Development of *Pheidole megacephala* (Hymenoptera: Formicidae) Colonies Following Ingestion of Fenoxycarb and Pyriproxyfen

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ABSTRACT Laboratory-reared *Pheidole megacephala* (F.) colonies were fed 2% pyriproxyfen (Sumitomo S-31183) or 2% fenoxycarb, ethyl [2-(4-phenoxyphenoxy)ethyl] carbamate, in soybean oil. Queens in colonies treated with pyriproxyfen laid significantly fewer eggs than controls 2 wk after treatment. Oviposition ceased in all pyriproxyfen-treated queens at 6 wk after treatment, with no recovery during the 15 wk of the study. Brood volumes in pyriproxyfen-treated colonies declined after 2 wk because of pupal death. No brood was found after 4 wk. Fenoxycarb-treated queens laid significantly fewer eggs than controls after 2 wk. Fenoxycarb-treated colonies shifted caste differentiation from workers to males at 3 wk after treatment. At 5 wk, 90% of larvae were male; the remaining 10% developed into intercastes.

KEY WORDS Insecta, *Pheidole megacephala*, fenoxycarb, pyriproxyfen

THE BIGHEADED ANT, *Pheidole megacephala* (F.), is a tramp species which occurs throughout the world tropics (Brown 1973). In Hawaii, it causes problems in agricultural, urban, and native environments (Reimer et al. 1990). *P. megacephala* is especially serious in pineapple in Hawaii (Rohrbach et al. 1988) because it tends the mealybugs *Dysmicoccus neobrevipes* Beardsley and *D. brevipes* (Cockerell) for honeydew. These mealybugs are associated with a wilt disease (Carter 1933) which has completely destroyed pineapple crops in Hawaii (Rohrbach et al. 1988). The disease has been successfully controlled in Hawaii since the 1940s through the elimination of ants from fields with chlorinated hydrocarbons. These insecticides are no longer available because of restrictions by the Environmental Protection Agency; therefore, alternative insecticides and control methods are being researched.

Corncob grit baits coated with a soybean oil attractant and a formicide were originally developed for use against the red imported fire ant, *Solenopsis invicta* Buren (Lofgren et al. 1963). Similar baits incorporating mirex or hydramethylnon as the active ingredient have successfully controlled bigheaded ant in pineapple (Su et al. 1980, Reimer & Beardsley 1990).

New toxicants are being sought for use in insecticidal baits. To be effective, these chemicals must satisfy the requirements outlined by Lofgren (1986). Some insect growth regulators (IGR) meet these requirements and are well suited for incorporation into insecticidal baits (Banks et al. 1983). Studies have shown that two materials, fenoxycarb, ethyl [2-(4-phenoxyphenoxy)ethyl] carbamate (Maag Agrochemicals, Vero Beach, Fla.) and pyriproxyfen (S-31183 of Sumitomo America, New York), produce insect growth regulator (IGR) activity when fed to *S. invicta* (Banks et al. 1983, 1988; Glancey et al. 1990). This paper reports on the effects of feeding these compounds to laboratory colonies of *P. megacephala*.

Materials and Methods

Thirty-six bigheaded ant colonies were collected from pineapple stumps in a ratoon field at Kunia, Oahu. They were brought back to the laboratory and extracted by beating the stumps into Berlese funnels. The ants and all debris were then evenly spread out in plastic boxes (26.5 by 33 by 9 cm). Plastic vials (55 ml) containing pieces of rolled-up black cloth were placed in each box to extract the ants. The workers moved all brood and queens into these vials. The colonies were then easily removed and transferred to transparent plastic shoe boxes (15 by 29 by 9 cm). Each shoe box contained food (modified Banks diet) (Banks et al. 1981), water, and a nest chamber (modified Bishop cell) (Bishop et al. 1980). Each colony was reduced to comparable numbers of workers, brood, and soldiers and allowed to stabilize for 2 wk at 25°C and a photophase of ≈10 h. At the end of 2 wk, each colony consisted of 1 queen, 25–50 eggs, 1–2 ml brood, 200–400 workers, and 20–40 soldiers.

At this time, each colony was starved for 4 d but still given access to water. Each of 12 colonies was then fed 0.5 ml of 2% fenoxycarb in soybean oil, 12 colonies were fed 0.5 ml of 2% pyriproxyfen,

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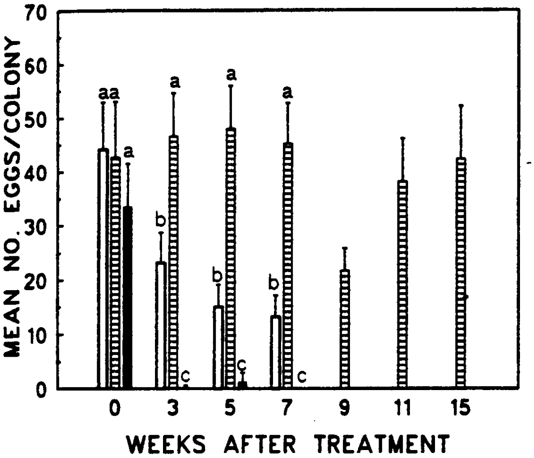

Fig. 1. Mean (±SE) number of eggs in laboratory-reared bigheaded ant colonies. Treatments: □, fenoxycarb; ▤, control; ■, pyriproxyfen. Significant differences among treatments are indicated by letters atop the bars for 0, 3, 5, and 7 wk after treatment ($P < 0.05$, Duncan's [1951] multiple range test). Differences between control and pyriproxyfen treatments were significant at 9, 11, and 15 wk after treatment ($P < 0.01$, ANOVA).

and 12 colonies were fed pure soybean oil as controls. All colonies were returned to their regular diet after 24 h, and the soybean oil was removed. The number of eggs, volume of brood, and number of workers and soldiers were counted for each colony at weekly intervals. The queens from three replications of treated and control colonies were removed at 5, 6, and 7 wk each for histological examination (Glancey et al. 1990) after egg, brood, worker, and soldier counts were made. Additionally, three queens were removed from fenoxycarb-treated colonies at 8 wk because the full effects of this material were not apparent in the earlier samples. The effects on the remaining three pyriproxyfen-treated colonies and controls were followed until 15 wk after treatment. Therefore, the pyriproxyfen and control colonies had 12 replications for weeks 1–5, 9 for week 6, 6 for week 7, and 3 for the remaining weeks. Studies with fenoxycarb-treated colonies were terminated after 8 wk because of the lack of colonies with queens.

Differences among treatments were analyzed using analysis of variance (ANOVA) (Sokal & Rohlf 1981), and means were compared using Duncan's multiple range test (Duncan 1951).

Results and Discussion

Eggs. There were significant differences among treatments at 3 through 15 wk after treatment but not at week 0 (Table 1). Pyriproxyfen-treated queens showed a rapid decrease in oviposition to levels significantly below controls within 3 wk after treatment (Fig. 1). One-third of the queens produced a few eggs at this time, but the remaining 66% of the queens had stopped ovipositing and never recovered. No eggs were laid in any pyriproxyfen-treated colonies after week 6. Some of these queens lived for 5 to 6 mo but did not recover from the single application of pyriproxyfen to the colony.

The decrease in the oviposition rate of fenoxycarb-treated queens was more gradual than in queens treated with pyriproxyfen. Fenoxycarb-treated queens laid significantly fewer eggs than control queens at weeks 3, 5, and 7 but laid significantly more eggs than pyriproxyfen queens through week 7 (Fig. 1).

The decreased egg production in pyriproxyfen- and fenoxycarb-treated bigheaded ant colonies can be explained by the effects of these materials on egg development in the queen. Histological studies determined that both of these IGRs caused egg

Table 1. Results of an ANOVA of a completely randomized design testing the effects of fenoxycarb, pyriproxyfen, and control treatments on laboratory colonies of *P. megacephala*

	Wk after treatment						
	0	3	5	7	9	11	15
Eggs							
F	1.56	186.36	224.1	113.2	182.5	333.23	333.23
df	2, 33	2, 33	2, 33	2, 15	1, 4	1, 4	1, 4
P	0.3	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Brood							
F	1.23	155.1	161.7	232.17	100.0	961.0	961.0
df	2, 33	2, 33	2, 33	2, 15	1, 4	1, 4	1, 4
P	0.3	<0.01	<0.01	<0.01	<0.01	<0.01	<0.01
Workers							
F	1.17	0.25	2.06	1.75	51.57	88.2	204.24
df	2, 33	2, 33	2, 33	2, 15	1, 4	1, 4	1, 4
P	0.32	0.78	0.14	0.21	<0.01	<0.01	<0.01
Soldiers							
F	0.85	0.24	0.18	0.19	1.43	0.45	2.8
df	2, 33	2, 33	2, 33	2, 15	1, 4	1, 4	1, 4
P	0.43	0.79	0.83	0.83	0.29	0.54	0.17

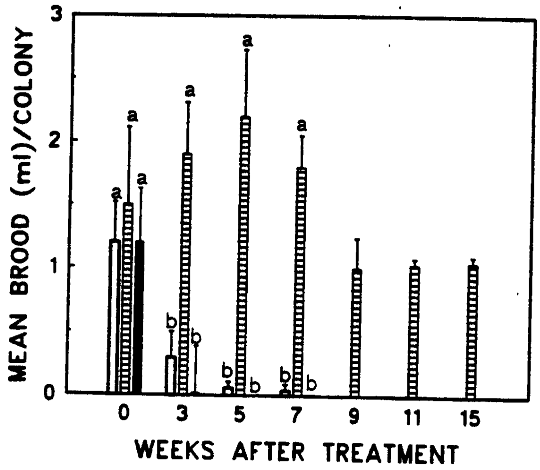

Fig. 2. Mean (±SE) volume (ml) of brood in laboratory-reared bigheaded ant colonies. Treatments: □, fenoxycarb; ⊟, control; ■, pyriproxyfen. Significant differences among treatments are indicated by letters atop the bars for 0, 3, 5, and 7 wk after treatment ($P < 0.05$, Duncan's multiple range test). Differences between control and pyriproxyfen treatments were significant at 9, 11, and 15 wk after treatment ($P < 0.01$, ANOVA).

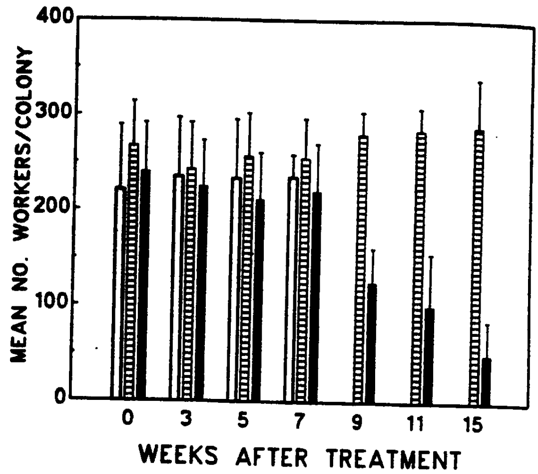

Fig. 3. Mean (±SE) number of minor workers in laboratory-reared bigheaded ant colonies. Treatments: □, fenoxycarb; ⊟, control; ■, pyriproxyfen. Treatments were significantly different at 9, 11, and 15 wk after treatment but not at 0, 3, 5, or 7 wk after treatment ($P < 0.01$, ANOVA).

resorption with decreased egg production and development of smaller eggs by *P. megacephala* queens at 6 wk after treatment (Glancey et al. 1990). The mode and site of action of these materials, however, was not determined and is still not known.

Brood. The volume of brood in the pyriproxyfen- and fenoxycarb-treated colonies declined to levels significantly (Table 1) below control levels at 3 wk after treatment and remained below controls throughout the study (Fig. 2). No brood were found in any pyriproxyfen-treated colonies after 4 wk. The rapid decline in pyriproxyfen brood volumes was due to the death of all pupae and the lack of replacement of larvae that had pupated. A few larval queens were observed after 10 d; however, none of these survived to pupation.

The decline in brood volume in fenoxycarb-treated colonies was due to the high mortality in worker larvae. These colonies produced male larvae at 2 wk after treatment; production of worker larvae decreased concurrently. At 5 wk after treatment, 90% of the larvae were males; the remaining larvae developed into intercastes between soldiers and males. No male, queen, or intercaste larvae were produced in control colonies; only worker and soldier larvae were produced. Similar results were found in *P. megacephala* colonies in pineapple fields treated with fenoxycarb (Reimer & Beardsley 1990). Colonies in treated plots had shifted brood production from worker to male pupae by 11 wk after treatment. Reimer & Beardsley (1990) found that 96.1% of the pupae in fenoxycarb-treated plots were male and 3.9% were workers compared with 100% worker pupae in control plots. The apparent differences in the time it took to produce males in treated colonies (3 wk in the laboratory versus 11 wk in field studies) can be explained by the fact that no samples were taken between weeks 3 and 11 in the field studies of Reimer & Beardsley (1990). Therefore, if the field colonies had produced males between weeks 3 and 11, they would not have been observed until week 11. Banks et al. (1988) found a similar response in *S. invicta*. Colonies treated with fenoxycarb shifted brood production from workers to reproductives between 4 and 8 wk after treatment.

Workers. There were no significant differences among treatments in worker numbers during the first 7 wk (Table 1). Worker numbers in colonies treated with pyriproxyfen declined to levels significantly (Table 1) below controls during weeks 9 through 15 (Fig. 3). This decline was caused by the death of worker larvae and pupae. The result was a gradual decrease in worker numbers due to the lack of replacement of older workers that had died from natural causes. A similar trend would also have been observed in fenoxycarb-treated colonies because no worker larvae were produced by these colonies after 5 wk. However, these colonies were moribund at 8 wk because of the death of all queens. Each colony was excluded from the experiment after its queen had died.

Soldiers. Mean numbers of soldiers per colony were not significantly different among treatments during any of the weekly counts (Table 1, Fig. 4). Few soldiers died during this study. The soldier caste is longer-lived than the workers (N.J.R., unpublished data); therefore, their numbers were not significantly affected by the lack of replacement during the relatively short duration of this study. The number of soldiers began to decline in the pyriproxyfen-treated colonies after 10–11 wk (Fig.

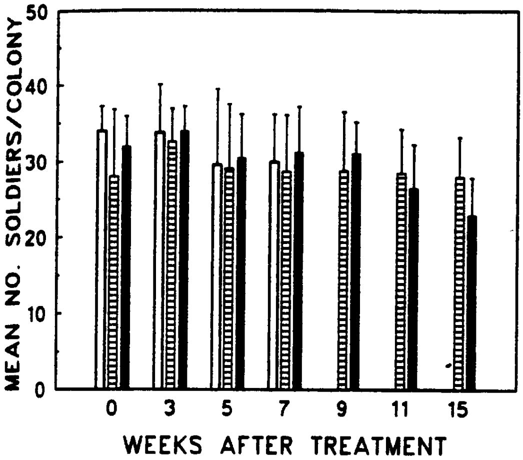

Fig. 4. Mean (±SE) number of major workers (soldiers) in laboratory-reared bigheaded ant colonies. Treatments: □, fenoxycarb; ⊟, control; ■, pyriproxyfen. Treatments were not significantly different at each week after treatment ($P < 0.01$, ANOVA).

4) but not to levels significantly lower than controls (Table 1).

Solenopsis invicta and *S. richteri* Forel fed fenoxycarb or other IGRs shifted caste differentiation from worker to sexual castes, reduced or stopped egg production, and suffered high mortality during the larval stadia (Banks et al. 1983, Troisi & Riddiford 1974, Vinson & Robeau 1974). Similar responses were observed in *P. megacephala* colonies fed either fenoxycarb or pyriproxyfen, which demonstrated that these materials also acted as IGRs on the bigheaded ant. Unlike *Solenopsis* colonies, *P. megacephala* colonies treated with fenoxycarb in this study and in field studies (Reimer & Beardsley 1990) never produced alate females. Instead, *P. megacephala* queens laid unfertilized (male) eggs after ingesting fenoxycarb. In field studies (Reimer & Beardsley 1990), however, *P. megacephala* queens recovered from the fenoxycarb treatment and stopped laying male eggs at 13 wk after treatment. Only workers and soldiers were produced by the colonies at that time. A similar recovery was not observed in these laboratory studies because of the premature death of all queens after 8 wk. Recovery of *S. invicta* queens has been observed as early as 2–3 wk after treatment with fenoxycarb (Glancey & Banks 1988) but, unlike the results with *P. megacephala*, those queens were isolated from the treated workers, of which the majors act as repletes in *S. invicta* colonies (Glancey et al. 1973).

Both fenoxycarb and pyriproxyfen showed potential for use in insecticidal baits. They caused a decrease or cessation of egg production, high mortality to immatures, and a shift in caste differentiation from workers to males within 2–6 wk after treatment. The effect of these materials on worker and soldier forager populations was much slower. Effects were not observed until 9–12 wk after treatment. This time lag needs to be considered when trying to control *P. megacephala* with these IGRs in small plots surrounded by thriving ant colonies. Healthy colonies will move into treated areas and displace the treated colonies within the 12-wk period after treatment.

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