Performance of Altriset™ (Chlorantraniliprole) Termiticide Against Formosan Subterranean Termites, *Coptotermes formosanus* Shiraki, in Laboratory Feeding Cessation and Collateral Transfer Trials, and Field Applications

by

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ABSTRACT

Chlorantraniliprole represents the first compound to be registered as a termiticide by the Environmental Protection Agency (EPA) in over a decade. This novel termiticide is currently registered as a 'reduced-risk pesticide' by the EPA. Laboratory and field trials were conducted to quantify mortality of Formosan subterranean termites (FST), *Coptotermes formosanus* Shiraki resulting from chlorantraniliprole treated soil, the degree to which the termites curtail feeding intensity post-exposure to chlorantraniliprole treated soil, collateral transfer of chlorantraniliprole among nest mates, and the effectiveness of chlorantraniliprole as a remedial treatment against structural infestations of FST. Termites which were exposed to chlorantraniliprole treated soil consumed significantly less paper than unexposed FST. The mean percent mortality of those termites exposed to chlorantraniliprole treated soil was significantly greater than that of unexposed FST. Depending on donor:recipient ratios, the mean mortality of recipients ranged from 14.65 – 90.00 % in the collateral transfer trials. There was a positive correlation between increased donor density and recipient mortality. Through 24 mo post-treatment, 27.3% of the structures which were treated in field trials were observed to have infestations of termites that required re-treatment; however, no FST were observed during the 30 and 36 month post-treatment inspections. Additionally, a novel scoring rubric was developed that will allow standardization of field study sites with respect to dissimilarity in site variables, and will allow for more consistent comparison of results across disparate field experiments. An explanation for

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the lack of successful remediation of many of the structures involved in the field trial is proposed and is based on our novel scoring system.

Key Words: *Coptotermes*, Chlorantraniliprole, Termiticide, Reduced-Risk, Invasive Species

INTRODUCTION

Formosan subterranean termites (FST) *Coptotermes formosanus* Shiraki (Isoptera: Rhinotermitidae), are an invasive insect pest in the United States and elsewhere. While this termite species is endemic to mainland China (Kistner 1985), their introduction into the Hawaiian archipelago is believed to have been the result of maritime activity originating from the Island of Taiwan (formerly Formosa), with subsequent introductions to continental United States originating from Hawaii (Su and Scheffrahn 1998; Cabrera *et al*. 2000; Hawthorne *et al*. 2000; Howell *et al*. 2000; Scheffrahn *et al*. 2001; Jenkins *et al*. 2002). However, Austin *et al*. 2006 suggests two distinct FST introductions to Hawaii, and then the continental United States, both origi-nating from mainland China.

FST represent one of the most economically important pest insects in the United States. Estimates indicate that the cost associated with monitoring, control, and repair of damages caused by FST exceeds $1 billion per year (Paudel *et al*. 2010). FST control methods include physical and chemical barriers to prevent termites from gaining access to structures, biological con-trols (including nematodes, bacteria, fungi, and botanical extracts), chemical treatment of soil and wood, and baits (Verma *et al*. 2009). The general public is educated regarding many of these termite control tactics, but when surveyed they cite (in descending order of acceptability) perimeter treatment with a liquid termiticide, bait treatment, and liquid + bait treatments at the most preferred methods of termite control (Paudel *et al*. 2010). However, there is an increasing emphasis towards the development of least-risk chemical treat-ments, as well as non-chemical tactics for termite control (Lewis 1997).

Belonging to a new class of chemical insecticides, chlorantraniliprole (Al-triset™) was recently developed and marketed by DuPont Crop Protection. Chlorantraniliprole is currently classified as a 'reduced-risk pesticide' by the Evironmental Protection Agency (EPA, 2008). The compound is an anthra-nilic diamide, and exhibits a novel mode of action in which insect ryanodine

receptors are activated, resulting in rapid paralytic muscle dysfunction (Hannig *et al.* 2009, Cordova *et al.* 2006, Cordova *et al.* 2007, Lahm *et al.* 2005, and Lahm *et al.* 2007). Regulation of the release of internal cell calcium is affected by activation of ryanodine receptors. The downstream physiological effect of this disruption of calcium homeostasis results in feeding cessation, and eventual death of the insect (Teixeira *et al.* 2008). The effectiveness of this compound has been demonstrated in mortality trials against a variety of insect species belonging to the orders Coleoptera, Diptera, and Hemiptera (Kuhar *et al.* 2008, Palumbo 2008, and Schuster 2007).

We designed laboratory trials to study the efficacy of chlorantraniliprole on FST feeding rates, collateral transfer of chlorantraniliprole among FST nestmates, and field trials to determine the effectiveness of chlorantraniliprole to control infestations of FST in structures and to protect those structures from reinvasion through time. Gautam & Henderson (2011) described the effects of chlorantraniliprole on FST in laboratory trials. Our work represents a synthesis of data related to the effectiveness of this new compound on FST in field applications correlated with laboratory trials.

MATERIAL AND METHODS

Laboratory Trials:
Termite Feeding Cessation

FST were field-collected from Beaumont, TX approximately 2 weeks prior to the initiation of this study. Sandy-loam soil was prepared for these trials by treating it with chlorantraniliprole using the following procedures. A 1,000 ppm stock solution of chlorantraniliprole and water was made by adding 0.10 g of technical grade chlorantraniliprole to 100 ml of deionized water. Next, a serial dilution of the stock solution was accomplished by adding 15 ml of deionized water to 15 ml of stock solution. This final solution was added to 270 g of soil and distributed by mixing the soil with a stir rod within a 750 ml plastic beaker. The treated soil was allowed to rest for a period of 24 hrs. Glass test-tubes (10 cm in length and open at both ends) served as arenas for this experiment and followed the design of Gold *et al.* 1996 (Fig. 1). After preparation, the tubes were stored vertically in test-tube racks (untreated-agar end at top). Tubes were stored at 25°C overnight to allow moisture to become uniformly distributed in the matrix. There were 10 replications
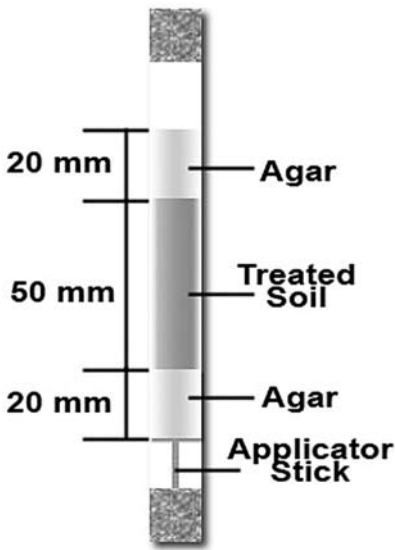
Fig. 1. Schematic showing components of glass-tube arenas used in FST feeding cessation experiment.

of each treatment and untreated control groups.

After the 24 hr period had elapsed, 20 FST workers and 5 soldiers per replication were randomly selected from laboratory stock and introduced into the "untreated-agar end" of the test-tube arenas, and the introduction time was recorded. The time at which the tunneling termites reached the treated soil was recorded as the exposure 'start' time. Cohorts of termites were allowed to tunnel in the soil for four different time periods (1, 2, 4, & 8 hr). These four time periods represent four distinct treatments. After the termites had tunneled for the pre-determined time period, the rubber stoppers were removed and termites and soil were carefully tapped out through the untreated end of the arena into a clean Petri-dish. To measure feeding intensity, pre-feeding digital images of 5 X 5 cm pieces of brown paper towel (Cormatic-Georgia Pacific, Atlanta, GA) were taken for comparison to post-feeding images at the end of the trial. Using soft forceps, the termites were then carefully moved to a smaller Petri dish containing the paper towel, half of which was covered by 50 g of play sand, moistened with 10 ml of water. This arrangement of paper towel and moistened sand provided ambient humidity within the arenas, as well as constantly moist paper towel, on which the termites fed. Petri dishes were sealed with ParaFilm , transferred to an environmental chamber, and maintained at 25 ± 5° C and 85 ± 5% RH. Untreated control tubes were constructed in the same manner as the treatment tubes, but the soil remained untreated. Termite cohorts were allowed to tunnel for the same time as the treatment groups periods (1, 2, 4, and 8 h). Termites were transferred to

identical feeding dishes, and feeding rates were then calculated in the same manner as in the treatment groups. These control groups are referred to as 'Tunneling Controls' (TC). An additional set of controls which were not allowed to tunnel, were established in feeding dishes (as above) to compare the amount of feeding for the same time period as the treatment and TC groups. Twenty termite workers and 5 soldiers were introduced directly into these Petri dishes without subjecting them to tunneling, and observation periods were identical to the treatment and untreated control groups. This control group is referred to as 'Feeding Controls' (FC).

Termite mortality was recorded daily for 12 d post-exposure. At the end of the trial, the remaining paper towel was allowed to dry in the laboratory, after which, digital images of the paper were taken with a Canon EOS 50D 15.1 megapixel digital camera fitted with a 28-135 mm lens (Canon U.S.A., Inc. Lake Success, NY). Pre-feeding and post-feeding images were then compared using SigmaScan PRO v.5.0 photo-editing software, and the surface area (cm^2) differential was calculated and statistically analyzed (calibration of the photography technique was made for each image prior to area measurement).

Collateral Transfer

As in the feeding cessation study detailed above, termites used in this study were field-collected from Beaumont, TX approximately 2 weeks prior to the initiation of the trial. Ten replications of each treatment (donor:recipient ratios), and untreated controls were conducted for this trial. Arenas consisted of a 15 cm Petri dish, each with a a 7.6 X 7.6 cm piece of brown paper towel (Cormatic-Georgia Pacific, Atlanta, GA) placed on the floor of the Petri dish and moistened with 8-10 droplets of water from a 25 ml Samco Scientific Corporation pipette (San Fernando, CA). The paper served as food and harborage for termites. Donor:recipient ratios in this trial included 0:20 (untreated controls), 1:19, 5:15, 10:10, 15:5, 19:1, and 20:0. Donor FST were marked using orange Rust-Oleum Marking Paint (Vernon Hills, IL). The methodology used to mark the the donors was similar to that described by Forschler 1994. A stock solution of 50 ppm was made using formulated chlorantraniliprole. Donor FST were treated on the thorax with 0.3 μl of 50 ppm chlorantraniliprole using a Hamilton 700 series micro syringe pipette (Reno, NV). Two untreated FST soldiers were added to each arena. Addition-

ally, two sets of post-treatment observations were made to observe mortality at 4 h, then daily through 7 d. Feeding intensity was measured in these trials by taking pre-feeding images of the 7.6 X 7.6 cm brown paper towel and post-feeding images at the end of these trials (7 d after treatment). As described above, pre- and post-feeding images were compared using SigmaScan PRO v.5.0 photo-editing software, and the surface area (cm^2) differential was the metric used for statistical analysis. Calibration of photography was made for each image prior to area measurement. A separate trial was established simultaneously to determine if marking had a significant deleterious effect on the termites. This trial included ten replications each of untreated-unmarked controls and untreated-marked controls.

Field Trials:

For the purposes of this field trial, Center for Urban and Structural Entomology (CUSE) and DuPont personnel jointly inspected and agreed upon 11 structures with monolithic slabs or pier and beam construction, each of which had at least one active FST shelter tube on the exterior of the structure. The minimum area of any structure included in the trial was 78.97 m^2. Pre-treatment inspections also included the interior of structures and bath traps. A diagram of each structure was prepared by CUSE personnel, who measured the size and recorded the shape of the foundation, and areas of termite activity. Live termites were collected from each structure, preserved in 100% ethanol, and stored as voucher specimens. All of these structures were located in southeast Texas. Chlorantraniliprole treatments were made by a pest management professional at each property according to the manufacturer's label directions. These treatments were overseen by CUSE personnel. All structures were treated between May and July 2008. Nine of the structures were of monolithic slab construction, and the two were pier and beam.

Post-treatment exterior (and interior when possible) inspections were made on or about 2 wk, and then at 1, 3, 6, 12, 18, 24, 30, and 36 months. This monitoring schedule is more robust than that which would generally be incorporated into professional pest management protocols. If active termites were discovered during post-treatment inspections, termite samples were collected and preserved in 100% ethanol as voucher specimens. DuPont authorized personnel were notified of any post-treatment activity before any supplemental treatments (ST) or re-treatments (RT) were performed.

A supplemental treatment (ST) in this study is defined as:

(a). Chlorantraniliprole spot treatments to active termite areas that were not originally treated at the initial application. This is not a failure of chlorantraniliprole, since the termiticide was not applied to that area, and termites penetrated through an untreated zone.

(b). Treatment of infested elements of construction where termites survived due to conducive conditions, such as leaking pipes, unusual construction elements that did not allow application to reach the infested area, or an isolated above ground colony with no soil contact.

(c). Treatment where a conducive condition existed which contributed to or allowed termites to remain active and penetrate the treated zone. Thus, it was not considered a chlorantraniliprole failure.

A re-treatment (RT) in this study was defined as: the application of chlorantraniliprole as a spot treatment to active termite areas that were originally treated at the initial application. This was considered a failure of chlorantraniliprole. That is, termites were able to penetrate through the treated zone; however, if that area of penetration had conducive conditions (or other issues as above) that allowed termites to penetrate, and the condition was not corrected, then this was considered a supplemental treatment (ST) as described above.

In this study, all structures were ranked based on several parameters related to the difficulty of the structure-specific treatment procedures. The parameters used to populate the ranking rubric included: termite species (1-desert termite, 2- drywood termite, 3-*Reticulitermes*, and 6-*Coptotermes*); number of mud tubes and location; the number of conducive conditions present at each structure; and, construction type (1 monolithic slabs, 2 pier and beam, and 3 floating slabs). The Total Difficulty Score (TDS) of each structure was calculated by summing all points assigned for each category. It is presumed that difficulty to control termites at structures is positively correlated with higher TDS.

RESULTS

Termite Feeding Cessation

Consumption: All treatment cohorts that were exposed to chlorantraniliprole treated soil (regardless of exposure time) consumed significantly

less paper ($F = 21.37$; df = 8,89; $P < 0.01$) than the 'Tunneling Controls' (TC) and 'Feeding Controls' (FC) (Fig. 2). Additionally, the mean amount of paper consumed by FST after exposure to chlorantraniliprole treated soil was negatively correlated with time of exposure (Fig. 2). A similar trend was not observed in the TC (Fig. 2).

Mortality: With the exception of the 1 and 4 hr Tunneling Control groups, the mean % mortality of FST remained below 10% in the untreated controls through 11 d after exposure to untreated soil (Fig. 3). At and beyond 3 d post exposure, the mean % mortality of all treatment cohorts that were exposed to chlorantraniliprole treated soil (regardless of exposure time) was significantly greater ($F = 9.64$; df = 8,89; $P < 0.01$) than the TC and FC (Fig. 3). Additionally, with the exception of the 12 d observation period, the mean % mortality of treatment cohorts that were exposed to chlorantraniliprole treated soil was positively correlated with time of exposure.
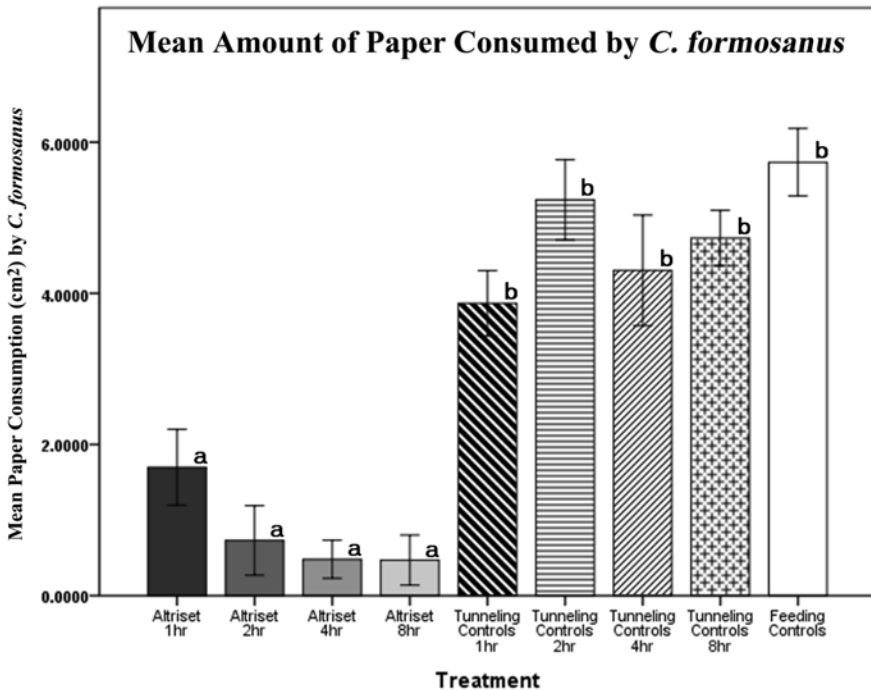

Fig. 2. Mean paper consumed (cm2) by FST through 12 d after exposure to chlorantraniliprole treated soil, untreated soil, or no soil. Treatments were replicated 10 times. Bars with the same letter are not significantly different using Analysis of Variance (ANOVA) and Tukey's HSD mean separation test at P < 0.05.

Collateral Transfer

Mortality- Percent donor mortality in all of the treatment ratios was significantly different ($F = 17.73$; df $= 8,89$; $P < 0.01$) than that of the 0:20 donor:recipient ratio (untreated controls) at 7 d post-treatment (Table 1). However, there were no significant differences in donor mortality between the different treatment ratios at the 7 d observation period, and mean mortality ranged from 50.00 – 76.00 % (Table 1). There were significant differences ($F = 17.73$; df $= 8,89$; $P < 0.01$) in recipient mortality levels among the different treatment ratios at 7 d post-treatment, and mean mortality ranged from 14.65 – 90.00 % (Table 1). Regarding total mortality (donors and recipients) for each donor:recipient ratio, there were significant differences ($F = 7.58$; df $= 8,89$; $P < 0.01$) at the 7 d post-treatment observation period (Table 1). The greatest percent total mortality (74.5 %) occurred in the 15:5 ratio at 7 d, followed by 67.5% mortality in the 19:1 donor:recipient ratio (Table 1). Mortality in the untreated and unmarked group was 9.00 %, and was not significantly different from that of the untreated and marked group (21.00 %) starting at 24 h thru 120 h post-treatment ($F = 0.067$, df$=19$, $P = 0.31$).

Consumption- There were significant differences in consumption of paper among the different treatment groups and the 0:20 (untreated control)
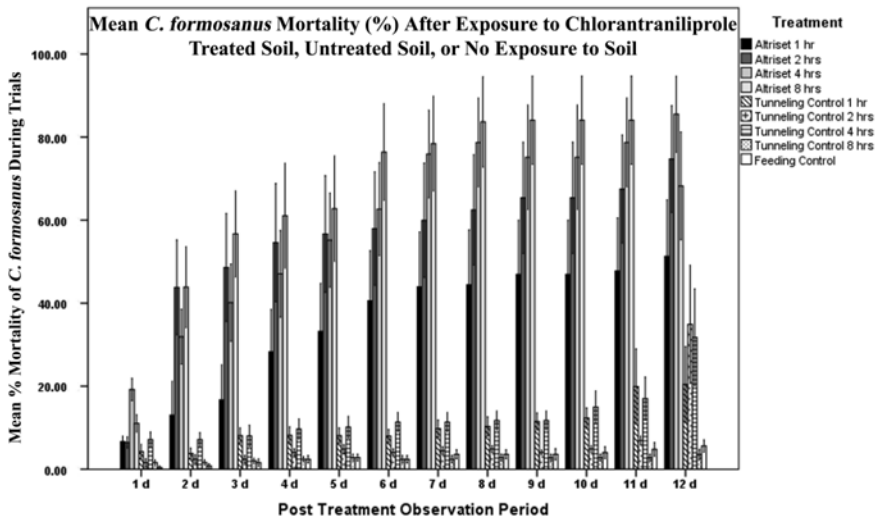

Fig. 3. Mean accumulated FST % mortality through 12 d of observation.

Table 1. Mean percent mortality at 7 d post-treatment of FST donors and recipients when donors were treated with 50 ppm chlorantraniliprole.

	% Mortality		Total
Ratio	Donor	Recipient	% Mortality
0:20 (untreated control)	0.00 (a)	42.50 (bcd)	42.50 (abcd)
1:19	70.00 (b)	48.93 (cd)	47.50 (bcd)
5:15	76.00 (b)	14.65 (abc)	29.50 (abc)
10:10	68.00 (b)	62.00 (de)	65.00 (cd)
15:5	69.98 (b)	88.00 (e)	74.50 (d)
19:1	65.76 (b)	90.00 (e)	67.50 (bcd)
20:0	50.00 (b)	0.00 (a)	50.00 (bcd)

*Means followed by the same letter(s) in the same column are not significantly different (Tukey's HSD, P=0.05)
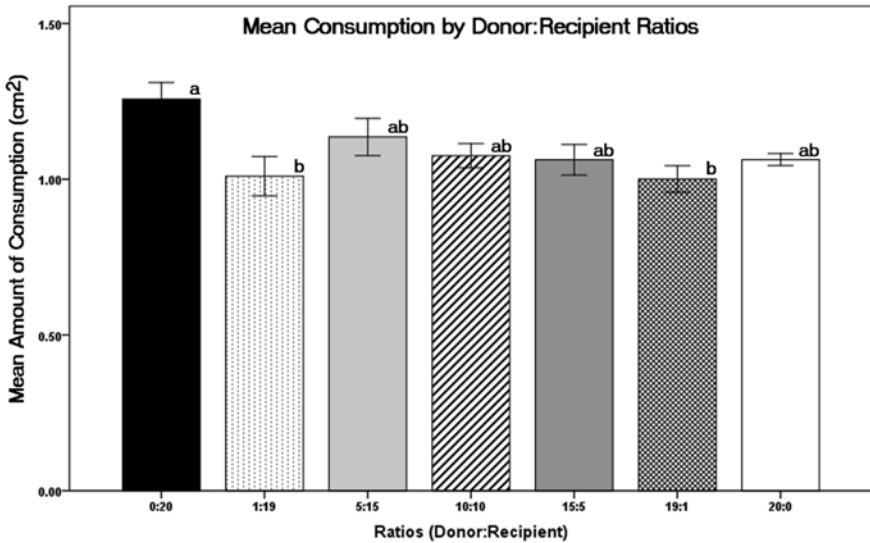

Fig. 4. Mean amount of consumption of substrate (cm2) by FST (donors and recipients) in each donor:recipient ratio through 7 d post-treatment. Bars with the same letter are not significantly different using Analysis of Variance (ANOVA) and Tukey's HSD mean separation test at P < 0.05.

Table 2. Mean, Standard Deviation, and Range of exterior and interior mud tubes, perimeter length, and amount of chlorantraniliprole applied to the exterior and interior of structures associated with the field study during initial observations and treatment.

	Exterior Mud Tubes	Interior Mud Tubes	Perimeter Length (m)	Product Applied to Exterior (l)	Product Applied to Interior (l)
Mean and S.D.	3.91 (3.28)	0.18 (0.40)	57.05 (55.90)	302.15 (25.37)	10.33 (1.62)
Range	1 - 10	0 - 1	30.48 – 91.44	151.42 - 454.25	0.00 -18.93

donor:recipient group ($F = 15.33$; df = 8,89; $P < 0.01$) (Fig. 4). The greatest consumption occurred in the 0:20 ratio, followed by the 5:15, and then the 10:10 (Fig. 4).

Field Trial

The mean number of pre-trial exterior termite mud tubes per structure was 3.91, and ranged from 1 – 10 (Table 2). The mean volume of finished solution applied to structure exteriors was 302.15 L, and 10.33 L on the interiors (Table 2). During the first 24 mo of the 36 mo trial duration, 27.27% of the structures were infested with FST and required re-treatment (RT), and 18.18% required supplemental treatment (ST). This includes Structure #3 in which FST were not completely controlled until a re-treatment (RT) was made after the 1 mo inspection, when FST were discovered on the exterior at the original site of infestation (Fig. 5 and Table 3). The re-treatment (RT) was performed with 15.14 L of chlorantraniliprole. Due to damage caused by hurricane Ike, access to eight of the structures was limited at the 3 mo post-treatment inspection, and only three of the eleven structures were inspected. No termite activity was found at those three structures. At the 6 mo inspection, two structures were found to have termite activity (Fig. 5 and Table 3), including Structure #2, which had active termites on the exterior and received a re-treatment (RT) with 15.14 L of chlorantraniliprole. Additionally during the 6 mo inspection, active termites were found swarming from an interior wall within structure #9. After further investigation of the swarm, a previously unknown cold joint was discovered and this structure received a supplemental treatment (ST) with 37.85 L of chlorantraniliprole. At the 12 mo inspection, Structure #8 had active FST swarming from a previously treated bath trap. Upon further examination, it was determined that there was a water leak in

Table 3. Data-populated scoring rubric developed for, and used in these trials to standardize the total difficulty score (TDS) associated with treatment of each structure.

Structure #	Termite Species	# of Mud Tubes	Construction Type	# of Conducive Conditions	Total Difficulty Score (TDS)	# of ST's	# of RT's
1	*C. formosanus*=6	10 Exterior	Monolithic=1	1	18.0	0	0
2	*C. formosanus*=6	10 Exterior	Monolithic=1	1	18.0	0	2
3	*C. formosanus*=6	1 Exterior 1 Interior	Monolithic=1	1	10.0	0	1
4	*C. formosanus*=6	3 Exterior	Pier & Beam=2	2	13.0	0	0
5	*C. formosanus*=6	2 Exterior 1 Interior	Pier & Beam=2	1	12.0	0	1
6	*C. formosanus*=6	4 Exterior	Monolithic=1	1	12.0	0	0
7	*C. formosanus*=6	5 Exterior	Monolithic=1	1	13.0	0	0
8	*C. formosanus*=6	3 Exterior	Monolithic=1	1	11.0	1	0
9	*C. formosanus*=6	2 Exterior	Monolithic=1	1	10.0	2	0
10	*C. formosanus*=6	2 Exterior	Monolithic=1	1	10.0	0	0
11	*C. formosanus*=6	1 Exterior	Monolithic=1	1	9.0	0	0
Mean & (S.D.)	6 (0.00)	3.91 (3.24)	1.18 (0.40)	1.09 (0.30)	12.36 (3.07)	0.27 (0.64)	0.36 (0.67)

NOTE: All structures were ranked on the difficulty of the treatment based on several parameters which included, termite species, number of mud tubes and location, and the number of conducive conditions present at each structure. The table details the ranking of termite species based on colony size: 1 desert termite, 2 drywood termite, 3 *Reticulitermes*, and 6 *Coptotermes*. Each structure received one point for every mud tube located on the structure, and one point for each conducive condition found to be present at the structure. Construction type was also given a treatment difficulty ranking as follows: 1 monolithic slab, 2 pier and beam, and 3 floating slab. The Total Difficulty Score (TDS) of each structure was calculated by summing all points assigned for each category.

the bath trap area, this leak was repaired, and the area received a supplemental treatment (ST) using 22.71 L of chlorantraniliprole. No subterranean termite activity was found during the 18 mo inspections. At 22 mo post-treatment, Structure #9 homeowners notified us of FST swarming again from the same cold joint, but in a different area. This area received a supplemental treatment

(ST) with 189.27 L of chlorantraniliprole. This ST is included in the 24 mo post-treatment observation period on Fig. 5. Also during the 24 mo inspection, active FST were discovered at Structures # 2 and 5 (Fig. 5 and Table 3). Both structures had active termites on the exterior, and both were re-treated (RT) with 15.14 L of chlorantraniliprole. No subterranean termite activity was found on any of these structures at the 30 or 36 mo inspection. After ranking, using the rubric described in the Material and Methods section, the mean total difficulty score (TDS) of the structures in this study was 12.36, and ranged from 9 – 18 (Table 3). At least one ST was required in 18.18% of the structures, and all were made to structures that ranked below the mean TDS. Of the re-treated structures (RT), all but one occurred in structures which ranked above, or just below the mean TDS (Table 3).
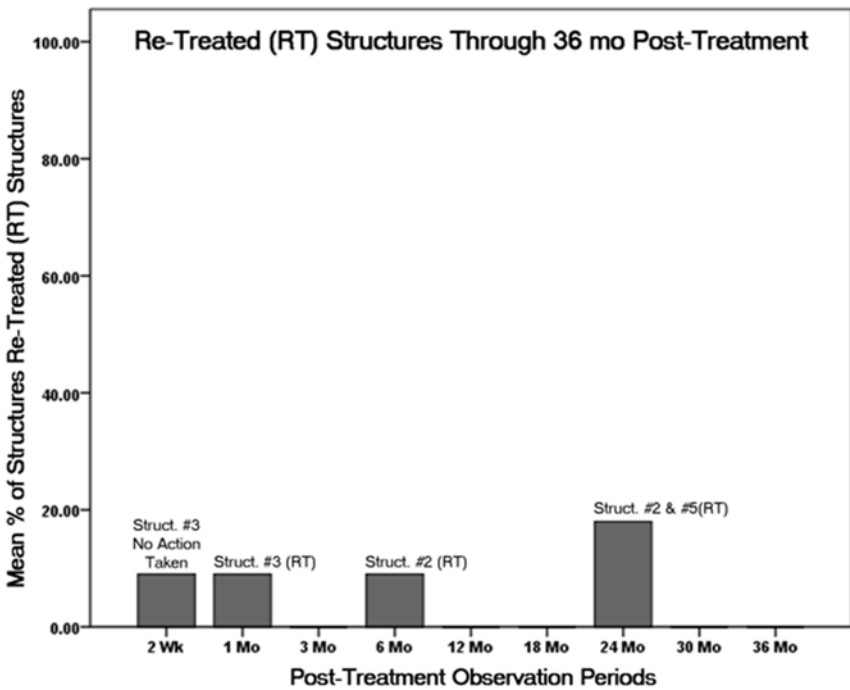

Fig. 5. Mean % of re-treated (RT) structures infested with FST after a post-construction treatment with chlorantraniliprole through 36 mo post-treatment. Nomenclature above bar: Individual structure number and RT (re-treatment). NOTE: Active termites were found at Structure #3 at 2 weeks post-treatment, but no corrective action was taken until 1 month post-treatment.

DISCUSSION

In laboratory trials, exposure of FST to chlorantraniliprole treated soil resulted in significantly greater mortality and significantly less consumption of food than either; 1) FST exposed to untreated soil, and 2) FST which were not exposed to soil at all. These data suggest that chlorantraniliprole could be considered an appropriate soil barrier-treatment for use against FST. However, it should be noted that the minimum termite exposure time investigated in these trials was 1 hr. It is not known if it would be realistic to presume that foraging termites would remain in a 'treatment zone' for this period of time in actual situations in which chlorantraniliprole is used to provide perimeter protection to a structure against subterranean termite infestations. Thus, we intend to initiate further investigations to determine the minimum temporal exposure threshold required for significant mortality and consumption cessation.

The significantly greater mortality of chlorantraniliprole-exposed termites relative to that of the TC and FC is a noteworthy aspect of this study. However, it is important to note that mortality of termites which were exposed to chlorantraniliprole ranged from a minimum of 51.31% (1 hr of exposure) to a maximum of 85.51% (4 hrs of exposure) after 12 d of post-treatment observation. It is likely that the mortality exhibited by these termites was partially the result of deprivation of nutrients resulting from feeding cessation. This effect of treatment is significant, and demonstrates a potential mode of action of chlorantraniliprole, and subsequent cascade of physiological alterations in termites that is more nuanced than that of many other classes of termiticides.

No attempt was made in these trials to determine the minimum concentration required to illicit the observed feeding inhibitory effects. We intend to design experiments to investigate this aspect of chlorantraniliprole. Additionally, the effects of chlorantraniliprole on other important subterranean termite species should likewise be investigated.

The positive correlation (with exception of 5:15 Donor:Recipient ratio) between increased donor numbers and recipient mortality provides evidence of the transfer effect of chlorantraniliprole (Altriset™) among FST nestmates, and suggests that this compound could affect mortality of FST in

field applications via insect-to-insect transfer. This density-dependent effect is dependent on the density of chlorantraniliprole-exposed insects (donors) relative to the density of recipients. This of course calls into question the feasibility and efficiency of reliance on this insect-to-insect transfer effect in order for the use of this product to provide effective remedial or prophylactic treatments on FST populations. That is, the density of chlorantraniliprole-exposed foragers (donors) in field treated colonies would likely not reach a level which would be analogous to the donor:recipient ratios which were required in these trials to significantly effect the density of recipients, and thus the colony. Interestingly, mortality of the donor population remained lower than expected and, on average (66.62 % overall mean mortality of donors) was similar to that observed in FST after exposure to chlorantraniliprole treated soil for 4 and 8 hrs in the feeding cessation trial. When considered in their totality, these results suggest that the chlorantraniliprole concentration to which FST were exposed in these trials (50 ppm) may be less than that required for acceptable termite control in field applications. However, Altriset™ is labeled for application at 500 ppm, or 10 times the concentration used in this laboratory trial.

Evidence of the slow acting nature of chlorantraniliprole when applied to infestations of FST is provided by the fact that several of the structures (27.27%) which were treated and monitored over the course of these trials were observed to have continuing infestations of FST. These structures required re-treatment (RT) during the first 24 mo of the monitoring phase of the study. In one case, RT became necessary twice within the same structure. However, by 30 months post-treatment, no FST activity was observed at any of the structures. This suggests that while chlorantraniliprole effectively controlled FST at structures in these trials, the compound requires a longer period of time to reduce populations of structure-infesting FST than alternative termiticides, but the end result of structure protection is the same.

Comparisons of field experiment results (such as those discussed in the Field Trial section of this document) across treatment environments are greatly complicated by the myriad variations associated with treatment scenarios and structure variables (French 1988; French and Ahmed 2005). Differences in treatment sites such as termite species, degree and location of infestation, degree and location of conditions conducive to termite infestation, foundation

type, construction materials, and soil type result in great difficulty in standard-ization of the finite number of treatment environments that are available to researchers. We have proposed a rubric for scoring treatment environments which allows for enhanced ability to compare and assess data related to field experimentation such as that documented herein. Careful examination of the results of the utilization of this rubric in this work revealed an interesting bifurcation of the mean total difficulty scores (TDS) of those structures that required either a supplemental treatment (ST) or re-treatment (RT). That is, the TDS of all ST's fell below the mean of 12.37, and a trend was noted in which RT's scored greater than or just below the mean TDS. This more thorough characterization and comparison of our treatment environments provides a refined and more complete picture of the challenges involved with treatment for FST in all study structures, and presents explanatory variables that we believe led to the lack of successful remediation and protection of RT structures with regards to FST. It is our hope that others will attempt to use this system in future work, and that after subsequent model refinement, eventually this will provide a consistent method to compare results across disparate, but related field research involving subterranean termites.

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