Attractiveness of Gel, Granular, Paste, and Solid Formulations of Ant Bait Insecticides to the Little Fire Ant, *Wasmannia auropunctata* (Roger) (Hymenoptera: Formicidae)

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Abstract. The little fire ant, *Wasmannia auropunctata* (Roger) (Hymenoptera: Formicidae), was first detected in plant nurseries in the Puna district of Hawaii island in 1999. *W. auropunctata* has since spread throughout Hawaii island, and is reported in homes, landscapes, plant nurseries and orchards, and forested areas. This study evaluated: 1) the attractiveness of several granular, liquid, gel, and paste insecticidal ant baits for homeowner and commercial use as compared with the standard granular baits containing hydramethylnon known to be attractive to and effective against *W. auropunctata*, and 2) the effects of weathering on granular bait attractiveness. Field attractiveness choice tests were conducted in an infested 37.2-m^2 plot, and worker ant foraging and recruitment were recorded at 15-min intervals for 2 h. Granular and paste products that were as attractive as standard granular baits (Amdro Fire Ant Bait, Probait) included others formulated with hydramethylnon, abamectin, hydramethylnon and S-methoprene, indoxacarb, fipronil, and metaflumizone. None of the gel or liquid ant bait products evaluated (active ingredients hydramethylnon, sodium tetraborate pentahydrate, thiamethoxam, fipronil or indoxacarb) were attractive to foraging workers. Attraction of these baits could possibly be improved with inclusion of preferred food sources, such as peanut butter or animal-based protein. Attractiveness of granular ant baits exposed to 7 and 14 days of weathering fell by 40 to 96% as compared to fresh deposits. Corn grit baits should be formulated to preserve attractiveness in tropical environments with high rainfall.

Key words: *Wasmannia auropunctata*, ant control, hydramethylnon, abamectin, S-methoprene, weathered bait

Introduction

The little fire ant, *Wasmannia auropunctata* (Roger) (Hymenoptera: Formicidae), native to South and Central America, is considered to be one of the most destructive global invasive ant species, and has been introduced to tropical and subtropical localities including the Caribbean and Pacific islands, West Africa and Australia (Lowe et al. 2000, Holway et al. 2002, Wetterer and Porter 2003). Little fire ant was first reported in Hawaii in 1999 at a plant nursery (Conant and Hirayama 2000) and has since spread throughout Hawaii Island. High populations of *W. auropunctata* have been correlated to reductions of other ant species, especially where they are not native and

have been introduced (Clark et al. 1982, Lubin 1984, Ulloa-Chacón and Cherix 1994). Field studies suggest that *W. auropunctata* use interference competition, resource competition, and predation to eliminate other ant species (Clark et al. 1982, Meier 1994, Achury et al. 2008). The little fire ant delivers a painful sting (Wetterer and Porter 2003), and agricultural workers suffer hazardous conditions when *W. auropunctata* "rain" onto them from arboreal colonies in trees where ants are tending honeydew-producing insects (aphids, mealybugs, soft scales). Arboreal colonies can exist without any foraging on the ground. (Spencer 1941, Clark et al. 1982, Ulloa-Chacón and Cherix 1990,Williams and Whelan 1992, de Souza et al. 1998, Wetterer and Porter 2003, Le Breton et al. 2004). Little fire ant stings to the eye have been linked to keratopathy in pets and other animals (Theron et al. 2007).

Typically, insecticidal ant bait consists of a slow-acting toxicant, formulated with an attractive food source and carrier for dispersal, that worker ants effectively transfer to the colony's queen or queens to reduce or eliminate their capacity to produce new offspring (Williams et al. 2001). One of the first highly effective granular ant bait insecticide was mirex bait for control of the red imported fire ant *Solenopsis invicta* Buren (Hymenoptera:Formicidae) in southeastern United States. Mirex ant bait consisted of 0.3% mirex in 14.7% soybean oil mixed with 85% corncob grits (Kaiser 1978). After mirex was banned in 1978, it was replaced with hydramethylnon in 1980, in a soybean oil–defatted corn grit carrier, to control *S. invicta* (Kaiser 1978, Williams et al. 2001, Causton et al. 2005); subsequently, ant baits were developed with other effective active ingredients, such as abamectin, borates, fipronil, methoprene, and spinosad (Williams et al. 2001). Broadcast applications of granular ant bait insecticides developed for *S. invicta* effectively reduce ground colonies of *W. auropunctata*, however, arboreal colonies may not be effectively treated with granular bait insecticides, and may require applications with aerial equipment or as a gel or paste ant bait formulation that is delivered into the tree from the ground (Souza et al. 2008, Vanderwoude and Nadeau 2009, Vanderwoude et al. 2010). Recently, a gel formulation of methoprene (Tango), consisting of corn oil, xanthan gum and peanut butter, was developed by Vanderwoude and Nadeau (2009) that can be applied in ant-infested trees and shrubs with a foliar sprayer or spray bottle (Vanderwoude et al. 2010, Hawaii Ant Lab 2012)

W. auropunctata has spread throughout east Hawaii island via movement of infested plant and construction material (Krushelnycky et al. 2005). Effective control products for large-scale application in plant nurseries and for homeowners are in high demand in these infested areas. There are, however, no attractiveness or efficacy data available for most commercially-available granular, gel, and paste bait insecticides except for Amdro (hydramethylnon), Extinguish Plus (hydramethylnon and S-methoprene), Esteem (pyriproxfen) and Siesta (metaflumizone) (Hara et al. 2011, Souza et al. 2008, Williams and Whelan 1992).

Previous research has demonstrated that several bait insecticides developed for *S. invicta* are also effective for *W. auropunctata* colony elimination (Williams and Whelan 1992, Hara et al. 2011). If a bait insecticide is accepted by foraging workers, then by trophallaxis, the toxicant should be transferred throughout the nest and ultimately cause the demise of the workers, brood and queen(s). This study evaluated the attractiveness of several commercially-available granular, liquid, gel, paste and solid ant bait insecticides to *W. auropunctata* workers, and assessed the effects of weathering on the attractive-

ness of several granular ant baits exposed to sunlight, high humidity, and rainfall conditions that stimulated mold growth on the baits.

Materials and Methods

Field choice bait tests. Attractiveness trials were conducted at the University of Hawaii at Hilo, College of Agriculture, Forestry and Natural Resource Management (CAFNRM) Instructional Farm near Hilo, Hawaii, within a 37.2-m^2 area that was infested with *W. auropunctata*. Ant bait products were obtained from several retailers (grocery, hardware, garden stores) and evaluated in four trials.

In trials 1 and 2, indoor ant baits were compared with two soybean oil-infused corn grit granular baits known to be attractive to and effective against *W. auropunctata*, Probait and Amdro Fire Ant Bait, respectively: 1) Amdro Kills Ants Stake (1.0% hydramethylnon), Combat Source Kill Ant (0.01% fipronil), Hot Shot MaxAttrax Ant $Bait_2$ (0.05% indoxacarb), Probait (0.73% hydramethylnon), Raid Ant Baits III (0.01% avermectin B1), and Raid Double Control Ant Baits II (0.05% avermectin B1), and 2) Amdro Fire Ant Bait (0.73% hydramethylnon), Grant's Kills Ants Ant Control (1.0% hydramethylnon), PIC Liquid Ant Bait Killer Killing System (5.0% sodium tetraborate pentahydrate), Raid Ant Gel Precision Placement Bait (0.003% thiamethoxam), and TomCat Ant Killer Gel Bait (0.03% indoxacarb).

In Trials 3 and 4, outdoor granular ant baits were compared with Amdro Fire Ant Bait and Probait, respectively: 3) Advance 375 (0.11% abamectin), Amdro Ant Block Home Perimeter Ant Bait (0.88% hydramethylnon), Amdro FireStrike Yard Treatment (0.0360% hydramethylnon and 0.0172% S-methoprene), Amdro Fire Ant Bait, and Green Light Fire Ant Control with Conserve (0.015% spinosad), Extinguish Plus (0.365% hydramethylnon and 0.25% S-methoprene), Maxforce FC Fire Ant Bait (0.00045% fipronil), and Siesta (0.063% metaflumizone); and 4) Advion Fire Ant Bait (0.045% indoxacarb), Extinguish Plus, Extinguish Professional Fire Ant Bait (0.5% S-methoprene), Maxforce Complete (1.0% hydramethylnon), and Probait.

Potential trial plots within the 37.2-m^2 area were visually surveyed with a nontoxic peanut butter lure for presence of foraging worker ants just before conducting each trial. Separate plots for each trial were used to avoid any carryover effects on foraging worker ants. Ant bait products in prepackaged bait stations, stakes or syringes were extricated, and approximately 1.1 g of each paste and solid bait or 1.2 ml of each liquid and gel bait were weighed and placed onto a semi-transparent plastic lid (45 mm diameter, L100 PC Lids, Fabri-Kal, Kalamazoo, MI). The contours on the inner surface of the lid allowed foraging worker ants to access the bait but deterred them from carrying off the bait during the observation period. In each trial, lids with different baits were randomly placed 3 cm apart at the base of rainbow shower trees (*Cassia fistula* x *javanica*) where *W. auropunctata* activity was observed. Each trial consisted of no more than six choices to ensure sufficient worker recruitment to all treatments. Each treatment was replicated at 10 sites within the trial plot, and treatment placement was randomized at each site. Field choice tests were conducted between 0800 and 1100 when ants were more likely to be actively foraging. Each baited lid replicate was digitally photographed at 15-min intervals for 2 h. The digital images were later enlarged on a computer monitor in the laboratory, and the ants on each lid were counted and recorded as number of ants attracted to the bait per observation time interval.

Weathered bait attractiveness. Extinguish Plus (0.365% hydramethylnon,

and 0.250% S-methoprene), Maxforce Complete (1.0% hydramethylnon), and Probait (0.73% hydramethylnon) baits were weathered by exposing the baits to sunlight and simulated rainfall for 7 or 14 d. Approximately 50 g of each bait were spread onto wire screens (1x1 mm mesh) and placed on top of a layer of potting media (volcanic cinder topped with peat moss, perlite, and vermiculite mix) in a wooden box (34.3 L x 34.3 W x 7.6 H cm), set on a bench in a greenhouse enclosed with 60% shade cloth, and exposed to sunlight and overhead irrigation (5.7 L min^{-1} for 5 min daily) for 7 or 14 d (8 or 15 d for Extinguish Plus). During bait exposure, average temperature was 22.1°C (range 17.5–28.7°C) and average relative humidity was 84.9% (range 74.7–90.5%) (HOBO Model H8, Onset Computer Corporation, Bourne, MA). Approximately 1.1 g of 7–8-d exposed (DE), 14–15-DE, and fresh (unweathered) bait were placed onto individual plastic lids (n=10 per treatment), and field choice tests were conducted as previously described. Peanut butter was included as a treatment to confirm the presence of *W. auropunctata* at sites where the trials were conducted.

Data analysis. For each trial, ant counts were log-transformed and analyzed using one-way ANOVA, and means were separated by Tukey's test (Roberts 2008, Minitab Version 16, Minitab Inc., State College, Pennsylvania). Non-transformed means are presented in tables. For choice test Trials 3 and 4, data for each trial were first subjected to two-way ANOVA to determine that there were no differences between peanut butter control replicates or standard granular hydramethylnon bait replicates (Amdro Fire Ant Bait or Probait in Trial 3 and 4, respectively) before consolidating all data within each trial for analysis.

Results

Field choice bait tests. Among household ant baits for indoor use evaluated in Trial 1 (Table 1), the paste formulations, both avermectin products (Raid Double Control Ant Baits II and Raid Ant Baits II) containing peanut butter and sucrose, and an indoxacarb product (Hot Shot MaxAttrax Ant $Bait_2$) that exuded a peanut butter scent, attracted as many ants as granular Probait (hydramethylnon) ($P > 0.05$). A semi-solid, gelatinous formulation containing hydramethylnon (Amdro Kills Ants Stake) and a solid fipronil bait (Combat Source Kill Ant) were the least attractive ($P < 0.05$) to *W. auropunctata* (Table 1).

None of the household liquid or gel ant baits for indoor use evaluated in Trial 2 (Table 2) attracted as many *W. auropunctata* workers as the peanut butter lure or granular Amdro (hydramethylnon) ($P < 0.05$). All but one (Green Light Fire Ant Control with Conserve (spinosad)) of the granular ant baits for outdoor use evaluated in Trial 3 (Table 3) attracted as many *W. auropunctata* as Amdro Fire Ant Bait ($P < 0.05$).

Comparing two products with similar concentrations of hydramethylnon but different concentrations of methoprene, Amdro FireStrike Yard Treatment (0.36% hydramethylnon and 0.0172% S-methoprene) attracted 6X as many ants ($P < 0.05$) in 2 h after placement as compared with Extinguish Plus (0.365% hydramethylnon and 0.25% S-methoprene) with nearly twice the concentration of S-methoprene (Table 3). Similarly, in Trial 4 (Table 4), significantly fewer *W. auropunctata* workers ($P < 0.05$) were attracted to Extinguish Professional Fire Ant Bait (0.5% S-methoprene) as compared to Extinguish Plus (0.25% S-methoprene and 0.365% hydramethylnon).

Weathered bait attractiveness. Comparing fresh to weathered hydramethylnon

Table 1. Field attractiveness of household ant baits for indoor use against little fire ants, *Wasmannia auropunctata,* as compared with Probait, 2 h after placement.

Ant Bait	Active Ingredient	Formulation	Number of ants at 2 h (±SEM)
Peanut butter (control)			151.6 ± 26.5a
Raid Ant Double Control Ant Baits II	0.05% abamectin	Paste	126.8 ± 19.8b
Raid Ant Baits III	0.01% abamectin	Paste	125.8 ± 10.1ab
Hot Shot MaxAttrax Ant Bait$_2$	0.05% indoxacarb	Paste	103.5 ± 10.6b
Probait	0.73% hydramethylnon	Granular	98.6 ± 14.0b
Amdro Kills Ants Stake	1.0% hydramethylnon	Gel	12.6 ± 3.5c
Combat Source Kill Ant	0.01% fipronil	Solid	9.4 ± 1.3c

Means in the same column followed by same letter do not differ statistically (Tukey's test) ($P \leq 0.05$). ($F = 33.83$; df = 6; $P = 0.0001$)

Table 2. Field attractiveness of household ant baits for indoor use to little fire ants, *Wasmannia auropunctata,* as compared with Amdro Fire Ant Bait, 2 h after placement.

Ant Bait	Active Ingredient	Formulation	Number of ants at 2 h (±SEM)
Amdro Fire Ant Bait	0.73% hydramethylnon	Granular	141.1 ± 18.2a
Peanut butter (control)			70.2 ± 15.7b
TomCat Ant Killer Gel Bait	0.03% indoxacarb	Gel	9.0 ± 4.6c
Grant's Kills Ants Ant Control	1.0% hydramethylnon	Gel	4.6 ± 1.6c
PIC Ant Killing System	5.0% sodium tetraborate pentahydrate	Liquid	0.5 ± 0.3c
Raid Ant Gel Precision Placement Bait	0.003% thiamethoxam	Gel	0.2 ± 0.1c

Means in the same column followed by same letter do not differ statistically (Tukey's test) ($P \leq 0.05$). ($F = 54.63$; df = 5; $P = 0.0001$)

granular baits (Probait, Extinguish Plus, Maxforce Complete), *W. auropunctata* workers were highly attracted to the fresh baits and the peanut butter control within 15 min of placement, and this preference over the weathered baits ($P < 0.05$) persisted for the entire 2-h observation period (Table 5). Of the three granular ant baits evaluated, less discoloration and mold growth were observed on Extinguish Plus at 8- and 15-DE, which continued to attract more than 50 workers per replicate, whereas mold growth was more extensive on 14-DE Maxforce Complete and 7- and 14-DE Probait, resulting in less worker recruitment ($P < 0.05$) over the 2-h observation period as compared with the respective fresh baits. Samples of moldy bait cultured for identification of microorganisms indicated the presence

Table 3. Field attractiveness of granular ant baits for outdoor use against little fire ants, *Wasmannia auropunctata,* as compared with Amdro Fire Ant Bait, 2 h after placement.

Ant Bait	Active Ingredient	Number of ants at 2 h (±SEM)
Amdro FireStrike Yard Treatment	0.360% hydramethylnon, 0.0172% S-methoprene	85.8 ± 17.7a
Amdro Ant Block Home Perimeter Ant Bait	0.88% hydramethylnon	68.9 ± 17.0a
Peanut butter (control)		54.5 ± 10.3a
Amdro Fire Ant Bait	0.73% hydramethylnon	48.1 ± 9.7ab
Advance 375A	0.011% abamectin	39.9 ± 10.8ab
Maxforce FC Fire Ant Bait	0.00045% fipronil	32.0 ± 8.6ab
Siesta	0.063% metaflumizone	18.2 ± 6.7bc
Extinguish Plus	0.365% hydramethylnon, 0.25% S-methoprene	13.6 ± 6.4bc
Green Light Fire Ant Control with Conserve	0.015% spinosad	0.2 ± 0.1c

Means in a column followed by the same letter do not differ statistically (Tukey's test) ($P \leq 0.05$). ($F = 6.82$; df = 8; $P = 0.0001$).

of predominantly *Curvularia*, *Penicillium* and *Fusarium* spp.

Discussion

Field choice bait tests. Peanut butter, as a lure, verified the presence of *W. auropunctata*, and at 4 of the 5 trial sites attracted as many or more *W. auropunctata* workers as the most attractive insecticidal bait(s) being evaluated. In addition, peanut butter-based household ant bait products, (Raid Double Control Ant Baits II, Raid Ant Baits II, Hot Shot MaxAttrax Ant Bait$_2$) attracted as many ants as the soybean oil based granular Probait (hydramethylnon). Peanut butter provides carbohydrates (sugar and fiber) as well as leguminous protein, and fat in the form of peanut oil (Özcan and Seven 2003, Ayoola et al. 2012); therefore, peanut butter provides essential macronutrients for all life stages in a *W. auropunctata* colony.

The soybean oil–infused, defatted corn grit–based granular baits (Amdro Ant Block Home Perimeter Ant Bait, Amdro Fire Ant Bait, Amdro FiresStrike Yard Treatment, Advance 375A, Extinguish Plus, Probait, Siesta) tested in this study were developed for *S. invicta* and found to be generally attractive to *W. auropunctata,* with the exception of one bait formulated with spinosad, averaging less than one ant per bait placement, perhaps due to a concentration that was repellent to *W. auropunctata*. Likewise, significantly fewer *W. auropunctata* workers were attracted to Extinguish Professional Fire Ant Bait that had a higher concentration of methoprene as compared with Extinguish Plus. Evidently, methoprene at higher concentrations is repellent to *W. auropunctata*, similar to observations by Drees and Barr (1998) with laboratory colonies of *S. invicta* where attractiveness of soybean oil was reduced as concentra-

Table 4. Field attractiveness of granular ant baits for outdoor use against little fire ants, *Wasmannia auropunctata*, as compared with Probait, 2 h after placement.

Ant Bait	Active Ingredient	Number of ants at 2 h (±SEM)
Peanut butter (control)		208.4 ± 19.4a
Maxforce Complete	1.0% hydramethylnon	165.3 ± 17.0ab
Probait	0.73%hydramethylnon	154.6 ± 17.7ab
Extinguish Plus	0.365% hydramethylnon, 0.25% S-methoprene	93.8 ± 15.3b
Advion Fire Ant Bait	0.045% indoxacarb	92.4 ± 12.7b
Extinguish Professional Fire Ant Bait	0.5% S-methoprene	6.4 ± 2.0c

Means followed by same letter in a column (Tukey's test) do not differ statistically ($P \leq 0.05$). ($F = 50.02$; df = 5; $P = 0.0001$)

Table 5. Field attractiveness of fresh versus 7- and 14-day-exposed (DE) Maxforce Complete, Probait, and Extinguish Plus ant baits to little fire ants, *Wasmannia auropunctata*, 2 h after placement.

Ant Bait[1]	Active Ingredient	Number of ants at 2 h (±SEM)
Fresh Maxforce Complete	1.0% hydramethylnon	97.7 ± 16.0b
7-DE Maxforce Complete		18.8 ± 7.2c
14-DE Maxforce Complete		5.5 ± 2.0c
Peanut butter (control)		292.4 ± 21.0a
Fresh Probait	0.73% hydramethylnon	184.6 ± 22.1b
7-DE Probait		6.8 ± 2.6c
14-DE Probait		13.1 ± 4.7c
Peanut butter (control)		264.8 ± 26.8a
Fresh Extinguish Plus	0.365% hydramethylnon, 0.25 S-methoprene	135.7 ± 14.1b
8-DE Extinguish Plus		53.8 ± 6.4c
15-DE Extinguish Plus		82.2 ± 11.0bc
Peanut butter (control)		337.7 ± 28.0a

[1]Weathered baits were irrigated with 28.5 l water per d in a 60% shade greenhouse.
For each bait formulation, means of fresh and weathered baits and control followed by the same letter (Tukey's test) do not differ statistically ($P \leq 0.05$). Maxforce Complete ($F = 34.47$; df = 3; $P = 0.0001$); Probait ($F = 63.26$; df = 3; $P = 0.0001$); Extinguish Plus ($F = 43.29$; df = 3; $P = 0.0001$).

tion of methoprene increased (0.1, 1, 5%).

In this study, MaxForce Complete (1% hydramethylnon) was consistently attractive and had a unique bait matrix of proteins, simple sugars, fats and complex carbohydrates (Bayer Environmental Science 2014). It is likely that MaxForce Complete is able to provide essential nutrients to all life stages in *W. auropunctata* colonies at any given time. Worker ants generally forage for their own nutritional needs, predominantly carbohydrates for energy, and for the needs of the colony, which include carbohydrates for new workers, and protein for brood development and the queen(s) (Cook et al. 2010, Mathieson et al. 2012).

Liquid, gel, and solid ant baits that were not attractive to *W. auropunctata*, including Amdro Kills Ants Stake, Combat Source Kill, PIC Ant Killing System, TomCat Ant Killer Gel Bait, Raid Ant Gel Precision Placement Bait, and Grant's Kills Ants Ant Control, did not claim or were not listed to be attractive to or effective for fire ants, indicating that *W. auropunctata* may similarly not be attracted to these types of baits.

Weathered bait attractiveness. Evaluation of the weathered bait insecticides among granular hydramethylnon ant baits (Probait, Extinguish Plus, Maxforce Complete) indicated a dramatic drop in attractiveness after 7 to 8 d of exposure due to the presence of mold in simulated tropical humid climatic conditions. Less discoloration and mold growth on a granular bait attracted more ants as compared with granular baits with more extensive mold growth. Most granular bait labels state that excessively wet conditions (prolonged high humidity, frequent and prolonged rainfall and heavy mist) can reduce effectiveness or render ant baits unattractive and not to apply baits when the ground is saturated with water, heavy dew or when rains are expected within 12 hours. Oliver et al. (2010) also reported that wet bait is not attractive to fire ants. Manufacturers of ant bait insecticides may consider adding a mold inhibitor and hydrophobic carriers to preserve the attractiveness and physical integrity of their baits.

Other ant bait formulations should be developed for treating *W. auropunctata* infestations in tropical environments with high rainfall and humidity levels, including encapsulation in alginate pellets, which were used to deliver *Beauveria bassiana* to *S. invica* colonies (Bextine and Thorvilson 2002), or saturating polyacrylamide hydrogels with effective insecticides, as was done to deliver thiamethoxam to *L. humile* colonies (Buczkowski et al. 2014). Attraction of gel formulations of baits could possibly be improved by inclusion of preferred food sources, such as peanut butter or animal-based protein.

Acknowledgments

We thank Jade Miyashiro for data collection and the University of Hawaii at Hilo, College of Agriculture, Forestry and Natural Resource Management (CAFNRM) Instructional Farm for allowing us to conduct the trials in an infested plot. This study was funded in part by Plant Pest and Disease Management and Disaster Prevention Programs - Farm Bill administered by the United States Department of Agriculture, Animal and Plant Health Inspection Service, Plant Protection and Quarantine and the Hawaii Department of Agriculture.

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