PROCEEDINGS OF THE 1986 IMPORTED FIRE ANT CONFERENCE

April 3-4, 1986
Austin, Texas

Hosted by
The Texas Department of Agriculture

Compiled by
Michael E. Mispagel, Ph.D.

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Dr. Michael E. Mispagel
Department of Physiology and Pharmacology
College of Veterinary Medicine
The University of Georgia
Athens, GA 30602

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(NS indicates that a contribution to this Proceedings was Not Submitted)

Recent Chemical Studies on the Imported Fire Ants, *Solenopsis invicta* and *S. richteri*

Robert K. Vander Meer
Insects Affecting Man and Animals Research Laboratory
ARS, USDA
P. O. Box 14565
Gainesville, Florida 32604

Two of the most important phenomena occurring with the imported fire ants of the southern United States are (A) the increasing populations of multiple queen colonies and (B) the discovery of viable hybrids from *Solenopsis invicta* and *S. richteri*. Both of these recently discovered forms pose serious questions for present and future control programs. This paper discusses what is currently known about the hybridization problem.

Last year we published the first report of hybridization between *S. invicta* and *S. richteri*. We used species specific venom alkaloids and hydrocarbons as biochemical characters to distinguish the two species. Sampling and analysis of *S. invicta* in Louisiana, Alabama, and Florida and *S. richteri* in the northern most extent of its range in northeastern Mississippi, demonstrated the consistency of these biochemical characters. In addition, we found that the Dufour's gland (the source of fire ant trail pheromones) is also an excellent species-specific character. All three biochemical markers clearly distinguished hybrid colonies. As would be expected, the gas chromatograph (GC) traces of hybrid colonies were intermediate between those identified as pure species.

Based on the venom alkaloid and hydrocarbon GC patterns of the pure species, we devised an algorithm that enabled us to calculate a biochemical similarity index. This provided a means of objectively evaluating colony GC data. Over 90% of 100 suspected hybrid colonies had an index value of 0.33-0.77 (1.0 = *S. invicta* and 0.0 = *S. richteri*).

Dr. Ken Ross and colleagues at the University of Georgia, in collaboration with our laboratory, carried out isozyme studies of hybrid and parental populations. The isozyme method shows excellent agreement with the biochemical methods described above. The isozyme systems used by Dr. Ross showed a clinal distribution with hybrids close to the *S. invicta* border similar to *S. invicta* and hybrids found close to the *S. richteri* border more similar to *S. richteri*. A plot of the biochemically derived index showed a similar cline.

Based on the isozyme studies of Dr. Ross and our own work, the hybrid is reproductively viable. Collections made by Mr. Stan Diffie, U of Georgia (Tifton), and our chemical evaluation have, surprisingly, shown the hybrid population to extend all the way across northern Alabama and into northwestern Georgia. Since S. richteri does not border this population, it is assumed that they were brought in with nursery stock some time ago. Therefore, the hybrid occupies a much larger area than originally suspected. Additional work in collaboration with Professor E. O. Wilson, Harvard University, has shown that hybridization between S. invicta and S. richteri occurred as far back as 1949. Hybridization is not a recent event. Its detection was complicated by the morphological similarity of the hybrid to S. richteri.

This extensive hybrid population may pose serious problems to fire ant control programs. For example, will biocontrol methods found for S. invicta also work on the hybrid? Is the hybrid more competitive than the pure forms? There are also taxonomic implications such as; should S. invicta have been given species status (depends on your definition of species). There is a great deal of work to be done in order to define hybrid population dynamics, and behavioral, biochemical, and physiological differences from the parent species.

SOLENOPSIS INVICTA PREDATION ON SUBTERRANEAN TERMITES

Deborah A. Waller and Jeffery P. La Fage

Entomology Department

Louisiana State University

Baton Rouge, Louisiana 70803 USA

Subterranean termites comprise up to 16% of fire ant diets in Louisiana (Wilson and Oliver 1969). The native Reticulitermes spp. and the introduced Coptotermes formosanus Shiraki (both Rhinotermitidae) are common in fire ant habitats. Both termite genera have soldier castes specialized for defense, but it is unknown whether soldiers are able to deter fire ant predation. In the present study, we investigated the effects of C. formosanus soldier secretion on S. invicta and the small ant, Monomorium minimum, and we tested the hypothesis that sodiers were unpalatable to ant foragers.

Ten ants of each species were immersed in ca. 0.4 ml C. formosanus secretion after determining that this amount was normally expelled by medium-sized termite soldiers after disturbance. Ants were chilled prior to treatment to facilitate handling. Ten control ants were chilled and wiped on a wet surface. There was no difference in survival after one day for treated and control S. invicta (Fisher's exact test, $p > .05$), and both groups of ants groomed similar lengths of time upon recovery (Mann Whitney U test, $p > .05$), indicating that the secretion does not contain noxious chemicals. In contrast, 90% M. minimum died from the treatment, while all controls recovered (Fisher's exact test, $p < .01$). The secretion probably acts as a glue to asphyxiate small ants, but is ineffective against larger ants on topical application.

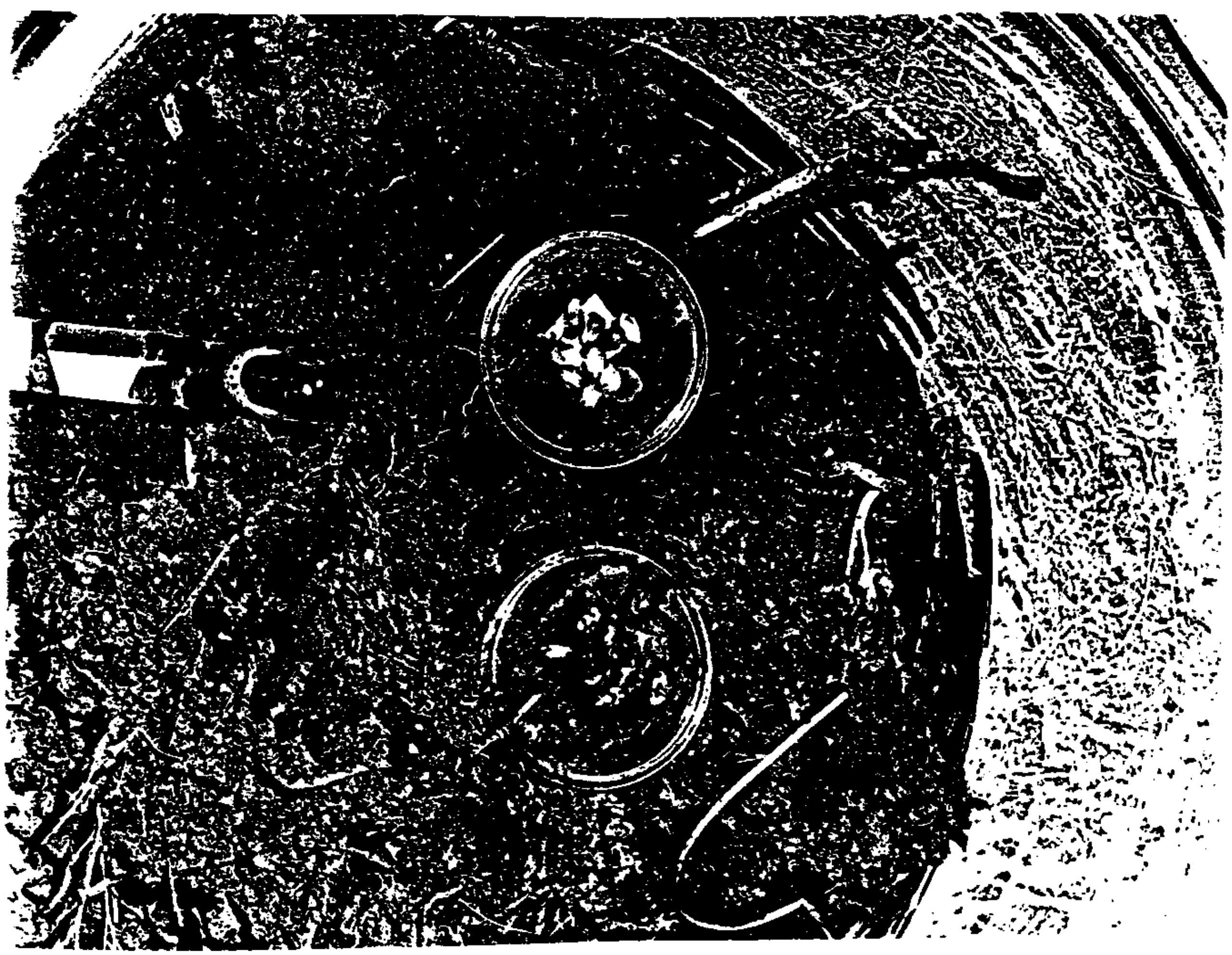
Frozen C. formosanus workers and soldiers were presented to M. minimum and S. invicta colonies. M. minimum removed workers and rejected soldiers on the first test date. S. invicta rejected soldiers after several days of presentation, so that there was a delayed response to soldier unpalatability. This difference may have resulted because S. invicta directly carried off prey items, while M. minimum carved up termites before removal, thus contacting internal chemistry while still in the foraging arena.

Solutions of homogenized C. formosanus workers attracted significantly more ants than soldier solutions when applied to popcorn kernals and placed in ant nests (Figure 1). Mixtures of worker and soldier solutions attracted as many ants as worker solutions, so that soldier solution probably does not contain any repellent chemicals. A comparison among solutions of C. formosanus workers and soldiers, and Reticulitermes virginicus worker and soldier solutions revealed that both soldier solutions attracted low numbers of ants, while both worker solutions were attractive. These results indicate that soldier chemistry is responsible for soldier unpalatability to ants.

Overall our results suggest that while C. formosanus soldiers are not directly repellent to S. invicta, soldiers are unpalatable in that they do not stimulate recruitment. R. virginicus soldiers also appear to be unpalatable. Soldier unpalatability may protect termite colonies if ants are deterred from recruiting nestmates to prey on the colony after encountering only unpalatable soldiers.

Figure 1

Solutions of homogenized C. formosanus workers and soldiers (40 termites/ ml deionized water) on soaked corn kernels presented to five S. invicta colonies.



ABSTRACT

A Long-term Study of a Single-queen/Multiple-queen Transect

Les Greenberg and S.B. Vinson
Department of Entomology
Texas A&M University, College Station, TX 77843

In 1982 we began a study of a 500 x 100 ft. transect at the Texas A&M University Farm. This farm has both single- and multiple-queen forms of the red imported fire ant. The transect includes both types. The first 100 ft. of the transect has a high density of multiple-queen mounds. The remaining 400 ft. has a much lower mound density and consists mostly of single-queen colonies. Workers were sampled by pushing a vial containing alcohol into the mound to catch a sample of workers. Worker head width was measured in the laboratory. Since there is a negative correlation between the number of queens in a colony and the size of their workers(1), samples of workers from all the mounds in the transect were measured over a period of 4 years to determine which were single- or multiple-queen colonies and whether one form of the fire ant was displacing the other.

The single- and multiple-queen forms are separated by a slight drainage area that floods during heavy rain. In the Spring of 1986 there was an unusual dry spell. Samples taken at that time showed that the multiple-queen form had crossed the drainage barrier and had moved 150 ft. into the single-queen population, as evidenced by their much smaller workers and higher mound densities. During the course of this study the number of single-queen mounds has remained static, while multiple-queen mounds have increased dramatically. Generally speaking, in College Station and surrounding areas it has become increasingly difficult to find single-queen colonies.

(1)Greenberg, L., D.J.C. Fletcher and S.B. Vinson, 1985. Differences in worker size and mound distribution in monogynous and polygynous colonies of the fire ant *Solenopsis invicta* Buren. J. Kansas Entomol. Soc., 58: 9-18.

Seasonal Changes In Bait Preference And Foraging Activity Of Red Imported Fire Ants

Stein, M.B. and H.G. Thorvilson

ABSTRACT

Using a bait attraction plot sampling regime, seasonal changes in foraging activity and bait preference of red imported fire ants were investigated. Foraging activity decreased and preference for the carbohydrate bait increased as preference for the protein bait decreased, as summer progressed into winter.

SUPERCOOLING POINTS OF RED IMPORTED FIRE ANTS

FROM LUBBOCK, TEXAS

BY

STEPHEN W. TABER

AND

JAMES C. COKENDOLPHER

DEPARTMENT OF ENTOMOLOGY
TEXAS TECH UNIVERSITY
LUBBOCK, TEXAS 79409

ABSTRACT

Supercooling points were measured for red imported fire ants from Lubbock, Texas. Ten minor workers from each of three marked colonies and one colony held at constant conditions in the laboratory were tested every two weeks from 14 October to 17 February. The supercooling abilities of the ants were altered by conditions in the laboratory. Differences between mean supercooling temperatures of ants in the field were found among sampling periods, but not among colonies. These differences may not be due to acclimatization. The maximum difference between mean supercooling points was less than 2 C^{o}, and the lowest mean supercooling point was slightly higher than -6^{o} C.

INTRODUCTION

We first became aware of the presence of red imported fire ants in Lubbock, Texas, during August 1985. Homeowners from a single subdivision reported colonies which had apparently been present for several years. This information was of immediate interest to us because Lubbock, at 33.5^{o} N latitude, then represented the extreme northwestern record of Solenopsis invicta. Furthermore, at 3200 feet elevation, the Lubbock population is among the highest on record. At least one prediction of the ultimate range of this species in Texas excluded the Panhandle area north of the 0^{o} F minimum temperature isotherm because of low winter temperatures in that area. Lubbock lies at the base of the Panhandle, and we were therefore in an ideal position to examine some features of Solenopsis invicta related to cold-hardiness or the ability to survive winter-kill.

Cold-hardiness in insects may be divided into three general categories: (1) cold-acclimation and acclimatization, (2) supercooling, and (3) freezing-tolerance. Cold-acclimation requires some physical or physiological preparation to avoid injury at temperatures too low for continued growth. We refer to short-term, laboratory adjustments as acclimation and gradual, naturally occurring adjustments as acclimatization. Supercooling is an adaptation in which insects avoid injury by resisting the freezing process. If the organism is able to withstand bodily freezing, it is described as being freeze-tolerant.

Supercooling occurs when the temperature of a liquid is lowered, without freezing, below its expected freezing point. Highly purified water may be rapidly cooled to approximately -40^{o} C without freezing.

but significant differences were found among castes (F (2,81 = 9.15, p<0.001). We found that bodily freezing is lethal for fire ants maintained under the laboratory conditions. Nothing was previously known, however, of the acclimatization abilities of Solenopsis invicta. Perhaps the onset of decreasing winter temperatures would trigger a physiological response which would decrease their supercooling points enough to ensure survival of some individuals and colonies. We decided to investigate the supercooling abilities of fire ants during the fall and winter months because the process is both indicative of cold-hardiness and easily observable.

RESULTS AND DISCUSSION

Every two weeks from October 1985 until March 1986 we collected 10 minor workers from each of three designated mounds and determined their supercooling points. There were a total of 10 such samples taken during the 18 week period. The previous study demonstrated that supercooling abilities differ among caste members and we decided to use minors in the present experiments because they were reported to have the lowest supercooling points. We measured these temperatures using a Bailey Instruments thermocouple microprobe and a digital thermometer with a 0.1 C^{o} resolution. A single minor worker was affixed with petroleum jelly to the tip of the microprobe which was then sealed within a glass shell vial. The assembly was placed inside a freezer and cooled at approximately 5 C^{o}/minute. Supercooling points were easily recognized by a sudden increase in temperature due to the release of the latent heat of fusion. After obtaining data from field colonies, we measured the supercooling points of 10 minor workers from

This phenomenon is enhanced by the elimination of molecular aggregations known as nucleators which are conducive to ice formation. The melting point or expected freezing point may also be depressed by the production of anti-freeze substances, dehydration, or gut evacuation. These modifications are important because many insects, including certain ant species, are unable to survive bodily freezing. For example, the formicine ant Camponotus herculeanus supercools, freezes, and dies at almost -30^{o} C but is able to survive less extreme exposures. Among the myrmicine ants, Leptothorax acerovorum and Myrmica rubra freeze at -7.1^{o} and -16.1^{o} C, respectively.

Previous work dealing only with laboratory colonies of North American fire ant species revealed no significant differences between mean supercooling temperatures of Solenopsis aurea, S. richteri, and S. xyloni within the minor caste. Neither was a significant difference found between Solenopsis invicta and S. geminata. These two groups were significantly different at the 5% level using Duncan's multiple range test. From the same study the following generalizations were made: (1) worker ants have a slightly lower supercooling point than reproductives, (2) within a given species, immature ants have lower supercooling temperatures than adults, and (3) pupae have lower supercooling temperatures than larvae. The effects of acclimation on the supercooling temperature of Solenopsis invicta were observed by maintaining major, medium, and minor workers at each of three temperatures: 12^{o}, 22^{o}, and 32^{o} C. Analysis of variance revealed no significant differences among treatments ($F_{(2,81)} = 1.19$, $p>0.05$), nor in the interactions between castes and treatments ($F_{(4,81)} = 2.11$, $p>0.05$), a laboratory colony previously removed from the field locality. The mean supercooling points ± standard errors for minor workers are shown in Fig. 1. The explanation for the large standard errors is not immediately apparent. Diet may be a factor; the laboratory colony was supplied with cockroaches, mealworms, and water. Temperature and photoperiod may also affect the supercooling point. The laboratory colony was kept in complete darkness at a constant 22^{o} C, whereas the field colonies experienced normal photoperiods and fluctuating temperatures. These comparisons are interesting; but because the primary goal of this study was to observe the behavior of mean supercooling points in the field colonies over time, no further investigation of the laboratory colonies was pursued.

In the present study of field colonies we found no overall significant differences between mean supercooling points from the three colonies using a one-way analysis of variance ($F_{(2,297)} = 0.45$). Analysis of covariance confirmed this but revealed significant differences among samples taken throughout the season ($F_{(1,294)} = 115.38$). The mean supercooling temperature ± standard error for each sampling period is shown in Fig. 2. Note the small standard errors which were recorded. The data seem to show oscillatory changes in the supercooling points over the eighteen week period, and there does not appear to be a uniform trend of acclimatization in minor workers of *Solenopsis invicta*. It is important to note that the maximum mean supercooling temperature difference between samples is less than 2 C^{o}.

In summary the following points are important to retain: (1) supercooling abilities of the red imported fire ant are altered by conditions in the laboratory, (2) differences between supercooling abilities of ants in the field were noted among samples, but not among colonies, (3) differences between sampling periods might not be due to acclimatization, and (4) the mean supercooling points for field colonies varied less than 2 C^{o} from October through February, but never fell below -6^{o} C.

Recent soil temperature measurements obtained in the vicinity of the field colonies indicate that the temperature at 30 cm between January 6 and February 20 never fell below 4^{o} C. If the supercooling temperature of Solenopsis invicta is the primary measure of cold-hardiness, such a soil temperature would cause little mortality.

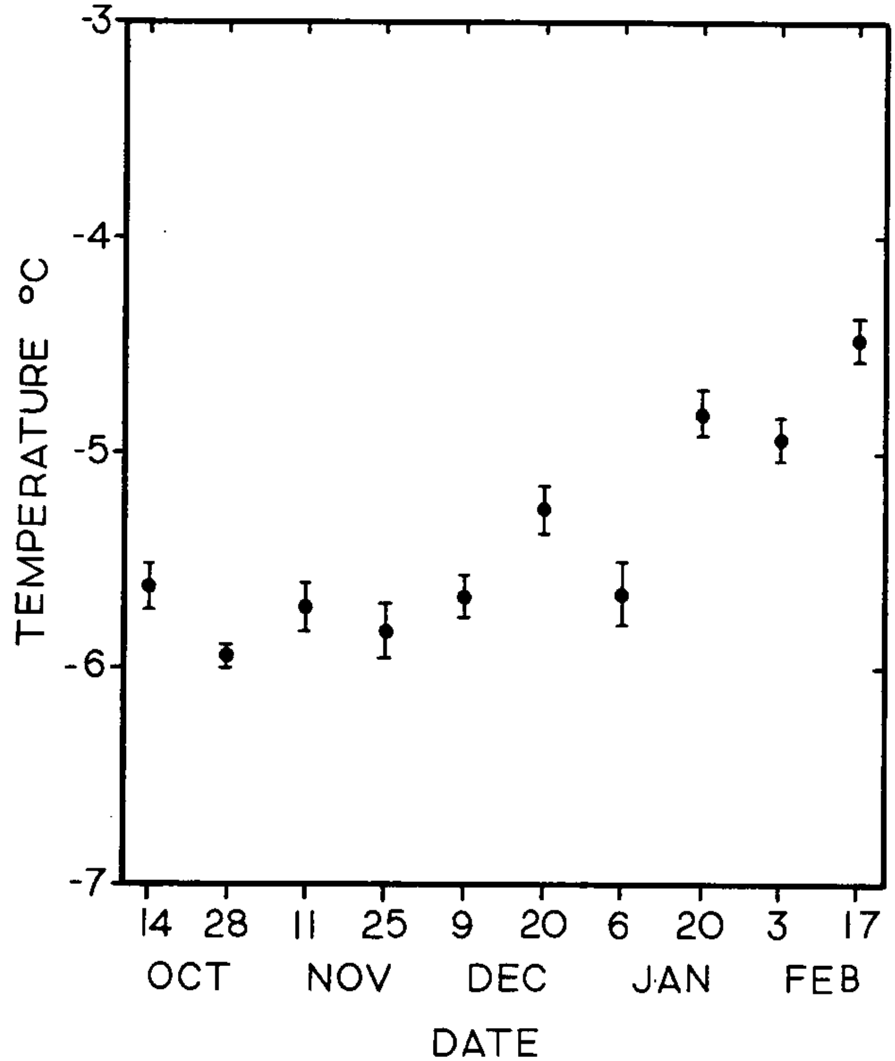


Fig. 1 - Mean supercooling points ± standard errors of *Solenopsis invicta* in field samples from Lubbock, Texas.

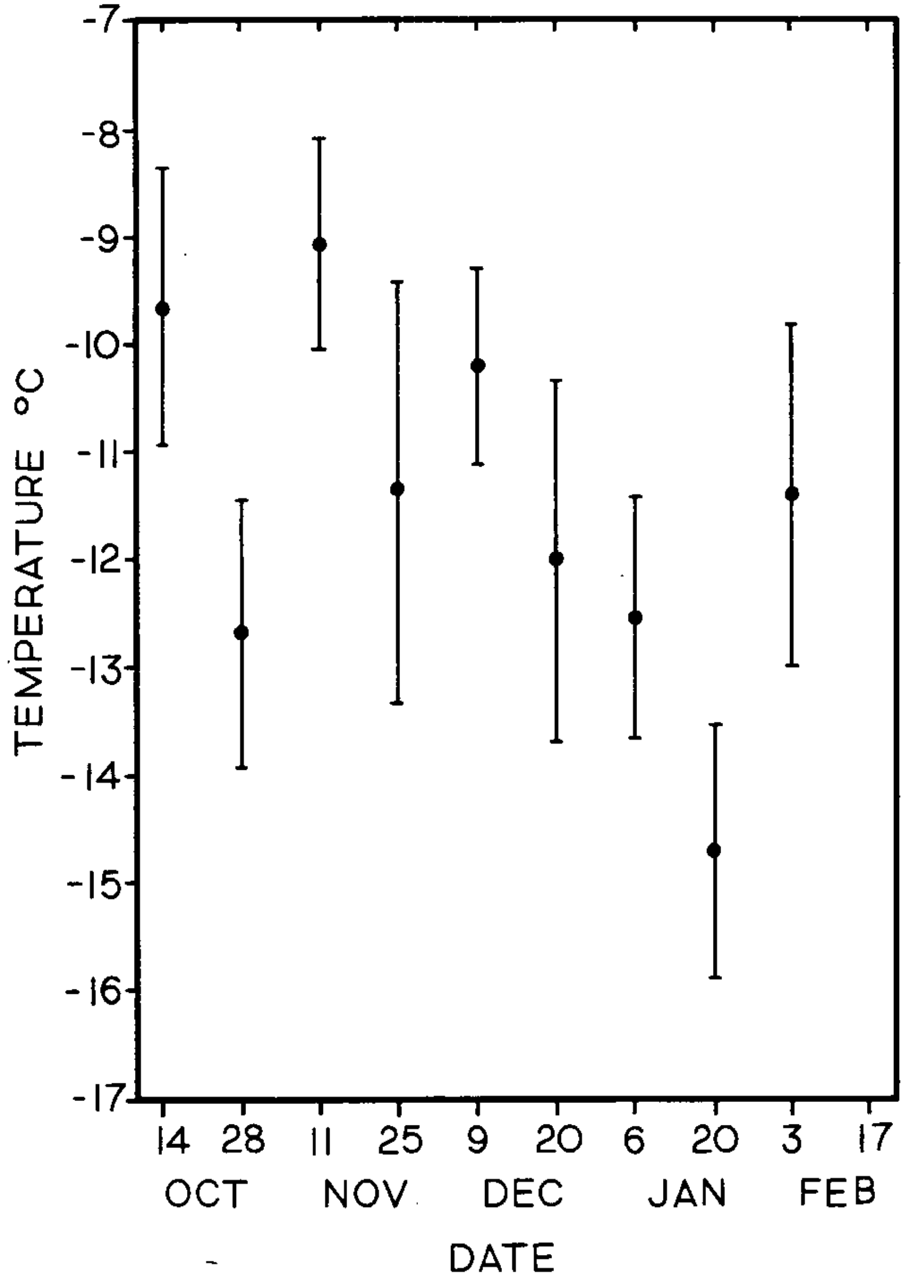


Fig. 2 - Mean supercooling points ± standard errors of *Solenopsis invicta* from laboratory maintained colony from Lubbock, Texas.

MOVEMENT OF MARKED ANTS BETWEEN THE FIRE ANT COLONIES IN A TEXAS PASTURE HABITAT

Awinash P. Bhatkar
Agricultural Experiment Station
Department of Entomology
Texas A&M University
College Station, Texas 77843

A technique was developed to mass-mark ants in the field, to study the traffic of workers between the mounds in a pasture habitat of the Texas A&M University Research Farm. Two 3/4 acre transects of monogynous and polygynous colonies were selected and mapped during May-June and August, 1985. The transect of polygynous colonies contained 368 mounds/acre while the monogynous transect contained 62 mounds/acre during the summer. The dispersion pattern of the mounds in 3 of 4 sectors of 0.2 acre central section of the polygynous transect was regular [Clark & Evans (1954) $R < 1$] and in 1 sector, it was random, similar to that of the transect of monogynous colonies. The pattern seems to be seasonal and R provides an useful index of the carrying capacity of the habitat. The 10,000-20,000 ants of 5-7 mounds in each transect were marked with pretested adhesive paints (Bhatkar & Vinson, in preparation). The worker movement was monitored by sampling ants from all the marked and surrounding colonies. The monogynous colonies did not show worker movement and were self-contained, monodomous colonies. Polygynous colonies showed the movement of workers to most of the neighboring mounds, such that there was a linear relation between the ratios: marked to total ants sampled, from the marked mounds during a consecutive 5 day sampling program (regression coeff. $r = 0.90-0.98$). Similar linear regression was observed when the data for all the mounds in the transect studied was pooled. The marking was selectively done, such that the random samples had ants of a certain ethological caste that appeared in a steady state with the others collected. Polygynous colonies were proved to be a polydomous supercolony, polycalic in nature, that may be limited by the dispersion of monodomous colonies in the pasture.

Research Highlights for 1985

B.Michael Glancey
USDA,ARS, Imported Fire Ant Project
Gainesville, Fl

We once did a study that showed that ants could not ingest particles larger than .8 microns. Every now and then, something comes along that reminds you that nothing is written in granite.

Case in point: we were collecting queens this past year and dissecting out the poison sacs for the queen recognition pheromone. During one of the dissections, we discovered in the crop of one of the queens some particles that were 100-200 microns in size. Here's a slide of the crop and a close up. There are about 10 of the large particles plus a mass of material in the distal part of the crop. The particles are not parasites, pathogens or spores, but they are particulate. It is quite possible that they are an accretion of crystals that all solidified to the same size. Since the crop is only a storage organ and not a site of synthesis or metabolism, this kind of picture is difficult to understand. If any of you have seen anything like this in your own researches, I'd like to talk to you about it.

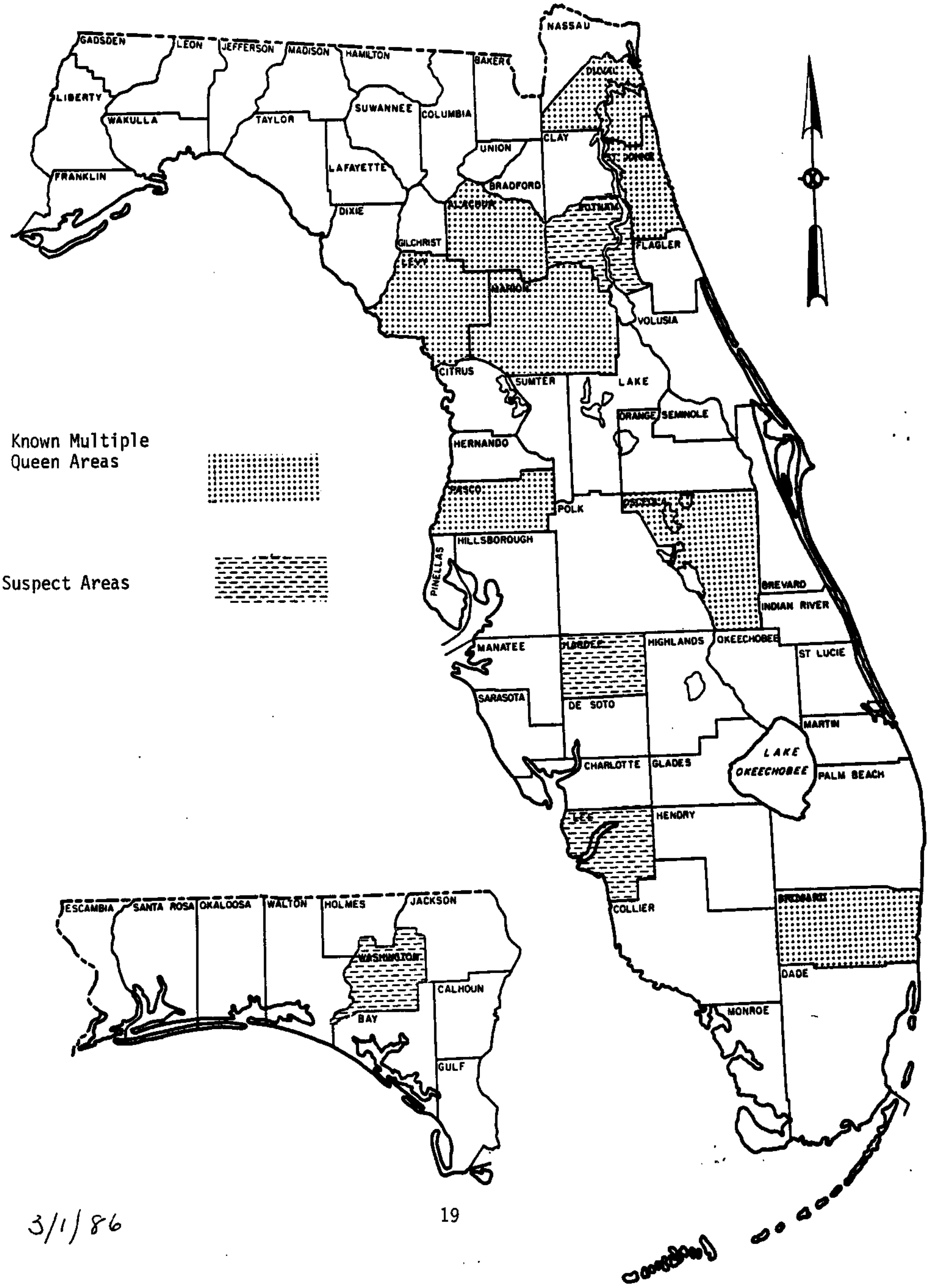
The Multiple Queen Problem

Another monster which we are paying close attention to is of course the problem of the spread of multiple queen colonies in the state of Florida.

In 1984, the polygynous condition was first found by Williams and Lofgren in Marion County (Ocala) . From this first reported finding 2 years ago, we can now report that the polygynous condition has been positively found in 8 other counties and is suspected to occur in another 4 counties.(Slide).

Some recent reports by the Florida Department of Agriculture, Division of Plant Industry, indicate that in areas where control failures with Amdro bait have occurred, the populations were found to be of the polygynous type. This has happened in at least 3 cases in various parts of the state. A very serious aspect of the problem is that once single-queen colonies have been eliminated from an area consisting of mixed populations, the surviving polygynous colonies are free to expand without any competitive restraints.

One point which may interest you is that in an area of Alachua county which was recently found to be in the polygynous condition, 9 queens were removed from a single mound. Other queens were left with the parent mound. When the queens were weighed before dissection, it was discovered that the range of weights went from 13mg to 19.5 mg. In addition, the workers did not exihibt the uniformity of size usually associated with the multiple queen condition. At this time we do not know if this represents the beginning of the multiple queen condition or the end of one in this particular area.



NASSAU
GADSDEN
LEON
JEFFERSON
MADISON
HAMILTON
BAKER
DUVAL
LIBERTY
WAKULLA
TAYLOR
SUWANNEE
COLUMBIA
CLAY
UNION
FRANKLIN
LAFAYETTE
BRADFORD
ALACHUA
DIXIE
GILCHRIST
FLAGLER
LEVY
MARION
VOLUSIA
CITRUS
SUMTER
LAKE
ORANGE
SEMINOLE
HERNANDO
PASCO
POLK
HILLSBOROUGH
PINELLAS
BREVARD
INDIAN RIVER
MANATEE
HIGHLANDS
OKEECHOBEE
ST LUCIE
SARASOTA
DE SOTO
MARTIN
LAKE OKEECHOBEE
CHARLOTTE
GLADES
PALM BEACH
LEE
HENDRY
COLLIER
DADE
MONROE
ESCAMBIA
SANTA ROSA
OKALOOSA
WALTON
HOLMES
JACKSON
WASHINGTON
CALHOUN
BAY
GULF
Known Multiple Queen Areas
Suspect Areas

3/1/86

Effect of Logic Upon the Queen

The Environmental Protection Agency granted registration in late 1985 to a new bait called Logic for control of the red and black imported fire ants, Solenopsis invicta Buren and S. richteri Forel. The active ingredient in Logic is an insect growth regulator, fenoxycarb (Ethyl [2-(p-phenoxyphenoxy)]carbamate)(Maag Agrochemicals RO 13-5223). Fenoxycarb is one of a group of chemicals synthesized with a carbamate moeity that has shown juvenile hormone activity against a variety of insects [1,2]. The compound has shown excellent activity against the red imported fire ant in the laboratory and field [3,4]. Although the exact mode of action of the chemical against fire ants is unknown, 3 factors given by Banks et al. [4] contribute to its efficacy, (1) direct toxicity upon existent brood or disruption of the life processes of developing larvae or pupae, (2) drastic reduction or cessation of egg production by the colony queen, and (3) a shift in caste differentiation from worker to sexual form.

Our present study involves the question of whether suppression of egg production is due to direct effects of fenoxycarb on the reproductive system of the queen or to other extrinsic factors impinging on the queen.

Materials and Methods

Twenty-seven laboratory colonies of red imported fire ants, consisting of a queen, 20 to 30 ml of immatures, and 40-60,000 worker ants, were each fed 0.5 ml of once-refined soybean oil containing 2.0% fenoxycarb (10 mg/colony active ingredient). Three additional colonies of comparable size were used as untreated controls and were given an equal volume of neat soybean oil. The oil solution was administered in micropipets from which the ants drank. Ingestion of the oil solution was complete within 24 hr after which the ants were returned to the normal laboratory diet [5]. The colonies were maintained in the laboratory at 27 $\pm$ 2 C.

At weekly intervals through 8 wk the queens from 3 treated colonies were sacrificed, the ovaries removed and observed, and the ovaries fixed in Kahle's fixitive. After fixation, the ovaries were embedded in paraffin (mp:57 C). The tissue was sectioned at 5um and stained with Harris's hemotoxylin and eosin. Three normal colony queens were also sacrificed and fixed to serve as controls. The remaining 3 colonies were held for observation of treatment effects on the colonies.

Results

The ovarioles of queens of the red imported ant are considered to be the merioistic type(=polytrophic)[6]. In this type of ovariole, each follicle contains a number of nurse cells together with one oocyte. Generally, the nurse cells and oocyte are derived from one oogonium. The many ovarioles (60 +) are tapered at the proximal end and all are attached to the body wall by means of the terminal filament.

The germarium(Slide 1) is an area of the ovariole that contains the oogonia which differentiate into the primary oocyte and the nurse cells(trophocytes) . These areas of the ovarioles are packed with heavily stained nuclei indicating an intense region of mitotic activity. The older developing oocyte has the nurse cells attached to the apical end and the entire mass is surrounded by the follicular epithelium.

As the oocyte ages, it moves down the ovariole to the area of differentiation (upper vitellogenic). In this area the nurse cells accompanying the oocyte continually deposit cytoplasm around the oocyte (Slide 2). This constant deposition of cytoplasm causes an enlargement and as you view the ovariole, you see an increasing egg size as you look distally toward the oviduct(Slide 3_). As the egg enters the lower vitellogenic zone, spheres of yolk are added to the cytoplasm and the nurse cells become squashed between the enlarging eggs (Slide 4). Eventually, the nurse cells disappear completely(Slide 5) .

There may be up to 7-10 very large heavily yolked eggs in the lower vitellogenic zone waiting for the moment of deposition. It is in this area that the chorion is laid down. There are another 10-20 smaller eggs above the yolked eggs, with some yolk spheres deposited within the cytoplasm. In sum, between the yolked eggs, the smaller eggs and the developing eggs, the queen may have at any given moment, about 70-80 eggs within a given ovariole.

Under gross examination, the ovaries show the typical string of pearls effect with the ovarioles containing the various sized eggs. The ovaries are quite large and fill the entire abdominal cavity.

The ovaries of queens subjected to fenoxycarb for only one week do not show any gross or microscopic signs of pathology. The nurse cells are quite obvious, there is still a zone of differentiation, lots of cytoplasm, lots of yolk and many eggs ready for deposition.

At 2 weeks after treatment with fenoxycarb, the first obvious differences can be seen microscopically. The proximal end of the ovariole shows the lack of a zone of differentiation. There is very little cytoplasm surrounding the oocyte and only a small amount of cytoplasm within the nurse cells. There are fewer yolked eggs ready for depostion and there is a reduced number of oocytes moving down the ovariole. There are some eggs in the ovarioles, but they are developing with a reduced number of nurse cells, or without the nurse cells completely. Some of the ovarioles show large areas which are vacuolated, with the absence of any developing eggs. All of these conditions are not present in all of the ovarioles at the same time or in all the ovaries examined. Some ovarioles show the damage while others appear quite normal.

Three weeks after treatment, gross examination shows the first effects of the chemical. The individual ovarioles appear to contain fewer eggs and the proximal end takes on the appearance of a thin piece of spaghetti. The ovaries are much reduced in size and do not fill the abdominal cavity. Microscopically , only about 5 eggs in each ovariole seem to have yolk deposited in them and the amount of yolk deposited seems to have been reduced. In the zone of differentiation, there is no normal sequence of nurse cells, oocyte, nurse cells, oocyte, etc. Structural intergrity seems to be lost in the germarial end. There is just a gimish of tissues which are undifferentiated. All that can be seen is a series of egg nuclei in a row, without cytoplasm, and without the accompanying nurse cells.

By the fourth week, only 1 or 2 yolk eggs are present in the ovarioles. And these are much reduced in size. The zone of differentiation is just about gone, only egg nuclei are present and these show a lack of cytoplasm surrounding them as well as a lack of cytoplasm in the follicular epithelial cells. There appears to be a lack of trophocytes and its covering of follicular epipthelium.

The ovarioles ~~from weeks~~ 5 through 8 show a continual reduction in the number of yolk eggs present until there is only one or two small eggs in the entire ovary. Normal eggs which would measure 50 X 25 units are now reduced in size to eggs of a 20 X 10 unit size. There has been a continual shrinkage of the ovariole so that in gross examination the entire ovary occupies an area of just one abdominal segment. Some ovarioles have been reduced to hollow tubes, some merely to strings of tissue. Some ovarioles show the chorion present, but no eggs. Examples of the effect at 2-8 weeks are seen in Slides 5-20.

Discussion

In the treatment of colonies with fenoxycarb, the ensuing death of a colony can be attributed to a number of interrelated effects. The drastic mortality of the brood results in a greatly reduced worker population . Such reductions in the worker caste are reflected in lower rates of food gathering, reduced brood tending and queen tending, and finally reduced colony maintainanence. All of these reductions can lead to the death of the queens and eventually to the death of the colony itself.

The effect of fenoxycarb upon the reproductive system is very similiar to that reported for the effect of Altosid (a juvenile hormone analogue) on the ovaries of the pharaoh's ant, Monomorium pharaonis (L.). Gross examination made of this ant's ovaries showed ovarioles reduced in size and containing no developing oocytes or trophocytes[7]. No microscopic examination of the ovarioles was made over the period of the experiment(30wks). At the microscopic level, the effect of Logic is very apparent. It is possible to photograph only one or 2 mature eggs from the ovary of an untreated queen. No other structures can be seen in the ocular because of the great size of the ovary. The ovaries of a treated queen at 8 weeks are so small and stringy that the entire ovary can be seen with the ocular.

Fenoxycarb appears to have a sequence of effects upon oogenesis. Firstly, instead of a normal area of differentiation into oocyte, nurse cells and follicular cell, there appears an area where discrimination is lost. Since control of differentiation is assumed to be in the corpora allatum [8], one may infer that one of the modes of action could be the reduction in the output of hormones by the CA. Secondly, there is a failure of nurse cells to develop and thus a failure to nourish the developing oocyte. This effect upon the nurse cells points again to an effect on the CA and/or the neurosecretory cells [9]. The nurse cell not only fail to add cytoplasm to the existing egg, but, in later stages, they fail to add the necessary yolk protein globules to the egg. Since the control of yolk synthesis is indirectly under the control of the CA[9], one again must suspect that the mode of action is at the hormonal level. Tests with chemosterilants and insecticides have shown that neurosecretory cells can be interfered with; however the mechanism of this interference is unknown. Riviere[10] found that vitellogenesis in the roach Blatella germanica could be prevented by the application of the insecticide propoxur. The reduction in vitellogenesis in the fire ant can be so total that the eggs are composed solely of the chorionic membrane.

The final sequence in the ant appears to be the total reabsorption of the ovariolar tissues until there remains only very thin strands of tissue. It is unknown at this time whether such a process of resorbtion in ants is under the control of the CA.

One other piece of data points to fenoxycarb affecting the CA/neurosecretory cells. Although a treated queen may not be producing any viable eggs and her ovaries may be reduced to very thin structures, she is capable of being restored to a viable egg laying queen simply by the removal from the treated workers. Banks[11] has shown that the replete workers store the IGR and thus serve as a reservoir for the constant introduction of the chemical. If the treated queen can be removed to a "chemically clean" colony, viable egg laying can be restored. This is not to imply that this occurs in all cases. But it does point out that the effect of fenoxycarb is not on the germ tissues per se, but rather, its effects are upon another dynamic metabolic system.

A Naturally Occurring Teratology in RIFA

Two aberrant forms of the red imported fire ant *Solenopsis invicta* Buren have been reported in the literature. The first was a gyandromorph (an individual having both male and female characteristics) found by Hung(1975) and the second, an intercaste (an individual with characteristics intermediate between a worker and a female alate) by Glancey et al.(1980). We now report the finding of a new morphological form which was apparently caused by environmental factors.

In October 1984, some field colonies were collected along State Road 100, about 2 miles south of Starke, Florida. The colonies were separated from the soil using our standard drip technique (Banks et al. 1981) and placed in rearing cells. While

we were collecting brood from the cells, we noticed some unusual pupae in one of the colonies. Examination of the pupae revealed that the forms were quite unlike anything we had seen before. The antennae and tarsi of these forms were completely deformed.

In a normal pupae, the antennal scape extends outward at an angle of 45° from the mid-line of the frons past the insertion of the mandible. The funiculus is bent back at a 45° angle so that the tips of the antennae approach each other. In the deformed pupa, the scapes were close together and extended along the mid-line of the frons. The funiculus was bent back at an angle of 90° toward the mid-line.

A total of 31 aberrant pupae were collected from the entire colony, representing about 2 percent of the total number of brood collected. The pupae were segregated along with five normal brood tenders to care for them. Fifteen of the pupae successfully eclosed while the remainder died.

The pupal deformities were also expressed in the newly eclosed workers. The antennae maintained the 90° bend of the funiculus, even after eclosion and, because of the antennal bend, the ants had difficulty in touching the substrate with the tips of the antenna. The femurs of the first legs were shortened and the tarsi of the second and third legs appeared to have two or three full twists, resulting in a spiral condition. It was extremely difficult for the ants to walk on these deformed legs.

Bioassays were conducted to determine the behavior of the deformed ants toward trails, brood and the queen.. A trail was streaked using 0.25 worker equivalents of a hexane extract of the Dufour's gland. The deformed ants were able to move along the trail by pulling themselves along with their spiral tarsi. Contact with the trail pheromone was maintained by the ants bending their heads down to allow the antennal tips to touch the trail. The ants were tested for brood-tending response by allowing them to contact pieces of brood taken from their own nest. The deformed ants did not respond at all to the brood, whereas normal siblings responded in 4 to 6 seconds. When the deformed workers were exposed to their mother queen, they neither groomed nor fed her.

Dissections of the deformed ants showed no deformities of the crops, esophagii, or intestines.

According to Berndt and Wisniewski(1984), deformities such as we found in RIFA, are easily induced in the Pharaoh's ant by altering the humidity. They demonstrated several years ago that low temperatures had no effect on the development of deformities. They were able to induce spiral legs and deformed antennae if they subjected prepupae to RH's of 30-50% under laboratory conditions and if they excluded any workers or young larvae from the experiment.

The eclosing workers were found to have only the second and third pair of legs affected by the humidity. Berndt and Wisniewski(1984) also state that the natural occurrence of such deformities is extremely rare. They cite two reasons for this, 1) brood tenders seek optimum environmental conditions within the nest for rearing the young, and 2) deformed larvae or pupae are usually removed. Our observations differ from these in that we did find the deformed ants under natural conditions, all 3 pairs of legs were affected, and the pupae were found with other workers and brood.

Berndt's and Wisniewski's explain the deformities by concluding that final development of both the antennae and tarsi occurs during a very narrow window of time,usually 20-30 min(under laboratory conditions). Under stress conditions of low RH, fluid which normally would be used to extend and harden the extremities is lost to the dryness of the environment. Lacking the needed fluid, the cuticle then collapses back upon itself into the so-called "deformities". It is difficult to imagine that under natural conditions workers would place prepupae in a separate part of the nest in which there was very low humidity. It is equally difficult to imagine that the workers forgot to move a set of the prepupae from an area of dryness. If Berndt and Wisniewski are correct, then exposure of the pupae to the low

RH must have occurred at some time when the mound was disturbed. Since the mounds were located along a roadside, this could happen if they were impacted by moving equipment or moving vehicles.

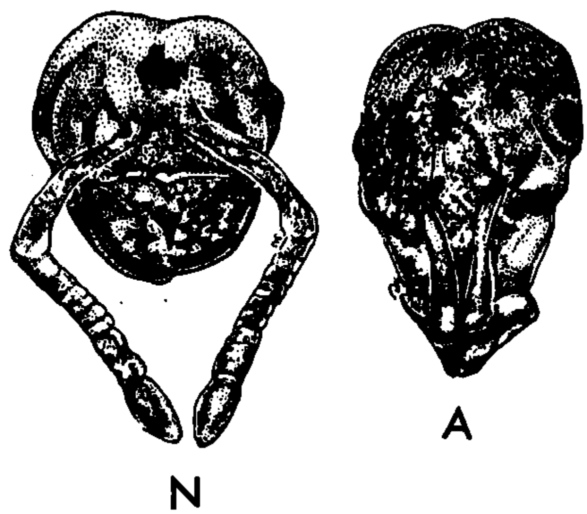


Figure 1. Drawing of a normal (N) and abnormal worker pupa showing the relative positions of the antennae.

Figure 2. Drawing of the antennae of an eclosed normal (N) and abnormal (A) worker ant. The abnormal antennae extended straight out from the frons,could not be bent or flexed, and were permanently held parallel to the floor of the nest.

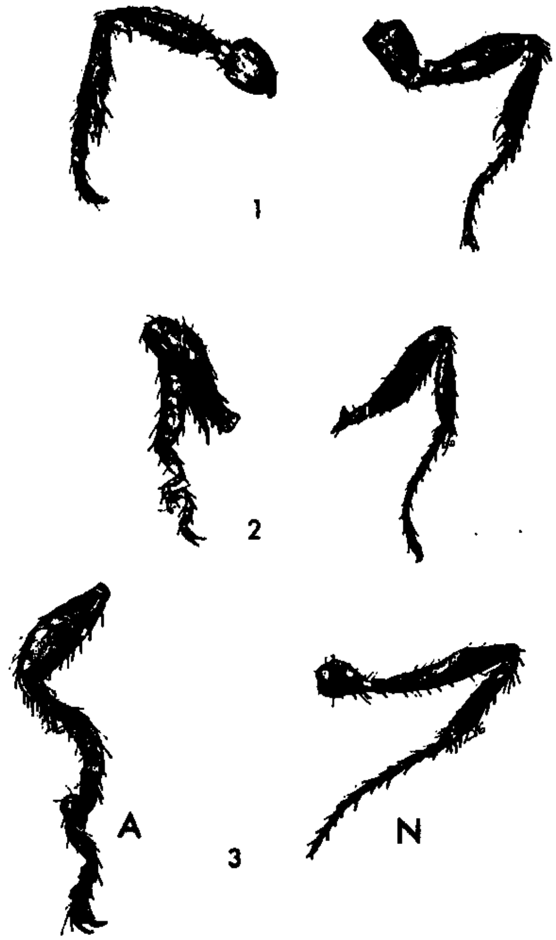


Figure 3. Drawing of the legs of eclosed normal (N) and abnormal (A) worker ants. The twisted condition of the 2nd and 3rd pair of legs prevented the ants from normal locomotion.

Status of the Search for Natural Control Agents in South America[a]

Daniel P. Wojcik

USDA-ARS
Insects Affecting Man and Animals
Research Laboratory
P.O. Box 14565
Gainesville, Florida 32604

The information presented here is the result of the combined efforts of Mr. Antonio C.C. Pereira, our resident Entomologist in Caceres, Mato Grosso, Brazil, Dr. Donald P. Jouvenaz, Mr. W. A. Banks, Dr. David F. Williams, Dr. Cliff S. Lofgren, and myself. These data are complete as of 24 March 1986 when my recent trip to Brazil terminated. The distribution of the fire ant species in South America as shown on a map by Buren et al. (1974) is still generally accepted. However, the recent identification of *Solenopsis invicta* from the Sao Paulo area of Brazil (J.C. Trager, Univ. Florida, unpublished) may radically alter our concepts of fire ant distribution in South America. The taxonomic studies of Trager (unpublished) and the finding of hybrid crosses of between *S. invicta* and *S. richteri* in the United States (Vander Meer et al. 1985) will probably also alter our concepts of fire ant taxonomy and distribution in South America.

In February 1984, as result of a cooperative agreement between the USDA-ARS and EMBRAPA (Empresa Brasileira de Pesquisa

[a]This paper presents the results of preliminary research only and will be published later through established channels with appropriate editing. This paper and data should not be referred to in literature citations until they appear in press.

Agropecuaria), we established a laboratory at the EMPA-MT (Empresa de Pesquisa Agropecuaria do estado de Mato Gross S/A) Research Station in Caceres, MT, Brazil. The main emphasis of the Brazilian project is to survey for and evaluate the natural biological control agents of fire ants, primarily S. invicta in Brazil. The survey is conducted by examining fire ant nest samples for pathogens and myrmecophiles. Standard size samples of 2-1/2 liters of tumulus excavated from fire ant mounds are placed in buckets. The ants and myrmecophiles are separated from the soil by floatation with water. The ants are examined under the microscope for pathogens (Jouvenaz et al. 1977). Myrmecophiles are preliminarily identified, labeled and preserved. Labeled taxonomic samples of all ant colonies examined are routinely preserved.

In addition to the regular natural enemy survey, we have three small studies in progress. The first study consists of 3 plots which are only surveyed for pathogens. Talc-vial samples of ants from each mound in each plot are taken every other month and examined for pathogens. The second study consists of 3 plots in which the ant mounds are examined for pathogens and myrmecophiles every 6 months. These samples are taken with the same methods as the regular survey samples. The third set of 3 plots are for the ecology study which are sampled every month. These plots are sampled for changes in fire ant populations by using mound counts, pitfall traps, and bait stations. These studies will be discussed at future IFA conferences.

To date (April 1986) 1000 fire ant colonies have been processed in the natural enemies survey in Brazil. Most of the colonies were collected within 150 km of Caceres. Numbers of samples were also collected around Cuiaba, along the Transpantaneira highway from Pocone to Porto Jofre in MT, and around Campo Grande, MS. Multiple collections of pathogens and myrmecophiles are common and a single colony may contain several pathogens and/or myrmecophiles. Although the data have not been analyzed statistically, we have thus far found no evidence of a correlation between any pathogens and/or myrmecophiles. When sufficient myrmecophiles are found in a nest sample which also contains a pathogen, samples of the myrmecophiles are also examined for pathogens. These obligate myrmecophile - fire ant host relationships would, of course, constitute an ideal mechanism for alternate host transmission of fire ant diseases.

The totals and percent of each pathogen and myrmecophile collected during the 21 months (July 1984 through March 1986) since the survey was conducted are given in Table 1. Each pathogen or myrmecophile collected during the survey will be discussed briefly.

The microsporidium, Thelohania solenopsae (Protozoa: Microspora), is dimorphic with 2 distinct spore types. The known biology is summarized by Jouvenaz (1986). Both types of spores are found in immature and adult ants. It is not known at this time, if the same species of pathogen is present in all fire ant species in South America as was previously assumed (Jouvenaz et al. 1980), or if sibling species are present (Jouvenaz 1986). We do not know how this disease is transmitted in nature and we have not been able to transmit it in the laboratory. The 2.7% collection rate was lower than expected as previous work had indicated higher levels of this disease (Jouvenaz et al. 1980, Jouvenaz unpublished).

A second undescribed microsporidian species, Vairimorpha sp. (Jouvenaz and Ellis in press), was found in 5.2% of the collections. Attempts to transmit this dimorphic pathogen have not been successful. Both kinds of spores are found in immature and adult ants.

The neogregarine, Mattesia geminata (Protozoa: Neogregarinida), infects S. geminata in Florida and S. invicta and other Solenopsis spp. in Brazil. The known biology is summarized by Jouvenaz (1986). The spores are only found in the immature stages of the ant as the infected individuals die in the pupal stage. Attempts to transmit this species have not been successful. The 6.4% infection rate is the highest ever recorded for this pathogen.

A nematode recently discovered in S. invicta in Mato Grosso (Jouvenaz et al. in press), has only been found in 1.1% of the collections. This nematode, apparently in the family Tetradonematidae (Mermithoidea), was found for the first time in February 1985. Little is known about its biology at this point. Curiously, all of the fire ant colonies containing this nematode have been collected along roadsides.

Several other pathogens including virus-like particles, an undescribed protozoan (probably a neogregarine), a fungus, and a bacterium are known from fire ants in Brazil, but have not been collected during this survey. The information on these are summarized in Jouvenaz (1986).

Beetles of the commonly collected myrmecophilous Scarabaeidae (Coleoptera) of the genus Martinezia were previously placed in the genus Myrmecaphodius (Chalumeau 1983). This genus contains at least 5 species which are present in Mato Grosso. One of these species, Martinezia dutertrei, is present in North America and is known to be predaceous on fire ant brood (Wojcik unpublished). The small numbers of brood eaten and the low numbers of beetles usually found in fire ant nests make the usefulness of this group in a biological control program questionable despite the fact that this group was present in 27.4% of the collections.

The myrmecophilous Histeridae (Coleoptera) collected from fire ant mounds are unidentified. Nothing is known about these common beetles found in 23.1% of the collections.

The myrmecophilous wasps of the genus Orasema, family Eucharitidae (Hymenoptera), were very common being found in 47.6% of the collections. Table 2 lists, by month, the percent of colonies collected which contained this parasite. Although the average number of specimens collected were less than 20 per nest, one colony contained 634 Orasema sp. larvae, pupae, and adults. The biology of this species has not been studied, but is assumed to be similar to the other species which have been studied (Williams 1980). The females lay their eggs in plant tissue, apparently near flowers. After the eggs hatch, the planidia (first instar larvae) are phoretic on worker ants. In the nest, the planidia leave the worker and attach to an ant larvae or pupae. The ant pupae never become adults as a result of feeding by these ectoparasites. The parasites are tended and protected by the host ants. The problem with using this parasite in a biological control program is that it lays its eggs in plant tissue and can cause cosmetic damage to fruit. This information is summarized by Wojcik (1986).

The workerless obligate parasitic ant, Solenopsis (Labauchena) sp. (Hymenoptera: Formicidae), was collected once during the present project in Brazil. We had collected this group previously from only 2 nest collections, once in 1974 and once in 1975 (Banks et al. 1985). The fire ant colony containing this parasite was collected near Campo Grande in March 1986. The host colony was identified as S. invicta by Dr. James Trager and confirmed by Dr. Robert Vander Meer using gas chromatography for cuticular hydrocarbons and venom alkaloids. The colony containing the parasitic ants was brought to Gainesville, Fl under USDA-APHIS permit and is currently in maximum security in the Quarantine Laboratory in Gainesville. This is the first confirmed collection of this group of parasites from S. invicta. What is known about this group is summarized by Williams (1980).

Myrmecophilous Thysanura were collected from only 1.9% of the ant nests. These unidentified Thysanura are similar to the species found in fire ants nests in the United States. The North American species is known to be predacous on ant brood (Wojcik unpublished). These insects are probably under represented in our samples because the ants are highly antagonistic towards the Thysanura and they do not survive the floating procedure well when large amounts of ants are present.

Myrmecophilous millipedes (Diplopoda) were collected from 16.2% of the fire ant nests in Brazil. A similar species is found in Florida and Georgia (Naves and Wojcik in manuscript). The species in the United States is a scavenger.

Another group which is associated with fire ants in South America are flies of the family Phoridae (Diptera). Species of the genera, Pseudacteon and Apodicrania, are apparently endoparasitic on fire ants (Williams 1980). One phorid puparium was dissected out of an alate S. invicta female (Wojcik et al. in press). This collection is not included in the data in Table 1, as we do not routinely dissect ants as part of the survey. The delicate morphology of the flies and the extreme antagonism of the ants to the flies makes it unlikely that these flies would be found in floated nest samples. These flies can readily be collected hovering over worker ants attracted to baits. In fact, these flies are the only myrmecophile of fire ants which can be collected readily without using floatation to separate samples from fire ant nests.

A great variety of other myrmecophiles are known from fire ant nests in South America but have not been collected during the present study. These other groups of arthropods are listed in the review by Wojcik (1986).

In the coming year we will continue the pathogen and myrmecophile survey and the other studies already in progress. We will emphasize the collection and colonization of the nematode and the Labauchena to study these organisms for possible introduction into the United States. The ultimate goal of our South American project is to establish, in the United States, a complex of specific natural enemies as a biological control component of an integrated pest management program for fire ants. Hopefully, these introduced natural enemies would exert continuing stress on Imported Fire Ant populations and reduce the necessity for chemical control measures in some areas.

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Table 1. Summary of collection records for pathogens and myrmecophiles found in fire ant nests in Mato Grosso and Mato Grosso do Sul, Brazil, July 1984 through March 1986.

Organism	Number of fire ant nests with organism	% of total fire ant nests with organism
Pathogen		
Thelohania	27	2.7%
Vairimorpha	52	5.2
Mattesia	64	6.4
Nematode	11	1.1
Myrmecophile		
Scarabaeidae	274	27.4
Histeridae	231	23.1
Eucharitidae	476	47.6
Formicidae	1	0.1
Thysanura	19	1.9
Diplopoda	162	16.2
Total Fire ant colonies	1000	100.0

Table 2. Percent of fire ant colonies which contained _Orasema_ spp. ectoparasitic wasps (collections made from July 1984 through March 1986).

January	34%	July	39%
February	56	August	36
March	58	September	32
April	56	October	28
May	42	November	38
June	25	December	32

Status of Research on the Economic Impact of Imported Fire Ants on Citrus, Potatoes, and Soybeans

C. S. Lofgren, C. T. Adams, and W. A. Banks

USDA-ARS
Insects Affecting Man and Animals Research Laboratory
P.O. Box 14565
Gainesville, Florida 32604

Studies of the impact of foraging by the red imported fire ant (RIFA), *Solenopsis invicta*, on citrus, potatoes, and soybeans have been conducted over the past 10 years. In citrus, the ants often build their nest around the base of young citrus trees (1-3 yrs old). They are attracted to the sap of the trees for food and chew off portions of the bark, often times girdling the tree and thereby killing it. They have also been observed clipping off the tips of new growth and feeding on flowers or developing fruit. To evaluate the impact of this foraging, we conducted a series of tests in a 30-acre grove near Titusville, Florida. The grove was divided into 3 10-acre plots that were planted with navel or Hamlin oranges or pink grapefruit. Each of these areas was divided in half; one-half was treated twice-a-year with Logic® for RIFA control while the other was left untreated. Ant density was monitored with pitfall traps and bait traps. Tree mortality was determined in October 1984 and 1985 of the 2-year test. Costs of replacing dead trees was obtained from a citrus economist at the University of Florida. The data are presented in Tables 1 and 2.

Ant collection data showed that Logic was effective in suppressing and maintaining low populations of RIFA. The mean monthly population varied from block to block and from year to year; however, the differences between treated and untreated plots were highly significant in both years.

Statistical comparison of tree loss data with Duncan's multiple range test showed that there is a strong correlation ($p = 0.0005$) between ant population levels and mortality of citrus trees. Tree mortality was consistently low in the plots treated for RIFA suppression whereas mortality in the untreated plots was significantly higher. Further evidence of the relationship between ant populations and tree mortality was obtained in 1985 when tree mortality for both Hamlin orange and pink grapefruit was higher than in 1984, which corresponded with increased RIFA populations in both treated and untreated areas.

Tree losses in both 1984 and 1985 were higher in Hamlin orange and pink grapefruit than in navel orange, suggesting that navel oranges may be less susceptible to damage by RIFA.

Replacement costs for the Logic-treated plots ranged from $79.50 to $264.75 compared to $250.25 to $1,036.60 for the untreated controls.

Table 1. Correlation of IFA density with die off and replacement costs of 1 to 4-year-old citrus trees (1984).

Ant index	No. of trees	% die off	Reset cost
	Navel		
57.3	484	0.6	$ 79.50
459.0	470	4.3	385.00
	Hamlin		
38.5	411	0.7	79.50
281.2	434	8.1	567.00
	Pink grapefruit		
71.1	477	0.6	79.50
535.0	451	8.4	648.00

Table 2. Correlation of ant density with die off and replacement costs of 1 to 4-year-old citrus trees (1985).

Ant index	No. of trees	% die off	Reset cost
	Navel		
11.1	484	0.6	$ 79.50
460.3	470	2.8	250.25
	Hamlin		
107.1	411	1.9	189.80
406.0	434	8.8	731.50
	Pink grapefruit		
95.2	477	2.7	264.75
620.6	451	14.2	1036.60

Reports of feeding damage by RIFA on commercial plantings of Irish potatoes were received through the University of Florida Agricultural Research and Extension Center (AREC) at Hastings, FL in late 1982. Wetumpka Fruit Company, one of the largest growers in the Hastings area, reported that one load of potatoes (var. Sebago) had been rejected at the produce market in New York City and a second accepted only after a negotiated price reduction of $1000. In both cases, the potatoes were heavily infested with RIFA. These reports of damage to Irish potatoes from the Hastings area and information derived from reports from the late 1940s and early 1950s prompted us to conduct studies to document the type of damage to Irish potatoes caused by RIFA and to correlate ant populations with its severity.

The studies were conducted on two small plots (10 x 30 m) at the USDA-ARS laboratory, Gainesville, FL and in commercial potato plantings at Hastings, FL. The potatoes were planted in late January or early February of 1983, 1984, and 1985. One of the small plots was maintained essentially ant-free by judicious use of Amdro® fire ant bait while the other was infested with RIFA at the equivalent of ca. 100 mounds/ha. Assessment of RIFA damage to commercial plantings was made in fields managed by the Wetumpka Fruit Company, Hastings, FL in 1984. Two fields, one abandoned by the grower because of ant problems, and the other essentially ant-free, were selected for study. In 1985, additional observations were made in a different field northwest of Hastings. At both sites, ant populations were monitored with bait traps. Potatoes were harvested in May on randomly selected 1-meter sections of row (15 to 25 sites per plot). Potatoes with more than 5 scars or entrance holes from RIFA feeding were considered damaged (unsalable). The data on ant density and damage are recorded in Table 3 and show clearly that loss of salable potatoes (weight) ranged from 12.2 to 26.1%.

A graphic plot of damaged potatoes (% by wt.) versus RIFA infestation (as reflected by ants per bait trap) revealed that there was a direct relationship between percent of potatoes damaged and ants per trap.

Our research on soybeans in relation to RIFA was terminated several years ago (see Adams et al. 1983, J. Econ. Entomol. 76: 1129-1132) when we reported that losses in yield of 6 to 8 bushels per acre were found with ant colony (mound) densities of about 40 to 60 per acre. The primary causes of the differences in yield were attributed to reductions in soybean stand due to RIFA feeding on germinating seeds and lack of other food for the ants at a time of maximum food requirements associated with brood rearing (May-June). A missing link in this research was data on RIFA density in soybean fields across the South. In late 1985, this data was obtained during a survey of soybean fields in 37 counties in 6 states. The data are recorded

Table 3. Density of red imported fire ants in relation to damage to Irish potatoes.

Year	Variety[a]	RIFA-Infested Sample weight (g)	RIFA-Infested % loss in weight of salable potatoes	RIFA-Infested Avg. RIFA workers per trap	RIFA-Free Sample weight (g)	RIFA-Free % loss in weight of salable potatoes	RIFA-Free Avg. RIFA workers per trap	Difference in % loss
Small plot tests (Gainesville, FL)								
1983	S	1588	27.1	205	2020	2.9	10	24.2
	R	1526	26.1	205	2091	0	10	26.1
1984	S	1346	22.3	191	912	1.9	2	20.4
	R	655	22.2	191	658	1.2	2	21.0
1985	S	1327	15.6	151	1489	0	4	15.6
	R	1259	12.2	82	1323	0	4	12.2
Commercial fields (Palatka, FL)								
1984	S	3364	36.6	231	3622	1.9	5	34.7
1985	S	3518	0	36	3323	0	0	0

[a]S=Sebago; R=Russett Centennial.

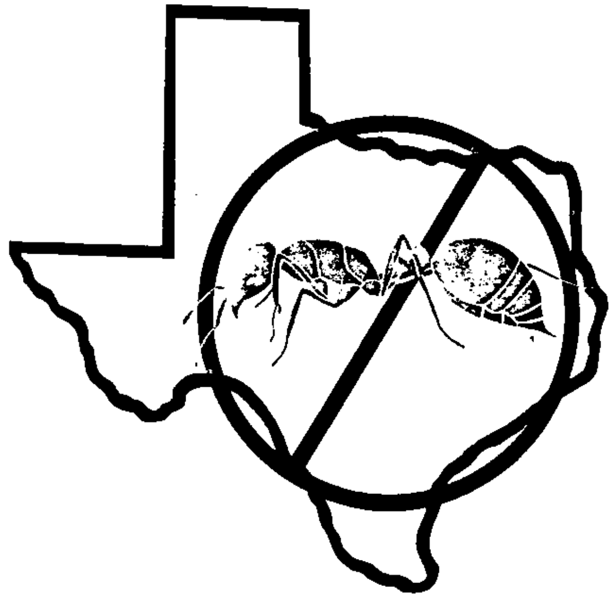
in Table 4 and show that average mound density per field (2 per county) ranged from 34.5 to 52.4. These data confirm that serious losses in yield of soybeans could be occurring throughout the South unless some method is used to control the fire ants.

Table 4. IFA mound densities in soybean fields.

State	Counties surveyed	Mound density/acre
Alabama	10	48.7 (9.0-77.5)
Florida	6	51.1 (38.5-74.5)
Georgia	13	52.4 (29.0-84.0)
Louisiana	1	34.5
Mississippi	5	49.7 (32.5-61.5)
South Carolina	2	46.5 (46.0-47.0)

NOTE: This paper presents the results of preliminary research only and will be published later through established channels with appropriate editing. This paper and data should not be referred to in literature citations until they appear in press.

Answers to Your Questions About Imported Fire Ants



FIRE ANT PROJECT
TEXAS DEPARTMENT OF AGRICULTURE

This brochure was prepared by Dr. A. Ann Sorensen, IPM Specialist, and Mark Trostle, Imported Fire Ant Specialist, of the Texas Department of Agriculture. Some information was provided by Extension Entomologists, Dr. Phil Hamman and Dr. Bastiaan Drees, from the Texas Agricultural Extension Service's publication L-2035 "Fire Ants and Their Control" (1985).

Where can I get more information on imported fire ants?

Contact the Agricultural and Environmental Sciences Program, Texas Department of Agriculture, P. O. Box 12847, Austin, Texas 78711, (512) 463-7540.

The Texas Department of Agriculture (TDA) has initiated an aggressive program aimed at controlling the imported fire ant. Upon request, specially trained TDA inspectors are prepared to give detailed presentations demonstrating the proper methods of treating fire ant mounds. TDA believes this formidable problem can be brought under control, if we all pay more attention to proper treatment procedures.

In addition, pamphlets are being prepared which give individuals detailed information on how to treat various forms of fire ant infestations, whether they are found in the yard, in the garden, in the field or in a greenhouse. Texans plagued with fire ants can now arm themselves with step-by-step instructions which best suit their situation.

TDA continues to make fire ant baits, as well as education and training on their proper uses, accessible to county and city governments so individuals can obtain them at reduced prices. The baits are available to schools, parks and other public facilities free of charge.

TDA currently funds fire ant research programs and works to ensure that treatment programs are regionalized so that infestations in various parts of the state are addressed individually.

We at TDA are committed to an integrated fire ant program in tune with your needs. This brochure gives some of the most commonly asked questions.

1. What is the imported fire ant?

The imported fire ant is similar to ordinary house and garden ants in appearance. It is about 1/8 to 1/4-inch long and reddish-brown to black in color. Colonies are readily identified by the large crusty conical nests or mounds that they build. Nests are usually constructed in open, warm and grassy areas such as pastures, new roads, lawns, unused cropland, and areas with up to 50 mounds per acre are common. The ant is widely known for its vicious sting and aggressive behavior.

2. Why are fire ants a problem?

Fire ants are a problem for many reasons. The primary problem is that they are aggressive and will readily defend their mound. As a result they will sting any intruder. The ants can sting repeatedly, and as they sting, they release a chemical that incites other ants to sting as well. Their sting is painful and results in a pustule which itches for a few days and may leave a scar. Some people are sensitive to fire ant venom and may go into shock after being stung. Small birds and animals may also be harmed by fire ants. In addition to the damage done by their stings, the mounds they build may damage harvesting equipment. The presence of fire ants in crops or gardens may prevent hand picking of fruits and vegetables because of the threat of stings. Imported fire ants may also cause plant damage, either by feeding or by tending aphids. They can also cause problems by damaging electrical equipment, and occasionally their mound building may cause sidewalks or roadways to collapse.

3. Why is a fire ant sting painful?

When a fire ant stings, first it anchors itself to its victim by biting. This causes a pricking sensation. Then the ant pushes its sting into the skin and injects a venom. The ant can sting repeatedly and will often leave a circle of pustules. When venom is injected, an intense burning sensation is felt and the area immediately reddens. Later a pustule forms, the result of injected alkaloids. These alkaloids are toxic to cells, and the resulting dead cells attract the body's defensive white blood cells, which accumulate at the venom site forming pus. The pus irritates the tissues and causes itching. If the skin is broken by scratching, bacteria may enter and an infection may develop. For most people the pustule dries up in a week or so, but for some people the pustule may leave a brown scar that lasts for months. In addition to the alkaloids, fire ant venom also contains a small amount of protein. For people who are sensitive to these proteins, a sting can lead to anaphylactic shock. The symptoms of shock include dizziness, sweating, swelling of the affected area, headache and shortness of breath. Immediate medical attention is required since anaphylactic shock can lead to death. For relief of stings, a simple application of a half-and-half solution of chlorine bleach and water, if used immediately, can reduce the pain, itching and pustule formation. A paste of meat tenderizer and water may also be effective. For relief of itching, products containing cortisone are recommended.

4. How can I avoid getting stung?

Fire ants are most aggressive near their mound, where many workers are available to sting intruders. Avoiding mounds and structures near mounds is the best way to avoid fire ants. If working in an area heavily infested with fire ants, wear a long-sleeved shirt and long pants and pull your socks up over your pant legs. The ants will attempt to sting through the cloth, giving you time to brush them off. Insect repellants sprayed onto the skin or clothing may be temporarily effective. Talcum powder sprinkled around the ankles on the socks or pant legs may deter the ants from climbing up your legs.

5. Where does the fire ant come from?

The imported fire ant comes from the state of Mato Grosso in Brazil, South America. It makes its home there in the Pantanal, the large, flat flood plain of the Paraguay River. It entered the United States sometime between 1933 and 1945 on a boat shipment to Mobile, Alabama, perhaps in the dirt carried for the ship's ballast.

6. Is there more than one kind of fire ant?

There are 5 species (or types) of fire ants in the United States. Most of the publicity has been given to the two imported species, *Solenopsis invicta*, the red imported fire ant and *Solenopsis richteri*, the black imported fire ant. The latter ant is presently confined to Mississippi and Alabama. There are also three native species. *Solenopsis xyloni*, the southern fire ant, is similar to *S. invicta* in its appearance, but nests in open areas are not dome-shaped and soil is thrown from the nest in irregular crater-shaped piles around the entrances. *Solenopsis aurea*, the desert fire ant, is the least common species of fire ant in Texas, occurring in only a few counties. It doesn't form a mound. *Solenopsis geminata*, the tropical fire ant, until the introduction of the imported fire ant, was the most economically important fire ant in Texas. However, *S. invicta* appears to be replacing both *S. geminata* and *S. xyloni* as it moves westward. All of the Texas fire ants sting, but only the imported fire ant leaves a pustule.

7. How are fire ant colonies started?

During the spring and summer months, fire ant colonies start producing reproductives. These are winged males and females that are responsible for starting new colonies. Periodically during these months these reproductives leave the mound, usually following a rain. The males and females mate in the air and the female then seeks a moist, protected area in which to start a new colony. The male dies shortly after mating. When the female lands she digs a small burrow and lays 10-15 eggs. Females will normally land 1/4 to 1 mile away from the mound where they were reared. The new queen tends the eggs and larvae that emerge. Within 20-28 days workers emerge and begin to forage for food. The colony slowly grows and after a year may contain 30,000 workers or more and continues the cycle by producing its own reproductives.

8. How widespread is the imported fire ant in the United States?

The imported fire ant currently infests all of Florida, Alabama and Louisiana, most of Georgia, South Carolina and Mississippi, and part of North Carolina, Arkansas, Texas and Puerto Rico. In Texas, the imported fire ant is found in 113 counties, mainly east of a line running from Jim Wells County in South Texas north to Montague County in North Central Texas. Atascosa, Bandera, Blanco, Comal, Frio, Gillespie, Kendall, Kerr, and Medina counties are also included in the infested areas. All total, more than 230 million acres of land in the United States are infested with fire ants, with over 50 million acres in Texas alone.

9. How do fire ants spread?

The imported fire ant is spreading by four methods: (1) Natural mating flights occur periodically during the late spring, summer and early fall. Female reproductives will commonly fly 1/4 to 1 mile away from the mound in which they were reared to start a new colony, but flights up to 12 miles have been documented; (2) Sometimes mated female reproductives land in trucks, train beds, or in other open containers that are moved long distances; (3) Colonies or mated queens can be inadvertently transported in nursery rootstock, sod or in soil clinging to construction equipment; (4) Whole colonies may be flooded out of their mounds during heavy rains and form ant rafts that float to new locations.

10. How far could the fire ant go?

The natural rate of fire ant spread is 5 to 12 miles per year. Unless an active control and regulatory program is maintained, the fire ant could continue to spread throughout the rest of the South, northward up the East Coast and westward to the Pacific Coast states. The fire ant cannot withstand long periods of dry or cold conditions. Based on current information, the fire ant is expected to someday occupy nearly one fourth of the United States in the areas south of the 10° isotherm (the areas where mean monthly temperatures do not go below 10° F). Mounds are expected to be widely scattered in those areas that receive less than 10 inches of rain per year.

11. What do fire ant mounds look like?

Mature fire ant mounds are normally conical in shape, 1 to 2 feet in diameter and 1 to 1 1/2 feet high. Their shape and size, however, depend largely on soil type. In sandy soils their mounds are flatter and more spread out. In some clay soils they form distinctive conical mounds with a solid crust of soil on the surface. The inside of the mound is filled with tunnels and chambers. These tunnels extend down into the ground as deep as 3-6 feet. Radiating out from the mound are shallow tunnels, 3-4 inches deep, that extend as far as 15-25 yards away. In dry compacted soil with deep cracks, the ants may not build a mound but instead colonize these natural tunnels. While mounds are important, they do not appear to be essential. All that is needed is a small protected space and a source of food and water. Thus the ants may nest in walls of buildings, in logs or under sidewalks.

12. How many ants live in a mound?

The typical fire ant mound contains 80,000 workers, one queen and several hundred reproductives. Mounds can contain up to 300,000 workers. The growth of new colonies can be rapid. After 60 days, workers number about 90. In 90 days they number 225. At 5 months there are over 1,000 workers. After 7 months they contain 6,500 to 14,000 workers, after 1 year they average 11,000 workers, after 1 1/2 years they average 30,000 workers, and after 2 1/2 - 3 years they average from 60-80,000 workers. A small percentage of fire ant mounds contain multiple queens. They are different from the more typical single-queen colony in that their workers are smaller and lighter in color and their mounds are much closer together. Multiple-queen colonies commonly contain 20-60 queens, but some contain 300 or more queens.

13. What do larvae (immature ants) look like?

Immature fire ants are legless, grublike, white and look like tiny rice grains inside the colony. They are helpless and must be fed and tended by the workers (adult ants). They pass through 4 larval stages or instars and shed their skin after each stage. This allows them to grow. Only the fourth instar larvae can digest solid food. All other instars are fed liquids. Adult workers, queens and males can also eat only liquid food. The fourth instar larvae digest the solid food particles brought in by the workers, making the resulting partially-digested liquid available for all of the other colony members.

14. How long do fire ants live?

The larger a fire ant worker, the longer it lives. The smallest fire ant workers are called minims and are produced only in a newly founded colony for the first month or so. They live 20-30 days. The typical small workers in a mature colony are called minors and live 30-60 days. The medium-sized workers are called medias and live 60-90 days. The largest workers are called majors and live 90-180 days. It takes 25-30 days for an egg to pass through the larval and pupal stages and emerge as a worker. Males live 1 to 3 months and die immediately after they fly on a mating flight. Queens live 2-5 years, so the life expectancy of a typical fire ant mound is also 2-5 years.

15. What do fire ants eat?

Fire ants will feed on almost anything. Their primary diet is insects, spiders, earthworms and other small invertebrates. They are attracted to plant sap and will sometimes girdle the stems of small seedlings. They will tend aphids and feed on the honeydew the aphids excrete. Food for the colony is collected by the foraging workers (10-20 percent of that colony) who leave the mound through foraging tunnels that radiate from the mound and are 3-4 inches under the soil surface. A mound usually will have 5-6 such tunnels, which branch repeatedly and extend as far as 15-25 yards from the mound. There are openings to the soil surface at irregular intervals, and a worker will emerge and forage at random until a food source is located. The worker then returns to the tunnel, laying a trail with a chemical it excretes through its sting. Other workers will follow this chemical, establishing a steady stream of ants between the food source and the nest.

16. Can fire ants damage my plants?

Fire ants only recently became a problem on many crops grown in the U.S. They can feed on germinating seeds, causing damage to corn and soybeans. They also feed on the buds and developing fruit of crops such as beans, berries, okra and citrus. They may girdle young trees such as citrus and pecan seedlings in an attempt to find a source of water. Fire ants also tend aphids which, in turn, damage plants through their feeding activities. In home gardens fire ants may damage young plants, especially during droughts.

17. What is the safest way to get rid of fire ants?

This question is a difficult one to answer since all of the methods currently available to control fire ants have an element of risk involved. Any pesticide is dangerous if the user does not first read the label and then **carefully follow the instructions provided.** In this manner, risks can be minimized. Most of the insecticides labeled for fire ant control are toxic to fish (heptachlor, pyrethrins, chlorpyrifos, amidinohydrazone). A few are also toxic to birds and other wildlife (chlorpyrifos). Most are harmful if inhaled or absorbed through the skin (chlorpyrifos, 1,1-trichloroethane, pyrethrin, diazinon). All are harmful if swallowed. The use of boiling water or biological control (mites) has the advantage of not leaving chemical residues, but both methods can be hazardous to the user. If you are not sure of what product to use in a particular situation, contact your local county extension agent or the TDA fire ant specialist in your district. They can tell you which products are appropriate for your needs.

18. Why can't I get rid of fire ants permanently?

Although homeowners can successfully treat all of the mounds on their property, fire ants quickly reappear for a number of reasons. Many mounds may be small or inconspicuous and thus avoid treatment. Mounds in neighboring areas that have not been treated will continue to produce reproductive forms that can fly into treated areas and quickly establish new colonies. A mature colony can produce 4,500 potential new queens during the year, and as many as 97,000 queens are produced per acre each year in infested areas. Although as many as 90-99 percent of these queens are unsuccessful, the remaining queens are more than enough to reinfest treated areas. Widespread treatment with chemicals may result in a more severe infestation since other competing species of ants may be killed along with the fire ants.

19. What kinds of products are available for individual mound treatments?

If you have only a few mounds, individual mound treatments are feasible. These may include mound drenches, granular products, dusts, injectable products and bait formulations. Mound drenches are usually formulated as liquid concentrates and are mixed with water before applying. The solution is sprinkled gently over the mound and may require several days to be effective. Granular products are sprinkled on top and around the mound, followed by an application of water to allow the product to penetrate the mound. Again, several days may be required before the entire colony is killed. Dusts must be evenly distributed over the mound and take about one week to work. Methyl chloroform is a liquid that rapidly volatilizes into a gas. It is poured into a hole drilled into the mound and will immediately kill the ants inside of the mound. Injectable products include pyrethrins and chlorpyrifos. They are sold in special aerosol containers to which a rod can be attached. The rod is repeatedly inserted into the mound and the pesticide injected for about 1 minute total time per mound. Bait formulations consist of processed corn grits coated with insecticide-laced soybean oil. Since soybean oil will quickly become rancid and less attractive to the ants, these products should be used as quickly as possible after opening and stored in a cool dry place. Baits should only be used when the grass and soil are dry and no rain is expected. To treat individual mounds, the bait should be sprinkled 1-3 feet away from the mound when the ants are actively foraging. This can be determined by leaving either tuna fish or peanut butter beside the mound and checking it for fire ant activity. During hot summer days, the worker ants forage during the night and are inactive during the day, so baits should be applied in the evening hours.

20. Are there non-chemical methods available to control fire ants?

There are several non-chemical control methods available to the homeowner. Hot water (almost boiling) can be poured on individual mounds. Approximately 3 gallons of water should be used, and since this treatment eliminates only 60% of the mounds treated, surviving mounds may require retreatment. Continual disturbance to a mound may cause the ants to move to a new location, but caution is advised since the ants will readily sting when disturbed. A biological control agent has recently been marketed as the "fire mite." This is a parasitic mite (scientific name: *Pyemotes tritici*) that feeds on fire ant larvae and queens. Several applications, 2 weeks apart, may be required to kill a colony, and colony death may take a month or so. These mites can also attack people, causing a rash similar to poison ivy. Although not dangerous, care should be taken to avoid skin contact with these mites.

Other products are also available but may be impractical for most situations. They include an electrical grid-like device that electrocutes fire ants and an explosive cartridge device that is used to blast apart the mound. Several home remedies are **not** recommended. These include use of gasoline or other petroleum products. These substances are dangerously flammable and may pollute the environment. Grits sprinkled over the mound are often suggested--the theory being that the grits swell when swallowed and rupture the ants' stomachs. Since only the last larval stage of the fire ant can eat solid food, there is no way that the grits can kill the ants. They may, however, cause the colony to move. Crushed or grated citrus peels have also been reported to kill fire ants. In this case, testing in the field has not yet produced results.

21. When and how do I treat mounds?

In order to kill a fire ant mound, you must kill the queen. The queen is the only individual in a colony that can lay eggs. All of the rest of the workers are sterile females that forage for food, feed the immature ants (the larvae), take care of the queen, repair and defend the mound. The queen is normally located deep inside the mound. When using a mound drench it is best to treat the mound in the early morning hours during warm weather. Approach the mound quietly to minimize disturbance to the ants. The worker ants are very sensitive to vibrations, and they will immediately lead the queen down deep inside the mound when disturbed. Since mounds may be as deep as 1-3 feet, mound drenches won't reach the queen if the mound has been disturbed. If using baits, sprinkle them evenly around the mound about 1-3 feet away. Apply the bait when worker ants are actively foraging. This can be determined by placing peanut butter or tuna fish near the mound and watching for ant activity.

22. How do I control fire ants in my yard?

Fire ants can enter your yard in two basic ways--first by whole colonies migrating from neighboring yards and second, from mating flights. If you have only a few well-established mature mounds, it might be wiser to leave them if they aren't causing problems. They will keep new colonies from establishing near them. If you do treat, new colonies may move in, and you'll have more mounds to contend with. In small areas of less than 1 acre or where there are only a few mounds, repeated individual mound treatments are recommended. In larger areas, an initial broadcast application of baits or granular pesticide (chlorpyrifos) followed by a clean-up program of individual mound treatments is suggested. Products containing isofenphos, used to control turf pests, are labeled to prevent reinfestation of fire ants after initial fire ant mounds have been eliminated.

23. How do I control fire ants in my garden?

In home gardens, pre-plant soil-incorporated insecticides for soil insect control may temporarily control fire ants. These chemicals include diazinon and chlorpyrifos. However, for mature mounds, control options are limited. Hot water (3 gallons per mound, 90° C) may be poured over the mound, but treatments may have to be repeated since only 60 percent success can be expected. Unfortunately, few chemicals are labeled for use in home gardens. Methyl chloroform may be used, but care must be taken to avoid plants. Amdro may be used around the perimeter of the garden only. If ants are foraging outside of the garden they may pick up the bait and carry it back to the mound. A chemical barrier around the garden may help keep fire ants out. Either diazinon or chlorpyrifos can be used.

24. How do I control fire ants inside my house?

Fire ants will often enter houses looking for either food or water. Sometimes they will nest inside wall voids, potted plants or in other suitable areas. For this reason, you should try to locate the nest by following the trail of ants. Ant trails can be spot-treated with products containing either carbaryl, chlorpyrifos, diazinon, propoxur, or resmethrin. Colonies in wall voids can be treated with dust formulations of chlorpyrifos (Dursban). Amdro is not labeled for use inside houses. Fire ants can also cause problems inside electrical housings and water meters. A short piece of dichlorvos strip (Vapona Pest Strip) can be placed inside electrical switch boxes to control the ants. Around water meters, pyrethrin aerosols or methyl chloroform provide immediate control.

25. How do I control fire ants in pastures and rangegrass?

Fire ant mounds in pastures and rangegrass can damage harvesting equipment and pose a threat to animal life. Dragging a heavy object (e.g. railway tie) across the pasture will knock down tall mounds but will not eliminate the fire ants. Broadcast applications of baits will offer short-term relief (6-9 months). However, avoid area-wide applications if the number of mounds on your property is less than 15 mounds/acre since treatment will probably result in an increased number of mounds after the initial knockdown. Mounds can also be treated individually using products like acephate (note that the number of mounds which can be treated per acre is restricted by the label), diazinon, chlorpyrifos, amidinohydrazone or methyl chloroform. Products labeled for rangeland insect control (DZN diazinon AG500 or DZN diazinon 50W) will kill foraging ants and suppress fire ant populations, but mounds will not be killed.

A state quarantine currently prohibits the shipment of hay from fire ant infested counties to uninfested counties. Treatment of hay prior to shipment may be desirable. Texas Department of Agriculture district inspectors will inspect and certify hay shipments. If free of imported fire ants on inspection, a TDA form Q499 will be issued, permitting hay movement out of the quarantined area. For more information, call TDA at 1-800-342-5429 (DIAL-HAY).

26. How do I control fire ants inside barns or poultry houses?

The presence of fire ants can bother poultry and domestic animals. To control the ants near livestock enclosures, take the following measures: (1) Remove all food sources such as spilled feed, trash, broken eggs, etc.; (2) Remove all potential nest sites such as stacks of lumber, old equipment, manure piles, weeds and tall grass; (3) If ants are nesting inside the structure, litter can be treated with products containing carbaryl. However, take care to avoid contamination of feed or water supplies and follow label directions; (4) If ants are foraging outside the structure, spray around the outside of the building with chlorpyrifos or diazinon to create a barrier; (5) Use either a broadcast or individual mound application technique to kill mounds adjacent to the structure.

27. How do I control fire ants in my field crops?

Fire ants will sometimes damage seedlings or remove planted corn, peanut, or soybean seeds before germination. Seed treatments, pre-plant or at-plant insecticides applied to control soil insects will generally prevent fire ant damage, although these products are not specifically labeled for fire ant control.

Later in the season, fire ants may feed on roots, blossoms, stems or fruit. They may also tend and protect aphids which feed on plants. However, the ants rarely cause problems severe enough to justify treatment, and they may reduce populations of plant feeding insects such as boll weevils, bollworms and sugarcane borers.

28. How do I control fire ants around bee hives?

Fire ants will readily enter bee hives and feed on developing bee larvae. Weakened colonies are especially vulnerable. Fire ant control around bee hives must be done carefully since the bees themselves are susceptible to most insecticides.

First treat the area around the hive to remove any mounds close by. Bait formulations are safest; dust formulations should be avoided. A chemical barrier can be painted onto the base of the hive or sprayed on the ground underneath the hive. Use either diazinon or chlorpyrifos. Apply the insecticide late in the evening or early in the morning when bees are inactive. Since these chemicals break down with time, repeated applications should be made periodically to keep barriers effective.

THE EFFECT OF ABAMECTIN ON FEMALE ALATE FERTILITY OF THE RED IMPORTED FIRE ANT (*Solenopsis invicta*)

Michael E. Mispagel, Ph.D.
Dept. of Physiology and Pharmacology
The University of Georgia
Athens, GA 30602

INTRODUCTION

In 1982, Glancey et al. conducted a laboratory investigation of the effects on the morphology of ovaries of Red Imported Fire Ant (RIFA) queens which had been sterilized by Avermectin B1a (Abamectin). Twenty-two weeks posttreatment, hypertrophy of the squamous epithelium sheathing the ovariole and pycnosis of nurse cell nuclei were observed in queens displaying either total sterility or a reduction in the number and size of eggs produced.

Glancey et al. (1982) further observed that colonies which were fed Abamectin in concentrations less than 0.0025% had decreased brood production which returned to normal after 16 weeks. No observations were made on unfertilized female alates.

The current study was designed to determine the effect of abamectin on the fertility of female RIFA alates.

METHODS

The formulated fire ant pesticide, Affirm, containing 0.011% of Abamectin in a pregelled defatted corn grit carrier with soybean oil as an attractant was supplied by Merck Sharp & Dohme Research Laboratories. A quantity of Affirm was dyed with Sudan III by addition of dyed soybean oil or an acetone mixture. The study was begun on May 20, 1985. After thorough evaporation of the acetone, about 3 g of dyed bait in one ounce plastic containers was offered to each of 15 functionally monogynous queen-right colonies of RIFA. Six monogynous queen-right colonies were maintained as untreated controls. Unconsumed bait was not removed from the colonies except during normal cleaning procedures to transfer the colonies to fresh trays. In general, bait was available throughout the study period though because of rancidity, not necessarily attractive to the ants.

Reproductive brood present prior to exposure was removed from all treated colonies under the assumption that all subsequent reproductive brood would be produced by a queen which had been exposed to the bait. The presence of red dye in all brood and alates was observed daily for two weeks after the initial exposure (PE).

At least four randomly selected female alates were removed from each treated colony on days 9, 16, 29, 37, 66, and 131 PE for ovariole examination. On June 3, 14 days PE, surviving queens from treated colonies were removed for examination of ovariole development. Queens were not removed from controls.

Alate ovarioles were prepared for examination by two methods. In the first method, collected alates were cut at the petiole and immersed in McDowell and Trumps solution. Two or 3 tergites and sternites were dissected from the gaster to assure maximum penetration of the McDowell and Trumps fixative. The preparations were rinsed twice in 0.1 M phosphate buffer, osmicated in 1% OsO_4 in 0.1 M phosphate buffer for 60 minutes, rinsed 3 times in water, soaked in ethanol at 50%, 75%, 95% and 100% concentrations to remove all water, mixed in 50% Spurr plastic with 100% acetone and embedded in 100% Spurr plastic. Tissues were stained with 1% toluene blue in 1% sodium borate and basic fuchian stain and sectioned at 1 um.

The second method (Tarpley and Mispagel, in prep.) utilized histopathological techniques employing fixation in methacarn, soaking in a chitin and kerotin softening agent for 48 h (Luna, 1968), and storing in methacarn until gaster bisection. The gasters were infiltrated with glycol methacrylate for 4-7 days and embedded in plastic using a JB-4 Embedding Kit from Polysciences. Polymerization in plastic processing molds continued for 8-12 hours at room temperature. Excess plastic was trimmed and specimens were sectioned at 2 um using Ralph glass knives. Sections were dried at 45 C for 20 min and stained using the Turner method with a 4.2 pH acetate buffer in the stain. A Lillie-Mayer hematoxylin stain for nucleii and eosonate of polychrome methylene blue stain were used. This method produced much better specimens for detailed examination.

RESULTS

COLONY OBSERVATIONS: Within 48 h after exposure to the dyed Affirm bait, red dye was visible in about half of the larvae and among ants of the nurse caste. No dye was observed at this or any other time in crushed alates. Within two weeks PE, up to 50% of the colonies' workers were killed by exposure to Affirm and, in 9 out of 15 treated colonies, the queen was killed as well. Eight days after exposure, dealates were observed in the queenless colonies. The other six treated colonies had queens that no longer appeared physogastric and appeared to be sterile as no further eggs were produced and the number of brood in these colonies declined through normal eclosion.

Fifty d PE, alates from three queenless colonies were producing eggs. Eighty-four d PE (August 12), six colonies (10,13,18,19,22,23) had eggs though only four had eggs in abundance because of the large number of alates in those particular colonies. At that time, none of those eggs showed signs of further development or maturation. Three of the treated colonies (17,18,22) had died completely by 172 d PE (November 8).

By the middle of November (180 d PE), reproductive brood was observed in colonies numbered 4 and 16. On January 7, 1986 (232 d PE), two treated colonies (10,16) had many female alates and a great number of reproductive brood. Four other colonies (4,5,13,23) had mostly female alates with only a few workers and a few eggs. Two days later, female alates in colonies 10 and 16 displayed hyperactivity indicative of pre-mating flight activity.

No male alates were produced from any of these colonies. Rather, all the brood developed into female alates. Some colonies produced well over a hundred female alates. The number of workers slowly declined by attrition through this period and only the largest workers seemed to survive.

By April, 1986 (315 d PE), the majority of the alates had died and no further reproductive brood was produced. However, on day 332 PE, it was observed that a single colony (5) had two dealates alive of which at least one was producing worker brood. At least 50 eggs and a number of worker pupae were observed. All the workers which eclosed were small, minim-sized. A month later, both dealates had died but about 100 small workers had been produced. By June, 1986 (1 year PE), all treated colonies were dead.

OVARIOLE OBSERVATIONS: The first technique for preparing and sectioning ovarioles proved unsatisfactory because of the presence of lipids within the abdomen and poison gland. Moreover, because of cuticular properties, the plastic did not hold the embedded specimen well for sectioning. A second technique which did not require dissection of gasters and which removed much of the oil, proved more satisfactory.

Ovarioles of treated queens showing signs of sterility (n=6) were sectioned. As Glancey et al. (1982) observed, there was definite and obvious hypertrophy of the squamous epithelium surrounding the ovarioles of queens which had been exposed to Affirm. However, pycnosis of the nurse cell nucleii was not as readily apparent since some clumping of chromatin was observed in control as well as in treated specimens.

Among female alates which had been exposed to the Affirm bait, some squamous epithelial cell hypertrophy was present as early as two weeks after treatment. This condition did not encompass the entire ovariole as observed in treated queens, but, when present, was observed most often in the well developed ovariole adjacent to the calyx rather than in the developing germarium. Many portions of the ovaries which showed this condition in specific ovarioles looked normal otherwise.

The effect of Affirm on the size or number of developing oocytes could not be determined since the number of oocytes in unfertilized alates is low under normal conditions and is probably affected by a number of factors including age, various colony pheromones, position of cut section, etc.

DISCUSSION

The dose of Affirm bait administered directly to the colonies in this study was less than that recommended for single mound treatment (5 Tbl/mound=20 g). Most of the bait presented to the colonies was left unconsumed by the ants. The result of this dosage was worker mortality and the death of 9 out of 15 queens within two weeks (the remaining treated queens were sacrificed). No alates were observed feeding directly on the bait.

Queen sterility was apparent among those surviving both by the lack of brood and by the observation of extensive hypertrophied squamous epithelial cells sheathing the ovarioles. The reversibility of queen sterility was not the object of this study and was not investigated.

Suppression of oogenesis by female alates was apparent for the first six months after treatment. This suppression was, however, temporary as eggs, though apparently not viable at this time, began to appear. Soon after, eggs developed into larvae and reproductive brood was observed. This brood developed exclusively into female alates. No males were produced from these treated alates.

Colony founding by pleometrosis in the fire ant is not an uncommon phenomenon (Tschinkel and Howard 1983). These authors (1978) have shown that orphaned colonies, i.e. colonies in which the physogastric and functioning queen had been removed, produced fertile replacements which, it was hypothesized, were inseminated, surviving foundresses from pleometrotic colony founding. Such colonies would be polygynous but functionally monogynous. These authors also noted that some uninseminated replacement queens produced worker pupae presumably by thelytokous parthenogenesis.

In our study, it is possible that such inseminated queens were present and survived the treatment when the functional queen did not. The lack of dye in alates suggests that the alates do not receive as much nourishment via trophallaxis as do workers and the functional queen. For example, in laboratory colonies where food is readily available, alates are often observed foraging on their own rather than relying solely upon trophallaxis as does the queen. This may explain the incomplete effect of Affirm on the squamous epithelial cells of the ovarioles among the treated alates. It was often difficult to distinguish between sections of ovarioles from control specimens and those from alates which had been subjected to the treatment.

If inseminated replacement queens had been present among the treated colonies, the production of female reproductive brood may have been a seasonal phenomenon slightly out of synchrony. In field colonies of imported fire ants, the development of large numbers of reproductive brood is common in the spring of the year. The constant and artificial conditions of the laboratory may have induced that exclusive production of reproductives earlier than would be observed in the field.

Although the functional queens from the control colonies designated for this study were not removed, other "queenless" colonies are maintained in our laboratory. Among these colonies which contain many female alates as well as a few dealates, exclusively female reproductive brood has also been produced though not in large quantities. This suggests that this is a natural phenomenon and not the result of treatment by Affirm.

The development of worker brood by colony 5 could have been the result of either an inseminated replacement queen or the result of thelytokous parthenogenesis (Haskins and Enzmann 1945). There is no reason to believe that exposure to Affirm had any effect on this production.

In conclusion, the effect of Affirm on the functional queens was as expected and as reported by Glancey et al. (1982). However, the effect on female alates was less clear microscopically. The development of brood by inseminated replacement queens or by thelytokous parthenogenesis further indicates that Affirm because of the very small doses to which the alates are exposed via trophallaxis, has only limited and temporary, if any, effect on the fertility of female alates or replacement queens.

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PRELIMINARY ELECTROPHYSIOLOGICAL EVALUATION OF THE EFFECTS OF AVERMECTIN AND A FLUORINATED SULFONAMIDE ON THE RED IMPORTED FIRE ANT

David D. Wilson and John M. Bowen
Department of Physiology and Pharmacology
University of Georgia, Athens

ABSTRACT

Two potential fire ant toxicants, Avermectin, a macrocyclic lactone glycoside, and a fluorinated sulfonamide, both dissolved in dimethyl sulfoxide (DMSO) were bath-applied to preparations of fire ant indirect flight muscles. Electrophysiological recordings were made of the resting membrane potentials and miniature postsynaptic potentials (MPSP) of these dorsal longitudinal muscles. The records of the MPSP were then analyzed using a microcomputer program with signal averaging capabilities. Preliminary results indicate a lowering of fire ant resting potentials after application of Avermectin at doses up to 12 uM. Miniature postsynaptic potentials exhibited slight decreases in amplitude and frequency with application of Avermectin at the same doses. The fluorinated sulfonamide showed increases in resting potential and amplitudes of the MPSP at doses up to 10 uM. There were some increases in the frequency of the MPSP. The results, though preliminary, indicate some trends which are being examined in greater detail.

DEVELOPMENT OF IFA QUARANTINE TREATMENTS

FOR NURSERY STOCK

For Presentation at:
Annual IFA Research Conference
Austin, Texas

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By:
Homer L. Collins
USDA, APHIS, PPQ
Imported Fire Ant Station
Gulfport, MS

ABSTRACT

Development of IFA Quarantine Treatments for Nursery Stock

Homer Collins

USDA, APHIS, PPQ

Gulfport, MS 39503

Movement of infested nursery stock in interstate commerce is responsible for much of the artificial or accidental spread of imported fire ants. Chlorinated hydrocarbon insecticides were widely used as preventative treatments from the late 1950's until their cancellation of registration in the 1970's. Development of alternate treatments in recent years has led to the registration of several formulations of chlorpyrifos for various use patterns including treatment of potting soils, grass sod and certain types of woody ornamental plants.

Triumph 1G insecticide has shown potential as a potting soil treatment in preliminary trials. Combination treatments employing Amdro bait plus granular chlorpyrifos was an effective procedure for reducing IFA populations in field grown nursery stock in 1985 tests.

Although fire ants can become established by a variety of means, it is generally agreed that long distance movement of nursery stock is the most efficient method of artificial dispersal into new areas. The explosive spread of the IFA during the 1950's by movement of infested nursery stock is well documented in the literature (Bruce et al, 1949, Culpepper, 1953). Several of the recently detected isolated infestations [SLIDE] can be directly traced to movement of infested nursery stock (Elizabeth City, NC., Nashville, TN., Lubbock, TX., and Laredo, TX.)

In 1958 Federal Quarantine 301.81 was enacted in an effort to slow or prevent further spread. Chlorinated hydrocarbon insecticides were effectively utilized as preventative treatments throughout the next two decades. Cancellation of registrations of these insecticides has prompted the need to develop alternate quarantine treatments to prevent artificial spread of the IFA through movement of infested plant material. Efforts by our laboratory to fulfill this need can be divided into three basic areas of endeavor:

1. Development of treatments for containerized plants [SLIDE]

2. Development of treatments for grass sod [SLIDE]

3. Development of treatments for field grown woody ornamentals[SLIDE]

(Balled and Burlapped Plants) [SLIDE]

The most desirable treatment for containerized plants is an insecticide that can be premixed or incorporated into the potting media prior to potting the plants.

A screening program to evaluate all insecticides suitable for use as a preplant incorporated treatment for potting soil has been conducted since 1974. Due to the unique requirements [SLIDE] of a potting soil insecticide, relatively few compounds have been evaluated. The most effective product evaluated to date has been chlorpyrifos, and two formulations have been registered for this use, but only one is available today [SLIDE]. Several candidates were screened in 1985; results with Triumph 1G are very encouraging since we have obtained 9 months residual activity [SLIDE] when mixed in potting soil at a rate of 11.2 gms AI/yd3 (approximately 10 lbs AI/3" acre).

Development of treatments for grass sod has also been slow and tedious, but we were able to obtain registration of a Dursban formulation for this use also [SLIDE]. Due to its relative short residual activity this treatment does not eliminate large mature colonies. However the major pest risk associated with sod is transport of newly mated queens, not entire colonies. Granular Dursban applied at a rate of 10 lbs AI/acre provides a residual barrier for 10 weeks against IFA alate queens.

The classical quarantine treatment used to eliminate other soil insects such as Japanese beetle, whitefringed beetle, etc. from Balled and Burlapped plant material is a root dip treatment in which the root ball is immersed in an insecticidal solution, thereby achieving total saturation. Although we have a Dursban registration for this use pattern, the procedure is costly, labor intensive and not well suited for all types of plant material [SLIDE].

Treatments for field grown stock is our most urgent quarantine need. The industry has requested simple, inexpensive in-field treatments. In 1985 we evaluated two procedures in an effort to comply with this request.

I. SWEEP-SHANK INJECTION OF DURSBAN

As mentioned earlier treatments for grass sod and potting soil are based on chlorpyrifos due to its high toxicity as a contact myrmicide as well as its residual activity compared to other available insecticides. However, chlorpyrifos photodegrades rather rapidly when surface applied. Therefore, a means of subsurface application through the use of a sweep-shank injector system was investigated as a possible means of extending the residual activity of chlorpyrifos. To be successful, a minimum of 24 months of protection would be required. A sweep shank injector basically consists of 5 twelve-inch "sweep" plows modified to include a flat fan spray nozzel mounted behind each sweep, giving a total coverage of 60 inches (i.e., a 60" band) which is injected at a depth of 2-4 inches beneath the soil surface.[SLIDES]

We evaluated this procedure by treating entire 1-acre plots at rates of 10 and 20 lbs AI/acre and monitoring treatment effects according to procedures described by others (Harlan et al 1981 , Lofgren and Williams 1982). Evaluations conducted 5, 10, and 20 weeks after treatment indicated that the procedure was not effective [SLIDE]. GLC analyses of soil core samples collected 10 weeks after application indicated that the Dursban was concentrated at the 2-4" depth (1.46 ppm). We therefore speculate that the ants are able to tunnel through the relatively thin Dursban layer and/or perhaps line their tunnels with soil particles from above or below the "lethal zone". Based on these results we do not plan to conduct additional trials with this procedure.

II. SEQUENTIAL APPLICATIONS OF AMDRO AND DURSBAN

Another procedure for certification of field grown nursery stock which was investigated last season involved sequential applications of Amdro and Dursban. Rationale for the dual applications is that the Amdro would drastically reduce the population while Dursban, applied approximately 7 days later, would further weaken exhisting colonies and leave a residual barrier against reinfestation for a number of weeks. Four separate tests were conducted in 1985 to evaluate this procedure. In all tests, Amdro was applied at 1.5 lbs bait per acre with a modified Gandy (R) applicator. Dursban 10G was applied at 6.0 lb AI/acre with a Skibee spreader. At each evaluation period, each plot was rated according to the population index system described by Lofgren and Williams, 1982).

Although some trials have not yet been completed, all available evidence indicates that this procedure is effective in reducing the IFA population to an acceptable level of risk [SLIDE]. Additional tests will soon be initiated to confirm these results.

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ford's

SPECIMEN LABEL

FIRE ANT 2.5 G Insecticide

Controls the Imported Fire Ant in Nursery Bench and Potting Media
Intended Primarily For Use on Woody Ornamental Plants Grown Outdoors in Containers

ACTIVE INGREDIENT:
Chlorpyrifos [O,O-diethyl O-(3,5,6-trichloro-2-pyridyl) phosphorothioate 2.5%
INERT INGREDIENTS 97.5%

PRECAUCION AL USUARIO: Si usted no lee ingles, no use este producto hasta que la etiqueta le haya sido explicada ampliamente.
TRANSLATION: (TO THE USED: If you cannot read English do not use this product until the label has been fully explained.)

KEEP OUT OF REACH OF CHILDREN
CAUTION
PRECAUTIONARY STATEMENTS
Hazards to Humans
MAY BE HARMFUL IF SWALLOWED
Do Not Take Internally — Avoid Contact With Eyes, Skin, and Clothing
Wash Thoroughly After Handling
Wash Contaminated Clothing Before Reuse
Avoid Breathing Dust

STATEMENT OF PRACTICAL TREATMENT

If Swallowed: Drink 2 glasses of water and induce vomiting by touching back of throat with finger. Do not induce vomiting or give anything by mouth to an unconcious person. Get medical attention. **If on Skin:** In case of contact, remove contaminated clothing and immediately wash skin with soap and water. **If in Eyes:** Flush eyes with plenty of water and get prompt medical attention.
Note to Physician: Chlorpyrifos is a cholnesterase inhibitor. Treat symptomatically. Atropine only by injection is an antidote.

ENVIRONMENTAL HAZARDS

This product is toxic to fish, birds and other wildlife. Birds feeding on treated areas may be killed. Do not apply where runoff is likely to occur. Do not apply when weather conditions favor drift from areas treated. Do not contaminate water by cleaning of equipment or disposal of wastes. Apply this product only as specified on this label.

AGRICULTURAL CHEMICAL

Do Not Ship or Store with Food, Feeds, Drugs or Clothing

EPA Reg. No. 10370-141 **EPA Est. No. 10370-TX**

USE DIRECTIONS

It is a violation of Federal law to use this product in a manner inconsistent with its labeling.
Apply Fire Ant 2.5 insecticide for the control of imported fire ants in nursery bench and potting media. Thoroughly mix 1 pound of Fire Ant 2.5 insecticide per cubic yard of media. Use equipment suitable to give uniform distribution of granules.

USE PRECAUTIONS

Do not use plants for food or feed purposes which have been exposed to soil treated with Fire Ant 2.5 insecticide.

STORAGE AND DISPOSAL

Prohibitions: Do not contaminate water, food or feed by storage or disposal. Open dumping is prohibited.
Pesticide Disposal: Wastes resulting from the use of this product may be disposed of on site or at an approved waste disposal facility.
Container Disposal: Completely empty bag into application equipment. Then dispose of empty bag in a sanitary landfill or by incineration, or, if allowed by State and local authorities, by burning. If burned, stay out of smoke.
General: Consult federal state or local disposal authorities for approved alternate procedure such as limited open burning.
NOTICE: Seller warrants that the product conforms to its chemical description and is reasonably fit for the purposes stated on the label when used in accordance with directions under normal conditions of use, but neither this warranty nor any other warranty of MERCHANTABILITY or FITNESS FOR A PARTICULAR PURPOSE, express or implied, extends to the use of this product contrary to label instructions, or under abnormal conditions, or under conditions not reasonably foreseeable to seller, and buyer assumes the risk of any such use.

U. S. Patent No. 3,244,586

22.68 kg/50 lb net

MANUFACTURED BY
ford's **CHEMICAL & SERVICE, INC.**
2739 Pasadena Blvd.
Pasadena, Texas 77502

Table 8. Residual Activity of Candidate Potting Soil Toxicants, 1985.

Toxicant	Mean % mortality to alate queens after 7 day exposure				
	Posttreatment interval (Months)				
	1	2	3	6	9
Triumph 1G	100	100	100	100	100
Dursban 50WP	50	66.7	100	100	100
Marshal 5G	15	15	5		
Mocap 10G	50	60	75		
Broot 15G	5	5	0		
Ficam 10G	85				
Check	15	10	5	0	

ford's SPECIMEN LABEL

DURSBAN® FIRE ANT 10% Granular Insecticide

FOR USE ON NURSERY GRASS SOD TO PREVENT THE ARTIFICIAL SPREAD OF IMPORTED FIRE ANTS.

Only for use by, or under the direction of, Federal or State Fire Ant Quarantine Officers.

ACTIVE INGREDIENT:	
Chlorpyrifos [0.0-diethyl-0-(3,5,6-trichloro-2-pyridyl) phosphorothioate]	10.0%
INERT INGREDIENTS	90.0%
TOTAL	100.0%

*Dursban trademark of the Dow Chemical Company

U. S. Patent No. 3,244,588

KEEP OUT OF REACH OF CHILDREN
CAUTION
PRECAUTIONARY STATEMENTS

Hazards to Humans

MAY BE HARMFUL IF SWALLOWED

Do not Take Internally —
Avoid Contact With Eyes, Skin, and Clothing
Wash Thoroughly After Handling
Wash Contaminated Clothing Before Reuse
Avoid Breathing Dust

STATEMENT OF PRACTICAL TREATMENT

IF SWALLOWED: Drink 1 or 2 glasses of water and induce vomiting by touching back of throat with finger. Do not induce vomiting or give anything by mouth to an unconscious person. Get medical attention. **IF ON SKIN:** In case of contact, remove contaminated clothing and wash affected areas with soap and water. **IF IN EYES:** Flush eyes with water and get prompt medical attention.

Note to Physician: Chlorpyrifos is a choinesterase inhibitor. Treat symptomatically. Atropine only by injection is an antidote.

ENVIRONMENTAL HAZARDS

This product is toxic to fish, birds and other wildlife. Birds feeding on treated areas may be killed. Do not apply where runoff is likely to occur. Do not apply when weather conditions favor drift from areas treated. Do not contaminate water by cleaning of equipment or disposal of wastes. Apply this product only as specified on this label.

USE DIRECTIONS

It is a violation of Federal law to use this product in a manner inconsistent with its labeling.

1. Use protective equipment (rubber gloves and aprons) when handling treated sod.
2. Apply this product to nursery grass sod with application equipment capable of uniform distribution at a rate of 40 to 60 lbs. per acre. A 4-week certification period is provided when 40 lbs. per acre are applied. 60 lbs. per acre provides a 10-week certification period.
3. Immediately after treatment irrigate treated area with at least ½ inch of water or a sufficient volume to thoroughly soak below the cut line.

STORAGE AND DISPOSAL

Prohibitions: Do Do not contaminate water, food or feed by storage or disposal. Open dumping is prohibited.

Pesticide Disposal: Wastes resulting from the use of this product may be disposed of on site or at an approved waste disposal facility.

Container Disposal. Completely empty bag into application equipment. Then dispose of empty bag in a sanitary landfill or by incineration, or, if allowed by State and local authorities, by burning. If burned, stay out of smoke.

General Consult federal state or local disposal authorities for approved alternate procedure such as limited open burning.

NOTICE: Seller warrants that the product conforms to its chemical description and is reasonably fit for the purposes stated on the label when used in accordance with directions under normal conditions of use, but neither this warranty nor any other warranty of MERCHANTABILITY or FITNESS FOR A PARTICULAR PURPOSE, express or implied, extends to the use of this product contrary to label instructions, or under abnormal conditions, or under conditions not reasonably foreseeable to seller, and buyer assumes the risk of any such use.

AGRICULTURAL CHEMICAL

Do Not Ship or Store with Food, Feeds, Drugs or Clothing.

EPA Reg. No. 10370-142 **EPA Est. No. 10370-TX**

U. S. Patent No. 3,244,586

22.68 kg/50 lb net

MANUFACTURED BY

ford's CHEMICAL & SERVICE, INC.

2739 PASADENA BLVD.
PASADENA, TEXAS 77502

Table 9. Sweep Shank Injection of Dursban.

Treatment	RESULTS AT INDICATED WEEKS POSTTREAT					
	% Colony Mortality			% Change in Pretreat Pop. index		
	(5)	(10)	(20)	(5)	(10)	(20)
Dursban injection--10#AI/acre	17	0	0	-52	49	373
Dursban injection--20#AI/acre	0	0	0	31	50	447
Untreated check	21	21	0	-34	0	22

Results with Sequential Applications of Amdro and Dursban for IFA Control in Nursery Stock, 1985.

Test No.	Treatment	Population index at indicated weeks postreatment							
		Pretreat	1	4	8	12	16	20	24
I	Amdro + Dursban	135		0	0		0	0	
	Ck	72		77	43		48	62	
II	Amdro + Dursban	1802	670		10	0	4	98	350
	Amdro only	926	684		275	130	181	360	625
	Ck	807	446		305	287	349	180	593
III	Amdro + Dursban	185		0	0				
	Amdro only	135		43	22				
	Dursban only	80		3	0				
	Ck	115		132	110				
IV	Amdro + Dursban	185		2		15			
	Amdro only	250		90		55			
	Dursban only	270		68		9			
	Ck	385		372		272			

Straw Itch Mites as a Biocontrol Agent of Red Imported Fire Ants.

H. Thorvilson, S.A. Phillips, Jr., A. Sorenson, and M. Trostle.
Texas Tech University and Texas Dept. of Agriculture.

ABSTRACT

The straw itch mite, Pyemotes tritici (Lagreze-Fossat and Montague), is a parasitic mite that has generated considerable interest as a biocontrol agent of the red imported fire ant (RIFA). To evaluate the mite's efficacy in the field, a large trial was conducted at 3 pasture sites near Bandera, in southcentral Texas. A total of 244 RIFA mounds was mapped, located with labeled stakes, and the mound ratings recorded. During May 1985, two applications of mites cultured on Angoumois-infested wheat kernels were introduced into 103 RIFA mounds, whereas 141 control RIFA mounds were treated with wheat kernels not infested with mites. The mounds were also treated in October. Throughout the experimental period, the RIFA mounds were periodically inspected and rated. Analysis of the data utilizing the Student's t-test was unable to detect significant differences in mound ratings between mite-treated and control RIFA mounds.

INTRODUCTION.

The straw itch mite, Pyemotes tritici (Lagreze-Fossat and Montague), is a parasitic mite that attacks several stored-grain insect pests and also parasitizes the red imported fire ant (RIFA), Solenopsis invicta Buren (Bruce and LeCato, 1979). In Georgia and Florida field trials, P. tritici "inactivated" more than 50% of RIFA mounds upon which they were applied (Bruce and LeCato, 1980). However, a relatively small number of mounds were treated in the study and some nests were treated up to 10 times with mites. Nonetheless, the straw itch mite has generated considerable interest as a potential biological control agent of the RIFA. In response to continuing promotion of "fire mites", a large-scale field study was initiated in Bandera County, Texas in cooperation with the Texas Department of Agriculture. The objective of the study was to determine the effect of P. tritici applications on the survival of RIFA field colonies.

METHODS AND PROCEDURES.

Three pasture sites near Bandera in southcentral Texas were chosen for the study. At each site, RIFA mounds were located, mapped, and labeled with wooden stakes. The pre-treatment rating of colony size and brood characteristics (Lofgren and Williams, 1982) was recorded for each mound. On 16,17 May 1985, the first mound treatments were completed at sites A and B. Randomly-selected mounds were treated with wheat kernels infested by Angoumois grain moth larvae that were parasitized by straw itch mites (ca. 30,000 mites/cup). Control treatments consisted of unparasitized grain moth larvae on wheat. The experimental introduction of material into the mounds was as follows: Six shallow holes (ca. 7.5 cm deep and 5.0 cm dia.) were punched into each mound. Two 34-ml plastic cups of wheat were poured into the holes in each mound and the holes were covered with soil. About 2 weeks later, the mounds were again rated and the treatments were repeated. The condition of the mounds was also evaluated at 30, 44, and 86 days posttreatment. Autumn treatments of the colonies occurred on 4 Oct. 1985 and 18 Oct. 1985 in an identical manner. A third location (site C), consisting of 8 mite-treated and 19 control mounds, was also initiated on 4 Oct.1985. The total of 244 colonies (103 mite-treated and 141 control colonies) was evaluated through 21 Nov. 1985.

RESULTS AND DISCUSSION.

The differences between nean colony ratings of mite-treated and control mounds were analyzed using Student's t-distribution tests. Few significant differences ($P<0.05$) were detected between treatments. At site A (Table 1.), the mite-treated mounds had a significantly higher mean mound rating than did the control mounds on 8 Aug. Similarly, mite-treated mounds at site B (Table 2.) had significantly higher mean ratings on 21 Nov. than did control mounds. An explanation for this unexpected vitality of mite-treated mounds is not readily apparent. At site C (Table 3.), no significant differences in mound ratings were detected.

As expected, mean mound ratings declined from spring to autumn according to normal phenology. Mean ratings of mite-treated mounds were significantly ($P<0.05$) reduced during both time periods 14 May-8 Aug. and 4 Oct.-21 Nov. at site A, while control mounds declined significantly only during the former time period. In contrast, the control mounds at site B declined significantly during the 4 Oct.-21 Nov. period. At both sites A and B, the mean mound ratings on 21 Nov. were significantly less than those on 14 May in both treated groups. No significant differences were detected at site C.

Some uncomfortable side-effects to applying mites in the field were noted. Mite dermatitis appeared on the arms and torso of researchers that carried boxes containing mite-infested cups of wheat. These effects occurred eventhough insect repellent-impregnated cloths were wiped over exposed skin prior to mite application.

The results of this field experiment suggest that the use of "fire mites" as a biocontrol agent of RIFA in the manner described herein does not work. Unless new methodology is developed, our conclusion is that other tactics of RIFA control are more efficacious than P. tritici.

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Table 1. Mean mound ratings of mite-treated and control RIFA mounds at site A, Bandera County, Texas, 1985.

Treatment	Date, 1985 May 14[a]	30[a]	June 13	27	Aug 8	Oct 4[a]	18[a]	Nov 8	21
Mites (34)	21.9	16.1	12.4	8.1	14.2	13.8	14.5	10.4	7.9
Control (39)	19.5	15.6	12.7	7.1	10.0	14.2	16.0	13.8	10.3
t-value	1.9	0.2	0.2	0.5	2.1*	0.3	0.7	1.3	1.0

[a] = dates of treatment

numbers in parentheses are the number of mounds

* = mean mound ratings are significantly different (Student's t-test, $P<0.05$).

Table 2. Mean mound ratings of mite-treated and control RIFA mounds at site B, Bandera County, Texas, 1985.

Date, 1985

Treatment	May 15[a]	31[a]	June 13	27	Aug 8	Oct 4[a]	18[a]	Nov 8	21
Mites (61)	15.5	12.9	13.7	13.3	13.0	11.6	15.5	14.7	8.8
Control (83)	14.0	14.8	14.5	14.3	12.8	9.8	14.2	11.1	4.7
t-value	1.5	1.4	0.6	0.7	0.1	1.3	0.6	1.7	2.0*

[a] = dates of treatment

numbers in parentheses are the number of mounds

* = mean mound ratings are significantly different (Student's t-test, $P<0.05$).

Table 3. Mean mound ratings of mite-treated and control RIFA mounds at site C, Bandera County, Texas, 1985.

Treatment	Date, 1985 Oct 4[a]	18[a]	Nov 8	21
Mites (8)	11.3	11.8	11.4	7.8
Control (19)	12.6	16.5	14.6	9.5
t-value	1.1	1.0	0.7	0.4

[a] = dates of treatment

numbers in parentheses are the number of mounds

no significant differences (Student's t-test, $P<0.05$) were detected in mean mound ratings.

Control of Imported Fire Ants With New Insect Growth Regulator and Fluorocarbon Baits

W. A. Banks
USDA-ARS
Gainesville, Florida

Two experimental insect growth regulators (IGR) from Union Carbide, UC-84572 and UC-86874, produced excellent suppression of worker brood production in laboratory colonies of red imported fire ants (RIFA). Dosages of 10 and 20 mg/colony of either compound produced total suppression of worker brood production by 6 weeks posttreatment in queen-right colonies of 50,000 or more workers. The suppression of brood caused a reduction of 80-85.6 percent in the colony index. No further production of worker brood occurred through 32 weeks posttreatment and all colony indices had been reduced by greater than 98 percent at that time (Table 1). Field tests with UC-86874 are planned for summer 1986.

The IGR S-31183 from Sumitomo Chemical Company produced total suppression of worker brood production at 10 mg/colony in queen-right colonies of 50,000 or more workers within 4 weeks posttreatment. All colonies had been reduced to less than 500 workers by 24 weeks posttreatment when the test was terminated. Field tests with pregel defatted corn grit baits containing 0.5 or 1.0% active ingredient (AI) and applied at rates equivalent to 5.26 to 23.4 g/ha AI produced reductions of 96.0 to 98.2 % in the population index within 13 weeks posttreatment. Logic at 12.18 g/ha AI had reduced the population index by 97.7% at the same interval (Table 3).

A series of fluorinated sulfonamides from 3-M Company were shown by Vander Meer et al. (J. Econ. Entomol. 78: 1190-7, 1985) to possess good delayed-action toxicity to red imported fire ants. Griffin Corporation, Valdosta, Georgia, has selected one of the more promising of this group of chemicals for commercial development as a bait toxicant for control of fire ants. We initiated a study in the fall of 1985 to evaluate the effectiveness of this compound, designated by Griffin as GX-071, in baits against natural infestations of red imported fire ants. Tests were conducted on ungrazed permanent pasture near Lakeland, Florida with pregel defatted corn grit baits containing GX-071 at 0.3 and 0.6%. Applications were made at rates equivalent to 2.58, 3.75, and 5.16 g/ha AI with the 0.3% bait, and to 5.07, 7.20, and 10.14 g/ha AI with the 0.6% bait. Amdro was applied at 9.64 g/ha AI as a standard. After 13 weeks the GX-071 baits had produced reductions in the population index of 80.7 to 93.7%. The highest rate of application of the 0.6% bait was the most effective. Amdro had produced a reduction of 82.1% in the population index (Table 4).

Classification of Laboratory Colonies of Imported Fire Ants

Based On Estimated Number of Worker Ants and Quantity of Worker Brood

Estimated Number Workers	Index	Value	Quantity Worker Brood (ml)	Index	Value
<100	1	1	None	A	1
100-5000	2	2	1-5 ml	B	5
5000-20000	3	3	5-10 ml	C	10
20000-35000	4	4	10-20 ml	D	15
35000-50000	5	5	20-30 ml	E	20
>50000	6	6	>30 ml	F	25

TABLE 1. Effectiveness of Union Carbide Insect Growth Regulators Against Laboratory Colonies of Red Imported Fire Ants. Avg. of 3 replications.

Dosage mg/colony	Pretreatment Colony Index	Percent reduction in colony index after indicated weeks posttreatment					
		4	6	12	20	28	32
		AI3-29865(UC 84572)					
10	150	85.3	96.2	96.9	97.8	98.7	98.7
20	150	85.3	96.0	96.4	97.6	98.0	98.2
		AI3-29866(UC 86874)					
10	142	85.6	96.2	96.7	97.4	98.1	98.4
20	142	80.0	96.0	96.5	97.2	98.1	98.3
		Untreated Check					
-	150	0	0	-	32.4	32.7	32.7

Table 2. Effectiveness of the Insect Growth Regulator Sumitomo S-31183 (AI3-29835) Against Laboratory Colonies of Red Imported Fire Ants (Avg. of 3 replications).

Dosage mg/colony	Pretreatment Colony Index	Percent reduction in colony index after indicated weeks posttreatment					
		4	8	12	16	20	24
10	133	80.0	96.5	97.8	96.5	99.0	100
Check	100	11.7	+3.3	+3.3	5.0	+23.3	+1.3

RATING OF FIELD COLONIES OF IMPORTED FIRE ANTS

BASED ON WORKER POPULATION AND BROOD STATUS

NUMBER OF WORKER ANTS	WORKER BROOD ABSENT		WORKER BROOD PRESENT	
	FIELD RATING	INDEX	FIELD RATING	INDEX
<100	1	1	6	5
100-1000	2	2	7	10
1000-10000	3	3	8	15
10000-50000	4	4	9	20
>50000	5	5	10	25

Table 3. Effectiveness of the Insect Growth Regulator Sumitomo S-31183 (AI3-29835) Against Natural Infestations of Red Imported Fire Ants. Polk Co., FL. 1984-85.

Formulation	Rate of Application		Pretreatment		Percent Reductions after indicated weeks			
					No. nests		Pop'n Index	
	Bait(kg/ha)	AI(g/ha)	No. active nests	Pop'n Index	6	13	6	13
70% PDCG 29.5% ORSBO 0.5% S-31183	1.05 2.1	5.26 10.5	27 25	650 596	11.1 32.0	88.9 72.0	88.6 92.1	98.2 97.2
70% PDCG 29.0 % ORSBO 1.0% S-31183	1.17 2.34	11.71 23.39	25 25	554 593	44.0 12.0	68.0 80.0	92.3 88.2	96.0 96.6
70% PDCG 29.0 % ORSBO 1.0% fenoxycarb (standard)	1.22	12.18	31	731	22.6	80.6	90.7	97.7
Check	-	-	27	612	27.2	29.6	27.2	24.8

Table 4. Effectiveness of the Griffin Fluorocarbon GX-071 in Baits Against Natural Infestations of Red Imported Fire Ants, Polk Co., FL. 1985-86.

Formulation	Rate of Application		Pretreatment		Percent reduction in population index after indicated weeks posttreatment	
	Bait(kg/ha	AI(g/ha)	No. active nests	Pop'n Index	6	13
70.0% PDCG 29.7% ORSBO 0.3% GX-071	0.86	2.58	26	615	66.0	82.0
	1.25	3.75	25	533	63.2	81.8
	1.72	5.16	25	565	75.9	84.2
70.0% PDCG 29.4% ORSBO 0.6% GX-071	0.845	5.07	26	610	78.0	80.7
	1.20	7.20	27	615	80.8	83.9
	1.69	10.14	26	600	88.5	93.7
AMDRO 0.88% (standard)	1.096	9.64	24	560	82.1	82.1
Check	-	-	41	900	+3.6	13.3

EFFECTS OF TWO BAIT TOXICANTS ON NATURAL POPULATIONS OF NONTARGET ANTS

DAVID F. WILLIAMS

USDA,ARS, INSECTS AFFECTING MAN AND ANIMALS RESEARCH LABORATORY, P.O. BOX 14565, GAINESVILLE, FLORIDA 32604

Large scale control programs using chemical baits against the red imported fire ant, Solenopsis invicta began in the early 1960's.

Although the type of effect that all of the bait toxicants exert on a fire ant colony may differ,at the present time, all use a bait system that relies on the chemical being dissolved in an attractant which is soybean oil and applied to a corn grit carrier. A major problem with this system is the large number of nontarget insects, especially beneficial ant species, that are also oil feeders and consequently are affected by the application of these types of baits. For example, studies in Florida by B.Edmunson and by J.L.Stimac indicated that Amdro had an affect on populations of nontarget ant species.

Studies on the effects of Pro-Drone on nontarget ant species is confusing. A study in South Carolina by L.A.Lemke indicated that Pro-Drone caused no deleterious effects on nontarget ants while a study by S.A.Phillips indicated that Pro-Drone applied over a large area in Texas had no effect on either the nontarget or target ant populations. A third study also in Texas by J.M.Lammers and S.B.Vinson indicated that some detrimental effects were caused on nontarget ant species by Pro-Drone.

Because Logic (Maag Agrochemicals) and Affirm (Merck & Company), both fire ant bait toxicants, had shown control effects on fire ant colonies that were different than Amdro and Pro-Drone, we initiated a study with the following objectives:

(1) What effects if any, will these two fire ant bait toxicants have on natural populations of nontarget ants;

(2) If some ant species are affected by one or both of these baits, will their populations return to pretreatment levels; and

(3) Will plots treated with Logic and/or Affirm become infested with S. invicta.

MATERIALS AND METHODS

All plots in the study were set up on 36 acres of improved pasture with mixed pasture grasses on the Santa Fe Correctional Farm property in Alachua County, Gainesville, Florida. To our knowledge, this property has not had any insecticide treatments for at least 40 years and probably has never had insecticides applied to it. The study was began in May 1983 with the first of two pretreatment samples recorded. All baits were applied in August 1983.

There were nine 4-acre plots that were treated. Three plots were treated with Logic at a rate of lb of bait (4 grams AI)

per acre, three plots were treated with Affirm (1b of bait-50mg AI per acre), and three plots designated as the checks were treated with only the soybean oil-corn grit bait but without any toxicant (1b per acre)
Within the center of each 4-acre plot, a 1-acre (square) area containing three 1/8-acre circles was used as the evaluation area.

Evaluation of the ant populations was undertaken every two months using three sampling methods; nest counts, pitfall traps, and soil core samples.
Nest counts are made by having 4-5 individuals form a line along a measuring tape stretched from the center of each 1/8-acre circle out to its edge. The tape is then slowly moved in an arc while all individuals search for and identifies each ant nest and also measures the distance from the center of the circle of each ant nest. In addition, the diameter of all fire ant nests is recorded. The individual standing at the center of the circle, plots the location of each ant nest by using the compass-table device described by Brown et al. 1980 (Fla. Entomol. 63: 257-258).
Pitfall trap locations were determined by using a computer program designated to select random points. Three pitfall traps per 1/8-acre circle were operated for a week during the sampling month. In order to minimize disturbance during this long-range project, a PVC pipe (3 1/2 inch diameter) sleeve was inserted into each pitfall trap hole. The sleeve gave protection and reduced maintenance of the holes. A

rainfall cover was placed over each trap. An 8 oz plastic cup was inserted into the hole and filled with 6 oz of 70% isopropyl. At the end of the one week sampling period, the samples were brought back to the laboratory for sorting and storage.

Soil core samples were utilized to survey populations of Diplorhoptrum spp. only. During a sample period, 27 radii and 6 points along each radius were randomly determined for each 1/8-A circle by a computer program. At each one of the 6 points, a 6-in core sample was taken using a 3/4-in diameter soil sampler. These samples were then returned to the laboratory and the ants removed using the magnesium sulfate suspension method..

The data were analyzed using General Linear Models in the Statistal Analysis System (SAS) (Helwig & Council 1979). The effects of treatment and species X weeks(time) and sampling method were tested for all plots. Also, the interactions between these were analyzed.

Core sample analysis was based on the percent of each sample that contained Diplorhoptrom spp.

During two and one-half years of the study, a total of 18 species of ants were collected using nest counts and 23 species were collected using the pitfall sampling method. Two species of the 18 recorded in the nest count sampling method were not found in pitfall traps while 7 species

collected in the pitfall traps were not seen using the nest count method.
Of the total species collected, regardless of the sampling method used, only 5 species occurred frequently enough throughout the study to analyze their populations. The large majority of the species occurred so infrequent and in such small numbers as to render analyses of their populations difficult.

In conclusion, both Logic and Affirm reduced many of the nontarget ant populations. However, the populations of all ant species affected by these two toxicants have returned to pretreatment levels with some species occurring in greater numbers than before treatment. A few large _S. invicta_ colonies have appeared in three of the evaluation circles (2 treatment and 1 check) in the study area. These fire ant colonies were older, mature colonies and thus, were not from newly-mated queens but from migrations into the evaluation circles of nearby established colonies. Although numerous newly-mated _S. invicta_ queens from recent mating flights were observed landing in treated plots, no new colonies were found in these areas.

THE EFFECT OF AN INSECT GROWTH REGULATOR (PRO-DRONE) ON *SOLENOPSIS INVICTA* BUREN (HYMENOPTERA: FORMICIDAE) AND NON-TARGET ANTS

Sherman A. Phillips, Jr. and Harlan G. Thorvilson

ABSTRACT

Large-scale field trials of an insect growth regulator (Pro-Drone) were conducted in southeastern and southcentral Texas in 1983 and 1984 against the red imported fire ant (RIFA), Solenopsis invicta Buren. Two aerial applications of Pro-Drone at the rate of 11.86 g AI/ha in Kerr and Kendall Cos. had no significant effect on the RIFA nor on non-target ant species in four distinct ecological communities. The use of monthly pitfall sampling did not detect significant differences in species richness, mean number of species, or the number of species within paired habitat types between control and treated plots. However, two aerial applications of Pro-Drone at the rate of 11.86 g AI/ha significantly and effectively reduced RIFA infestations in Chambers and Jefferson Cos. In addition, observations on the rate of bait removal at the time of application revealed that the Pro-Drone bait was removed by RIFAs within the first few hours of application. Since the RIFA was the only arthropod observed foraging of the bait, we conclude that at least the majority of the bait was reaching the target ant.

INTRODUCTION

More than 3,000 chemicals have been evaluated for control of the red and black imported fire ants (IFA), Solenopsis invicta Buren and S. ricteri Forel, since Congress initiated the Federal-State Cooperative IFA eradication program in 1957 (USDA 1976). Of those compounds tested, the most effective were the diene-organo-chlorine insecticides. However, environmental persistence and harmful effects on non-target organisms led the Environmental Protection Agency to cancel registration of these cyclodienes, culminating with Mirex in 1977 (Banks and Schwarz 1980). Since then, intensive research aimed at finding alternative control agents has been implemented. Recently, insect growth regulators (IGRs), with juvenile hormone activity, have warranted further investigation. Though these compounds are non-toxic to S. invicta adults, they do cause deformities in the larvae which are expressed in the adult ant. These IGRs influence imported fire ant fecundity, metamorphosis, and caste determination (Troisi and Riddiford 1974, Vinson et al. 1974, Vinson and Robeau 1974, Robeau and Vinson 1976, Banks et al.1978, Banks and Schwarz 1980, Banks and Harlan 1982, Banks et al. 1983). IGR-induced deformities, death of developing larvae, and a shift in caste differentiation from worker to reproductive forms cause a cessation of worker replacement, resulting in death of the colony (Banks et al. 1983).

Of twenty-six IGRs tested for efficacy against laboratory colonies of S. invicta, one of these compounds (A13-36206) caused 75% colony mortality (Banks et al.1978). Therefore, this IGR [1-(8-methoxy-4, 8-dimethylnonyl)-4-(1-methylethyl) benzene], developed under the trade name of Pro-Drone, was subjected to large-scale field trials (ca. 53,000 ha) in southeastern Texas. In addition, large-scale field testing of this compound was also conducted in Kerr and Kendall Cos. during 1983 to determine the effect of Pro-Drone on non-target ant species. This paper presents the results of our findings.

MATERIALS AND METHODS

Effect on non-target ant species

Large-scale field testing (ca. 200,000 ha) of Pro-Drone was conducted in Kerr and Kendall Cos., Texas. A standardized bait formulation (Stauffer Chemical Co., Westport, CN) was aerially broadcast at a rate of 11.86 g AI/ha 10-20 June 1983 and a second time 26 Sept.-1 Oct. 1983. Based on accessibility, landowner cooperation, and differences in predominant vegetation, eight study sites were selected - four sites in the treated area and four correspondingly similar sites in the untreated area. The four habitat types were juniper-grassland, live oak-grassland, grazed pasture-grassland, and southern cypress-grassland communities. Three treated sites were in Kerr Co., whereas the fourth (southern cypress-grassland community) was located in Kendall Co. Although the control juniper-grassland and live oak-grassland communities were also in Kerr Co., the control southern cypress-grassland and grazed pasture-grassland communities were located in Bandera Co., immediately south of Kerr Co.

A line transect of 20 pitfall traps (437 ml plastic cups containing ethylene glycol placed ca. 5 m apart) was established in each site. Twenty traps were considered the necessary number to sample the ant fauna in each of the four vegetational communities (Wojcik, personal communication). An asphalt roofing shingle was secured above each trap to form a protective cover (Martin 1978). Traps were placed in the field every 3 wks for a period of 7 days from May 1983-April 1984. All ant specimens trapped were identified and tallied. However, several specimens could not be identified to species because key couplets require information as to whether the specimen was from a dimorphic or polymorphic colony (Creighton 1950). Identifications were verified from determined specimens in the Entomological Collection, The Museum, Texas Tech University. Voucher specimens have been preserved and deposited in that collection.

The complete block design consisted of 2,080 subsamples (260 traps/site: 8 sites) collected from the 13 sampling periods (July consisted of 2 sampling periods). Since all experimental units (number of species or number of individuals within a species) were subjected to a set of standardized conditions and therefore treated alike, the mean number of species or individuals within a species occuring each month within each of the four habitats (blocks) was plotted through time for both the treated and control areas (Gill 1978). Analysis of covariance was performed to test the homogeniety of regression coefficients. Data were next analyzed by simple regression followed by a Student's t-test for homogeniety of the two regression slopes. Also, the mean number of species occurring each month for the first six months in the treated and untreated areas were compared by Student's paired t-test, as was the mean number occurring the last six months (Steel and Torrie1980). Comparing species richness among treatments for the first six months and again tor the last six months allowed sufficient replications of samples over time, while minimizing variation resulting from phenological fluctuations (Samways 1983).

Efficacy on target ant species

A large-scale field test (ca. 53,000 ha) of Pro-Drone was conducted in Chambers and Jefferson Cos., Texas. A standardized bait formulation (Stauffer Chem. Co., Westport, CN was aerially broadcast at a rate of 11.86 g AI/ha during late October, 1983, and a second time during late March, 1984. The fall application was timed to weaken the colony before the onset of cold weather, whereas the spring application was timed to maximize product effect on colonies already weakened by the combined actions of the first application and the effect of winter. Based on accessibility and landowner cooperation, ten study sites were selected in grazed, coastal bermuda grass habitats; five sites in the treated area and five sites in the control area. Within each site, six 0.1 ha non-overlapping circles were randomly located. RIFA mounds within each circle were identified with an individual number and the pretreatment location of each mound was recorded according to compass heading (degrees) and distance (meters) between mounds. The control and treated circles contained a total of 289 and 291 RIFA mounds, respectively. Colonies were rated by opening the mounds with a shovel, estimating the number of observed workers, and noting the presence or absence of worker brood. The RIFA mound-rating system described by Harlan et al. (1981), as modified by Lofgren and Williams (1982), was used to obtain a population index for each circle.

The RIFA mounds were monitored for approximately one year. Each mound was rated pretreatment (21 - 22 Oct., 1983), and again at 3, 8, and 10 months after the first treatment (27 - 28 Jan.; 18 - 19 June; and 30 Aug. - 1 Sept., 1984). The sum of all mound ratings within each circle was calculated and designated as the population index (PI) for each site. Analysis of the differences between pretreatment and posttreatment ratings of circles was by Student's paired t-test (Steel and Torrie 1980). Also during the 10-month posttreatment rating, all 0.1 ha circles were again surveyed to determine if the original mounds had fragmented, or if new mounds had been established by newly-mated queens.

Rate of bait removal

Within the treated area, three disjunct sites, separated by 3 -8 km, were located in improved pasture. Approximately 5 h prior to aerial application of Pro-Drone, all vegetation was removed from eighteen 1-m^2 randomly-selected plots (six plots within each site). A standardized bait formulation of Pro-Drone (once-refined soybean oil + IGR incorporated into pregel, defatted corn grits) was aerially broadcast at the rate of 11.86 g AI/ha on 21 Oct. 1983. The number of bait particles falling within each cleared 1-m^2 plot was recorded. Colored pins were placed adjacent to each bait particle to mark its location. The rate of bait removal by foraging RIFAs was monitored at 30 min intervals until all bait was removed. The bait removal rate was expressed as percent of total grits remaining through time. Site A was treated at 1230 h and a total of 383 grits (x = 63.8) were located in the six plots. Since both sites B and C were treated at 1600 h by 9 aircraft, data from these two sites were combined and totaled 451 grits (x = 37.6) in all six plots. Data obtained from all sites were analyzed by simple linear regression.

RESULTS AND DISCUSSION

Effect on non-target ant species

Twenty-four taxa representing five subfamilies (Dolichoderinae, Dorylinae, Formicinae, Myrmicinae, and Ponerinae) were detected in this year-long study (Table 1). Each taxon identified from the eight study sites was found in at least one community from both the test and control area. Based on frequency of capture, S. invicta appears to be the dominant species, in that it was detected a total of 71 times. The most frequently detected non-target ants were Pheidole sp., Monomorium minimum (Buckley), Paratrechina melanderi (Wheeler), Pachychondyla harpax (F.), and Forelius pruinosus (Roger).

Table 1. List of the 24 ant species in descending order of occurrence detected in treated and untreated areas from May 1983 - April 1984. (260 collections/site: 4 sites/area). Each taxon identified from the study was detected in at least one community from both the test and control area.

	No. Times Detected[1]		
SPECIES	Treated	Untreated	Total
Solenopsis invicta Buren	35	36	71
Pheidole sp.[2]	29	35	64
Monomorium minimum (Buckley)	25	35	60
Paratrechina melanderi (Wheeler)	26	31	57
Pachychondyla harpax (F.)	20	25	45
Forelius pruinosus (Roger)	16	21	37
Atta texana (Buckley)	14	19	33
Crematogaster laeviuscula Mayr	12	20	32
Forelius foetidus (Buckley)	11	19	30
Solenopsis (Diploroptrum)	12	15	27
Solenopsis geminata (F.)	3	22	25
Pogonomyrmex barbatus (Smith)	13	10	23
Conomyrma bicolor (Wheeler)	12	10	22
Leptogenys elongata (Buckley)	11	10	21
Labidus coecus (Latreille)	7	13	20
Odontomachus clarus Roger	16	3	19
Conomyrma insana (Buckley)	8	5	13

(Continued)

Camponotus sp.[2]	5	7	12
Strummigenys louisianae Roger	6	6	12
Neivamyrmex opacithorax (Emery)	1	8	9
Tetramorium spinosus (Wheeler)	6	3	9
Crematogaster punctulata (Emery)	1	7	8
Brachymyrmex depilis Emery	2	3	5
Myrmicina americana Emery	3	1	4

[1] Maximum possible times detected: 96 (8 sites sampled monthly for 12 months). Each trap is a subsample of the transect.

[2] Unable to identify to species (See text).

Analysis of covariance indicated that the variance in species richness among subjects (blocks) was the same at each sampling point in time (months) and, therefore, the covariance for any two sampling points were homogeneous (Gill 1978). Because of this homogeniety of variance among blocks, the mean number of species occurring in each community for the test and control could legitimately be regressed through time (Fig. 1). Regression indicates that the relationship between species richness and time in both test and control areas was linear ($P<0.01$). In addition, the high coefficients of determination (control: $r^2 = 0.92$; test: $r^2 = 0.92$) indicate that the linear model describes the data well for both treatments. Test for homogeniety of regression slopes between test and control indicates no significant difference ($t = 0.059$; d.f. = 20; $P>0.05$). Therefore, species richness in the test area was no different than that of the control area through time.

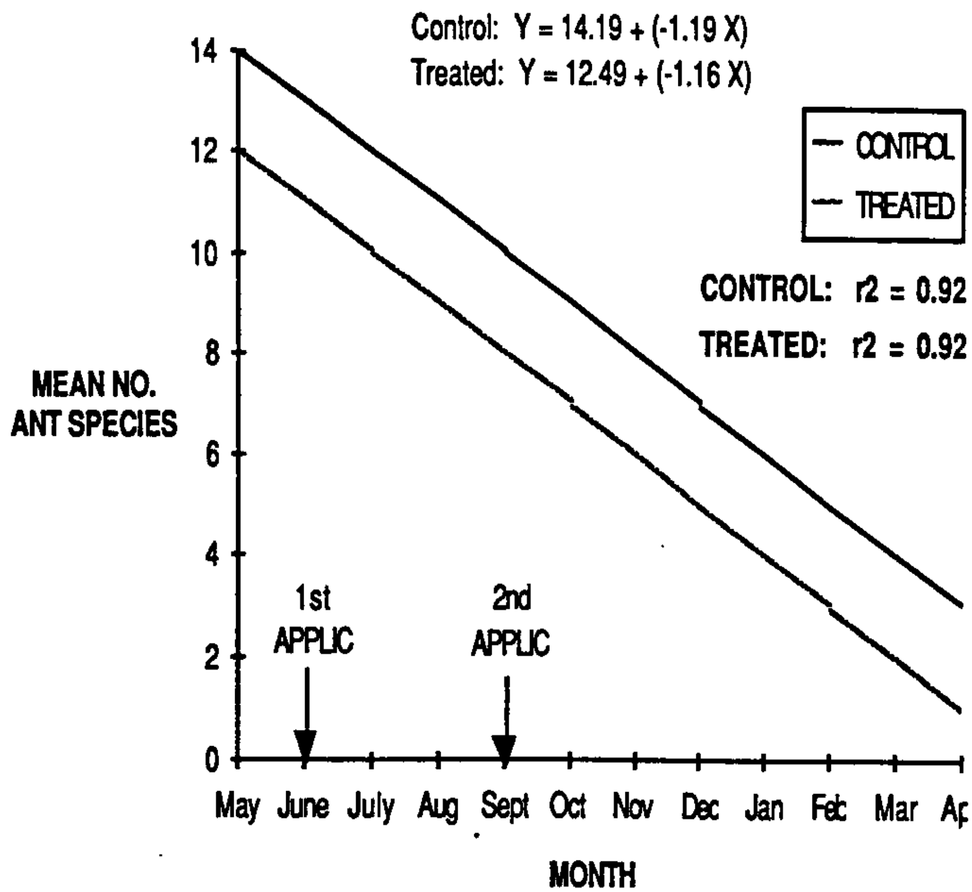

Fig. 1. Effect of Pro-Drone on mean number of ant species from May 1983-April 1984. Data analyzed by simple linear regression.

Replicating species richness through time (Table 2) and comparing means detected between test and control areas for the first six months, no significant difference was detected ($t = 1.07$; d.f. = 10; $P>0.05$). Additionally, no significant difference was detected in means between treatments for the last six months ($t = 2.04$; d.f. = 10; $P>0.05$). Finally, no significant differences were detected in species richness of the habitats or subfamilies detected in this study between the test and control areas through time, nor in the number of individual per species (including S. invicta) between the two treatments through time.

Table 2. The effect of Pro-Drone on species richness of ants in Kerr and Bandera Cos. Texas (1983-1984). Treated and control areas were compared to each other for the first six months as were the treated and control areas for the last six months (Student's paired t-test).

Mean No. Ant Species			
First 6 Months (May - Oct.)		Last 6 Months (Nov. - April)	
Treated	Untreated	Treated	Untreated
11.25	11.50	4.00	6.25
10.50	10.50	1.75	4.50
12.75	16.75	2.00	3.00
10.25	13.25	1.25	2.75
9.00	10.25	2.00	3.00
7.00	8.00	1.25	2.00
t = **1.07** N. S.		t = **2.04** N. S.	

Student's paired t-test (P>0.05; df = 10; **critical t = 2.228)**

Various researchers have utilized pitfall traps to study the foraging behavior of fire ants and the community structure of other ants (Apperson and Powell 1984; Samways 1983). Although various problems are associated with interpreting arthropod sampling by this technique, the method does provide reliable information as to annual periodicity of activity and species composition of a community (Adis 1979). In addition, pitfall traps allow for the relative comparison between different areas of a habitat within the same sampling periods. As noted from Table 1, species richness of the two treatments for the entire year were the same, indicating that the two localities were homogeneous. Four distinct habitats were chosen in each area to detect as many species of ground foraging ants as possible.

Although the posttreatment number of species detected each month was lower in the treated than in the control area, the numbers detected pretreatment were also lower. The largest numbers were detected at the beginning of the study probably because the greatest foraging activity occurred in May and June. This decline in number of species through time was most likely a lack of detection of the less common species as a result of an overall decline in foraging activity with the onset of cooler weather, and not the result of Pro-Drone application. Foraging activity continued to be negatively impacted by the unusually low winter temperatures and extremely dry spring. Therefore, the number of species detected did not increase with the onset of spring as would normally have been expected. However, since neither the number of individuals within each species nor the number of species in each habitat or in each subfamily was significantly lower in the treated area than in the control area, Pro-Drone apparently had no significant effect on the ant fauna (non-target and target species) in Kerr and Kendall Cos., Texas.

Efficacy on target ant species

Control sites

The 3- and 8-month posttreatment control ratings (Table 3) were each significantly different ($P<0.01$) from the pretreatment control ratings. These differences were expected because the 3- and 8-month readings occurred during the winter and spring months, respectively. The lower ratings were likely caused by reduced brood rearing during the cooler seasons of the year. Even the low ratings at 8-months posttreatment (as evidenced by PIs) indicate that colonies had not yet recovered from the effects of winter. However, a general increase in PI from the 3- to 8-month recordings was noted, indicating colony recovery at the control sites. Finally, the 10-month difference in individual mound ratings compared to pretreatment mound ratings was not significantly different ($P>0.05$). This recovery would be expected, since the final mound ratings occurred during the same season (different year) as the pretreatment ratings.

Table 3. Large-scale field test results of aerially applied Pro-Drone against the red imported fire ant, S. invicta, in Jefferson and Chambers Counties, Texas (1983-1984). Posttreatment PIs in control sites were compared to pretreatment PIs in control sites.

No. Mounds controls	Population index	Population index		
	Pretreatment	Posttreatment		
		3-months	8-months	10-months
(43)	198	87	451	100
(38)	265	125	211	76
(72)	788	201	218	782
(65)	628	173	437	712
(71)	865	249	538	785
Student's paired t-test (P<0.01; df=29)		t=6.48**	t=2.9	t=2.03ns

Treated sites

The 3, 8, and 10-month posttreatment ratings from the treated areas (Table 4) were significantly different (P<0.01) from pretreatment ratings. Although the 3- and 8-month PIs were expectedly lower because of cooler winter and spring weather, the 10-month reading is also significantly lower (P<0.01) than the pretreatment reading. Mound monitoring was unnecessary after the 10-month rating for the following two reasons: lower PIs would not be a function of treatment, but a function of colony phenology due to lower winter temperatures; and, effects of this IGR would be detectable within one year of initial treatment. Because colonies from the 10-month control locations (Table 3) fully recovered and the treated colonies (Table 4) had not recovered, the decline in PIs for the treated colonies is attributed to the treatment. In addition, 236 colonies in the treated area were dead, whereas only 70 dead colonies were recorded from the control area, resulting in 81.1% and 24.2% mortality, respectively. Colony death in the control area is attributed to natural mortality. Finally, no worker brood was detected in any of the treated colonies (0.0%), whereas 133 control colonies (46.0%) contained worker brood.

Table 4. Large-scale field test results of aerially applied Pro-Drone against the red imported fire ant, S. invicta, in Jefferson and Chambers Counties, Texas (1983-1984). Posttreatment PIs in treated sites were compared to pretreatment Pis in treated sites.

No. Mounds treatment	Population index	Population index		
	Pretreatment	Posttreatment		
		3-months	8-months	10-months
(42)	580	129	196	23
(95)	1087	337	171	9
(43)	428	115	103	18
(64)	626	210	327	25
(47)	364	138	196	8
Student's paired t-test ($P<0.01$; df=29)		t=6.9**	t=5.4**	t=7.8**

Rate of bait removal

RIFAs removed ca 50% and 70% of the IGR bait ca. 1 h after the 1230 h (early) and 1600 h (late) applications, respectively (Fig. 2). Four and one-half hours elapsed from the early application before 90% removal occurred, whereas 90% bait removal occurred within 2.5 h of the late application. Since higher ground and air temperatures existed at 1600 h (g = 27 C; a = 29 C) compared to 1230 h (g = 20 C; a = 23 C), greater numbers of foragers would be expected at that time. Therefore the steeper negative slope value (-0.46 as comparred to -0.20) indicates greater foraging activity. Test for homogeniety ($t = 2.17$; $df = 17$) indicates that the slopes are significantly different ($P<0.05$). The high coefficients of determination (Site A: $r^2 = 0.88$; Sites B and C combined: $r^2 = 0.87$) indicate that the linear model describes the data well during both time periods. Therefore, these coefficients indicate that the RIFA was thoroughly "canvassing" the sites and that bait removal was a function of time.

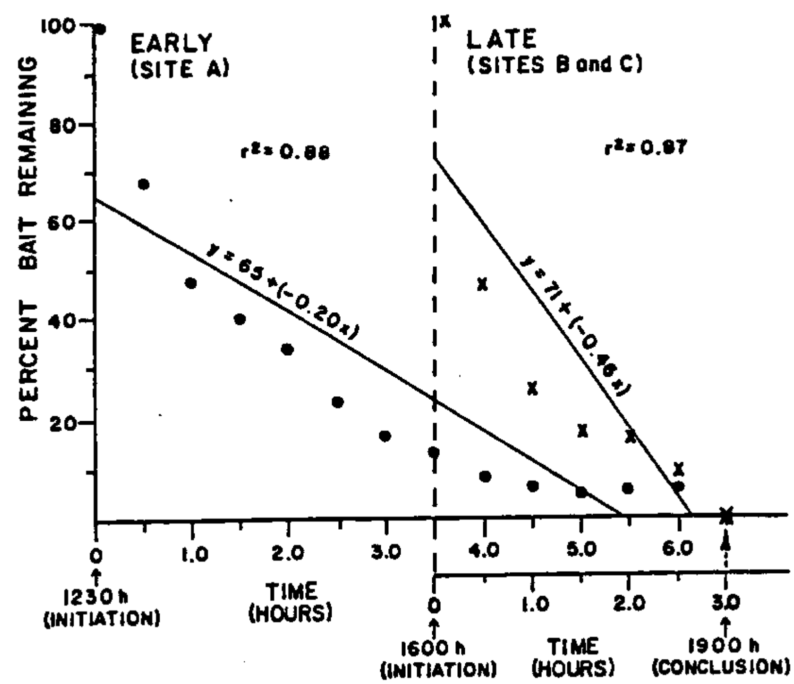

Fig. 2. Percent insect growth regulator (Pro-Drone) bait remaining every 30 min. at three disjunct locations in red imported fire ant, Solenopsis invicta, infested area, 21 October 1983 (data from sites B and C are combined--see text).

Conclusion

In conclusion, no effect was detected on either the non-target ant species or the target pest (RIFA) in Kendall and Kerr Cos., Texas, as a result of the Pro-Drone application of 1983. However, Pro-Drone significantly reduced the population index of the red imported fire ant, S. invicta, in Chambers and Jefferson Cos., Texas, between 1983 and 1984. In addition, no fragmentation of treated colonies was detected 10-months posttreatment. Finally, although Pro-Drone bait is formulated for RIFAs, other "oil-loving" ant species may be attracted to the bait. However, we observed no other invertebrate nor any vertebrate species foraging on the baits. Since all of the bait was collected by the foraging RIFA within a short time period, we suspect that at least a large majority of bait-formulated Pro-Drone reached the target organism. Therefore, these data indicate that Pro-Drone may be an effective control tactic for use against the red imported fire ant.

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APPENDIX A

PARTICIPANTS IN THE 1986 IMPORTED FIRE ANT CONFERENCE

Charles F. Adams
American Cyanamid Co.
Agricultural Division
Wayne, NJ 07470

David Austin
100 Shelmar Dr.
Euless, TX 76039

Richard Bagley
MAAG Agrochemicals
P. O. Box X
Vero Beach, FL 32961-3023

William Banks
USDA-ARS-Imported Fire Ant Research
2701 S.W. 100th St.
Gainesville, FL 32607

Charles Bare
USDA/APHIS/PPQ
6505 Belcrest Rd., Rm. 633
Hyattsville, MD 20782

Thomas Betz
Texas Dept. of Health
1100 W. 49th
Austin, TX 78756

Dr. Awinash Bhatkar
Texas A&M University
Dept. of Entomology
College Station, TX 78743

John Bowen
University of Georgia
Dept. of Physiology & Pharmacology
Athens, GA 30602

Marion Bradford
A.E. Staley
2200 East Eldorado St.
Decatur, IL 62525

Ralph A. Bram
USDA, ARS
Rm. 211, B-005, BARC-West
Beltsville, MD 20705

Ernest Bredefeld
Chevron Chemical Co.
6750 Poplar Ave., Suite 300
Memphis, TN 38138

Tom Bridges
MAAG Agrochemicals, Inc.
P. O. Box X
Vero Beach, FL 32901-3023

Roy Burton
Burton Pest Control
2007 North Mays, Suite 105
Round Rock, TX 78664

Ken Chisholm
PBI Gordon
6 Bent Ln.
Newark, DE 19711

James Cokendolpher
Texas Tech University
Dept. of Entomology
Lubbock, TX 79409

Homer Collins
USDA-APHIS-PPQ
Methods Development
Gulfport, MS 39503

John Connolly
The ANTSER Corporation
321 Folwell
Houston, TX 77024

Bobby Davis
Texas Dept. of Health
1100 W. 49th
Austin, TX 78756

Roxanne Davis
Plant Genetics Inc.
1930 Fifth St.
Davis, CA 95616

Dr. Bastiaan Drees
Texas Agricultural Ext. Service
P. O. Box 2150
Bryan, TX 77806

Ray Dunton
Texas A&M University
Dept. of Entomology
College Station, TX 78743

Dr. John Ellison
Texas A&M University
Dept. of Biochemistry
College Station, TX 78743

Robert Eplee
USDA-APHIS
P. O. Box 279
Whiteville, NC 28472

R. Terry Ervin
Texas Tech University
Dept. of Ag Economics
Lubbock, TX 79408

Chas. Wayne Evans II
SAC Research Center
5000 Belmont St.
Beaumont, TX 77707

Homer E. Fairchild
USDA-APHIS-PPQ
Methods Development
Hyattsville, MD 20782

Theodore Freeman
United States Air Force
9326 Lamerton Rd.
San Antonio, TX 78250

Ramon Georgis
Biosis
1057 East Meadow Circle
Palo Alto, CA 94303

B. Michael Glancey
USDA-ARS IFA Project
3925 NW 37th Place
Gainesville, FL 32606

Brooks Goodhue

SAC Research Center
5000 Belmont St.
Beaumont, TX 77707

John Goodhue
SAC Research Center
5000 Belcrest St.
Beaumont, TX 77707

Dr. Les Greenberg
Texas A&M University
Dept. of Entomology
College Station, TX 78743

Dr. Jane A. Greenblatt
Merck Sharp & Dohme Research Lab
12511 Hunters Chase
Austin, TX 78729

Monroe Hall
Illinois Cereal Mills
616 S. Jefferson
Paris, IL 61944

Dr. Phillip Hamman
Texas Ag. Extension Service
Texas A&M University
College Station, TX 77843

Don Harlan
503 Auburn
W. Memphis, AR 72301

Lance Hemsarth
Lehn & Fink Products Group
7 Mercedes Dr.
Montvale, NJ 07645

Richard Herlich
American Cyanamid
1 Cyanamid Plaza
Wayne, NJ 07470

Harry Howell
Texas A&M University
Dept. of Entomology
College Station, TX 77843

Jackie Jacobson
Lone Star Chapter Sierra Club

201 Skyline Drive
Austin, TX 78746

Dr. Frank K. James, Jr.
8601 Village Drive
Suite 202
San Antonio, TX 72817

Dr. Spencer Johnston
Texas A&M University
Dept. of Entomology
College Station, TX 78743

Ron Kuehler
EcoLogic Pest Mgmt.
3301 Hancock, Suite 6
Austin, TX 78731

Arthur Kushner
Biosis
1057 E. Meadow Circle
Palo Alto, CA 94303

Jim Lammers
University of California
Dept. of Entomology
Berkeley, CA 94720

Neil Lapp
Merck & Co., Inc.
7208 Madiera Ct.
Raleigh, NC 27609

C.W. Lawler
Rampart Industries
15102 Sommermeyer
Houston, TX 77041

Dr. Clifford Lofgren
USDA-SEA-AR
P. O. Box 14565
Gainesville, FL 32604

Roland Lovelace
Cotton's Fire Ant Killer
P. O. Box 1588
Rockdale, TX 76567

Jeff Luedke
Green Light Company
San Antonio, TX 78217

Barney Mahan
Barney Mahan Inc.
Rt. 8, Box 99-T
Waco, TX 76705

Pam Mavrolas
Texas Center for Rural Studies
Box 2618
Austin, TX 78768

Lynn McGuire
Cotton Products, Inc.
P. O. Box 1588
Rockdale, TX 76567

Susan McKeon
Research Technician - Fire Ants
P. O. Box CSB, 402 Baylor
Waco, TX 76798

Alex C. Mintzer
Dept. of Entomology
Texas A & M University
College Station, TX 77843

Michael E. Mispagel
University of Georgia
110 Flannigans Pl.
Athens, GA 30605

Paul Munroe
Texas Tech University
P. O. Box 4169
Lubbock, TX 79409

Dr. Ana Ortiz
United States Army
13109 Hunters Ledge
San Antonio, TX 78230

Clair Palmer
MAAG Agrochemical Inc.
P. O. Box 6430
Vero Beach, FL 32961

N. Robert Pasarela
American Cyanamid Co.
Box 400
Princeton, NJ 807

Sherman Phillips, Jr.
Texas Tech University
Dept. of Entomology
Lubbock, TX 79409

Don E. Rawlins
American Farm Bureau Federation
225 Touhy Ave.
Park Ridge, IL 60068

Dr. T.E. Reagan
Louisiana State University
Dept. of Entomology
Baton Rouge, LA 70803

Greg Rich
Chevron Chemical
6750 Poplar Ave., Suite 300
Memphis, TN 38138

Dr. Dick Richardson
University of Texas
Dept. of Zoology
Austin, TX 78712

David T. Rowlands
American Cyanamid
3197 Ferns Glen Dr.
Tallahassee, FL 32308

Max C. Sconyers
Mobay Chemical Corp.
5215 16th St.
Vero Beach, FL 32960

Craig Sheppard
University of Georgia
Dept. of Entomology & Fisheries
Tifton, GA 31794

John Shriver
Merck & Co., Inc.
P. O. Box 459
Paris, IL 61944

Paul Sisson
EcoLogic Pest Mgmt.
3301 Hancock, Suite 6
Austin, TX 78731

Ray Smith
Ciba-Geigy
2807 S. Wilderness
College Station, TX 77840

Dr. Ann Sorensen
American Farm Bureau Federation
225 Touhy Ave.
Park Ridge, IL 60068

Terrell Stamps
Texas A&M University
Dept. of Entomology
College Station, TX 78743

Martin Stein
Texas Tech University
Dept. of Entomology
Lubbock, TX 79409

Robert Stevens
American Cyanamid
P. O. Box 2577
Nacogdoches, TX 75961

Stephen Taber
Texas Tech University
Dept. of Entomology
Lubbock, TX 79409

Glenna Teltow
Texas Dept. of Health
1100 W. 49th
Austin, TX 78756

Dr. Harlan Thorvilson
Texas Tech University
Dept. of Entomology
Lubbock, TX 79409

Glynn Tillman
Ciba-Geigy

College Station, TX

Mark Trostle

Texas Department of Agriculture
P. O. Box 12847
Austin, TX 78711

Steve Turner
Griffin Corporation
P. O. Box 1847
Valdosta, GA 31601

Antonio Carlos C. Pereira
EMBRAPA
Antigo Posto Agropecuario - EMPA-Lacere/MT
Brasil - Laixa Postal: 191 78·700

Bill Van Eimeren
4209 Ave. B #109
Austin, TX 78751

Harold G. Fowler
University of Florida
Instituto De Biociencias, UNESP
Rio Claro, SP, Brazil 13500

Robert Vander Meer
USDA-ARS
1600 SW 23rd St.
Gainesville, FL 32604

A.D. Ali
University of California
Cooperative Extension
Riverside, CA 92521

S. Bradleigh Vinson
Texas A&M University
Dept. of Entomology
College Station, TX 78743

C. Ronald Carroll
University of California
2120 University Ave.
Berkeley, CA 94720

Deborah A. Waller
Louisiana State University
Entomology Dept.
Baton Rouge, LA 70803

Gerald King
Arkansas State Plant Board
P. O. Box 1060
Little Rock, AR 72103

Penny Wilkinson
Texas A&M University
Dept. of Entomology
College Station, TX 78743

Robert Reeves
USDA-APHIS-NMRAC
P. O. Box 3209
Gulfport, MS 39503

David F. Williams
USDA-SEA-ARS
P. O. Box 14565
Gainesville, FL 32604

Diane Weigle
University of Florida
6224 SW 8th Place
Gainesville, FL 32607

David Wilson
University of Georgy
Dept. of Physiology & Pharmacology
Athens, GA 30602

David Wiemer
University of Iowa
Dept. of Chemistry
Iowa City, IA 52242

Daniel Wojcik
USDA-SEA-AR
P. O. Box 14565
Gainesville, FL 32604