

ARGENTINE ANTS, *LINEPITHEMA HUMILE*, (HYMENOPTERA: FORMICIDAE) IN LOUISIANA: ANT-ANT COMPETITION AND TREATMENT WITH INSECTICIDES

LINDA M. HOOPER-BÙI, ZHISHENG JIANG AND JESSICA L. ROSSON

Department of Entomology, Louisiana State University, 404 Life Science Building LSU Baton Rouge, LA 70808

Abstract The Argentine ants nest in the ground and forage into hundreds of trees and houses in the area. We investigated the effect of competition between Argentine ants and red imported fire ants. When equal numbers of Argentine ants and fire ants interacted, fire ants were more successful than Argentines, which faced great mortality risk. However, when large numbers of Argentines interacted with fewer fire ants, entire colonies of fire ants were quickly decimated. We also tested several methods of controlling Argentine ants on a large scale. We tested contact insecticide barrier sprays, broadcasts of liquids and granules, and liquid and solid baits. Some of these efforts suppressed the ants for a short time, while others appeared to be ineffective. Several hypotheses were tested: contact insecticides were sprayed around the trees in conjunction with large-scale baiting to reduce ant populations; pesticide degradation or runoff was investigated by testing the soil for pesticide residue; and, ants were tested for pesticide detoxification enzymes. Contact insecticides around the trees reduced the number of ants foraging in the trees but did not eliminate. The granular bait that was broadcast combined with strategically placed liquid bait stations further reduced the population of Argentine ants. Some of the contact insecticides that were applied were not present in tested treated soil one month after treatment. Subsequent experiments provide evidence that there is a runoff. Argentine ants from Toledo Bend had 7.5 times more glutathione-S-transferase activity than ants from California, which have never been exposed to insecticides. These results may explain some of the difficulty in managing Argentine ants.

Key Words Insecticide toxicology, ant behavior, aggressive interactions

INTRODUCTION

Linepithema humile (Mayr) is a cosmopolitan ant from South America that has invaded the Mediterranean and subtropical habitats worldwide (Suarez et al., 2001). *Solenopsis invicta* Buren is also invasive and originally from South America and has invaded Puerto Rico, Australia, and New Zealand, as well as the entire southeastern United States and California. Competitive displacement of native ant species by each of these ants has been studied (Crowell, 1968; Hung and Vinson, 1978; Banks and Williams, 1989; Human and Gordon, 1997; 1999; Human et al., 1998; Holway, 1998a; 1999; Morrison, 2000; 2002). The interactions of these two invasive ant species in their expansion habitat has not been examined.

The study of invasive ants in ecology is important because invasions provide researchers unique opportunities to study community dynamics such as competition (Diamond and Case, 1986) and disruption of community structure (Fisher, 1998; Holway, 1999) because ants have many vital roles in the ecosystem, acting as mutualists, earth-movers, pollinators, predators, and herbivores. Abiotic factors such as temperature, humidity, vegetation type, and soil characteristics affect the microhabitat where a particular ant species will live and how it will behave and subsequently, the distribution and the rate of spread. Resource availability and the ability of ants to exploit resources faster and longer and to defend their territory also influence ant abundance and diversity. Determining what ecological, behavioral, and genetic factors are most likely to limit a population of ants or influence its growth and spread has been the focus of many ant invasion studies (Holway et al., 2002a).

L. humile and *S. invicta* have been widely studied due to their overwhelming success as single invaders. Both of these ant species have had devastating effects on native ant communities and vertebrate fauna. Superior exploitative and interference abilities, escape from natural enemies, successful reproductive strategies, and ties to human-mediated activities have made these ants top competitors. However, few studies have been conducted in invaded, non-native areas which include both Argentine ants and red imported fire ants, and further research should also examine Argentine ants and red imported fire ants in native areas (Holway et al., 2002a). Argentine ants spread 6-100 m/year through colony budding in riparian corridors (Holway, 1995; Holway, 1998b; Crowell, 1968; Erickson, 1971). Holway (1995) discovered Argentine ants steadily expand their territory in northern California, in short distances by natural diffusion through colony budding or in long distances through human-

mediated jump dispersal. Human-mediated jump dispersal has resulted in the establishment of this species on six continents (Africa, Asia, Australia, Europe, southeastern portion of North America, South America) and on many oceanic islands in the Atlantic, Mediterranean, and Pacific oceans (Suarez et al., 2001). The rate of invasion for *Solenopsis invicta* is remarkably similar to that of Argentine ants (Tsutsui and Suarez, 2003) but they also have mating flights that allow rapid natural range expansion. Fire ants have recently spread to coastal California, Hawaii, Puerto Rico, and Australia where Argentine ants have previously invaded (Taber, 2000). Two different forms of competition often occur in the ecosystems, exploitative competition and interference competition. Exploitative competition involves the ability of an ant species to locate a resource quickly and recruit high numbers of workers to the resource before other species of ants arrive. Argentine ants are better at exploiting food resources than native ants in California (Human and Gordon, 1996). Interference competition involves the ability of an ant species to defend a resource from other ant species or dominate it by attacking the ants already there. Successful invasive ants may be more aggressive than native ant species, as in Argentine ants, and are likely to initiate encounters with another ant species prompting that species to retreat (Human and Gordon, 1999).

The large numbers of Argentine ants, physical aggression, and chemical defensive compounds are also worth noting when considering Argentine ants' proficiency at interference competition (Holway 1999), which often result in the displacement of native ants especially epigeic species, creating less diverse communities (Crowell, 1968; Human and Gordon, 1997; 1999; Human et al., 1998; Holway, 1998a; 1999). As early as 1908, the Argentine ant had displaced a potentially beneficial ant species, *Solenopsis geminata* Fabr. and other ant species from the Audubon Park region of New Orleans (Foster, 1908).

Fire ants are also fierce competitors but are less efficient at foraging than some native ants; yet native ants may be out-competed in interspecific interactions because of large fire ant colony sizes (Jones and Phillips, 1990) creating less diverse communities (Hung and Vinson, 1978; Banks and Williams, 1989). As fire ant colony numbers increase, native ant populations are reduced due to direct predation or indirectly through competition for food or nest sites (Wojcik, 1983). Like Argentine ants, fire ants also adversely affect other arthropod assemblages in a variety of habitats (Porter et al., 1988; Porter and Savignano, 1990). Polygynous *S. invicta* resemble Argentine ants in its absence of intraspecific aggression (Human and Gordon, 1996; Holway, 1999).

The objective of the aggression laboratory study was to measure aggression, as a form of interference competition, occurring between Argentine ants and red imported fire ants. These studies of aggressive behavior offer insight into what occurs at the Argentine ant / fire ant interface. Although this study includes nestmate and non-nestmate interactions, nestmate recognition was not the focus of this study. Many aggression bioassays have been performed with the objective of determining the level of nestmate recognition (Roulston et al., 2003), because nestmate recognition is an important factor to consider when studying social insects such as ants (Vander Meer and Morel, 1998). We also examined interactions between large colonies of both species in the field.

Argentine ants have been subjected to large amounts of contact insecticides. However, it is unknown whether they are able to detoxify the insecticides to which they are exposed. Glutathione S-transferases (GSTs) are known to catalyze conjugations by facilitating the nucleophilic attack of the sulfhydryl group of endogenous reduced glutathione on electrophilic centers of a vast range of xenobiotic compounds, including insecticides. GSTs have been described to play a major role as a detoxification mechanism for insecticides, thus contributing to insecticide resistance in economically important pest species in diverse agronomic cropping systems as well as disease vectors such as mosquitoes. GSTs have been shown to be involved in the detoxification of several chemical classes of insecticides, i.e., organophosphates, pyrethroids, carbamates, and chlorinated hydrocarbons such as DDT, many of which are used for Argentine ant control all over the world. GST-based resistance to insecticides was described to be facilitated by the increase in the level of expression of one or more GSTs.

Esterases detoxify insecticides that contain ester-type groups and are considered a Phase I reaction that do not require high energy intermediates. There are two classes of esterases, A-type and B-type. The A-type are insensitive to OP inhibition and include aryl-esterases (phosphotriester hydrolases). The B-type are OP inhibition sensitive and hydrolyze a variety of aromatic aliphatic esters, and include cholinesterase, carboxylase, and lipases. Esterases are extremely important for the metabolism of ester-containing allelochemicals and insecticides in insects and often play key roles in the development of resistance.

MATERIALS AND METHODS

Aggression Study. Two different laboratory experiments designed to detect interspecific and intraspecific aggression in both species were used. The first was one-on-one interactions where individuals of the same or different species were observed together, and the second was ten-on-ten interactions where groups of ten ants from the same or different species were observed.

One-on-one Interactions. To investigate aggressive behavior between individual ants of the same or different species, bioassays were conducted to observe initial encounters between ants. This first experiment was conducted in small 35 x 10 mm plastic tissue culture dishes (Becton Dickinson, Falcon 35-3001, Easy Grip) lined with Teflon (fluoropolymer resin, Dupont, Wilmington, DE, production date 05-Jun-03) to prevent ant escape. Two ants were placed in each Petri dish and were immediately observed until their first encounter. The ant that initiated the encounter was noted, and the initial encounter was scored as retreat, neutral, or aggressive, and the response to this encounter was scored as retreat, neutral, or aggressive. This aggression rating scale is modified from Human and Gordon's (1999) study on behavioral interactions of *L. humile* with native ant species. Neutral behavior was characterized by the absence of retreat or aggressive behavior or one ant antennating another ant. Retreats were characterized by an ant running away after encountering another ant or by spasm which includes curling up and cleaning antennae or legs or moving legs and body spasmodically.

Aggressive behavior was characterized by chasing another ant, biting another ant, directing the gaster toward another, or stinging another ant. All interactions involved monogynous *S. invicta*, and both lab-reared colonies (> 6 months in rearing room) and recently collected (< 36 hours in laboratory) field colonies were used. Lab-reared *S. invicta* tend to be less aggressive toward humans than *S. invicta* collected directly from the field, possibly because the ants may be satiated (pers. obs.). All *L. humile* were collected from the field and placed in containers with sides coated with Teflon. Although *L. humile* were collected from different areas at a site, they may not actually be from different colonies since they are known to form supercolonies (Tsutsui et al., 2000; Giraud et al., 2002). There were 10 replications for each interaction.

Ten-on-ten Interactions. To observe aggressive behavior that occurs when larger equal-sized groups of competing ants (same species or different) are present, another set of bioassays was conducted with groups of ten ants of each type (lab-reared *S. invicta*, field-collected *S. invicta*, and *L. humile*). The second experiment was set up similar to the first. Rectangular acrylic containers (17.2 cm L x 8 cm W x 4 cm H) with Teflon on the sides were used as arenas for each of the interactions. Ten ants of each type were used for a total of 20 ants in each container. In instances when both types of ants were *S. invicta*, white non-toxic paint on the rounded end of an insect pin (Painters, Hunt Corporation, Statesville, NC) was used to mark one set of the ants. Preliminary tests showed no differences in behavior or mortality between painted ants and non-painted ants. Interactions with all *L. humile* were performed, but the *L. humile* were too delicate to paint. In these interactions, aggression ratings were assigned, but they were assigned to the whole group, not just to the ten ants belonging to the same group for all experiments. Aggression ratings and mortality were assigned at first contact, after 15 minutes, after 30 minutes, after 45 minutes, and after 60 minutes for each group of 10 ants. Ten replications were performed for each interaction. We also conducted small tests in the field where large colonies of *S. invicta* were brought into areas infested with *L. humile*. The colonies were observed for 1 hour and again after 24 hours.

Statistical Analyses. The one-on-one interactions were analyzed with mixed model analysis and chi-square tables to determine the significance of role (initiator or respondent), the type of ant (lab-reared *S. invicta*, field-collected *S. invicta*, or Argentine ant), and aggressive behavior between intraspecific and interspecific interactions. Ten-on-ten interactions were analyzed with mixed model analysis and chi-square tables to determine the significance of aggressive behavior in intraspecific and interspecific interactions, type of ant, and aggressive behavior of initiating ant. Mortality data from the ten-on-ten interactions were analyzed using chi-square tables, logistic regression, and mixed model analyses. All analyses were performed using the SAS version 9.0 software package (2002, SAS Institute Inc., Cary, NC).

Large-scale Treatment

Argentine ants were targeted for a large-scale treatment in August 2004. We applied malathion and cypermethrin around the base of pine and hardwood trees 1m up and 0.3 m out. Care was taken to ensure that the cracks and crevices in the bark were treated. We attempted to treat every tree in this heavily wooded area. With the cooperation of the residents, we broadcast MaxForce (hydramethylnon) granular bait over the entire area (1.5pounds/acre). We treated approximately 50 hectares.

Pesticide Application. In a separate study, we applied liquid bifenthrin (1 oz/1000 ft²) as a barrier around the foundation of houses in Toledo Bend, Louisiana. At some of the houses, we also broadcast granular bifenthrin or liquid bifenthrin over the entire yard. We removed soil and tested the soil for presence of bifenthrin monthly for four months.

Pesticide Extraction from Soil. For the extraction phase of the experiment, we spiked 25 g of soil with a known amount of permethrin to record percent recovery, then we also spiked 25 g of soil with a known amount of bifenthrin to discover percent recovery. Once percent recovery is known, extraction for both bifenthrin were similar and were as follows. 125 ml flasks were washed with TWEEN 20 (Polyoxyethylene-20-sorbitan Monolaurate, 98% purity) and dried. After weighing each flask, 25 g of soil was added and heated over night at 100°C. The weight of the soil in each flask was recorded again. 40ml HPLC-grade acetone was added to the soil and Cypermethrin (CY-40) technical standard was added in the solution. The top of flasks were covered with Reynolds Wrap[®] aluminum foil to prevent evaporation of acetone. The soil mixed with acetone was placed on a shaker (InnOva Platform Shaker 2100) at 200 rpm for 4 hrs. One COORS USA 60240 ceramic funnel was placed in each 125 ml flask and then one Whatman Glass Microfibre Filter (GF/F Circles 55mm Ø Cat No 1825055) into the ceramic funnel. One Whatman Folded Filter Paper 2V (Qualitative 100 circles 125mm Ø) was placed into the funnel and the soil sample was poured into the filter and all liquid was completely drained into the flask. The next step was to place one F Prep Sep[™] Extraction Column Florisil into a 125 ml flask and the filtrate was poured into florisil column and allowed to drain into 125 ml flasks. The 125 ml flask was placed under UHP Nitrogen evaporator until sample is dry and then 1 ml of HPLC grade Acetonitrile-water (80% CH₃CN and 20% H₂O) was poured into 125 ml flasks containing dry sample. At this point the sample was transferred into 1.5 ml HPLC vials for analysis. The HPLC was Summit[™], with a P580 pump, a Dionex ASI automated sample injector for the auto sampler, and a photodiode array detector.

Enzyme Analysis

Collecting and Maintaining the Ants. Workers of Argentine ants, *L. humile*, were a gift by Dr. Michael K. Rust from University of California, Riverside, CA. Several colonies were collected from Toledo Bend, La. They were maintained in the lab with fresh crickets, water, and 20% sugar (v/v) in water. Temperature and humidity were controlled at 23 ± 2°C and 65 ± 5%.

Chemicals. Reduced glutathione (GSH), CDNB (1-chloro-2,4-dinitrobenzene), Coomassie brilliant blue G250, Fast Blue B salt, α -naphthyl acetate (α -NA), and bovine serum albumin were purchased from Sigma Chemical Company, St. Louis, MO. All other reagents were purchased from Sigma, and all other chemicals and solvents used were of analytical grade.

Preparation of Tissue Homogenates. Twenty abdomens of Argentine ant workers were homogenized in 0.4 ml of ice-cold 1.15% KCl using an all-glass homogenizer. The homogenate was centrifuged at 4°C for 10 minutes at 10,000g (~11200 rpm). The resulting supernatant was transferred into microcentrifuge tube by filtering through glass wool, and was used as the enzyme resource for enzyme activity assays within 3 hours of preparation. Keep all reagents and enzymes on ice until ready to start the assays.

Enzyme Assays. Glutathione S-transferase (GST) activity was measured using 1-chloro-2,4-dinitrobenzene (CDNB) and reduced glutathione (GSH) as substrates with slight modifications according to Ottea et al. in 96-well microplates (Costar, Cambridge, MA), which were prewashed with 2.5% Tween 20 (v/v in water). Substrate solution (0.75 mM) was prepared by mixing stock solution of substrate (50 mM CDNB in DMSO) with buffer containing 15% glycerol. Buffers used included: Tris-HCl (50 mM, pH 7.2-8.8), ammonia-ammonium chloride ($I=0.2$, pH 8.2-10.0). Typical reaction mixtures in individual wells consisted of 200 μ l CDNB substrate solution (0.5 mM final concentration), 10 μ l of enzyme homogenate, and 30 μ l GSH (8 mM final concentration). The corresponding buffer was added to wells to make a pre-reaction volume of 100 μ l, and reactions were initiated by adding 200 μ l substrate solution. Incubations without protein were used as non-enzymatic controls. Each treatment has three replicates. Rate of change in optical density at 340 nm during the initial 10 min of the reaction was measured and first-order reaction rates were converted to nmol/min using the experimentally derived "extinction" coefficient of 10.09 mM⁻¹ 300 μ l⁻¹.

the *S. invicta* nestmate interactions. *L. humile* were not aggressive toward other *L. humile*, and these interactions were significantly different from the nestmate *S. invicta* interactions because no instances of aggression were ever observed. The 15-minute time group was significantly different from all other time groups. The 30-minute time group was not significantly different from the 45-minute time group, but it was significantly different from the 60-minute time group. The 45-minute time group was not significantly different from the 30-minute time group or the 60-minute time group.

Table 1. Ant interactions and aggression observed in one-on-one experiment and in ten-on-ten experiment.

Analysis	One-on-One			Ten-on-Ten		
	Test Value	Df	p-value	Test Value	Df	p-value
χ^2 interaction / aggressive behavior	$\chi^2=32.02$	8	<0.0001	$\chi^2=153.11$	8	<0.0001
χ^2 interaction / retreat behavior	$\chi^2=17.01$	8	<0.03	$\chi^2=132.71$	8	<0.0001
χ^2 interaction / neutral behavior	$\chi^2=46.2$	8	<0.0001	$\chi^2=212.05$	8	<0.0001
Mixed model- aggressive behavior						
interaction	F=5.34	8,168	<0.0001	F=37.98	8,349	<0.0001
ant type (Arg / LFA / FFA)	random variable			F=49.09	2,349	<0.0001
role (initiator / respondent)	F=3.67	1,168	<0.0437	N/A		
Mixed model- retreat behavior						
interaction	F=5.2	8,168	<0.0001	F=43.07	8,349	<0.0001
ant type (Arg / LFA / FFA)	F=12.34	2,168	<0.0001	F=83.12	2,349	<0.0001
role (initiator / respondent)	F=1.61	1,168	0.2068	N/A		

Arg=Argentine and; LFA=lab-reared red imported fire ant; FFA=field collected red imported fire ant in the one-on-one interactions, the ant type was used as a random variable to achieve a better fit statistic (-2 Res Log Likelihood = 96.1). In ten-on-ten interactions the role (initiator or respondent) was not recorded.

When large colonies of fire ants were placed among large colonies of Argentine ants, the Argentine ants immediately began raiding the brood of the fire ants and attacked the workers. For all three colonies studied, all of the *S. invicta* brood was removed by *L. humile* within the first hour. After 24 hours, all fire ant colonies were decimated.

Large-scale Treatment

We treated >1500 trees with a power sprayer; approximately 50% were treated with cypermethrin and the rest with malathion in Toledo Bend, La. Immediate reductions in ants foraging in the trees were observed, however, foraging trails were not eliminated. Argentine ants immediately began foraging on the granular bait that was broadcast immediately following the treatment of trees. Trails of ants on the ground and in the trees were reduced but not eliminated. The residents reported that the combination of treatments reduced the ant population on each of their properties. Many reported that for the first time in a long time, they did not have ants in their houses, cars, and boats for several months.

Pesticide Application and Extraction

Liquid and granular applications of bifenthrin suppressed Argentine ants by more than 50% (Figure 1). The amount of bifenthrin extracted from soil near the foundations of the houses was more than 10 times higher than that extracted from the soil from the yards (Figures 2 and 3). Argentine ant populations were initially suppressed in the yards but within two months were nearly equal to the numbers measured before pesticide application. These data reflect the decreased amount of bifenthrin extracted from yard soil two months after treatment.

Effect of Temperature on GST Activity in Argentine Ants from California

According to our previous results (data not published), we define pH 8.8 Tris-HCl (50 mM), and pH 10 ammonia-ammonium chloride (AMCl, $I=0.2$) as optimal pH in Argentine ants from California. Under these conditions, GST activities in different temperatures were measured (Table 2). The result indicated the optimal pH in both Tris-HCl and AMCl buffers is 25C.

Table 2. GST Activity Against CDNB in different temperatures.

Temperature (°C)	20	25	27	30	35	37	40
Tris pH 8.8	682.62	1189.5	376.35	364.80	64.52	80.40	260.01
AMCl pH 10	614.23	2244.28	625.38	225.99	67.74	0	150.67

GST activity unit is $\text{nmol}\cdot\text{min}^{-1}\cdot\text{mg}^{-1}$ protein

Effect of Protein on GST Activity in Argentine Ants from California

Difference in protein/activity relationship was also measured in assays with Argentine ants from California. In test with CDNB, GST activity in pH 8.8 Tris-HCl buffer in Argentine ants was linear within the range of protein concentrations examined (0-6 μg) (Table 3). The linear regression equation between protein content and GST activity is $y = 0.861 + 0.344x$, and the regression coefficient is 0.9921 ($R^2 = 0.9842$).

Table 3. GST Activity vs. Protein in Argentine Ants from California

Protein content (g)	0.71	1.52	3.18	4.33	5.37	6.36
GST activity*	1.05 ± 0.06	1.39 ± 0.02	1.94 ± 0.09	2.48 ± 0.09	2.79 ± 0.04	2.91 ± 0.11

* Unit is $\text{nmol}\cdot\text{min}^{-1}$

Difference Of GST Activity Between Two Colonies

Both Tris – HCl (pH 8.8) and AMCl (pH 10) buffers were used to measure GST activity from two kinds of ants colonies, one is from California, and another is from Toledo Bend. In the same colony, there was significant difference in GST activity between different buffers. There were also significant differences in GST activity between two kinds of colonies (Table 4).

Table 4. GST activities in Argentine ants from different colony

Colony	Tris-HCl pH 8.8	AMCl pH 10
California	0.221 ± 0.081 b	12.661 ± 0.647 b
Toledo Bend	1.662 ± 0.446 a	15.337 ± 0.631 a

GST activity unit is $10^3 \text{ nmol}\cdot\text{min}^{-1}\cdot\text{mg}^{-1}$ protein.

Means followed by the different letter in the same column are significantly different (DMRT, $P < 0.05$)

DISCUSSION

Both one-on-one interactions and ten-on-ten interactions had similar results. All intraspecific nestmate interactions demonstrated neutral behavior as expected. The intraspecific nestmate and non-nestmate interactions of Argentine ants confirms other reports in the literature, because all results indicated neutrality. Argentine ants are capable of forming supercolonies (Tsutsui et al., 2000; Giraud et al., 2002) and have lost their intraspecific aggression (Barber, 1922; Holway, 1998b; Holway et al., 1998). In the southeastern United States, isolated colonies of Argentine ants have exhibited high aggression levels (Roulston et al., 2003), but since all Argentine ants were collected from a single field in Baton Rouge, they were not isolated and may have been a very large colony with several nests. With *S. invicta*, high aggression levels were not expected between nestmates, but they were expected between non-nestmates. These results confirm low aggression levels between nestmates, but non-nestmates from lab-reared colonies also had relatively low aggression levels. This low aggression level between non-nestmates may be because they occupy a single small rearing room and are fed the exact same diet, and they may all have a common "odor" even though they are kept as separate colonies (Vander Meer and Morel, 1998). The lab-reared fire ants were expected to have lower levels of aggression compared to field-collected fire ants because of their observed loss of aggression toward humans and because of their continuously available food supply. Lab-reared fire ants were even more aggressive toward Argentine ants than field-collected fire ants were toward Argentine ants, and lab-reared fire ants were also especially aggressive toward field-collected fire ants. Although Argentine ants were likely to initiate encounters with other ants in our study and Human and Gordon's study (1999), they were also more likely to retreat than fire ants. The ability of Argentine ants to quickly assess the situation, then act, may give Argentine ants an advantage when competing with other ants. Argentine ants typically use chemical defensive compounds when engaging in interference competition, but in these bioassays, the RIFA showed no reaction to defensive compounds.

The mortality data showed that Argentine ants suffered high mortality compared to fire ants. Interactions involving Argentine ants have a higher mortality early on whereas interactions with lab-reared fire ants have a very low mortality throughout the duration of the experiment. The Argentine ants were quick to engage in interference competition with fire ants, but they were often not the victorious survivor. These data support the idea that Argentine ants may not be the better competitor at interference competition with fire ants when interacting with equal numbers of each species. Nevertheless, unequal numbers may explain shifts between these two species. In preliminary study, large fire ant colonies were relocated to an area where extremely large infestations of Argentine ant colonies existed in Toledo Bend, Louisiana. Within one hour, the Argentine ants had raided all the brood. In 24 hours, there was no evidence of live fire ants. Although many studies have documented the interaction between Argentine ants and native ants and fire ants and native ants (Crowell, 1968; Hung and Vinson, 1978; Banks and Williams, 1989; Jones and Phillips, 1990; Human and Gordon, 1996; Human and Gordon, 1996, 1997, 1999; Human et al., 1998; Holway, 1988a, 1999; Morrison, 2002), no other study has looked at interactions between fire ants and Argentine ants and the behavior at this interface between these two species. Both Argentine ants and fire ants occupy similar habitats, but what allows one species to dominate a territory is still unknown.

We investigated the effect of competition between Argentine ants and red imported fire ants. When equal numbers of Argentine ants and fire ants interacted, fire ants were more successful than Argentines, which faced great mortality risk. However, when large numbers of Argentines interacted with fewer fire ants, entire colonies of fire ants were quickly decimated. Suppression of Argentine ants by baits may allow fire ants to adequately compete and then dominate an area.

We also tested several methods of controlling Argentine ants on a large scale. We tested contact insecticide barrier sprays, broadcasts of liquids and granules, and liquid and solid baits. Some of these efforts suppressed the ants for a short time, while others appeared to be ineffective. Many diligent homeowners also repeatedly applied pesticides to suppress the ants. Contact insecticides around the trees reduced the number of ants, but did not eliminate the ants from foraging in the trees. At the same time, the granular bait that was broadcast combined with strategically placed liquid bait stations further reduced the population of Argentine ants. Some of the contact insecticides that were applied, were not present in tested treated soil one month after treatment. Subsequent experiments provide evidence that there is runoff. Additionally, Argentine ants from Toledo Bend had 7.5 times more glutathione-S-transferase activity than ants from California, which have never been exposed to insecticides. Together, these results may explain some of the difficulty in managing Argentine ants. Also, the combination of contact insecticides barriers to block the ants' food sources and broadcast of granular and liquid bait may be an effective large-scale method of suppressing troublesome Argentine ant populations:

CONCLUSION

Ant distribution can be determined by abiotic and biotic factors. A previous field study showed that certain abiotic factors may affect ant abundance. Additionally, competition, both exploitative and interference, can shape ant communities and change their dynamics. In the late 1930's, Argentine ants were the major ant pests in East Baton Rouge Parish, La, and presently, fire ants are the major ant pests in the area. No environmental factors were discovered which would explain this shift, however the aggression bioassays offer an explanation for this shift. When even numbers of fire ants and Argentine ants interact, Argentine ants face a higher risk of mortality. When numbers of fire ants are low relative to Argentine ant numbers, then the Argentines have the opportunity to win. The loss of intraspecific aggression and the ability Argentine ants have to build supercolonies does not appear to be enough of a competitive advantage to displace fire ants. Invasive species are often characterized by their loss of intraspecific aggression and their ability to form large populations. Although monogyne fire ants have high intraspecific aggression, this aggressive ability has enabled these ants to displace native ant species.

Heavy applications of contact insecticides may further exacerbate this ecological imbalance. We present preliminary evidence that Argentine ants workers may produce high levels of insecticide detoxification enzymes. The presence of these enzymes may help explain why residents report out-of-control Argentine ant populations and why they say that insecticidal treatments “do not work.”

ACKNOWLEDGMENTS

The authors would like to thank Thabit Folami, Rebecca Baliff, and Kathryn O'Brien for their help.

REFERENCES CITED

- Banks, W. A. and D. F. Williams. 1989.** Competitive displacement of *Paratrechina longicornis* (Latreille) (Hymenoptera: Formicidae) from baits by fire ants in Mato Grosso, Brazil. *Journal of Entomological Science* 24: 381-391.
- Barber, E. R. 1922.** The Argentine Ant as a Household Pest. *Farmers Bulletin* 1101. USDA, Washington, D.C., USA.
- Crowell, K. 1968.** Rates of competitive exclusion by the Argentine ant in Bermuda. *Ecology* 49: 551-555.
- Diamond, J. and T. J. Case. 1986.** Overview: introductions, extinctions, exterminations, and invasions. Pages 65-79 in J. Diamond and T. J. Case, editors. *Community Ecology*. Harper and Row, New York, New York, USA.
- DMAP Disease (Distance) Mapping and Analysis Program. 1997.** University of Iowa, Department of Geography. Iowa City, IA, USA.
- Environmental Systems Research Institute. 2002.** Arview 3.2a. Redlands, CA, USA.
- Erickson, J. M. 1971.** The displacement of native ant species by the introduced Argentine ant *Iridomyrmex humilis* Mayr. *Psyche* 78: 257-266.
- Fisher, B. L. 1998.** Insect behavior and ecology in conservation: preserving functional species interactions. *Annals of the Entomological Society of America* 91: 155-158.
- Foster, E. 1908.** The introduction of *Iridomyrmex humilis* (Mayr) into New Orleans. *Journal of Economic Entomology* 1: 289-293.
- Giraud, T., J. S. Pederson and L. Keller. 2002.** Evolution of supercolonies: the Argentine ants of southern Europe. *Proceedings of the National Academy of Sciences* 99: 6075-6079.
- Holway, D. A. 1995.** Distribution of the Argentine ant (*Linepithema humile*) in northern California. *Conservation Biology* 9: 1634-1637.
- Holway, D. A. 1998a.** Effect of Argentine ant invasions on ground-dwelling arthropods in northern California riparian woodlands. *Oecologia* 116: 252-258.
- Holway, D. A. 1998b.** Factors governing rate of invasion: a natural experiment using Argentine ants. *Oecologia* 115: 206-212.
- Holway, D. A., A. V. Suarez and T. J. Case. 1998.** Loss of intraspecific aggression in the success of a widespread invasive social insect. *Science* 282: 949-952.
- Holway, D. A. 1999.** Competitive mechanisms underlying the displacement of native ants by the invasive Argentine ant. *Ecology* 80: 238-251.
- Holway, D.A., L. Lach, A. V. Suarez, N. D. Tsutsui and T. J. Case. 2002.** The causes and consequences of ant invasions. *Annual Review of Ecology Systematics* 33: 181-233.
- Hooper, L.M. 1998.** The nutritional ecology and effects of toxicants on colonies of Argentine ants, *Linepithema humile* (Mayr). 1-157.
- Human, K. G. and D. M. Gordon. 1996.** Exploitation and interference competition between the invasive Argentine ant, *Linepithema humile*, and native ant species. *Oecologia* 105: 405-412.
- Human, K. G. and D. M. Gordon. 1997.** Effects of Argentine ants on invertebrate biodiversity in northern California. *Conservation Biology* 11: 1242-1248.
- Human, K. G., S. Weiss, A. Weiss, B. Sandler and D. M. Gordon. 1998.** Effects of abiotic factors on the distribution and activity of the invasive Argentine ant (Hymenoptera: Formicidae). *Environmental Entomology* 27: 822-833.
- Human, K. G. and D. M. Gordon. 1999.** Behavioral interactions of the invasive Argentine ant with native ant species. *Insectes Sociaux* 46: 159-163.
- Hung, A. C. F. and S. B. Vinson. 1978.** Factors affecting the distribution of fire ants in Texas (Myrmicinae: Formicidae). *The Southwestern Naturalist* 23: 205-214.
- Jones, S. R. and S. A. Phillips, Jr. 1990.** Resource collecting abilities of *Solenopsis invicta* (Hymenoptera: Formicidae) compared with those of sympatric Texas ants. *Southwestern Naturalist* 35: 416-422.

- Knight R.L. and Rust M.K. 1990.** Repellency and efficacy of insecticides against foraging workers in laboratory colonies of Argentine ants (Hymenoptera: Formicidae Econ. Entm. 83: 1402-1408.
- Morrison, L. W. 2000.** Mechanisms of interspecific competition among an invasive and two native fire ants. *Oikos* 90: 238-252.
- Morrison, L. W. 2002.** Long-term impacts of an arthropod-community invasion by the imported fire ant, *Solenopsis invicta*. *Ecology* 83: 2337-2345.
- Newell, Wilmon. 1908.** Notes on the Habits of the Argentine or "New Orleans" Ant, *Iridomyrmex humilis* Mayr. *Journal of Economic Entomology* 1: 20-35.
- Nasfield, G. 1968.** Argentine Ants. *Australian Natural History* 16: 12-15.
- Porter, S. D., B. Van Eimeren and L. Gilbert. 1988.** Invasion of the red imported fire ants (Hymenoptera: Formicidae): microgeography of competitive replacement. *Annals of the Entomological Society of America* 81: 913-918.
- Porter, S. D. and D. A. Savignano. 1990.** Invasion of polygyne fire ants decimates native ants and disrupts arthropod community. *Ecology* 71: 2095-2106.
- Roulston, T. H., Buczkowski, G. and Silverman, J. 2003.** Nestmate discrimination in ants: effect of bioassay on aggressive behavior. *Insectes Sociaux* 50: 151-159.
- Rust M. K., Reiersen D. A and Klotz J. H. 2003.** Pest management of Argentine ants (Hymenoptera: Formicidae) *Entm. Sci.* 38: 159-169.
- Sanders et al. 2001.** Long-term dynamics of the distribution of the invasive Argentine ant, *Linepithema humile*, and the native ant taxa in Northern California. *Oecologia* 127: 123-130.
- SAS Institute Inc. 2002.** Version 9.0, Cary, NC.
- Skaife, S. H. 1961.** The Argentine Ant. *The Study of Ants*: 1-13.
- Suarez, A.V., D. A. Holway and T. J. Case. 2001.** Patterns of spread in biological invasions dominated by long-distance jump dispersal: insights from Argentine ants. *Proceedings of the National Academy of Sciences* 98: 1095-1100.
- Taber, S.W. 2000.** Fire ants. Texas A&M University Press, College Station, TX, USA.
- Tsutsui, N. D., A. V. Suarez, D. A. Holway and T. J. Case. 2000.** Reduced genetic variation and the success of an invasive species. *Proceedings of the National Academy of Sciences* 97: 5948-5953.
- Tsutsui, N. D. and A. V. Suarez. 2003.** The colony structure and population biology of invasive ants. *Conservation Biology* 17: 48-58.
- Vander Meer, R. K. and Morel, L. 1998.** Nestmate recognition in ants. In, *Pheromone Communication in Social Insects*. Eds. R. K. Vander Meer, M. Breed, M. Winston, and K.E. Espelie. Westview Press, Boulder, CO, p. 79-103.
- Wilson, N.L., Diller, J.H. and Markin, G.P. 1971.** Foraging territories of imported fire ants. *Annals of the Entomological Society of America*. 64:660-665.
- Wojcik, D. P. 1983.** Comparison of the ecology of red imported fire ants in North and South America. *Florida Entomologist* 66: 101-111.