

Areawide Suppression of Fire Ant Populations in
Pastures: Project Update^{1,2}

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ABSTRACT The red imported fire ant, *Solenopsis invicta* (Hymenoptera: Formicidae), is an invasive species that creates serious medical and agricultural problems, damaging many crops and cattle production. Its annual economic impact to the US economy is approximately \$6 billion. The integration of chemical bait insecticides and biological control agents is used in an areawide management program for fire ants coordinated by USDA-ARS Center for Medical, Agricultural, and Veterinary Entomology in Gainesville, Florida. Control sites, where no biological control agents were released, and biocontrol treatment sites, where both decapitating flies and the microsporidium *Thelehanthia solenopsae* were released, have been established in five states (Florida, Texas, Mississippi, Oklahoma, and South Carolina). All sites received chemical bait applications of a 1:1 mixture of hydramethylnon and methoprene baits applied at a rate of 1.7 kg per ha. To evaluate the effect of the treatments on the arthropod biodiversity, pitfall traps were used twice a year. Decreases in fire ant populations have been observed at the different demonstration sites. The decapitating fly *Pseudacteon tricuspis* (Diptera: Phoridae) has been established at demonstration sites in three states (Florida, Texas, and South Carolina); *P. curvatus* was established in Mississippi, Florida, and South Carolina. The disease *T. solenopsae* has been established in four states (Florida, South Carolina, Texas, and Oklahoma), and continues to spread. This new approach to fire ant control may have a significant impact on the future management of fire ant populations.

KEY WORDS Fire ant, *Solenopsis invicta*, Hymenoptera, Formicidae, integrated pest management, areawide control, biological control.

The red imported fire ant, *Solenopsis invicta*, is an invasive species inadvertently introduced into the United States. The ant currently infests over 130 million ha in 12 southeastern states and Puerto Rico (Callcott & Collins 1996, Code of Federal Regulations 2001). The fire ant thrives in disturbed habitats where humans are likely to be present and its potent sting and large populations pose serious medical and agricultural problems. Approximately 40% of the human

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population in infested areas is stung each year. Medical attention is required in many severe cases, and some result in death as a result of strong allergic reactions (deShazo et al. 1999). Fire ants also reduce populations of native ants and other insects, as well as many other animals (Porter & Savignano 1990, Jusino-Atresino & Phillips 1994, Wojcik 1994).

Recently, imported fire ants have become established in some areas in California, Arizona, and New Mexico. The economic impact of fire ants to the American economy is estimated at approximately \$6 billion annually in damage, control measures, and medical treatments (Pereira et al. 2002). Chemical treatments can provide fast control in limited areas but are costly and provide only temporary fire ant suppression. Fire ants damage many crops and create problems for the cattle industry. Treatment of pastures is usually cost-prohibitive and a rare practice in the United States.

There are only two commercial bait treatments available for fire ant control on pastures. These baits are nonspecific, costly, require multiple treatments per year, and must be used repeatedly as long as fire ants are present. The continuous treatment/re-invasion process perpetuates the ecological imbalance originally upset by the imported fire ant invasion. Imported fire ant densities in the United States are about 5X greater than those found in their indigenous South America, most likely because in the United States they have escaped from their native natural enemies (Porter et al. 1992, Porter et al. 1997). Natural enemies of fire ants, such as parasitic phorid flies and a microsporidian pathogen, are being evaluated to develop self-sustaining, biologically based integrated pest-management strategies for permanent or long-term suppression of fire ants in the United States.

The decapitating flies, *Pseudacteon tricuspis* (Diptera: Phoridae) and other species in the same genus, and the microsporidian ant pathogen, *Thelohania solenopsae*, have been successfully established in several states in the past few years (Williams et al. 2003). The prevalence of these biocontrol agents continues to increase at many release sites where they have been established. These self-sustaining biocontrol agents are important stressors to fire ant populations and can impact the pest populations over wide areas. *Pseudacteon* flies are very host-specific flies (Porter et al. 1995, Porter & Alonso 1999) that complete their development within the fire ant heads, causing them to fall off. The flies cause direct mortality of fire ants, but their most detrimental effect may be the reduction of foraging opportunities and consequent weakening of fire ant colonies (Mehdiabadi & Gilbert 2002). *Thelohania solenopsae* is also very host-specific, infecting both red and black imported fire ants. It causes a chronic disease in worker and reproductive ants that can be transmitted transovarially to the brood and horizontally to other fire ants. The disease causes a slow death of the colony (Williams et al. 1999) and makes ants more susceptible to the insecticide hydramethylnon (Valles & Pereira 2003).

The primary objective of the Areawide Imported Fire Ant Project is to demonstrate that fire ant populations can be reduced and maintained at very low levels over a large area using a strategy that integrates chemical baits and biocontrol agents. The combination of these elements is a new strategy that was recently validated at a military installation near Columbia, South Carolina (Williams et al. unpublished data). In that study, excellent results in fire ant population reduction were obtained and populations have been maintained at very low levels

(greater than 95% reduction) for more than 2 years after application of a formulation containing fipronil, and the release of the decapitating fly, *P. tricuspis*, and the pathogen, *T. solenopsae*. Decapitating flies have been found attacking fire ant workers, and *T. solenopsae* has spread over most of the treated area. The South Carolina experiment was conducted in small, 2-ha nonagricultural areas. Despite the encouraging results, there is a need for replication of similar experiments over larger agricultural areas, such as improved pastures.

A combination of baits containing hydramethylnon and methoprene was chosen for use in the areawide project. This combination has been tested extensively with excellent results in Texas (Barr 2002). Choice of these products was heavily influenced by the need for registered products that farmers could later adopt for applications in grazed cattle pastures. Also, it was important to have products that would not impact the non-target fauna in the demonstration sites, since the rebound of the natural population of arthropods is an expected result of the fire ant management program. A "Special Local Needs" label (24C) was needed in Oklahoma, where the use of doses lower than the label rate is illegal. In other states (Florida, Mississippi, South Carolina, and Texas), there were no restrictions on the use of lower individual doses of these two pesticides when applied in combination for control of fire ants.

The integration of chemical baits and biological control agents is used in this areawide management program for fire ants in pastures. Applications of chemical baits are used to reduce the fire ant population. Application of the biocontrol agents to the peripheral area will serve to prevent, limit, or slow reinfestation of the chemically treated area. These biocontrol agents will be introduced using the inoculative approach that has been successful in other areas. The objectives of this project are to: 1) maintain low fire ant populations using the combination of self-sustaining fire ant biological control agents and bait toxicants, 2) assess the economic impact associated with fire ants and the benefits of fire ant control, 3) monitor the environmental impact of fire ants and the effects of their control on native ant fauna, and 4) develop materials to educate the public on fire ant control options, including the establishment of a web site (<http://www.ars.usda.gov/fireant/>; accessed 12 May 2004).

The Areawide Suppression of Fire Ant Populations in Pastures project is funded through special allocations by the USDA-ARS National Program Staff. Project team members include: David Williams, Robert Vander Meer, Sanford Porter, David Oi, Steven Valles, and Roberto Pereira at the USDA-ARS Center for Medical, Agricultural and Veterinary Entomology (CMAVE), in Gainesville, Florida. State collaborators include Douglas Streett and J. T. Vogt (USDA-ARS Stoneville, Mississippi); Anne-Marie Callcott (USDA-APHIS Gulfport, Mississippi); Bart Drees, Charles Barr, and Curtis Lard (Texas A&M University); Russell Wright and Wayne Smith (Oklahoma State University); Mac Horton and Tim Davis (Clemson University, South Carolina); and Philip Koehler (University of Florida).

In addition to the demonstration component reported here, the areawide project also involves research activities at CMAVE. The research supported under this project has the objective of supporting the activities in the demonstration sites and improving the control methods integrated into management of the fire ant population at the demonstration sites. Research activities sponsored by the areawide project include the development of: 1) *S. invicta*-specific bait system

using semiochemical technology, 2) decapitating fly detection-trap system using fire ant semiochemicals, 3) new techniques for rearing decapitating flies, including new species not yet released in the United States, and 4) methods for detection of *T. solenopsae* and other fire ant diseases using immunological or genetic methods.

Materials and Methods

Treatments and demonstration site selection. Demonstration sites have been chosen in five states to represent the diversity of climatic conditions where fire ants exist in the United States. Florida is a subtropical environment, and fire ants have mating flights throughout the year. Oklahoma is the northern extreme of fire ant invasions, where populations may be limited by cold winter temperatures and may be relatively unstable. Texas is the western extreme of contiguous infestations in the southeastern United States, where a semiarid climate may favor the impact of parasitic flies on fire ant populations. South Carolina is the eastern extreme of the fire ant range in the United States, where fire ant populations are still expanding. Mississippi's sites are established in areas infested with the black imported fire ant (*Solenopsis richteri*) and the red/black hybrid and will allow comparison of the same IPM approach on a different species from the common red imported fire ant (*Solenopsis invicta*).

In each state, at least two demonstration sites were established: a control site, where no biological control agents were released, and a biocontrol treatment site, where both decapitating flies and the microsporidium *T. solenopsae* were released and/or established in the fire ant population. Control sites were at least 48 km away from the biocontrol treatment sites to prevent spread of the biocontrol agents into the control site over the expected 5-year duration of the demonstration project. In Florida, the biocontrol site is a pasture in Levy County, between Gainesville and Williston; the control site is in Jackson County, south of Marianna. In Texas, the biocontrol site was established 8 km north of Caldwell in Burleson County, and the control site is 16 km north of Bryan in Brazos County. In Mississippi, two control sites were established: one near Torrance Landing, and another near Holcomb, both in Grenada County. Two treatment sites were established in separate farms near West Point, Clay County. In Oklahoma, the control site is in the extreme southeast near Eagleton, McCurtain County. Two biocontrol sites were established one in northern Bryan County, near Kenefic, and another at the southern end of the county is near Colbert. In South Carolina, the control site is near Calhoun Falls, Abbeville County, near the Georgia border, whereas the biocontrol site is near White Oak, Fairfield County.

Sites used in the areawide fire ant project are improved pastures infested with polygyne red imported fire ant populations (Texas, Oklahoma, Florida, and South Carolina) or black/hybrid fire ants (Mississippi). Sites were selected in areas with population high enough (256–659 mounds/ha) to allow assessment of population reduction, as well as the economic and environmental impact of the fire ant management program. Sites were selected as large pasture sites, with boundaries represented by natural elements (woods, bodies of water, etc.) as much as possible. In states with high fire ant populations (Texas, South Carolina, and Florida) sites were a minimum of 120 ha in pastures to be treated with chemical baits (treatment area), plus a variable number of hectares surrounding the treat-

ment area to serve as a buffer and as release site for the biocontrol agents (peripheral area). Treatment areas in Mississippi and Oklahoma were smaller (approximately 100 and 60 ha, respectively) because of the difficulties in finding large areas with adequate levels of fire ant populations comparable with other states. The treatment areas were also chosen in areas where fire ant baits could be spread by aerial application. The peripheral area, where the biocontrol agents were released, were selected according to characteristics known to facilitate establishment of the biological control agents. For example, decapitating flies require bodies of water and forested areas to be available nearby, and the microsporidium *T. solenopsae* spreads more rapidly when introduced into polygynous colonies (Oi & Williams 2002).

Fire ant and other arthropod population estimates. To characterize the fire ant populations and the populations of native ants and other arthropods present in the demonstration sites, 500-m² circular plots were permanently marked in the demonstration areas. For each plot, the center location was georeferenced using handheld geographic positioning system device. In the area to be treated with chemical baits, 20 circular plots were established, whereas 30 circular plots were established in the peripheral areas surrounding the bait-treated area. In Oklahoma and Mississippi, the numbers of plots were adjusted proportionally to the size of the demonstration areas. The USDA population index (Lofgren & Williams 1982) was used to characterize each mound in the plots. This index varies between 1 and 10, with ratings 1–5 corresponding to nests of increasing sizes but with no brood, whereas ratings 6–10 are given to nests of corresponding sizes which contain fire ant worker brood. Fire ant populations were estimated in the spring and the fall of each year and approximately 4 weeks after chemical bait applications to determine how treatments affected the pest population.

Baits were used to monitor the activity of fire ants and other ants in the demonstration sites. Ten baits (per circular plot) consisting of hot dog slices (approximately 1-cm thick) were placed directly on the ground in a circular array distributed concentrically at two thirds of the plot radius away from the plot center. Baits were evaluated after 30–60 min by using a scalar system (0, 1, 10, 50, 100) corresponding to the approximate number of fire ants or other ants at the baits. Hot dog baits were used whenever mound counts were conducted.

To evaluate the effect of the treatments on the arthropod biodiversity, pitfall traps were used twice a year (spring and fall) to collect crawling arthropods. Eight pitfall traps were used per circular plot. Pitfall traps consisted of 7-dram plastic snap-cap vials one-third filled with propylene glycol anti-freezing fluid. Traps were left in the field for 24–48 h, collected, labeled, and capped. The contents of each trap were identified and quantified in the laboratory. Ants were identified to the species level whereas other arthropods were identified to higher taxonomic levels according to their numbers in the samples, ecological importance and difficulty in identification. Although these procedures do not guarantee samples that include all the arthropod biodiversity present in the sites, they capture a significant number of the most abundant species. These procedures allow comparisons of the most abundant populations of arthropods present before and after fire ant control.

Treatment procedures. The bait used to control fire ants in the demonstration areas was a 1:1 mixture consisting of Amdro Pro or Siege Pro (both from

BASF, Research Triangle Park; 0.73% hydramethylnon) and Extinguish (Wellmark, Bensenville; 0.5% methoprene), applied at a rate of 1.7 kg of mixture per ha. In Oklahoma where the use of below label rates is restricted by law, 2.2 kg of the mixture were used, so the application of each product was according to the label rate. Treatment decisions are triggered when the mound counts reached a level of 50 mounds/ha, or an average of 2.5 mounds per 500-m² circular plot, a threshold level determined by the researchers involved in the project.

The decapitating flies (*P. tricuspis*) were released at the biocontrol sites over a 2-wk period by disturbing 5–10 mounds and releasing 30–60 adult flies at each mound. After the release, the demonstration sites were inspected for flies to determine whether new releases were needed. The presence of *Pseudacteon* flies was determined by disturbing fire ant mounds, and inspecting them for hovering flies every few minutes over a 20- to 30-min period. Decapitating flies were determined to be established at a location when active flies were observed in the spring or summer after surviving the winter in the field. *Thelohania solenopsae* inoculations were made within active fire ant mounds by introducing 3 g of live, *T. solenopsae*-infected brood per mound, in areas where the protozoan was not yet established at the start of the demonstration project (South Carolina and Mississippi). *Thelohania solenopsae* infections were determined by inserting a vial with inner walls coated with Fluon® (Asahi Glass Co., Chadds Ford) into each mound and collecting worker ants. The worker ant samples were subsequently returned to the laboratory and examined for *T. solenopsae* spores using a phase contrast microscope (Williams et al. 1999). *Thelohania solenopsae* was considered established at a location when spores were observed in samples more than 6 months after inoculations of sites. After 6 months, infected ants added to the mounds would have been eliminated from the ant population and observed infections would necessarily represent new infections acquired in the field.

Results and Discussion

As of November 2002, all sites in the Areawide project had received at least one application of fire ant baits, and biological control releases had been conducted in all treatment sites. All sites are now at the same level of project development, except for sites where the biological controls could not yet be confirmed as established. After initial applications in the spring 2002, applications of chemical baits were repeated in fall 2002 in Florida and Texas in sites where fire ant populations were above established threshold levels (2.5 mounds per 500-m² circular plot). New applications were also necessary in Oklahoma, Mississippi and South Carolina in spring 2003 so the fire ant populations could be reduced to below threshold levels. Decreases in fire ant populations (85–99% in number of fire ant mounds) have been observed at the different demonstration sites where the areawide fire ant control, including biocontrol agents and chemical baits has been implemented. Whether these reductions can be sustained by the presence of the biocontrol agents requires further investigation.

The decapitating fly (*P. tricuspis*) has been established in demonstration sites in three states (Florida, Texas and South Carolina), whereas a *S. richteri* biotype of the *P. curvatus* species was established in Mississippi. New releases of *P. tricuspis* were necessary in Texas and South Carolina where only few (0–5 flies observed over 8–10 mounds over 20–30 minutes) parasitic flies were observed

after overwintering in the field. The decapitating fly populations were not considered sufficient at the start of spring 2003, but since then higher fly populations (>10 flies observed over 8–10 mounds over 20–30 minutes) have been observed in both locations. A new biotype of *P. curvatus* has been released in the Florida and South Carolina demonstration sites. This biotype differs from the *P. curvatus* biotype established previously in Mississippi and in Alabama (Graham et al. 2003), and is more apt to attack the red imported fire ant *S. invicta*, and better adapted to cooler climates.

The disease *T. solenopsae* has been established in four states (Florida, South Carolina, Texas, and Oklahoma), and continues to spread to populations well beyond the demonstration sites. The control site in Texas, has now been invaded by this fire ant pathogen, and will force the relocation of the control site. In Mississippi, where the fire ant population consists of black and hybrid fire ants, the microsporidian disease has not yet been established, since no infection has been detected in inoculated sites. New releases of infected brood were conducted during the spring 2003, but establishment of the disease has not yet been observed. Previous attempts to introduce the US strain of *T. solenopsae* to black and hybrid fire ants have also been unsuccessful, although the disease is common in South America populations of the black fire ant (Briano et al. 1996).

Ecological and economic components of the areawide project are not yet completely implemented. Ecological evaluation continued at all sites with collection and processing of information on the arthropod population, with special attention to the ant fauna in the demonstration sites. Results of this evaluation are preliminary due to the limited time the fire ant population has been under control. However, in Florida the non-fire ant populations seem to be increasing at the demonstrations sites where fire ants are under control. Economic surveys have been forwarded by the state cooperators to the economic team directed by Dr. Curtis Lard from the Texas A&M University. These surveys are being analyzed and the data will be used to estimate economic impact of the fire ants on the US agriculture, and the costs related to the integrated control approach used in the areawide project.

The project web site (<http://www.ars.usda.gov/fireant/>) is continuously updated with new information. Educational videos describing the fire ant disease caused by *T. solenopsae* and the decapitating flies are in their final editing phases and should be released soon to extension personnel and the public as part of the educational component of the areawide project. Part of the decapitating fly video was the subject of an article by the nationally syndicated columnist Dave Barry. This caused a high-traffic volume at the areawide site, which needed to be moved to a larger and faster server that could handle the sudden increase in public interest. A new brochure describing the biocontrol agents used in the areawide project has been distributed to state collaborators and will soon be incorporated in an improved areawide project web site. The impact of the project and its educational component (brochures, videos, and web site) has made the public more aware of the fire ant problem and the self-sustaining solutions available.

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