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Comparison of the Effectiveness of a Label-Rate Broadcast Treatment With Advion™, a Label-Rate Broadcast Treatment with Amdro®, and a Pre-Baiting Broadcast Treatment with Advion™ at Controlling *Solenopsis Invicta* (Hymenoptera: Formicidae)

by

Barry D. Furman & Roger E. Gold

ABSTRACT

Indoxacarb-containing Advion™ is a new bait that was developed for the control of the red imported fire ant (RIFA), *Solenopsis invicta* Buren. To date, only limited research has been conducted which compares the effectiveness of Advion™ to other RIFA baits, and no research has been conducted to analyze the effectiveness of Advion™ when used in a pre-baiting broadcast treatment. The concept of pre-baiting involves treatment with non-toxic bait in order to stimulate feeding activity prior to treatment with toxic bait. The objective of this research was to compare the effectiveness of label-rate broadcast treatment with Advion™, label-rate broadcast treatment with Amdro®, and pre-baiting broadcast treatment with Advion™, in terms of overall level and speed of RIFA control. For the treatments consisting of Advion™ or Amdro®, 0.13 ha plots were subjected to label-rate (1.7 kg/ha) broadcast treatment with either 0.045% Advion™ or 0.73% Amdro®. For the pre-baiting treatment, 0.13 ha plots were first subjected to broadcast treatment (1.7 kg/ha) with 0.0% Advion™ (no indoxacarb), followed 1 h later by label-rate broadcast treatment with 0.045% Advion™. Both pre-baiting broadcast treatment and label-rate broadcast treatment with 0.045% Advion™ resulted in the same overall level of RIFA colony mortality (98-99%), which was significantly greater ($P<0.05$) than that resulting from treatment with Amdro® (87%). Ultimately, label-rate broadcast treatment with 0.045% Advion™ was determined to be the most effective treatment due to the fact that it resulted in an LT_{90} of only 6.2 d, compared to the 11.7 d LT_{90} resulting from pre-baiting broadcast treatment.

Keywords: Advion™, indoxacarb, ant bait, *Solenopsis invicta*, red imported fire ant, fire ant control, broadcast treatment, pre-baiting.

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INTRODUCTION

Since the 1930's, a wide variety of chemicals and methods of delivering those chemicals have been utilized in an attempt to control the red imported fire ant (RIFA), *Solenopsis invicta* Buren (Banks 1990; Collins *et al.* 1992; Eden & Arant 1949; Lofgren *et al.* 1964; Lofgren *et al.* 1975; Phillips & Thorvilson 1989; Sauer *et al.* 1982; Vander Meer *et al.* 1982). Beginning in the 1960's, the effectiveness of baits as a method of chemical delivery was realized, and their use became increasingly popular. Over the years, a number of different chemicals have been delivered via baits in an attempt to control the RIFA. These chemicals range from the GABA-gated chloride channel antagonist, mirex, which was used in the 1960's and 1970's, to chemicals that are being used at the present time, such as the Type II electron transport inhibitor, hydramethylnon, and the juvenile hormone mimic, fenoxycarb (Collins *et al.* 1992; Lofgren *et al.* 1975; Phillips & Thorvilson 1989; Vander Meer *et al.* 1982). The newest chemical available for the control of the RIFA is indoxacarb. Discovered by E.I. DuPont de Nemours and Company in 1991, the oxadiazine indoxacarb is a novel insecticide that must first be metabolized in order to become acutely toxic (McCann *et al.* 2001). Following ingestion by the insect, metabolic breakdown of indoxacarb occurs, ultimately producing an N-decarbomethoxyllated metabolite. It is this metabolite that is toxic to the insect, functioning as a potent, voltage-dependent sodium channel blocker that ultimately results in the rapid death of the insect (Wing *et al.* 2000).

Registered by the United States Environmental Protection Agency in August of 2004 (EPA Registration #352-627), one of the newest baits available for RIFA control is indoxacarb-containing Advion™ (E.I. DuPont de Nemours and Company, Wilmington, DE). As with many RIFA baits, Advion™ can either be applied as a broadcast treatment, or as an individual mound treatment. While both methods have the potential to effectively achieve control of the RIFA, individual mound treatments are generally more expensive and more labor intensive than broadcast treatments. Though there are some circumstances in which individual mound treatments are more practical and appropriate, broadcast treatments are widely considered to be more efficient than individual mound treatments in terms of the cost/labor to benefit ratio (Barr 1999; Barr & Best 1999; Barr *et al.* 1999; Drees *et al.* 1996).

The objective of this research was to compare the effectiveness of label-rate broadcast treatment with Advion™ to the effectiveness of two other broadcast treatments: label-rate broadcast treatment with the hydramethylnon-containing RIFA bait Amdro® (Ambrands, Atlanta, GA), and pre-baiting broadcast treatment with Advion™. To date, only limited research comparing the effectiveness of broadcast treatment with Advion™ to the effectiveness of broadcast treatment with other baits has been conducted and published (Barr 2003, Barr 2004). Furthermore, no research results have been published on the effectiveness of Advion™, or any other RIFA bait, when used in a pre-baiting broadcast treatment. Pre-baiting, which involves treatment with non-toxic bait in order to stimulate feeding activity prior to treatment with toxic bait, has already proven to be effective at controlling numerous pest species (Shumake *et al.* 2002; Sterner 1999).

MATERIALS AND METHODS

This experiment was conducted in the field on a private game ranch (N28°05'79" W98°05'75") located in Jim Wells County, Texas. The property was ~4856 ha, with the majority of the land groomed for dove, quail and deer hunting. A considerable portion of the land was utilized for cattle grazing, as well. Though there were a variety of different types of vegetation throughout the property, this experiment was conducted on a 5 ha field consisting of grass pastureland. No cattle grazing was allowed on this field immediately preceding the experiment, during the experiment, or immediately following the experiment. The grass comprising this 5 ha field was mowed ~1 wk prior to the beginning of the experiment and was then allowed to grow unimpeded for the duration of the evaluations.

RIFA mounds were selected for use in this experiment by first placing an individually numbered, 50 cm tall, metal wire, fluorescent colored flag in the center of a given mound. Once placed in the mound, the metal wire was vibrated to elicit a response from the RIFA and to determine the activity level of the colony. The following Lichert scale was used to determine the activity level: "0" = inactive (no ants responding), "1" = minor activity (1-50 ants responding), "2" = moderate activity (51-100 ants responding) and "3" = fully active (more than 100 ants responding). Only colonies with a Lichert scale rating of "3" were selected for use in this experiment (Gold *et al.* 1996a,

1996b). All RIFA mounds on the 5 ha field that was used for this experiment were measured for colony activity in this manner.

Next, 15 rectangular-shaped 0.13 ha plots were measured via the use of a Rolatape® M300 series measuring wheel (Rolatape Corp., Spokane, WA). Each plot was separated from the next nearest plot by at least 15 m. A 46 cm x 5 cm x 2 cm wooden stake was then hammered into the soil at each of the four corners of a given plot, and #16 polyester/cotton twine was used to connect one stake to the next, thus forming a clearly demarcated rectangular border. At least 10 RIFA mounds with an initial Lichert scale rating of "3" were located within the borders of each of these 15 plots.

For this experiment, there were four different treatment groups and a control group: Treatment 1 consisted of broadcast treatment with 0.0% Advion™ (contained no indoxacarb); Treatment 2 consisted of broadcast treatment with 0.73% Amdro®; Treatment 3 consisted of pre-baiting broadcast treatment comprised of broadcast treatment with 0.0% Advion™ (contained no indoxacarb) followed 1 hour later by broadcast treatment with 0.045% Advion™; Treatment 4 consisted of broadcast treatment with 0.045% Advion™; and the control group consisted of untreated colonies. Treatment 1 was included in this experiment due to the fact that there has previously been no research conducted to analyze any potential insecticidal qualities that the inert ingredients within Advion™ might exhibit. Therefore, this treatment was included to ensure that the mortality resulting from treatment with Advion™ resulted solely from indoxacarb. There were three replicates for each of the four treatment groups and the control group, thus a total of 15 plots were utilized for this experiment.

Experimentation was conducted for a total of 7 wk (June 4-July 23, 2004). Between approximately 7:00 a.m. and 9:00 a.m. C.S.T. on the first morning of the experiment, broadcast treatments were administered at label rates (1.7 kg/ha) via the use of Scotts® Handy Green II® hand spreaders (Scotts Company, Marysville, OH). Separate hand spreaders were used for each of the different treatments. For Treatment 1, 227 g of 0.0% Advion™ was broadcast over each plot, and 227 g of 0.73% of Amdro® was broadcast over each plot for Treatment 2. For Treatment 3, 227 g of 0.0% Advion™ was broadcast over each plot, and then ~1 h later 227 g of 0.045% Advion™ was broadcast over each of those same plots. For Treatment 4, 227 g of 0.045% Advion™ was broadcast over each plot. The plots for the control group were left untreated. The bait used for each treatment was carefully weighed using an Ainsworth® 6000 g electric scale, model APX-6001 (Denver Instrument Company, Denver, CO), and placed into separate, sealed, plastic bags the morning of treatment to ensure that each plot within a given treatment group received an identical amount of bait.

The next morning, ~24 h following treatment, the metal wire of the flag that had been placed in each mound was vibrated to elicit a response from the RIFA and measure the activity level of the colony. The response by the ants was graded according to the Lichert scale previously described, and colony mortality was assumed when a response of "0" was observed. Data was gathered in this manner each day for the first 7 d, with an additional reading for each colony being taken 2 wk later, and one final reading for each colony being taken 4 wk from the previous reading. Thus, ultimately, a data set of colony mortality was produced for each treatment. Longevity of control afforded by each treatment was also measured via daily observations of any new RIFA mounds appearing within the plots.

Additionally, the soil temperature and precipitation were measured during this experiment. Soil temperature was measured with a Sergeant-Welch 12.7 cm soil thermometer (Sergeant-Welch, Buffalo Grove, IL) each morning within each of the plots for the first 7 d of the experiment, and then again on the two subsequent mornings when additional colony activity readings were taken. Precipitation was measured with a Garden Treasures® 15.2 cm capacity rain gauge (Lowe's Companies Inc., Wilkesboro, NC) for each 24 h period for the first 7 d of the experiment. No 24 h precipitation readings were taken after the first 7 d.

Statistics. At the conclusion of this experiment, SPSS® software (SPSS 2001) was used to conduct statistical analysis of the data. First, however, Abbott's formula was used to correct all mortality data (Abbott 1925). Abbott's formula for correcting mortality data is:

$$[(X-Y) / X] \times 100 = \text{percent control}$$

where "X" is the percent colony survival in the control group, and "Y" is the percent colony survival in the treatment group. Next, ANOVA was conducted on the corrected mortality data sets. Finally, the LSD post hoc test

was conducted on those same data sets to determine significant differences among treatments. All tests of significance were evaluated at P = 0.05.

RESULTS

The mean morning soil temperature and the daily precipitation are shown in Table 1. The mean morning soil temperature was always 26.0° C or greater, and no precipitation occurred within 2 d of when the broadcast treatments were conducted. A comparison among treatments of the mean cumulative daily colony mortality is shown in Fig. 1 ($F=4.28$; $df=35$; $P<0.05$). For days 2-7 of the experiment, significantly greater ($P<0.05$) RIFA colony mortality resulted from broadcast treatment with 0.045% Advion™ than from any of the other treatments. Pre-baiting broadcast treatment resulted in significantly greater ($P<0.05$) colony mortality during that same time period than all other treatments except for the broadcast treatment with 0.045% Advion™. No colony mortality resulted from broadcast treatment with 0.73% Amdro® until day 5, and no colony mortality resulted from broadcast treatment with 0.0% Advion™ during the entire first week. By day 21, both broadcast treatment with 0.045% Advion™ and pre-baiting broadcast treatment had essentially eliminated all colonies within their respective plots, and there was no significant difference ($P>0.05$) in mortality between these two treatments. Additionally,

Table 1. Mean morning soil temperature and daily precipitation for the experiment that consisted of label-rate broadcast treatment with 0.045% Advion™, 0.73% Amdro®, and 0.0% Advion™ (contained no indoxacarb), and pre-baiting broadcast treatment consisting of treatment with 0.0% Advion™ (contained no indoxacarb) followed 1 h later by treatment with 0.045% Advion™.

Day	Mean morning soil temperature ± SD (°C)	Daily precipitation (cm)
0	28.4±0.7	0.0
1	27.7±0.6	0.0
2	27.1±0.9	0.0
3	26.6±0.7	<0.3
4	26.2±0.9	3.0
5	26.0±0.6	0.3
6	26.3±0.7	1.3
7	26.5±0.4	<0.3
21	26.6±0.4	—[a]
49	27.4±0.7	—[a]

[a] A precipitation reading was not taken on this day.

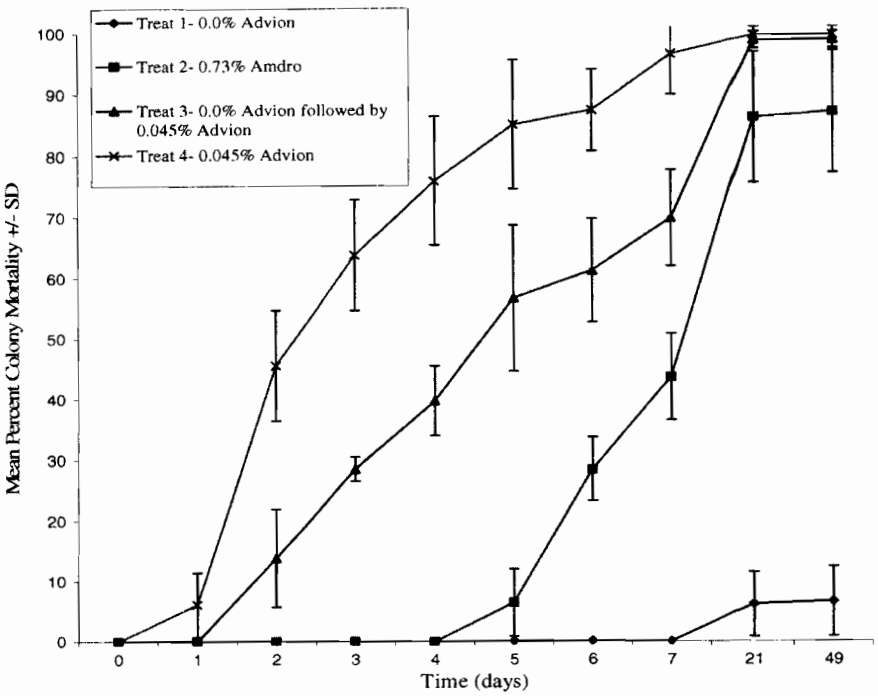

Fig. 1. A comparison of the mean percent RIFA colony mortality resulting from label-rate broadcast treatment with 0.045% Advion™, 0.73% Amdro®, and 0.0% Advion™ (contained no indoxacarb), and from pre-baiting broadcast treatment consisting of treatment with 0.0% Advion™ (contained no indoxacarb) followed 1 h later by treatment with 0.045% Advion™.

both of the previously mentioned treatments resulted in significantly greater ($P<0.05$) colony mortality by day 21 than broadcast treatment with 0.73% Amdro®, and there was no change in these data by day 49.

A comparison of the LT_{50}'s and LT_{90}'s among treatments is shown in Table 2. The LT_{50} resulting from broadcast treatment with 0.045% Advion™ (LT_{50}=2.4 d) was one half the corresponding LT_{50} resulting from pre-baiting broadcast treatment (LT_{50}=4.7 d), and both of those LT_{50}'s were considerably less than that resulting from broadcast treatment with 0.73% Amdro® (LT_{50}=9.7 d). Similarly, the LT_{90} resulting from broadcast treatment with 0.045% Advion™ (LT_{90}=6.2 d) was one half the corresponding LT_{90} resulting from pre-baiting broadcast treatment (LT_{90}=11.7 days). Broadcast treatment with 0.73% Amdro® did not attain an LT_{90}, and broadcast treatment with 0.0% Advion™ did not attain an LT_{50} or an LT_{90}.

A comparison of the mean number of new RIFA mounds appearing i
the plots of the four treatment groups and the control group at days 21
49 is shown in Table 3 (F=7.11
P<0.05). After 21 d, there was li
ference among the groups, as fo
the five groups exhibited no si
difference (P>0.05) in terms
number of new RIFA mounds p
Additionally, all four of those
had a mean of less than one ne
mound per plot after 21 d, and t
that were subjected to either b
treatment with 0.045% Advion™
baiting broadcast treatment co
no new RIFA mounds. However,
d, the control group and the tre
group consisting of broadcast tre
with 0.0% Advion™ had a signi
greater (P<0.05) number of new
mounds per plot than the othe
groups, with each having three ne
mounds per plot. Though the tre
group consisting of broadcast tre
with 0.73% Amdro® had signi
fewer (P<0.05) new RIFA mou
plot than both the control group
treatment group consisting of b
treatment with 0.0% Advion™,
significantly greater (P<0.05)
of new RIFA mounds per p
treatment groups consisti
treatment with 0.045
baiting broadcast
no significant di
of new RIFA m

Table 2. Comparison of the LT50's and the LT90's for RIFA colonies that were treated via label-rate broadcast treatment with 0.045% Advion™, 0.73% Amdro®, and 0.0% Advion™ (contained no indoxacarb), and via pre-baiting broadcast treatment consisting of treatment with 0.0% Advion™ (contained no indoxacarb) followed 1 h later by treatment with 0.045% Advion™.

Lethal time	LT50 and LT90 per treatment (d)			
	Treatment 1- plots treated with 227 g of 0.0% Advion™	Treatment 2- plots treated with 227 g of 0.73% Amdro®	Treatment 3- plots treated first with 227 g of 0.0% Advion™, then with 227 g of 0.045% Advion™	Treatment 4- plots treated with 227 g of 0.045% Advion™
LT50	—[a]	9.7	4.7	2.4
LT90	—[b]	—[b]	11.7	6.2

[a] An LT50 was not attained during this experiment.
[b] An LT90 was not attained during this experiment.

...ed Mean cumulative number of new RIFA mounds per plot [a,b] (Mean ±SD)

	Treatment 1- plots treated with 227 g of 0.0% Advion™	Treatment 2- plots treated with 227 g of 0.73% Amdro®	Treatment 3- plots treated first with 227 g of 0.0% Advion™, then with 227 g of 0.045% Advion™	Treatment 4- plots treated with 227 g of 0.045% Advion™	Control- plots left untreated
21	1.3±0.6a	0.3±0.6b	0.0±0.0b	0.0±0.0b	0.7±0.6ab
49	2.7±0.6a	1.3±0.6b	0.0±0.0c	0.3±0.6c	3.0±0.0a

[a] No new RIFA mounds were observed on any of the plots for days 1-7 of the experiment. Therefore, only new RIFA mounds observed on days 21 and 49 were used in this analysis.
[b] Means within a row with different letters are significantly different at P<0.05. Means were separated using the LSD test.

latter two treatments and, at 0.3 and 0.0 new mounds per plot respectively, they had significantly fewer (P<0.05) new RIFA mounds per plot than the other two treatment groups and the control group.

DISCUSSION

For this experiment, as with all experimentation conducted on RIFA in the field, it was important to ensure that all bait was broadcast when the soil temperature was within the optimal foraging range of the ants (22-36° C), and when a rainfall event was not expected for one or more days following treatment (Drees *et al.* 1996; Vinson 1997). As the data shown in Table 1 indicated, the bait was available during peak foraging periods, and the bait was not adversely affected by a rainfall event. Thus, the timing of treatment for this experiment was validated.

As was clearly shown by the data in Fig. 1 and Table 2, broadcast treatment with 0.045% Advion™ resulted in 90% colony mortality far more quickly than any of the other treatments. The next nearest treatment in terms of overall colony mortality and speed of mortality was pre-baiting broadcast treatment, which took nearly twice as long to achieve 90% colony mortality as broadcast treat-

ment with 0.045% Advion™. However, though it took longer to achieve, pre-baiting broadcast treatment ultimately resulted in the same overall level of RIFA colony control as the aforementioned broadcast treatment with 0.045% Advion™, as there was no significant difference ($P>0.05$) between the two treatments by the third week of the experiment. Essentially, though the total number of granules that is ultimately spread out over a given land area via label-rate broadcast treatment is relatively small even when making two applications of bait, as was done with the pre-baiting broadcast treatment, there appears to be a dilution effect associated with the pre-baiting applications. This was likely the reason that, even though pre-baiting broadcast treatment ultimately achieved the same level of RIFA colony mortality as the highly effective label-rate broadcast treatment with 0.045% Advion™, it took nearly twice as long to attain that level.

Broadcast treatment with 0.73% Amdro®, which ultimately achieved ~87% colony mortality over the duration of this experiment, resulted in significantly less ($P<0.05$) colony mortality than either of the two aforementioned treatments. Further, the LT_{50} resulting from treatment with Amdro® was more than twice that of the pre-baiting broadcast treatment, and more than four times that of label-rate broadcast treatment with Advion™. Broadcast treatment with 0.0% Advion™ resulted in no colony mortality for the first 7 d and, ultimately, RIFA colony mortality only reached ~6%, which was not significantly different than the untreated control group. The data from this broadcast treatment with 0.0% Advion™ indicated, therefore, that the insecticidal qualities of Advion™ did in fact result solely from the chemical indoxacarb. Resultantly, the label-rate broadcast treatment with 0.045% Advion™ was the most effective overall in terms of RIFA colony mortality and speed of mortality.

The data in Table 3 indicated that there was little difference between treatments in terms of the number of new RIFA mounds per plot observed 21 d after treatment. However, there were significant differences in the number of new RIFA mounds per plot 49 d after the broadcast treatments were conducted. Both the control group and the treatment group consisting of broadcast treatment with 0.0% Advion™ had a mean of three new RIFA mounds per plot by day 49, and this was significantly greater ($P<0.05$) than the number of new RIFA mounds per plot in any of the other three groups.

Though there were only 1.3 new RIFA mounds per plot by day 49 in the treatment group consisting of broadcast treatment with Amdro®, that was still significantly greater ($P<0.05$) than the number of new mounds per plot that were observed in the treatment groups consisting of broadcast treatment with 0.045% Advion™, and pre-baiting broadcast treatment. After 49 d, only one new RIFA mound was observed on the plots subjected to the latter two treatments and, resultantly, those two treatment groups had significantly fewer ($P<0.05$) new RIFA mounds per plot than any of the other treatment groups.

In conclusion, both pre-baiting broadcast treatment and label-rate broadcast treatment with 0.045% Advion™ ultimately resulted in the same high level of RIFA colony mortality (98-99%), which was significantly greater ($P<0.05$) than that resulting from the other treatments. Further, there was no significant difference ($P>0.05$) in the mean number of new RIFA mounds observed per plot over the course of this experiment for the two aforementioned treatment groups. However, though pre-baiting broadcast treatment and broadcast treatment with 0.045% Advion™ ultimately achieved the same level and duration of RIFA control, broadcast treatment with 0.045% Advion™ was determined to be the most effective treatment overall due to the fact that it resulted in an LT_{90} of only 6.2 d, which was approximately half the LT_{90} resulting from pre-baiting broadcast treatment.

In order to further validate these results, this experiment should be repeated. In future research, it would be ideal to use various quantities of 0.0% Advion™ in the pre-baiting experiment to determine whether pre-baiting is, in fact, less effective due to a dilution effect. Additionally, the length of time it takes for the RIFA to re-colonize an area that has been subjected to broadcast treatment with a bait is highly variable, and largely dependent upon the number of RIFA colonies located on the land adjacent or in close proximity to the treatment area (Drees *et al.* 1996). Although for this experiment there was only one new RIFA colony observed after 49 d on the plots that were treated in some manner (i.e. pre-baiting broadcast treatment or label-rate broadcast treatment) with 0.045% Advion™, numerous studies of varied duration in various environments need to be conducted to definitively determine the longevity of control afforded by label-rate broadcast treatment with 0.045% Advion™.

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