways not immediately apparent when viewing the tables of results. It was observed that within the first few days of initiating this bioassay, the termites with the water and solvent controls directed their activities to 'muddying-up' the sides of the container with substrate and faecal materials, rather than to feeding on the wood blocks. However, the termites exposed to the flurox-treated blocks exhibited considerably different behaviour. First, the termites clustered all over the treated blocks (Figure 3), and did not mud up the container. Second, the termites deposited virtually no faecal material on the blocks while feeding, in contrast to the water and solvent control blocks. The controls were coated with spots of faecal material. Furthermore, when turning on the light in the insectary to examine termite behaviour, termites on the flurox-treated blocks seemingly exhibited no reaction. They continued feeding on the blocks as if still in the dark. Whereas the termites in the water and solvent controls jars behaved as one would expect from such a disturbance, namely, they quickly moved off the surface of the blocks and disappeared into the substrate within the container.

Mortality

Feeding on leached flurox-treated blocks

The mean percentage mortality of *C. acinaciformis* after feeding on leached flurox-treated blocks is shown in Figure 1. After eight weeks, the mortality of the termites with the water and solvent control treatments was 12.4 and 9.4%, respectively, whereas in the treated blocks, termite mortality was virtually 100% (Figure 2). More differences occurred between treatments after just six weeks. All the termites feeding on flurox at 400 ppm died within six weeks, while at the same period of test, there were 95, 59, 50 and 39% mortality at the 200, 100, 50 and 25 ppm treatment levels, respectively.

Regardless of the level of flurox impregnated into the blocks, when mean mortality was between 35 to 40%, complete mortality then later occurred mostly within two weeks across all the treatment regimes under test (that is, from 25 to 400 ppm).

Feeding on unleached flurox-treated blocks

Table 2 shows the mean percentage mortality of *C. acinaciformis* after feeding on unleached flurox-treated blocks for eight weeks. There is no significant difference between the results for termite mortality under either the leached or unleached treatment regimes.

The similar result for both leached and unleached blocks suggests that flurox is firmly bound to the wood substrate so that toxicity is not diluted by leaching into water. While leaching is not of paramount importance for

Table 1. Mean weekly visual inspections results on bait block status and the rating used for termite activity with the bait block status (Results for leached and unleached blocks combined).

Number of weeks	Rating *						
	Controls		Flurox a.i. (ppm)				
	Water	Solvent	25	50	100	200	400
1	7	7	13	14	16	17	19
2	9	9	14	16	19	24	29
3	13	13	17	20	27	32	34
4	18	18	21	24	30	34	0
5	23	23	24	28	33	34	0
6	30	30	27	31	33	0	0
7	34	34	32	33	34	0	0
8	36	36	34	34	0	0	0

*Baits blocks status		V. low	Low	Normal	High	Higher
No termite activity	NA	0/1	2	3	4	5
Faecal deposit	FD	6	7	8	9	10
Termite activity	TA	11	12	13	14	15
Termite visit	TV	16	17	18	19	20
Termite nibble	TN	21	22	23	24	25
Slight attack	SA	26	27	28	29	30
Moderate attack	MA	31	32	33	34	35
Severe attack	VA	36	37	38	39	40

Termite activity = Termite walking across the wood block. Termite visit = staying at the block to investigate.

Table 2. Mean percentage mortality (standard deviation) of *C. acinaciformis* after feeding on unleached flurox-treated wood blocks over eight weeks.

Flurox	Weeks of termite bioassay							
Conc. ppm	WK1	WK2	WK3	WK4	WK5	WK6	WK7	WK8
400	0.0	18.6 (3.7)	40 (7.1)	77 (5.1)	99.6 (0.8)	100 (0)	100 (0)	100 (0)
200	0.0	9 (3.7)	22 (5.1)	44 (3.7)	69 (3.7)	96 (3.7)	100 (0)	100 (0)
100	0.0	7 (4)	16 (3.4)	27 (5.1)	52 (4)	68 (5.1)	97 (4)	100 (0)
50	0.0	5.8 (5.8)	14 (3.7)	20 (7.1)	40 (7.1)	52 (5.1)	69 (3.7)	97 (4)
25	0.0	0.0	2 (2.5)	11.2 (1.6)	42.6 (4.4)	47 (9.3)	60 (8.4)	94 (5.8)
SC	0.0	0.0	0.0	0.0	0.0	0.0	0.0	12 (2.3)
WC	0.0	0.0	0.0	0.0	0.0	0.0	0.0	10 (2.3)

WK1, 2, = week1, week 2; Conc., Concentration; ppm, parts per million.

potential bait toxicants, the performance of flurox-treated blocks in this bioassay suggests a possible role for this chemical as an active ingredient in the protection of timbers against subterranean termites when the treated timbers are exposed outdoors.

Termite feeding days

The mean consumption rates for termites feeding on leached and unleached flurox treated blocks, together with the time, the termites were feeding on the treated blocks prior to their demise are shown in Figure 1. There was little difference between the mean mass loss of leached and unleached treatment groups, irrespective of whether they were control or flurox treatments. Irrespective of the flurox treatment regime, the percentage mean mass losses ranged between 91 to 132 mg of wood consumed over the test period. However, the rate at which wood was consumed within the different treatment groups was different. The termites feeding on the highest loading of flurox (that is, 400 ppm, leached and unleached) consumed a similar amount of wood to those with the other treatments, but within 35 days. The

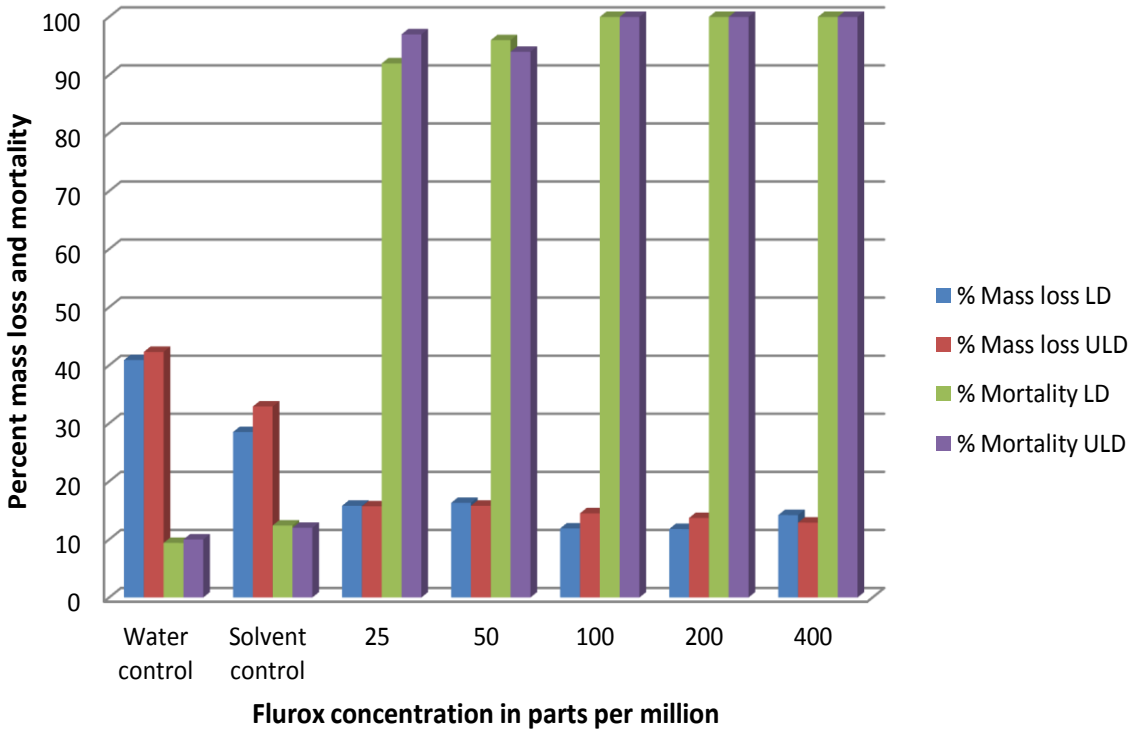

Figure 1. Termite percent mortality rate and percent mass loss of flurox treated wood blocks (six replicates/unit treatment) after eight weeks exposure.

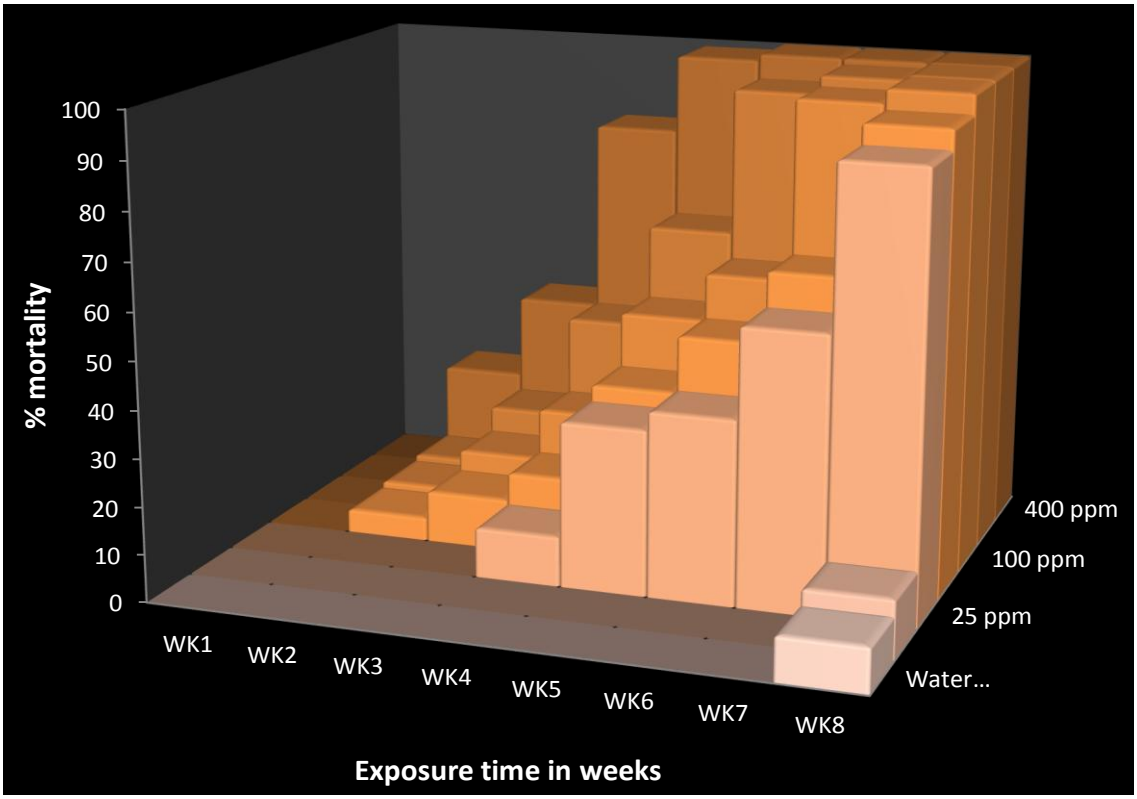

Figure 2. Mean percentage mortality of *C. acinaciformis*.

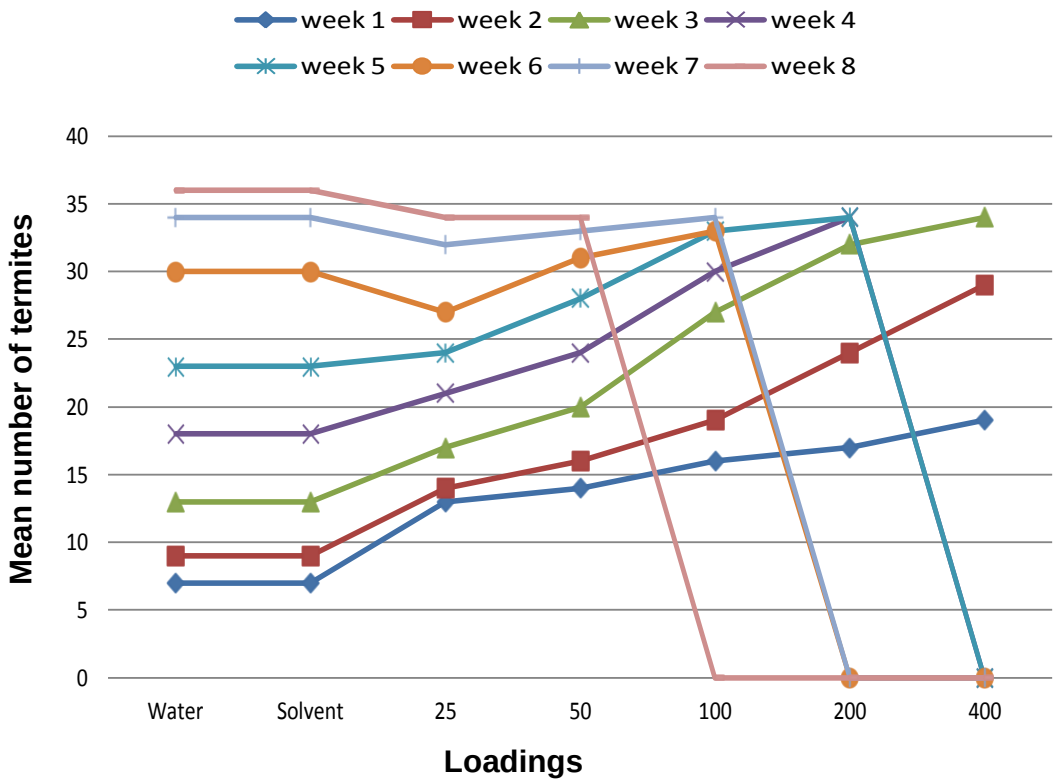

Figure 3. Mean number of termites visiting each bait block per week.

termites on the other treated blocks (25 to 200 ppm) took 49 to 56 days to consume the same amount. This suggests that the higher loading of flurox stimulated more feeding activity by the termites. The higher loading did not deter termite feeding.

Conclusion

Flurox as bait a toxicant

Termites actively attacked the flurox-treated wood blocks at all loadings (that is, 25, 50, 100, 200 and 400 ppm of flurox) in both the leached and unleached conditions. Termites were particularly active on blocks with the highest loadings in the first week of test. All levels of flurox treatments proved toxic to the test termite, *C. acinaciformis* within 8 weeks. This compares to maximum mean termite mortality on control blocks during the same period of only 10%. Flurox seems to act as a bait toxicant for *C. acinaciformis*.

Flurox stimulated active termite feeding

Termites tended to override their tendency to 'mud-up' their food source and surroundings. This suggests a strong 'attractancy-feeding response' induced by flurox in this laboratory evaluation.

Flurox is firmly bound to the wood substrate

The toxicity of treated specimens was not reduced by leaching in water. While leaching is not of paramount importance when viewing potential bait toxicants, the performance of flurox-treated blocks in this bioassay suggests a possible role for this chemical as an active ingredient in the protection of timbers against subterranean termites when the treated timbers are exposed to outside climatic conditions and hazards. However, there is likely to be some mass loss of wood in service before colony death, which may be unacceptable for appearance grade timbers.

Termite behavioural responses

It is essential to regularly observe termite behavioural responses during the whole period of testing in order to observe, record and understand more comprehensively the effects of flurox treatments.

These laboratory results strongly indicate that flurox has potential as a termite bait toxicant, particularly against *Coptotermes* species. More field trials are needed to confirm these laboratory results.

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REFERENCES

Anon (1995). Building industry adjusts to the phase out of organochlorines. Nat. Regist. Authority News. 2(2): 12. (URL location: http://www.dpie.gov.au/nra/nra2_082.html).

Archicentre (2007). “An Architect perspective”. Report published for Pest control and building industry.

Australian Standard (1995). Protection of buildings from subterranean termites. Part 1: New buildings. AS 3660.1.

Forschler BT (1996). Baiting *Reticulitermes* (Isoptera: Rhinotermitidae) field colonies with abamectrin and zinc borate-treated cellulose in Georgia. Sociobiology 28(3):459-484.

French JRJ (1986). Termites and their economic importance in Australia. In: Vinson, S.B., ed. Economic impact and control of social insects. New York, Praeger. pp. 103-129.

French JRJ (1994). Combining physical barriers bait and dust toxicants in future strategies for subterranean termite control (Isoptera). Sociobiology 24(1):77-91.

French JRJ (1996). Current developments in subterranean termite control in Australia. 25th For. Prod. Res. Conf., Melbourne, 18-21 November, 1996, P. 13.

Gold RE, Howell Jr, HN, Pawson BM, Wright MS, Lutz JC (1996). Persistence and bioavailability of termiticide to subterranean termites (Isoptera: Rhinotermitidae) from five soil types and locations in Texas. Sociobiology 28(3):337-363.

Grace JK, Yates III JR, Tome CHM, Oshiro RJ (1996). Termite-resistant construction: Use of a stainless steel mesh to exclude *Coptotermes formosanus* (Isoptera: Rhinotermitidae). Sociobiology 28(3):365-372.

Lewis VR, Haverty MI, Carver DS, Fouche C (1996). Field comparison of sand or insecticide barriers for control of *Reticulitermes* spp. (Isoptera: Rhinotermitidae) infestations in homes in Northern California. Sociobiology 28(3):327-335.

Myles TG (1996). Development and evaluation of a transmissible coating for control of subterranean termites. Sociobiology 28(3):373-457.

Pawson BM, Gold RE (1996). Evaluation of baits for termites (Isoptera: Rhinotermitidae) in Texas. Sociobiology 28(3):485-510.

Su N-Y, Scheffrahn RH (1996). A review of the evaluation criteria for bait-toxicant efficacy against field colonies of subterranean termites (Isoptera). Sociobiology 28(3):521-530.