Release and Attempts to Document the Establishment of Natural Enemies of the Red Imported Fire Ant
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A number of natural enemies that occur with the red imported fire ant, *Solenopsis invicta* Buren, in South America have been identified that can potentially reduce populations of this pest species in the southern United States. Among those biological control candidates is a group of parasitic flies in the genus *Pseudacteon* (family Phoridae). Thus far, only one of the species of *Pseudacteon* has been approved for release in the United States. Dr. Sanford Porter, USDA-ARS, Gainesville, FL, has determined that *Pseudacteon tricuspis* is host specific to *S. invicta* (Porter et al. 1995a). With the documentation provided by Dr. Porter, *P. tricuspis* has been approved for release in the United States. Releases have been made by Dr. Porter at several sites in Florida, South Carolina, Georgia and Alabama. Releases have also been made near Laredo, Texas by Dr. Larry Gilbert, University of Texas.

Pseudacteon tricuspis is a fly whose larvae develop in the head of the red imported fire ant (Porter et al. 1995b). The female *P. tricuspis* deposits an egg near the inter-segmental membrane of the head of a red imported fire ant worker ant. The egg hatches, and the fly larvae go through their larval development inside the head of their hosts. The fly larvae feed on the tissues of the fly head. After the larval stage is completed the fly pupates and causes the head of its host to fall off. The head is then removed from the ant colony by other worker ants. The pupal stage lasts about two weeks, after which the adult fly emerges. Mating occurs after emergence and flies then hover around fire ants until eggs are deposited (Morrison et al. 1997).

The effects of this parasitization can be detrimental to red imported fire ant populations in two ways. First there is some direct impact by the killing of worker ants, although not enough to alone manage fire ants. There is also a behavioral impact caused by this association. When phorid flies are attacking fire ants, they cause a behavioral change in the foraging activities of the worker ants (Orr et al. 1995, Porter et al. 1995c). Ants tend to not forage when the flies are around. This decreases the amount of resources provided to fire ant colonies and allows other ant species to better compete for existing resources. Thus, a more even competition exists between native ant species and red imported fire ants (Porter et al. 1997).

The control of red imported fire ants in the United States has thus far used chemicals in management efforts. However, there are several reasons to turn to biological control to help in this management effort. First, the use of large amounts of chemicals needed to control fire ants is potentially harmful to the environment and, therefore, undesirable. Second, the continued use of chemicals is a costly, and temporary, solution to the problem. Third, treatment with some chemicals eliminates not only fire ants, but also other ants. After treatment, it is fire ants that are the first to return to the area and they are then able to keep out other ants and monopolize available resources. In the long term, chemical treatments can favor the increase in populations of fire ants. Finally, the only solution to reducing populations of fire ants is to provide a sustained pressure that is specific to fire ants. The logical way of doing this is to bring the natural enemies of the fire ant into their environment. This project is an attempt to do just that. We have begun

to evaluate whether *P. tricuspis* can establish in Texas at the training sites used by the Texas Army National Guard. This is just one of several natural enemies that may be needed to actually provide management of red imported fire ants.

This is the first year of this project. The initial step of this program is to determine if *P. tricuspis* will establish in Texas. The reason for this evaluation is that it is not certain whether *P. tricuspis* will adapt to the Texas climate and be able to sustain a population using the polygyne form of the red imported fire ant. In Texas, fire ant colonies are predominantly polygynous, whereas in its native South America, populations are largely monogyne. One result of this situation is that ant workers tend to be smaller in polygyne colonies and *P. tricuspis* must have a certain size of worker in order to develop females (Morrison et al. 1999).

Materials and Methods

Pupae of *Pseudacteon tricuspis* were obtained from Dr. Sanford Porter, USDA-ARS, Gainesville, FL. Dr. Porter shipped about 5,000 heads to the Center for Urban Entomology, Texas A&M University. These pupae were still enclosed in their fire ant host's head capsule. The pupae were kept in 10 by 10 by 10 inch plexiglass cages with fine netted openings for ventilation. The pupae were placed in petri dishes filled with dental stone and the dental stone was kept moist. Other moist dental stone was placed in the cage to maintain a relative humidity of over 80 percent to facilitate emergence. The cages were kept at room temperature. Flies emerged begining about a week after the arrival of the pupae and upon emergence they were transported to the release site. Flies emerged and were released over a period of 3 weeks.

The release site was a site near permanent water at Camp Swift, near Bastrop, Texas. This site was a quarter acre plot, with a fire ant density of over 250 mounds per acre of polygynous imported fire ants. A second, similar site was set aside as an untreated control site to evaluate effects of the phorid flies, should they successfully establish. Prior to making releases, surveys were made of ant populations at both plots using bait traps and pit-fall traps. Baits were placed in test tubes with a piece of wiener as an attractant. After 30 minutes the test tubes were capped and taken to the laboratory for identification and counting of ants. A total of 25 bait stations were used at each site. Pit-fall traps used cups with propylene glycol that were left in the soil for three days and then collected for counting and identification of ants. A total of 15 pit-fall traps were used at each site.

Releases were made by placing a large plastic box (20 by 10 by 10 inch) over a fire ant mound and putting a canvas tarp above the box to shade the sun. The mound was disturbed and flies were introduced under the box. The mound was disturbed about every 15 minutes (or more frequently if fire ants became inactive) for a period of two hours. Observations were made to insure that the phorid flies were ovipositing on ants. All releases were started between 10:30 am and 12:00 noon.

One month after releases surveys were made by disturbing mounds and checking for phorid fly recruitment. These surveys were made for approximately one hour during each check and used several mounds for observation. Checks were made approximately twice a month for the first two months and once a month afterwards.

Results

Densities of the two plots were similar. The pit-fall samples have more native ants than do bait stations and will be used in evaluations in future years of this project, it the phorid flies are shown to establish. Bait station collections were used to show relative numbers of fire ants between the two plots and will also be used in the evaluation of effect on fire ants should the phorid flies establish. These results showed that the two plots were statistically similar (P = 0.399). The results of bait sampling are presented in **Table 1** (all counts are of *Solenopsis invicta*, except those noted as otherwise).

Phorid flies were released between April 8, 1999 and April 24, 1999. A total of 3,350 flies were released. **Table 2** gives the number of flies released on each date, along with release conditions.

Sampling for establishment and the occurrence of a field generation began one month after the final release. For the first two months of sampling, checks were made approximately bi-weekly. After that time, checks were made approximately monthly. **Table 3** provides the results of those checks.

Discussion

Dr. Sanford Porter has determined that at least 1,500 phorid flies are required to make a successful release (personal communication). The number that I released (3,350) easily was enough to establish phorid flies if conditions were right for them. I released flies over a period of approximately 3 weeks to allow for the possibility of a continuous population emerging. My idea was that if the flies that I released oviposited soon after their release, then a continuous population of phorid flies might result. The reason that these flies were released over a 3 week time-span was so that I could determine that later observations were from a field population and not a released population. *Pseudacteon tricuspis* adults live for only about 5 days and the development time is 5 or more weeks. The difference in development time is dependent on environmental temperature.

My results show that *P. tricuspis* established through at least one field generation. After this time, I have not observed flies for about two months. This does not mean that the population is still not active, but they have not been observed. Sanford Porter has had a population established for nearly 2 years in Florida and there are times when he does not observe them for some time (personal communication). Therefore, the success of this release can not be determined for some time and even if the population has been lost, there is still the chance for a future establishment of another population. The results of finding at least one field generation shows that there is the potential for establishment.

There are several factors that may be detrimental to the establishment of *Pseudacteon tricuspis*. First, *P. tricuspis* must have a certain size of ant do develop a female. In Texas, most red imported fire ant colonies are of the polygynous form, which have smaller workers than the monogyne form. There are some workers that are large enough to allow for females to develop in polygynous red imported fire ants, but that percentage is around 10 percent of the colony. One question that has not been researched is how good *P. tricuspis* is at searching for proper hosts to develop a sufficient number of females. Many insect parasites are very good at host

determination, but no work has been done on this phorid fly species. This is a project that should be investigated, but it would take funding for basic research. Second, Texas has a distinct dry period during each year and this is not encountered in *P. tricuspis*' native range in Brazil. There has been no research on whether this will affect their establishment. Third, *P. tricuspis* appears to attack almost always at disturbed mounds. This limits their encounters with red imported fire ants, especially in mid-summer, when most of the ant population is deep in the soil and active mostly at night. However, this is also the case in Florida, where Sanford Porter has populations that have established through this period. There are other phorid species that attack imported fire ants primarily on foraging trails, but these species have not yet been approved for release in the United States.

The evaluation of this release will become clearer in the next year and I plan to release more *P. tricuspis* to supplement this population, or try to re-establish it. There is also a possibility that other phorid fly species or strans may be approved for release and these could be added to the management plan.

References cited

Morrison, L. W., C. G. Dall'Aglio-Holvorcem, and L. E. Gilbert. 1997. Oviposition behavior and development of *Pseudacteon* flies (Diptera: Phoridae), parasitoids of *Solenopsis* fire ants (Hymenoptera: Formicidae). Environmental Entomology 26(3): 716-724.

Morrison, L. L., S. D. Porter, and L. E. Gilbert. 1999. Sex ratio variation as a function of host size in *Pseudacteon* flies (Diptera: Phoridae), parasitoids of *Solenopsis* fire ants (Hymenoptera: Formicidae). Biological Journal of the Linnean Society 66: 257-267.

Orr, M. R., S. H. Selke, W. W. Benson, and L. E. Gilbert. 1995. Flies suppress fire ants. Nature 373: 292-293.

Porter, S. D., H. G. Fowler, S. Campiolo, and M. A. Pesquero. 1995. Host specificity of several *Pseudacteon* (Diptera: Phoridae) parasites of fire ants (Hymenoptera: Formicidae) in South America. Florida Entomologist 78(1): 70-75.

Porter, S. D., M. A. Pesquero, S. Campiolo, and H. G. Fowler. 1995. Growth and development of *Pseudacteon* phorid fly maggots (Diptera: Phoridae) in the heads of *Solenopsis* fire ant workers (Hymenoptera: Formicidae). Environmental Entomology 24(2): 475-479.

Porter, S. D., R. K. Vander Meer, M .A. Pesquero, , S. Campiolo, and H. G. Fowler. 1995. *Solenopsis* (Hymenoptera: Formicidae) fire ant reactions to attacks of *Pseudacteon* flies (Diptera: Phoridae) in southeastern Brazil. Annals of the Entomological Society of America 88(4): 570-575.

Porter, S. D., D. F. Williams, R. S. Patterson, and H. G. Fowler. 1997. Intercontinental differences in the abundance of *Solenopsis* fire ants (Hymenoptera: Formicidae): escape from natural enemies? Environmental Entomology 26(2): 373-384.

Table 1. Counts of ants collected at bait stations left in plots for 30 minutes. All counts are of *Solenopsis invicta*, unless noted otherwise.

Sample Number	Control Plot Counts	Release Site Plot Counts
1	36 (*Dorymyrmex* sp.)	75
2	0	0
3	0	89
4	0	99
5	0	171
6	0	39
7	12	23
8	110	164
9	197	190
10	266	7
11	33	93
12	137	147
13	133	40
14	323	1
15	171	106
16	215	96
17	253	6
18	34	8
19	50	129
20	184	187
21	191	112
22	250	86
23	259	139
24	284	94
25	0	157

Table 2. Conditions for phorid fly releases and the numbers of flies released on given days.

Date	# of Flies	Air Temp. (C°)	Soil Temp. (C° at 4 in.)	Wind (mph)	Ant Depth (in.)
4/8	50	27	27	5-15	2
4/9	180	28-31	28	0-5	2
4/10	180	27	24	0	1
4/11	200	26-28	24	0-5	1
4/12	240	26-28	24	5	1
4/13	260	27-28	24	0-5	2
4/15	450	20-21	22	5-15	1
4/16	350	20	22	0-5	1
4/17	280	20-21	20	0-5	2
4/18	220	23-26	22	5-10	2
4/19	200	27-28	23	0-5	3
4/21	160	26-28	24	5-10	3
4/23	300	29-31	25	5-10	4
4/24	280	29-31	25	0-5	3

Table 3. Monitoring of phorid flies at the release site. Dates and conditions are shown along with numbers of phorid flies observed.

Date	# Mounds	# w/flies	M/F	Soil Temp (C°)	Air Temp (C°)	Wind (mph)
5/14	8	0	-	28	31	5-10
5/24	6	1	4/1	29	32	5-10
6/8	5	2	10/6	29	30	0-5
6/21	6	1	4/2	29	31	5-10
7/19	5	0	-	30	32	5-15
7/29	6	0	-	30	33	0-5
8/16	5	0	-	30	33	5-10