Pharaoh Ant (Hymenoptera: Formicidae) Colony Development After Consumption of Pyriproxyfen Baits

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ABSTRACT Pharaoh ant, *Monomorium pharaonis* (L.), colonies were effectively controlled following ingestion of pyriproxyfen formulated in peanut butter oil. Pyriproxyfen, a juvenile hormone analog, reduced egg production in the queens, decreased the amount of brood due to delayed death in the eggs and larvae, caused death of pupae about 3 wk after treatment, and decreased the number of workers due to attrition and toxic effects. Queens, which continued to produce a small amount of eggs, eventually died. Queen death may have been caused by lack of workers required to tend them, old age or toxic effects. At concentrations of 0.25, 0.5 and 1%, pyriproxyfen was more effective than the once commercially available bait, Pharorid (methoprene) for the control of the Pharaoh ant.

KEY WORDS *Monomorium pharaonis*, insect growth regulator, juvenile hormone

THE PHARAOH ANT, *Monomorium pharaonis* (L.), is a pervasive ant, having been carried by commerce to all regions of the world (Wheeler 1910). It is a major pest ant in Florida (Bieman and Wojcik 1990, Klotz et al. 1995), as well as other parts of the United States (Knight and Rust 1990) and the world. Infestations usually occur in large office buildings, houses, apartment complexes, factories, food establishments, and hospitals (Edwards 1986). In hospitals, it causes problems by contaminating equipment and sterile packaging, penetrating intravenous solutions and tubing, and feeding on dressed wounds. Worker ants can carry several pathogens including *Clostridium, Salmonella, Staphylococcus, Streptococcus,* and *Pseudomonas* (Beatson 1972).

Because *M. pharaonis* is a major pest, new active ingredients are sought for use in insecticidal baits. Insect growth regulators (IGR), such as methoprene and fenoxycarb, that were formulated in baits have provided control of this pest (Edwards 1975; Williams 1990; Williams and Vail 1993, 1994). Another IGR, pyriproxyfen (2-[1-methyl-2-(4-phenoxyphenoxy) ethoxy] pyridine [McLaughlin Gormley King, Minneapolis, MN]), has recently been shown to have IGR effects against the red imported fire ant, *Solenopsis invicta* Buren (Glancey et al. 1990, Banks and Lofgren 1991), and the big-headed ant, *Pheidole megacephala* F. (Reimer et al. 1991). Pyriproxyfen caused a reduction in oviposition by the queens, a reduction in worker brood caused by suspected low level toxicity and a caste shift from workers to reproductives in *S. invicta.* Queen oviposition by *P. megacephala* declined 2 wk after treatment and halted within 6 wk after treatment. Brood disappearance was attributed to pupal death. This study was conducted to evaluate the effects of pyriproxyfen baits on colony development of the Pharaoh ant.

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Materials and Methods

Laboratory colony rearing and maintenance were as described by Williams (1990).

Small Colony Evaluations. Evaluations were conducted with small laboratory colonies each containing 44.4 ± 6.5 (mean ± SEM) queens, 419.4 ± 46.4 workers, and a brood rating of 3.5 ± 0.2. Brood (eggs, larvae, and pupae) was rated by visually comparing a photograph of known quantities of brood with the brood in a cell. Each quantity of brood in the photograph was given a rating that was assigned as follows: 0.5 ≤ 50 pieces of brood, 1.5 = 0.012 g, 2.0 = 0.099 g, 2.5 = 0.187 g, 3.0 = 0.273 g, 3.5 = 1.317 g, 4.0 = 2.358 g, 4.5 = 3.4 g, and 5 ≥ 3.5 g.

Pyriproxyfen dissolved in the peanut butter oil, which served as an attractant and carrier, was evaluated at the following concentrations: 0.05, 0.5, 1, and 2.5% (AI) by weight. Pharorid (0.5% [AI], methoprene, Zoecon, Dallas, TX), a once commercially available insect growth regulator, was used for comparison purposes and the bait was formulated in liver powder:honey:sponge cake (2:1:1) by weight. The control colonies were fed only peanut butter oil. The bait solutions were offered to each colony in five 100-μl pipettes and Pharorid was offered to the colonies at an equivalent weight

in small plastic weigh boats. Colonies were held without food for 72 h before treatment. Formulations were introduced to the colonies twice at 1-wk intervals because previous results indicated that 1 baiting was insufficient for control when using fenoxycarb (Williams and Vail 1993). Formulations were offered to the colonies for 72 h after which they were removed and the colonies returned to the regular laboratory diet of adult mosquitoes and house flies, hardboiled chicken egg yolk, and honey-agar for 72 h. In addition, each colony was provided with cotton saturated with water for moisture. Food was removed for 24 h, then treatment formulations were again offered to each colony for 72 h after which the formulations were removed and the colonies given the regular laboratory diet 2 times a week. Percentage of bait consumption was determined by estimating the reduction in the solution levels in the pipettes and by ascertaining before and after weights of the methoprene baits. For each evaluation, three colonies were treated per dosage; 3 control colonies were given the same quantity of peanut butter oil alone.

Weekly observations were made on the status of the colony; queen number, type and quantity of brood, and estimated worker numbers (dead and live). Any obvious morphological anomalies were noted. The 3 main criteria for efficacy were the following: (1) the number of workers present, (2) the brood rating, and (3) the number of queens present. Colonies were evaluated once a week through 20 wk and at 24 and 28 wk or until the colony died or completely recovered and returned to normal. The colony was considered to be normal when the queens began normal egg laying, all stages of brood were present, and the amount of brood and number of workers were similar to or greater than before treatment. Once a colony returned to normal, it was removed from further evaluation.

Large Colony Evaluations. The most effective concentrations of pyriproxyfen in the small colony evaluations (0.5 and 1%), as well as 0.25%, and 0.5% methoprene, were evaluated against large laboratory colonies which contained (mean ± SEM) 126.0 ± 6.3 queens, 5107.9 ± 305.0 workers, and a brood rating of 4.5 ± 0.2. The pyriproxyfen was dissolved in peanut butter oil at 0.25, 0.5, and 1% by weight. Before initiation of the evaluation, colonies were starved for 72 h. Each colony was offered 1.0 ml of the treatment solution in micropipettes (ten 100-μl pipettes) for 72 h after which the solutions were removed and the colonies were returned to the regular laboratory diet for 72 h. Following 24 h of starvation, the baiting was repeated with each colony receiving 1.0 ml of solution for 72 h after which the colonies were returned to their regular diet. Also, 3 colonies were given methoprene formulated in peanut butter: honey according to manufacturer's recommendations for each baiting. Because 1 ml of peanut butter oil weighed ≈0.9 g, this amount of methoprene was used per colony. Consumption of each bait was recorded following the procedure used in the small colony evaluation. Three colonies were treated per dosage and 3 control colonies were given an equal amount of peanut butter oil without pyriproxyfen. Weekly observations similar to those in the small colony evaluations were made once a week for 8 wk and then every 2 wk through week 32.

Statistical Analysis. For the initial reading date, data (number of workers, queens, and brood rating) were transformed with the $\log(x + 1)$ and analyzed with the general linear model (GLM) procedures (SAS Institute 1993). Data were transformed to reduce variation and to generate a more normal distribution. For the successive readings, the percentage of reduction in each of the variables was calculated, arcsine transformed and analyzed with GLM. Tukey honestly significant difference (HSD) test was used for means separation ($P = 0.05$; SAS Institute 1993). The number of dead workers in the large colony evaluation was also $\log(x + 1)$ transformed before GLM analysis.

Results and Discussion

Small Colony Evaluations. All concentrations of pyriproxyfen were effective in reducing the colony size of Pharaoh ants based on the brood rating (Table 1), worker number (Table 2), and queen number (Table 3) throughout the study. Methoprene was not as effective as pyriproxyfen in reducing colony size. Brood was reduced by 33 and 50% at 2 wk and 4 wk, respectively, in colonies treated with methoprene (Table 1); however, by week 8, the brood rating for methoprene had increased and was no longer different from the control. Queen and worker numbers in the methoprene-treated colonies were never significantly different from the controls. By week 9, methoprene-treated colonies had returned to normal and were removed from further study.

Brood ratings for the pyriproxyfen-treated colonies were not significantly different from the controls on the initial reading date (0 wk) (Table 1). Three weeks after the 1st treatment and on all subsequent readings, the pyriproxyfen-treated colonies significantly reduced the brood rating except for 0.05% pyriproxyfen at 28 wk. The 0.05% treatment reduced the brood rating less than the other pyriproxyfen treatments throughout the study and the 0.05% colonies appeared to be recovering at the termination of the study. Brood was reduced most rapidly in the 0.5% pyriproxyfen treatment; by 12 wk, no brood was remaining. In the 1 and 2.5% treatments, no brood remained by 17 and 24 wk, respectively.

Initially, dead or dying brood was observed, followed by extended periods where only eggs were present in the pyriproxyfen-treated colonies. The eggs eventually shrivelled and died. It was not apparent if oviposition had ceased. Dead pupae and

Table 1. Mean percentage of reduction ± SEM in brood level by pyriproxyfen and methoprene baits (small colony evaluation)

% concn	Brood rating at week 0[a]	% reduction in brood rating at weeks indicated[b, c]									
		2	3	4	6	8	12	16	20	24	28
Pyriproxyfen											
0.05	3.5 ± 0.50a	24.1 ± 4.9ab	40.7 ± 7.4ab	46.3 ± 6.7a	72.2 ± 14.7ab	57.4 ± 14.5a	64.8 ± 17.7a	61.1 ± 20.0a	55.6 ± 22.2a	50.0 ± 25.5a	38.9 ± 33.8ab
0.5	3.0 ± 0.00a	22.2 ± 11.1ab	55.6 ± 14.7a	83.3 ± 0.0a	88.8 ± 5.6a	88.9 ± 5.6a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
1.0	3.7 ± 0.44a	8.5 ± 4.3ab	43.9 ± 6.4ab	65.3 ± 12.0a	57.9 ± 4.8ab	78.6 ± 6.0a	85.2 ± 14.8a	96.3 ± 3.7a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
2.5	4.3 ± 0.17a	7.4 ± 3.7ab	19.4 ± 4.2b	53.7 ± 1.9a	57.9 ± 6.8ab	57.9 ± 6.8a	69.9 ± 18.3a	77.3 ± 16.8a	81.0 ± 13.2a	100.0 ± 0.0a	100.0 ± 0.0a
Methoprene											
0.5	3.0 ± 0.0a	33.3 ± 0.0a	38.9 ± 5.6ab	50.0 ± 16.7a	27.7 ± 5.6bc	0.0 ± 0.0b	—	—	—	—	—
Control	3.5 ± 0.29a	−4.8 ± 4.8b	−15.9 ± 9.7c	−11.7 ± 13.3b	−20.6 ± 10.4c	−31.0 ± 15.6b	−35.1 ± 11.5b	−40.7 ± 15.7b	−44.8 ± 12.1b	−44.8 ± 12.1b	−166.27 ± 26.9b
P	0.0727	0.0217	0.0001	0.0005	0.0001	0.0001	0.0014	0.0007	0.0004	0.0001	0.0008
df; F	5, 12; 2.71	5, 12; 4.06	5, 12; 13.56	5, 12; 10.33	5, 12; 14.20	5, 12; 34.55	4, 10; 10.32	4, 10; 12.26	4, 10; 14.43	4, 10; 19.07	4, 10; 12.04

Means within a column followed by the same letter are not significantly different ($P < 0.05$, Tukey HSD test [SAS Institute 1993]). —, Colonies returned to normal and were removed from further study.

[a] GLM and Tukey HSD analysis performed on log(x + 1) transformed data; untransformed means presented.

[b] GLM and Tukey HSD analysis performed on arcsine transformed data; untransformed means presented.

[c] Weeks after 1st treatment.

Table 2. Mean percentage of reduction ± SEM in worker number by pyriproxyfen and methoprene baits (small colony evaluation)

% concn	Worker no. at week 0[a]	% reduction in worker number at weeks indicated[b, c]							
		2	4	8	12	16	20	24	28
Pyriproxyfen									
0.05	520.0 ± 156.2	19.0 ± 8.0ab	59.6 ± 12.3a	77.1 ± 9.3ab	84.4 ± 7.5a	90.3 ± 6.0a	90.1 ± 7.7a	85.3 ± 1.7a	81.0 ± 9.7a
0.5	330.0 ± 55.7	26.1 ± 4.9a	36.5 ± 7.1ab	91.2 ± 5.6a	98.5 ± 1.1a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
1.0	466.7 ± 111.0	11.1 ± 3.4ab	24.3 ± 6.4b	59.6 ± 7.8b	87.4 ± 11.7a	99.9 ± 0.1a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
2.5	610.0 ± 135.8	0.8 ± 2.2b	43.9 ± 7.7ab	68.0 ± 7.7ab	88.2 ± 7.4a	94.4 ± 5.6a	97.8 ± 2.2a	100.0 ± 0.0a	100.0 ± 0.0a
Methoprene									
0.5	220.0 ± 5.7	−4.8 ± 7.3b	−18.4 ± 4.5c	−31.8 ± 15.9c	—	—	—	—	—
Control	370.0 ± 45.1	−0.5 ± 3.3b	−13.2 ± 4.3c	−46.5 ± 11.7c	−76.4 ± 14.4b	−404.4 ± 25.4b	−144.7 ± 35.7b	−185.9 ± 45.7b	−246.8 ± 57.9b
P	0.0959	0.0037	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
df; *F*	5, 12; 2.44	5, 12; 6.52	5, 12; 24.17	5, 12; 45.21	4, 10; 25.30	4, 10; 56.71	4, 10; 74.41	4, 10; 86.4	4, 10; 64.89

Means within a column followed by the same letter are not significantly different ($P < 0.05$, Tukey HSD test [SAS Institute 1993]). —, Colonies returned to normal and were removed from further study.

[a] GLM and Tukey HSD analysis performed on log(x + 1) transformed data; untransformed means presented.

[b] GLM and Tukey HSD analysis performed on arcsine transformed data; negative numbers replaced by zero for arcsine transformation; untransformed means presented.

[c] Weeks after 1st treatment.

Table 3. Mean percentage of reduction ± SEM in queen number by pyriproxyfen and methoprene baits (small colony evaluation)

% concn	Queen no. at week 0[a]	% reduction in queen number at weeks indicated[b, c]						
		4	8	12	16	20	24	28
Pyriproxyfen								
0.05	36.0 ± 9.5a	15.2 ± 36.0a	25.7 ± 37.2a	31.7 ± 40.1ab	37.4 ± 36.9ab	42.5 ± 29.1ab	42.5 ± 29.1ab	31.4 ± 37.2ab
0.5	48.3 ± 31.1a	11.2 ± 5.9a	61.8 ± 18.9a	98.7 ± 1.3a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
1.0	40.7 ± 11.0a	−0.4 ± 18.5a	18.6 ± 14.5a	76.9 ± 19.4ab	90.0 ± 10.0a	99.4 ± 0.6a	99.4 ± 0.6a	99.4 ± 0.6a
2.5	63.3 ± 10.1a	19.7 ± 4.3a	26.3 ± 6.6a	39.2 ± 16.8ab	63.1 ± 18.5ab	69.2 ± 15.8a	90.4 ± 9.6a	100.0 ± 0.0a
Methoprene								
0.5	21.0 ± 6.03a	−41.7 ± 38.0a	−60.7 ± 27.3a	—	—	—	—	—
Control	57.0 ± 16.1a	−15.4 ± 19.4a	−21.8 ± 25.7a	−31.3 ± 34.4b	−41.5 ± 44.4b	−74.4 ± 66.8b	−91.8 ± 77.3b	−129.8 ± 97.4b
P	0.3998	0.5464	0.0554	0.0243	0.0162	0.0024	0.0011	0.0009
df; F	5, 12; 1.12	5, 12; 0.84	5, 12; 3.00	4, 10; 4.51	4, 10; 5.16	4, 10; 8.94	4, 10; 10.94	4, 10; 11.52

Means within columns followed by the same letter are not significantly different ($P < 0.05$, Tukey HSD test [SAS Institute 1993]). —, Colonies returned to normal and were removed from further study.

[a] GLM and Tukey HSD analysis performed on $\log(x + 1)$ transformed data; untransformed means presented.

[b] GLM and Tukey HSD analysis performed on arcsine transformed data; negative numbers replaced by zero for arcsine transformation; untransformed means presented.

[c] Weeks after 1st treatment.

newly emerged workers were observed in the piles outside of the nest cell.

Repellency of the baits, indicated by a decrease in consumption with an increase in concentration, could explain the delay in brood reduction with increased concentration. However, the percentage of the bait consumed was higher, although not significantly, at the 1 and 2.5% colonies than it was for the 0.5% pyriproxyfen-treated colonies (Table 4).

Consumption of the baits was reduced the 2nd wk when compared to the 1st. For week 1, 21.2 ± 2.5% (mean ± SEM) of the baits were consumed as compared with 8.6 ± 1.8% of the baits the 2nd wk ($F = 19.99$; df=1, 34; $P < 0.0001$). Several hypotheses could explain the reduction in consumption the 2nd wk. The colonies were only starved for 24 h the 2nd wk as opposed to 72 h the 1st wk. Switching from or feeding less on a bait after long-term exposure could be the result of satiation with the bait or a need to obtain a balanced diet (Edwards and Abraham 1990). The percentage of the bait consumed in the controls also decreased the 2nd wk (Table 4) eliminating the explanation that pyriproxyfen affected feeding.

Worker number reductions were significantly greater in all the pyriproxyfen-treated colonies than in the methoprene and control colonies by 4 wk and all subsequent weeks (Table 2). The number of workers was reduced more rapidly in 0.5%-treated colonies and workers were completely eliminated by week 13. Worker numbers were reduced to zero by 18 wk in 1%-treated colonies and by 24 wk in the 2.5%-treated colonies. This trend was similar to that found with the brood rating.

The number of queens in the pyriproxyfen-treated colonies gradually decreased (Table 3). At 8 wk, 60% of the queens had died in the 0.5% treatment and a 20% increase in queens had occurred in the control colonies, but these treatments were not significantly different. At 9 wk, 0.5% treated colonies contained significantly less queens than the control colonies ($F = 4.74$; df = 5, 12; $P = 0.0127$). The 1% pyriproxyfen-treated colonies achieved 99% or greater reduction in queen number by week 18. The 0.5% pyriproxyfen-treated colonies reached 100% queen reduction by week 13. The mortality of the queens resulted from natural mortality, a decrease in the number of workers tending them, or possible toxic effects.

Large Colony Evaluations. Pyriproxyfen was also extremely effective in reducing colony size when given to large colonies. All pyriproxyfen treatments significantly reduced brood by week 3; an effect which continued throughout the study (Table 5). Brood was reduced by >89% by week 12 and reduction eventually increased to 100% for all pyriproxyfen treatments.

Methoprene was more effective against large colonies than small colonies, but still not as effective as pyriproxyfen. Unlike the small colony evaluations, the large colony results indicated that

Table 4. Mean percentage of consumption ± SEM of methoprene and pyriproxyfen baits by Pharaoh ants

% concn	% consumption of baits[a]			
	Small colony evaluation		Large colony evaluation	
	Week 1	Week 2	Week 1	Week 2
Pyriproxyfen				
0.05	30.3 ± 7.4a	9.3 ± 5.4a	—	—
0.25	—	—	74.7 ± 12.9a	20.7 ± 8.8a
0.5	15.7 ± 3.7a	3.7 ± 0.7a	70.5 ± 6.8a	29.0 ± 14.7a
1.0	21.3 ± 4.7a	12.3 ± 8.8a	88.8 ± 5.4a	17.7 ± 12.8a
2.5	19.7 ± 5.8a	8.0 ± 4.0a	—	—
Methoprene				
0.5	9.7 ± 2.0a	5.2 ± 0.2a	86.7 ± 10.9a	16.7 ± 2.6a
Control	30.7 ± 3.7a	13.3 ± 1.7a	87.3 ± 12.7a	72.5 ± 14.9a
P	0.0548	0.5777	0.6392	0.0486
df; *F*	5, 12; 3.01	5, 12; 0.79	4, 10; 0.65	4, 10; 3.52

Means within columns followed by the same letter are not significantly different ($P < 0.05$, Tukey LSD test [SAS Institute 1993]).
[a] GLM and Tukey HSD analysis performed on arcsine transformed data; untransformed means presented.

these colonies receiving methoprene experienced a continued, reduced level of brood (Table 5). Worker levels initially increased, followed by a dramatic decrease at week 12 (57%), but reduction never reached 100% (Table 6). Brood was reduced 4 wk after treatment in the methoprene-treated colonies, although the brood rating was not significantly lower than the controls until week 6 (Table 5). Also, at 6 wk, brood reduction in the methoprene-treated colonies was only ≈½ of the brood reduction in the pyriproxyfen-treated colonies. Subsequent readings revealed a reduced brood rating of ≈50% in the methoprene-treated colonies to the end of the study. Methoprene-treated colonies responded variably. One colony was almost completely eliminated, 1 decreased in all variables but recovered, and 1 colony was not affected. This may indicate variable distribution of the methoprene in the peanut butter:honey.

Effects of pyriproxyfen on the brood were more obvious in the large colony evaluation. By week 3, most of the pupae were dead in the pyriproxyfen-treated colonies and were apparent in large piles in the tray. Pupae appeared normal and darkened, but did not eclose into adults. Pupal death was apparently caused by pyriproxyfen ingested during the larval stage and not the result of a toxic contact effect on the original pupae. Although eggs and some early larval instars were present after the pupal deaths occurred, only eggs were prevalent throughout the study until they shrivelled and died. We did not determine if the queens continued to oviposit, but eggs were present throughout the study until the colony completely died.

Worker number was not reduced as quickly as the brood by the 4-wk reading. In the pyriproxyfen treatments, worker numbers were reduced by at least 20% by 4 wk (Table 6). However, >88% and 99% of the workers were eliminated in these treatments by week 8 and 12, respectively. One hundred percent of reduction in the worker number occurred at week 16 in the 0.25 and 1%-treated and at week 26 for the 0.5%-treated colonies. Methoprene did not reduce the worker number until week 10 (4.4 ± 11.3%); at week 12, worker number was reduced to 50% or greater and did not significantly decrease as compared to the controls until week 14. Methoprene did not reduce worker number as quickly as pyriproxyfen.

The IGR had a toxic effect on the workers. At week 6, when worker death was the greatest, the number of dead workers was greater in the IGR treatments than the controls (Table 7). The number of live workers in the control colonies was much larger than the those in the pyriproxyfen-treated colonies. Hence, if natural mortality accounted for worker death, than a higher number of dead workers should be found in the controls.

The number of queens was gradually reduced in all treatments (Table 8); however, the reduction in the number of queens in the pyriproxyfen-treated colonies was significantly higher than the methoprene-treated colonies. By 12 wk, queen reduction was >86% in the pyriproxyfen-treated colonies and only 32.6% in the methoprene-treated colonies.

Consumption of the baits was not significantly different for any of the treatments in the 1st wk after treatment (Table 4). As observed in the small colony evaluation, there was a significant decline in the percent consumption of baits during the 2nd wk of baiting. For week 1, 81.6 ± 4.3% (mean ± SEM) of all baits combined was consumed as compared with 31.3 ± 7.2% of the baits the 2nd wk ($F = 27.39$; df= 1, 28; $P < 0.0001$). The overall ratio of percentage of bait consumed during week 2 to the percentage of bait consumed during week 1 was nearly the same in the small (0.41) and large colony evaluations (0.38). Consumption of the control bait remained high in the 2nd treatment for the large colony evaluations, possibly indicating that pyriproxyfen did have more of an effect on the foraging behavior of Pharaoh ants when fed to large colonies than small colonies.

Table 5. Mean percentage of reduction ± SEM in brood levels by pyriproxyfen and methoprene baits (large colony evaluation)

% concn	Brood rating at week 1[a]	% reduction in brood rating at week indicated[b, c]								
		3	4	6	8	12	16	20	24	28
Pyriproxyfen										
0.25	8.7 ± 0.3a	44.0 ± 3.6a	50.0 ± 0.0a	55.6 ± 3.2a	69.0 ± 4.6a	94.2 ± 0.2a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
0.5	8.3 ± 0.3a	44.0 ± 0.2a	43.8 ± 6.3a	66.0 ± 1.8a	78.0 ± 1.8a	89.8 ± 4.3a	93.8 ± 6.3a	97.9 ± 2.1a	97.9 ± 2.1a	100.0 ± 0.0a
1.0	8.8 ± 0.2a	41.5 ± 1.6a	52.8 ± 1.6a	68.0 ± 6.5	75.7 ± 10.2a	92.5 ± 1.8a	98.1 ± 1.9a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
Methoprene										
0.5	9.3 ± 0.3a	7.0 ± 1.5b	10.9 ± 3.4b	27.4 ± 8.9b	34.6 ± 9.8b	33.0 ± 11.9b	50.0 ± 25.1ab	48.1 ± 24.9ab	48.1 ± 27.3ab	50.0 ± 28.9ab
Control	9.2 ± 0.6a	0.0 ± 0.0b	−10.1 ± 7.6c	−10.1 ± 7.6c	−10.1 ± 7.6c	−10.1 ± 7.6c	−10.1 ± 7.6b	−10.1 ± 7.6b	−10.1 ± 7.6b	−10.1 ± 7.6b
P	0.4171	0.0001	0.0001	0.0001	0.0001	0.0001	0.0004	0.0002	0.0005	0.0008
df; *F*	4, 10; 1.08	4, 10; 133.58	4, 10; 55.73	4, 10; 31.53	4, 10; 25.3	4, 10; 56.69	4, 10; 14.07	4, 10; 15.91	4, 10; 8.85	4, 10; 9.82

Means within columns followed by the same letter are not significantly different ($P < 0.05$, Tukey HSD test [SAS Institute 1993]).
[a] GLM and Tukey HSD analysis performed on $\log(x + 1)$ transformed data; untransformed means presented.
[b] GLM and Tukey HSD analysis performed on arcsine transformed data; negative numbers replaced by zero for arcsine transformation; untransformed means presented.
[c] Weeks after 1st treatment.

Table 6. Mean percentage of reduction ± SEM in worker number by pyriproxyfen and methoprene baits (large colony evaluation)

% concn	Worker no. at week 1[a]	% reduction in worker number at weeks indicated[b, c]						
		4	8	12	16	20	24	28
Pyriproxyfen								
0.25	9,366.7 ± 1,133.3	26.6 ± 8.5a	88.9 ± 7.2a	99.8 ± 0.1a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
0.5	10,100.0 ± 305.5	20.0 ± 3.6ab	97.2 ± 1.5a	99.8 ± 0.1a	99.9 ± 0.1a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
1.0	10,433.3 ± 66.7	21.7 ± 3.5a	96.7 ± 1.7a	99.9 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
Methoprene								
0.5	10,583.3 ± 220.5	−11.7 ± 1.3b	−13.5 ± 7.9b	57.0 ± 28.6ab	69.9 ± 28.1a	70.3 ± 28.4a	65.4 ± 32.7ab	61.8 ± 31.2ab
Control	7,866.7 ± 895.0	−16.0 ± 4.3b	−55.6 ± 13.0b	−60.0 ± 14.3b	− 49.2 ± 24.2b	−67.0 ± 27.3b	−62.9 ± 17.4b	−65.4 ± 20.2b
P	0.1108	0.0032	0.0001	0.0001	0.0004	0.0009	0.0025	0.0017
df; *F*	4, 10; 2.49	4, 10; 8.31	4, 10; 234.36	4, 10; 11.78	4, 10; 14.07	4, 10; 11.69	4, 10; 8.85	4, 10; 9.82

Means within columns followed by the same letter are not significantly different ($P < 0.05$, Tukey HSD test [SAS Institute 1993]).
[a] GLM and Tukey HSD analysis performed on $\log(x + 1)$ transformed data; untransformed means presented.
[b] GLM and Tukey HSD analysis performed on arcsine transformed data; negative numbers replaced by zero for arcsine transformation; untransformed means presented.
[c] Weeks after 1st treatment.

Table 7. Comparison of the number of dead workers ± SEM at 6 wk after 1st treatment in Pharaoh ant colonies fed pyriproxyfen and methoprene baits (large colony evaluation)

% concn	No. dead workers[a]
Pyriproxyfen	
0.25	1,016.7 ± 316.7a
0.5	1,516.7 ± 109.3a
1.0	2,166.7 ± 417.7a
Methoprene	
0.5	720.0 ± 316.6a
Control	128.3 ± 34.2b
P	0.0002
df; *F*	4, 10; 16.07

[a] GLM and Tukey HSD analysis performed on $\log(x + 1)$ transformed data; untransformed means presented; means within columns followed by the same letter are not significantly different ($P < 0.05$, Tukey HSD test [SAS Institute 1993]).

The overall effects of pyriproxyfen on Pharaoh ants in laboratory colonies were a decline in the quantity of brood, in the worker force and gradual death in the queens. Brood reduction was caused by pupal death at week 3, a gradual death of the other immature stages and a decrease in oviposition. Reduction in worker number was due to attrition, older workers were dying and not being replaced, and to a possible toxic effect. Queens which initially continued to produce a small amount of eggs, eventually died. Queen death may have been the result of a lack of workers to tend the queens or a toxic effect.

It appears that worker numbers in pyriproxyfen-treated colonies are reduced faster than in colonies treated with other insect growth regulators, such as methoprene and fenoxycarb. For those concentrations of pyriproxyfen that were most effective in the large colony evaluations, 0.25, 0.5, and 1%, worker numbers were reduced by 99% or greater by week 12. Our results with methoprene were not as encouraging, even when there was an effect. Our most promising results included 1 colony in which worker numbers were reduced by >99%; however, this occurred 16 wk after treatment. Large Pharaoh ant colonies given 2 baitings of 0.25% fenoxycarb in peanut butter oil experienced >99% reduction in worker number at ≈15 wk (Williams and Vail 1993).

Pyriproxyfen given to Pharaoh ant colonies in baits also reduced brood faster than fenoxycarb or methoprene. Twenty weeks were necessary for complete reduction in brood for large colonies treated with 0.25% fenoxycarb (Williams and Vail 1993) as compared with 16 wk for large colonies treated with 0.25% pyriproxyfen. Subsequent research to this study indicated that pyriproxyfen also causes the development of intermediate castes, a characteristic of colonies treated with fenoxycarb. Intermediate castes are defined as individuals resembling queens but smaller, often possessing ocelli and vestigial wing buds. Therefore, our re-

Table 8. Mean percentage of reduction ± SEM in queen number by pyriproxyfen and methoprene baits (large colony evaluation)

% concn	Queen no. at week 1[a]	% reduction in queen no. at week indicated[b, c]						
		4	8	12	16	20	24	28	32
Pyriproxyfen									
0.25	201.7 ± 28.9a	13.0 ± 6.0a	41.6 ± 10.7ab	89.3 ± 3.1a	94.0 ± 3.6a	98.8 ± 1.3a	99.3 ± 0.7a	99.6 ± 0.4a	100.0 ± 0.0a
0.5	191.7 ± 23.5a	15.5 ± 8.9a	68.7 ± 2.3a	86.3 ± 6.0a	91.6 ± 6.3a	96.1 ± 3.2a	98.8 ± 0.8a	100.0 ± 0.0a	100.0 ± 0.0a
1.0	298.3 ± 39.4a	9.1 ± 7.0a	54.2 ± 16.2a	87.9 ± 2.4a	96.4 ± 1.3a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a	100.0 ± 0.0a
Methoprene									
0.5	250.0 ± 22.5a	1.2 ± 10.8a	29.5 ± 4.7ab	32.6 ± 3.2b	42.7 ± 28.1b	46.5 ± 11.1b	53.6 ± 17.6b	52.8 ± 22.0ab	57.0 ± 20.6ab
Control	251.7 ± 75.1a	2.0 ± 5.8a	0.5 ± 6.0b	2.7 ± 10.3c	−2.1 ± 20.6b	−3.3 ± 34.6b	−25.2 ± 50.6b	−20.1 ± 48.3b	−15.9 ± 42.2b
P	0.5226	0.7883	0.0052	0.0001	0.0001	0.0001	0.0003	0.0008	0.0005
df; *F*	4, 10; 0.85	4, 10; 0.42	4, 10; 7.28	4, 10; 80.08	4, 10; 26.77	4, 10; 22.96	4, 10; 15.64	4, 10; 11.95	4, 10; 13.55

Means within the same column followed by the same letter are not significantly different ($P < 0.05$, Tukey HSD test [SAS Institute 1993]).

[a] GLM and Tukey HSD analysis performed on $\log(x + 1)$ transformed data; untransformed means presented.

[b] GLM and Tukey HSD analysis performed on arcsine transformed data; negative numbers replaced by zero for arcsine transformation; untransformed means presented.

[c] Weeks after 1st treatment.

sults indicated that pyriproxyfen in baits is a more potent IGR than fenoxycarb or methoprene for Pharaoh ant control.

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