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Transfer of indoxacarb among workers of *Coptotermes formosanus* (Isoptera: Rhinotermitidae): effects of dose, donor:recipient ratio and post-exposure time

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Abstract: Horizontal transfer of indoxacarb among workers of the Formosan subterranean termite, *Coptotermes formosanus* Shiraki, was examined under laboratory conditions. The effects of dose (0, 10, 20, 50, 100 or 200 ng AI per donor), donor:recipient ratio (1:1, 1:4 or 1:9) and post-exposure time (2, 4, 8, 16, 20 and 24 days) on lethal transfer of indoxacarb were investigated using a donor/recipient model in groups of 100 workers. Transfer of lethal doses from donors to recipients was evidenced by significant recipient mortality in 13 out of 15 treatments within 24 days post-exposure. Dosage significantly affected indoxacarb transfer. Higher doses resulted in greater recipient mortality than lower doses. The highest dose tested resulted in 100% death of recipients and donors within 20 days. A dose of 100 ng resulted in recipient mortalities ranging from 68 to 100%, whereas doses ≤50 ng killed <60% of recipients within 24 days. Donor:recipient ratio also had considerable effect on indoxacarb transfer. At 24 days post-exposure, greater recipient mortalities were observed at ratios ≥1:4 at doses ≥20 ng, but only at a ratio 1:1 at dose 10 ng. Recipient mortality increased significantly as post-exposure time increased. The higher the doses applied on donors, the shorter were the times required for the onset of recipient mortality to occur and for recipients to reach maximum mortality. Significantly greater recipient mortalities were not observed at doses 20 and 50 ng until more than 20 days post-exposure, indicating a delayed activity of indoxacarb. Possible transfer mechanisms are discussed.

Keywords: indoxacarb; horizontal transmission; Formosan subterranean termite; *Coptotermes formosanus*; termite control

1 INTRODUCTION

New technologies developed in recent years for subterranean termite control include physical barriers, insecticide-impregnated polymer barriers, monitoring–baiting systems and non-repellent termiticides.[1–5] Non-repellent termiticides are usually slow-acting insecticides. Su *et al*.[6] defined slow-acting insecticides as those that require a longer time to kill termites at low concentrations or doses than at higher concentrations or doses and the level of mortality and the speed of death are concentration or dose dependent. Lack of repellence of such termiticides allows termites to move freely within treated soil before dying. Delayed activity allows transfer of the insecticides to occur from exposed to unexposed termites.[5,7,8] Hence slow-acting non-repellent insecticides have a greater potential impact on termite population than repellent insecticides such as synthetic pyrethroids and fast-acting non-repellent insecticides such as chlorpyrifos.[7–10]

Horizontal transfer of insecticides is a strategy presently being exploited for termite control.[11] The theorized horizontal transfer of non-repellent insecticides such as fipronil (Termidor®) and imidacloprid (Premise®) has been documented in *Coptotermes formosanus* Shiraki using laboratory studies. Fipronil-treated workers (topically dosed with 2.5 ng AI per termite) placed with untreated workers at ratios ≥10% significantly increased untreated worker mortality within 72 h.[12] In another study, workers exposed to fipronil- or imidacloprid-treated sand (0.1 g AI kg^{-1}) for 1 h and placed with untreated workers at a ratio 1:19 resulted in significantly greater mortality in untreated workers after a 15-day interaction.[13]

Indoxacarb, methyl (*S*)-*N*-[7-chloro-2,3,4a,5-tetrahydro-4a-(methoxycarbonyl)indeno[1,2-*e*][1,3,4] oxadiazin-2-ylcarbonyl]-4′-(trifluoromethoxy)carbanilate, is a neurotoxic oxadiazine insecticide discovered by DuPont.[14,15] Indoxacarb has been

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designated by the US Environmental Protection Agency a 'reduced-risk' pesticide owing to its relatively low mammalian toxicity and low impact on human health.[16] It is active both by contact and ingestion and is currently registered for several vegetables and crops under the trade names Avaunt® and Steward® and as bait against the red imported red fire ant, *Solenopsis invicta* Buren, under the trade name Advion™ (DuPont Crop Protection, Wilmington, DE, USA). It was reported as effective against German cockroaches, *Blattella germanica* (L.).[17] A recent study using a modified glass-tube bioassay showed that indoxacarb was effective and non-repellent against *C. formosanus* and the eastern subterranean termite, *Reticulitermes flavipes* (Kollar), when they were exposed to soil that had been treated at concentrations $\geq 50\,mg\,kg^{-1}$.[18] In the same study, indoxacarb also demonstrated a delayed activity which may facilitate transfer of the insecticide from exposed to unexposed termites.

The purpose of this study was to investigate the potential horizontal transfer of indoxacarb from treated to untreated workers of *C. formosanus*, using a modified donor/recipient model.[19] The effects of dose topically applied to donors, donor:recipient ratio and post-exposure time were considered.

2 MATERIALS AND METHODS

2.1 Termites

Two field colonies of *C. formosanus* were collected in April 2004, using a bucket trapping technique.[20] One colony was from Opelika, Lee County (central Alabama) and the other colony was from Fairhope, Baldwin County (southern Alabama). Termites were kept in plastic boxes (13.5 × 19 cm) supplied with moistened sand and spruce (*Pinus pungens* Engelm) wood blocks to allow 1-week acclimation at 22 ± 1 °C and 92 ± 3% RH before testing. Only healthy intact workers were used in this study. Voucher specimens are preserved in 100% ethyl alcohol and stored in the insect collection of the Department of Entomology and Plant Pathology, Auburn University, Auburn, AL, USA.

2.2 Termiticide

An experimental indoxacarb $150\,g\,liter^{-1}$ SC was provided by DuPont (Wilmington, DE, USA).

2.3 Experimental design

Termites designated to become 'donors' were stained by feeding them for 7 days on moistened filter-paper (Whatman, Clifton, NJ, USA) dyed with an aqueous solution of Nile blue ($0.01\,g\,liter^{-1}$; 2 ml per paper). Nile blue A (Aldrich, Milwaukee, WI, USA) is a suitable persistent stain for termites.[21] Termites designated to become 'recipients' were fed on similar filter paper moistened with water (2 ml) only.

The indoxacarb formulation was diluted with distilled water to obtain concentrations of 0.05, 0.1, 0.25, 0.5 or 1 g AI $liter^{-1}$, so that when 0.2-μl droplets were applied to termites, five doses (10, 20, 50, 100 and 200 ng AI per donor termite) were achieved.

A droplet (0.2 μl) of the required concentration was applied topically to the dorsal thorax of a stained termite isolated in a glass Petri dish, using a 1-μl micro-syringe (Hamilton, Reno, NV, USA). The same volume of distilled water was used as a control. A preliminary test showed that this volume provides full coverage of the dorsum of the thorax without liquid runoff. The topical application was conducted on termites without chilling them at low temperature or anesthetizing them with carbon dioxide to minimize possible negative impacts. In cases where the droplet ran off or was misplaced, the treated stained termites were excluded from testing. Henceforth, topically dosed stained termites were designated as donors and untreated unstained termites as recipients.

Donors were introduced into Petri dishes (60 × 15 mm, Fisher brand) containing 10 g sterilized sand moistened with deionized water (1.2 ml), food (a 30 × 30 mm Whatman filter-paper and a 20 × 20 × 5 spruce wood block) and sufficient recipients to provide three ratios of donor–recipient combination (1:1, 1:4 and 1:9) in groups of 100 termites. There were 15 treatments (five doses by three combinations) and three controls. Each treatment and control was replicated five times. Petri dishes were kept in three plastic boxes (13.5 × 19 × 11 cm) and held in an incubator at 22 ± 1 °C. A 30-cm long dental absorbent (Absorbal, Wheat Ridge, CO, USA) moistened with water (10 ml) was placed in each plastic box to retain humidity. Mortalities of donors and recipients were recorded at 2, 4, 8, 16, 20 and 24 days post-exposure by counting the numbers of survivors. Dead termites, including those moribund or partially consumed, were not removed from the Petri dishes.

2.4 Statistical analyses

Data were analyzed using Statistix® 8 software.[22] Percentages of donor and recipient mortalities were normalized using an arcsine square root transformation before being subjected to analysis of variance (ANOVA). Because the objective of this study was to examine whether indoxacarb could be transferred from donors to recipients and cause consequential death of recipients, significant differences of recipient mortality between doses at a ratio 1:1 and between ratios at the end of the 24-day test were determined by Tukey's honestly significant difference (HSD) test following ANOVA. Pearson's procedure was used to analyze the correlations between recipient mortalities and the variables of dose, ratio and post-exposure time. To determine how one variable affected the main effect of the other factors on recipient mortalities, means plots were conducted when two-factor interaction (dose × ratio, dose × post-exposure time or ratio × post-exposure time) was significant. The difference between donor and recipient mortalities were analyzed using a paired *t*-test at each specific combination of variables. All statistical analyses were

conducted at the $P = 0.05$ level of significance. Data are presented as actual percentage mortalities in all figures.

3 RESULTS AND DISCUSSION

3.1 Effects of dose

Dose had a significant effect on the mortality of both donors and recipients. Higher doses applied on donors caused more rapid and greater mortalities than lower doses on each observation day ($F > 98.8$; $P < 0.001$; df = 5, 20). Recipient mortality was correlated with dose topically applied on donors (correlation coefficient $r = 0.83$). The higher the doses applied on donors, the shorter were the times required for recipients to begin dying and to reach maximum mortality. At a ratio 1:1, doses of 100 and 200 ng killed all donors and recipients within 20 days and onset of recipient mortality occurred within 2 or 8 days, respectively (Fig. 1). However, doses of 10, 20 and 50 ng did not result in significant recipient mortality (12, 27 and 57%, respectively) until the last observation day of the 24-day trial. Additionally, onset of recipient mortality did not happen until after almost a 20-day interaction with donors.

In all the treatments, the mortality of donors occurred consistently sooner and greater than that of recipients before both reached 100% (Fig. 1). Of course, this may be explained because donors received lethal doses before recipients did and the doses directly applied on donors were presumably relatively higher than those acquired indirectly by recipients via interactive behavior. Dose dependence was also shown in a previous study when varying levels of mortality and speed of death of *C. formosanus* and *R. flavipes* were observed over several concentrations of indoxacarb from 10 to 100 mg kg^{-1}.[18] Shelton and Grace[13] observed a similar phenomenon when *C. formosanus* donors were exposed to fipronil- or imidacloprid-treated sand (0.1 g AI kg^{-1}) for 1 h and placed with recipients at a ratio 1:19. After a 15-day interaction, the mean donor mortality was 97.8 or 100%, while the recipient mortality was 38.6 or 61.5%, respectively. Further studies will address the amount of indoxacarb that each recipient acquires over time to determine the distribution pattern of indoxacarb within testing groups.

3.2 Effects of donor:recipient ratio

The donor:recipient ratio affected the mortality of donors and recipients (Fig. 2). At doses ≥20 ng, mortalities of both donors and recipients were significantly greater at ratios 1:1 and 1:4 than at a ratio 1:9 at 24 days post-exposure ($F_{5,20} > 30.45$, $P < 0.001$). There was no significant difference between ratios 1:1 and 1:4 at these doses. At a dose of 10 ng, a ratio 1:1 resulted in greater mortalities of donors and recipients than ratios 1:4 and 1:9, which were not significantly different from each other and control. However, the correlation between recipient mortality and ratio was not strong ($r = 0.38$) and a factor interaction (dose × ratio) was detected. Analysis

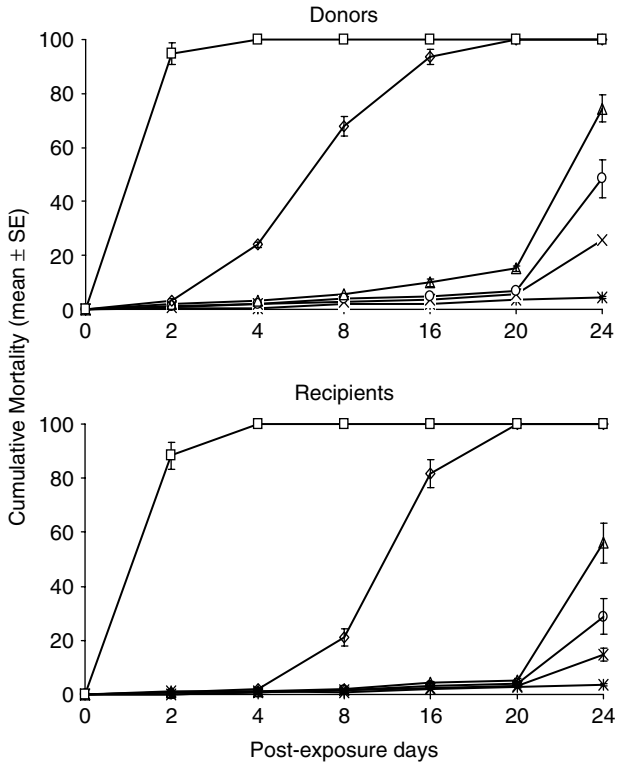

Figure 1. Mean cumulative mortalities of donors and recipients of *Coptotermes formosanus* at (□) 200; (◊) 100; (△) 50; (○) 20; (×) 10; (✱) 0 (control) ng of indoxacarb per donor when tested in groups of 100 workers at a donor:recipient ratio of 1:1 during the 24-day trial.

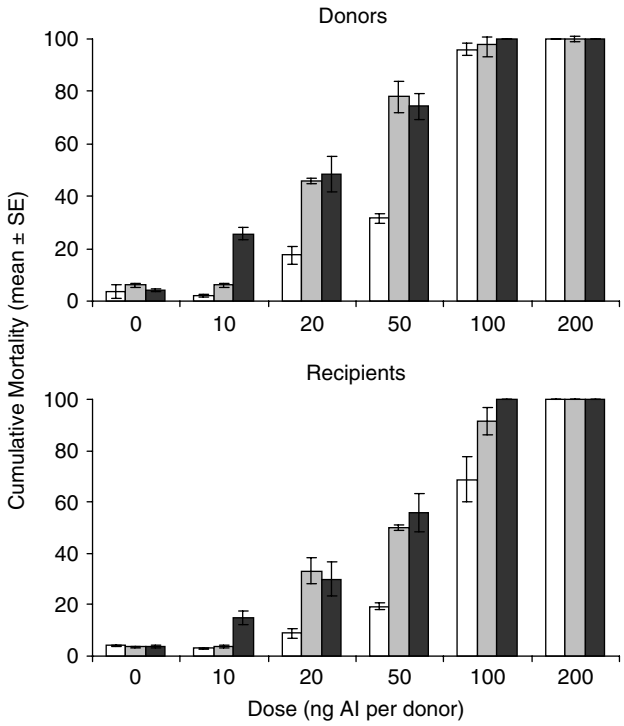

Figure 2. Mean cumulative mortality of donors and recipients of *Coptotermes formosanus* at donor:recipient ratios of (white) 1:9, (gray) 1:4 and (black) 1:1 after a 24-day interaction, when tested in groups of 100 workers at various doses of indoxacarb.

of mean plots indicated that the effect of dose on mortality overpowered the effect of ratio.

Ibrahim *et al*.[12] documented significant effect of donor:recipient ratio on fipronil transfer in laboratory groups of *C. formosanus*. Their data indicated that a minimum of 50% worker donors topically dosed with 2.5 ng of fipronil were required to obtain ≥95% recipient mortality within 72 h. Under the conditions of our study, at least 20% donors were needed to kill ≥94% of recipients within 24 days using a dose of 100 ng of indoxacarb per donor.

3.3 Effects of post-exposure time

The time period after donors were placed together with recipients was important for the transfer of indoxacarb (Fig. 3). As the post-exposure time increased, the mortalities of both donors and recipients increased. This can be explained by longer post-exposure times providing more opportunities for recipients to interact with donors and to acquire indoxacarb continuously, regardless of dosage or ratio. The value of the correlation coefficient (r) between donor mortality and post-exposure time was 0.82 and between recipient mortality and post-exposure time was 0.68. The relatively low value of the correlation coefficient between recipient mortality and post-exposure time may be explained by the delayed action of indoxacarb at low doses in particular. At 200 ng, minimum post-exposure times of at least 4, 8 and 20 days were necessary to achieve 100% recipient mortalities at ratios 1:1, 1:4 and 1:9, respectively. At 100 ng, recipients did not begin to die until after 4, 8 and 16 days post-exposure, although recipient mortalities reached as high as 100, 94 and 68% by 24 days at ratios 1:1, 1:4 and 1:9, respectively. Of interest was the significantly delayed recipient mortality in treatments with lower doses (20 and 50 ng), where cumulative recipient mortality remained unchanged until 20 days post-exposure, regardless of ratio. However, as soon as the onset of mortality began, recipient mortality increased considerably to levels significantly greater than control within 4 days (at 24 days post-exposure). Those data implied that the transfer of non-repellent termiticide from donors to recipients might not be a one-time event but rather a cascading event of repetitive transfers as suggested by others.[23,24] Our data showed that it might take a significant amount of time for recipients to acquire and accumulate indoxacarb to lethal levels, depending on the doses directly exposed to donors and donor:recipient ratios. Further study is needed to address the minimum threshold levels of indoxacarb required to affect mortality post-exposure.

3.4 Transfer of indoxacarb

The relative toxicity of indoxacarb was indicated by the mortality of directly exposed donors, while the lethal transfer of indoxacarb was demonstrated by the mortality of indirectly exposed recipients. By the end of the 24-day trial, the transfer of indoxacarb from donors

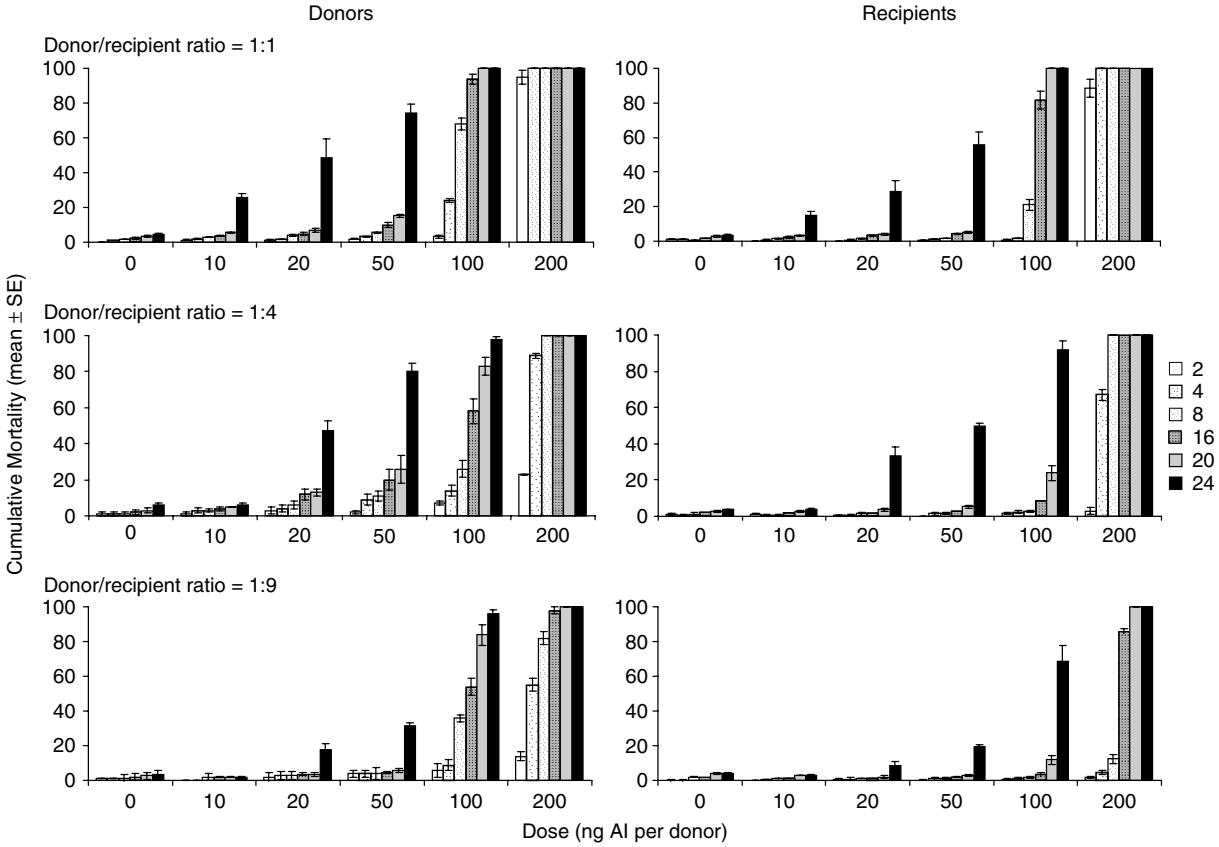

Figure 3. Mean cumulative mortality of donors and recipients of *Coptotermes formosanus* in groups of 100 workers at 2, 4, 8, 16, 20 and 24 days (see key) at various doses of indoxacarb and various donor:recipient ratios.

to recipients was evidenced by significant recipient mortalities at doses ≥20 ng regardless of ratios ($P < 0.05$). Transfer of non-repellent insecticides such as imidacloprid and fipronil from treated to untreated nestmates was reported in previous studies by other researchers.[12,13,25]

Among the mechanisms suggested or proved by previous studies, three mechanisms might have contributed to the transfer of indoxacarb in this study. As social insects, termites perform various interactive behaviors for communication, nestmate care and movement of nutrients, other chemical compounds or pathogens among nestmates.[8,11–13,23,25,26] Grooming is one of the most important and common forms of interactive behavior for communication within a colony. Using ^{14}C label and videotaping technologies, Tomalshi and Vargo[25] demonstrated that grooming of topically treated termites by untreated termites was a principal mechanism of transport and transfer of imidacloprid in *R. flavipes* groups. Untreated workers were observed acquiring [^{14}C]imidacloprid mainly by grooming it from the cuticle of treated workers and becoming debilitated. The intoxicated recipients then became secondary donors to other untreated workers (secondary recipients), who again acquired it via grooming. Although we did not investigate the exact mechanisms of transfer of indoxacarb in this study, grooming between donors and recipients was often observed at the times when mortality was recorded. Additionally, the significant increase in recipient mortality following donor mortality evidenced a pattern of secondary transfer. Nevertheless, our observation of grooming of donor by recipients and the death of recipients suggest that grooming could have played a significant role in the transfer of indoxacarb. These findings confirmed that indoxacarb at these concentrations was neither detectable nor repellent to *C. formosanus*.[18]

Casual contact among termites or with contaminated substrate in the test arena could be the second mechanism. It was possible for recipients to pick up indoxacarb on cuticles of termites (alive and dead, donors and contaminated recipients) or indoxacarb deposit on substrate in the test arena via casual contact. Ferster *et al*.[19] considered contact with carcasses of both directly and indirectly exposed termites to be a main mechanism for significant transfer of lethal doses of spinosad from nymph donors to nestmate recipients of drywood termites, *Incisitermes snyderi* (Light). Haagsma and Rust[23] believed that *R. hesperus* Banks recipients could acquire a small amount of [^{14}C]hexaflumuron by contacting with donors fed with [^{14}C]hexaflumuron-treated bait.

The third potential mechanism could be cannibalism of nestmates. Cannibalism was considered complementary to grooming and casual contact in the distribution of toxicant among termite nestmates by Thorne and Breisch.[27] Our data support this hypothesis. At each recording date, the total number of living and dead termites was often less than the original number of 100 termites. Some of the termites, alive or dead, were missing body parts partially or entirely. Recipients were also observed biting off legs or antennae of live donors. This was the main reason why we counted the number of living rather than dead termites for mortality assessment. However, owing to the limitation of the scope of this study, it is not known to what level the cannibalism contributed to indoxacarb transfer.

Other factors, such as trophallaxis and metabolism, which are considered to play significant roles in the transmission of ingested toxicants and nutrients between members of a termite colony, might have a trivial role in transferring insecticidal deposits on the cuticle.[23,27–29] Further investigations are required for studying the interactive behaviors among donors and recipients to understand the exact mechanisms of transport and transfer of indoxacarb.

4 CONCLUSIONS

The results of this study demonstrate the transfer of lethal doses of indoxacarb from topically dosed donors to untreated recipients of *C. formosanus*, as indicated by significant recipient mortality. When donors represented >10% members in groups of 100 workers, a minimum dose of 100 ng of indoxacarb per donor was necessary to achieve recipient mortalities ranging from 68 to 100% within 24 days post-exposure, where donor mortalities ranged from 95 to 100%. A previous study of toxicity and barrier efficacy showed that a soil barrier (2 cm) containing 50–100 mg kg^{-1} indoxacarb killed all tested *C. formosanus* termites within 21 days and allowed a free and complete penetration of the soil barrier.[18] Although both studies tested groups of termites in laboratory conditions, the results indicate that indoxacarb could be a good candidate as soil insecticide against subterranean termites. Currently, trials are under way to evaluate the effectiveness of indoxacarb for subterranean termite control under field conditions.

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REFERENCES

1 Lenz M and Runko S, Protection of building, other structures and materials in ground contact from attack by subterranean

termites (Isoptera) with a physical barrier—a fine mesh of high grade stainless steel. *Sociobiology* **24**:1–16 (1994).
2 Su N-Y, Scheffrahn RH and Ban PM, Uniform size particle barrier: a physical exclusion device against subterranean termites (Isoptera: Rhinotermitidae). *J Econ Entomol* **84**:912–916 (1991).
3 Grace JK and Su N-Y, Evidence supporting the use of termite baiting systems for long-term structural protection. *Sociobiology* **37**:301–310 (2001).
4 Su N-Y, Ban PM and Scheffrahn RH, Polyethylene barrier impregnated with lambda-cyhalothrin for exclusion of subterranean termites (Isoptera: Rhinotermitidae) from structures. *J Econ Entomol* **97**:570–574 (2004).
5 Lewis VR, Alternative control strategies for termites. *J Agric Entomol* **14**:291–307 (1997).
6 Su N-Y, Tamashiro M and Haverty MI, Characterization of slow-acting insecticides for the remedial control of the Formosan subterranean termite (Isoptera: Rhinotermitidae). *J Econ Entomol* **80**:1–4 (1987).
7 Kard B, Gulfport studies stay the course. *Pest Control* **69**:30–33 (2001).
8 Thorne BL and Breisch NL, Effects of sublethal exposure to imidacloprid on subsequent behavior of subterranean termite *Reticulitermes virginicus* (Isoptera: Rhinotermitidae). *J Econ Entomol* **94**:492–498 (2001).
9 Osbrink LA, Lax AR and Brenner RJ, Insecticide susceptibility in *Coptotermes formosanus* and *Reticulitermes virginicus* (Isoptera: Rhinotermitidae). *J Econ Entomol* **94**:1217–1228 (2001).
10 Kard BM, Integrated pest management of subterranean termites (Isoptera). *J Entomol Sci* **8**:200–224 (2003).
11 Myles TG, Development and evaluation of a transmissible coating for control of subterranean termites. *Sociobiology* **28**:373–457 (1996).
12 Ibrahim SA, Henderson G and Fei H, Toxicity, repellency and horizontal transmission of fipronil in the Formosan subterranean termite (Isoptera: Rhinotermitidae). *J Econ Entomol* **96**:461–467 (2003).
13 Shelton TG and Grace JK, Effects of exposure duration on transfer of nonrepellent termiticides among workers of *Coptotermes formosanus* Shiraki (Isoptera: Rhinotermitidae). *J Econ Entomol* **96**:456–460 (2003).
14 McCann SF, Annis GD, Shapiro RD, Piotrowski W, Lahm GP, Long JK, Lee KC, Hughes MM, Myers BJ and Griswold SM, The discovery of indoxacarb: oxadiazines as a new class of pyrazoline-type insecticides. *Pest Manage Sci* **57**:153–164 (2001).
15 Lahm GP, McCann SF, Harrison CR, Stevenson TM and Shapiro R, Evolution of the sodium channel blocking insecticides: the discovery of indoxacarb, in *Agrochemical discovery: insect, weed, and fungal control*, ed. by Baker DR and Umetsu NK, ACS Symposium Series no. 774, American Chemical Society, Washington DC, pp. 20–34 (2001).
16 US Environmental Protection Agency, *Indoxacarb, Pesticide Fact Sheet*. Office of Prevention, Pesticides and Toxic Substances (7505C), Washington, DC (2000).
17 Appel AG, Laboratory and field performance of an indoxacarb bait against German cockroaches (Dictyoptera: Blattellidae). *J Econ Entomol* **96**:863–870 (2003).
18 Hu XP, Evaluation of the efficacy and non-repellency of indoxacarb and fipronil treated soil at various concentrations and thicknesses against two subterranean termites (Isoptera: Rhinotermitidae). *J Econ Entomol* **98**:509–517 (2005).
19 Ferster B, Scheffrahn RH, Thoms EM and Scherer PN, Transfer of toxicants from exposed nymphs of the drywood termite *Incisitermes snyderi* (Isoptera: Kalotermitidae) to unexposed nestmates. *J Econ Entomol* **94**:215–222 (2001).
20 Hu XP and Appel AG, Seasonal variation of critical thermal limits and temperature tolerance in two subterranean termites (Isoptera: Rhinotermitidae). *Environ Entomol* **33**:97–205 (2004).
21 Su N-Y, Ban PM and Scheffrahn RH, Evaluation of twelve dye markers for population studies of the eastern and Formosan subterranean termite (Isoptera: Rhinotermitidae). *Sociobiology* **19**:349–362 (1991).
22 *Statistix® 8*. Analytical software, Tallahassee, FL (2003).
23 Haagsma KA and Rust MK, Effect of hexaflumuron on mortality of the western subterranean termite (Isoptera: Rhinotermitidae) during and following exposure and movement of hexaflumuron in termite groups. *Pest Manag Sci* **61**:517–531 (2005).
24 Suarez ME and Thorne BL, Rate, amount and distribution pattern of alimentary fluid transfer via trophallaxis in three species of termites (Isoptera: Rhinotermitidae, Termopsidae). *Ann Entomol Soc Am* **93**:145–155 (2000).
25 Tomalshi MD and Vargo EL, Chain reaction. *Pest Control* **May**:51–53 (2004).
26 Myles TG, Laboratory studies on the transmission of *Metarhizium anisopliae* in the eastern subterranean termite, *Reticulitermes flavipes* (Isoptera: Rhinotermitidae), with a method for applying appropriate doses of Conidia to trapped termites for release. *Sociobiology* **40**:265–276 (2002).
27 Thorne BL and Breisch NL, Effects of sublethal exposure to imidacloprid on subsequent behavior of subterranean termite *Reticulitermes virginicus* (Isoptera: Rhinotermitidae). *J Econ Entomol* **94**:492–498 (2001).
28 Spragg WT and Paton R, Tracing, trophallaxis and population measurement of colonies of subterranean termites (Isoptera) using a radioactive tracer. *Ann Entomol Soc Am* **73**:708–714 (1980).
29 Sheets JJ, Karr LL and Dripps JE, Kinetics of uptake, clearance, transfer and metabolism of hexaflumuron by eastern subterranean termites (Isoptera: Rhinotermitidae). *J Econ Entomol* **93**:871–877 (2000).