Release and establishment of fire ant parasitic flies, *Pseudacteon curvatus* and *Pseudacteon tricuspis* (Diptera: Phoridae) in Denton and Red River counties in 2008

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Biological control, combined with conventional chemical methods, are now being used to manage the red imported fire ant, *Solenopsis invicta* Buren (Hymenoptera: Formicidae) in the United States. Classical biological control has the potential to effectively control pest populations and in some cases it is more effective than conventional chemical methods. There are many natural enemies of the red imported fire ant that have been identified in South America (Knutson and Drees 1998). These natural enemies have the ability to reduce the number of fire ant colonies in an area, sustain themselves and spread over time. A parasitic fly in the genus *Pseudacteon* (Diptera: Phoridae) is one natural enemy from South America that attacks fire ants in the genus *Solenopsis*. All of these fly species are host-specific and do not affect any other ants or animals (Porter and Gilbert 2004). This is a logical approach to suppressing fire ants, since this fly will provide a sustained pressure that is specific to red imported fire ants.

Pseudacteon sp. is a parasitic fly whose larvae develop inside the head of a fire ant worker. The mated female fly lives for several days. She seeks foraging worker ants in the daytime. Upon finding a foraging worker fire ant, she lays her egg inside the thoracic region the fire ant worker. After the egg hatches, the larva digests the tissues within the thoracic region of the fire ant worker and then the fly larva moves into the head. The fire ant worker's head fall off and the larva continues to feed on tissues within the head and then pupates inside the head. The other fire ants within the colony will remove the parasitized ant from the colony, so the adult fly will be able to emerge from the head capsule outside the colony and begin seeking new fire ant hosts nearby. Male and female flies mate after their emergence and then the females hover over the fire ant workers until her eggs are deposited (Knutson and Drees 1998).

Phorid flies impact fire ant colonies in two ways. First, these flies selectively detect and kill fire ant workers through parasitism. This causes about 2% of the worker ants in the colony to become parasitized. Secondly, host-seeking female phorid flies causes shifting in fire ant foraging behavior. After a fly detects a fire ant worker, the fly hovers over the worker making the worker hide to avoid attack, thereby disrupting day-time surface activities of the worker ants. This causes the ants to spend more time avoiding the flies and less time foraging for food (Porter 1998).

Currently there are five candidate species within the genus *Pseudacteon* that attack fire ants. However, only two species have been released in Texas, *Pseudacteon tricuspis* and *Pseudacteon curvatus*. With *P. tricuspus*, which has been released and established in a number of release sites in Texas, the sex of the fly is determined by the worker ant head capsule size. Females are only produced in large major worker ants, and fly larvae developing in smaller minor fire ant workers became males. However in North Texas, multiple queen colonies are more prominent, so the average fire ant worker size is small. Therefore, many males and only a few females are produced in polygyne fire ant colonies and often fail to sustain a phorid fly population. Conversely, *P. curvatus* sex ratio is not

determined by worker ant head capsule size (Morrison and King, 2004). Both *P. tricuspus* and *P. curvatus* have also been released in other southern states such as Florida, South Carolina, Louisiana, Georgia, and Alabama.

This trial was a collaborative effort between Texas AgriLife Extension Service and the United States Department of Agriculture - Animal and Plant Health Inspection Service (USDA-APHIS) that supplied the phorid flies for all releases.

Materials and Methods

The October 2006 release site for *P. tricuspis* was below the dam of Lake Ray Roberts in Pilot Point, TX in Denton County (Figure 1). Active mounds were located along the sidewalk and marked with flags. A total of 2,906 flies were released onto 53 mounds. On April 15th and October 30, 2008, we used "pizza traps" at the release area to determine the expansion of the *P. tricuspis* population. The passive "pizza" traps consist of a large petri dish containing a smaller petri dish filled with ¼ cup of midden (decomposing fire ants) and an upside down pizza tri-stand covered with Tanglefoot® (Figure 2). The phorid flies are attracted to the midden and perch onto one of the prongs of the pizza stand. The flies become stuck to the trap, which allows the traps to be placed into certain locations for up to 48 hours. We collected the traps after 24 hours and then placed each trap under a microscope so the phorid flies could be properly identified. With the help of volunteers, the traps were placed out in each of the four cardinal directions for a total of 10 miles in each direction. The traps were placed out beginning at 11am and concluding at 1:00pm with temperatures at 72°F. The traps remained in the respective locations overnight and each one was collected on April 16th and October 31st from 12pm-2pm with temperatures at 76°F and 72°F, respectively (Table 1).

Another release of *P. tricuspis* occurred in Detroit, TX, Red River County in 2008. Thirteen active mounds were located within the 50 acre release site. A total of 1000 fire ant heads were shipped from Gainesville, Florida on May 10, 2008. A total of 422 flies were released onto 13 mounds. The GPS coordinates were taken for each active mound and releases were conducted daily from 12:00pm-2:00pm from May 12, 2008 through May 25, 2008 (Table 2). On September 24, 2008, "pizza traps" were placed into the release area to determine the establishment of a population. Eight traps were placed in the release area and seven traps were recovered. The traps remained in their respective locations overnight and each one was collected on September 25, 2008 from 12:30-1:30 with temperatures at 77°F and winds 0-3mph.

The *P. curvatus* release was conducted in Red River County on a 100 acre ranch (Figure 3) beginning on October 11, 2007 and concluded on October 26, 2007. On June 10th and September 24, 2008, ten "pizza traps" were placed into the release area to determine the establishment of a population. The traps remained in their respective locations overnight and each one was collected on June 11th and September 25, 2008 from 10:30am-11:30am with temperatures at 72°F and 71°F, respectively.

Results

For the 2006 release of *P. tricuspis* at Ray Roberts Park, we monitored the expansion of the population on April 15th and October 30, 2008 in all four cardinal directions. On April

16, 2008 (Table 3), five flies were recovered on trap 2, which is two miles west of the release site. On October 31, 2008 (Table 4), northbound, two flies were recovered on trap 2, one fly was recovered on trap 3, ten flies were recovered on trap 4 and two flies were recovered on trap 6; southbound, five flies were recovered on trap 1, two flies were recovered on trap 2, four flies were recovered on trap 3, thirty-two flies were recovered on trap 4, nine flies were recovered on trap 5, one fly was recovered on trap 6; eastbound, five flies were recovered on trap 1, three flies were recovered on trap 3, four flies were recovered on trap 5, 1 fly was recovered on trap 6, one fly was recovered on trap 8, one fly was recovered on trap 9; westbound, one fly was recovered on trap 1, two flies were recovered on trap 3, six flies were recovered on trap 6. We plan to continue monitoring the expansion of *P. tricuspis* at Ray Roberts Park by traveling 15 miles in each cardinal direction both in the spring and the fall of 2009.

For the 2008 release site of *P. tricuspis* (Table 5) in Red River County, we monitored the establishment on September 24$^{th\ by}$ placing out "pizza" traps. On September 25, 2008 we recovered one fly on trap 3. We will monitor the expansion of *P. tricuspis* by traveling 5 miles in each cardinal direction from the release site in both the spring and fall of 2009.

For the *P. curvatus* release site in Red River County, we recovered one fly on trap 6, one fly on trap 8 and two flies on trap 9 on June 11, 2008 (Table 6). On September 24th we were able to recover one fly on trap 1, nine flies on trap 2, ten flies on trap 3 and one fly on trap 4 (Table 7). In addition, we were able to recover *P. curvatus* on traps seven miles from the Ray Roberts release site of *P. tricuspis*. *P. curvatus* was released in 2004 at the MT Cole Ranch in Denton County, which is 25 miles from the Ray Roberts release. Over this four year period, *P. curvatus* was able to expand over 25 miles from the original release site.

Discussion

Little is known about the favorable environmental conditions *P.curvatus* and *P. tricuspis* must have in order to establish a population. *P.curvatus* is better adapted to survive in polygyne red imported fire ant populations found in Denton and in Red River counties. We were also able to recover *P. curvatus* on the traps at Ray Roberts, which is over 25 miles from the original release site at the MT Cole Ranch. Faculty members at University of Austin recently placed passive traps in the fall of 2008 in North Texas and were able to recover *P. curvatus* flies in the Hillsboro area. They believe these flies are decedents from the 2004 release in Denton County. We will continue to monitor the spread of *P. curvatus* in the spring and fall of 2009.

We were also able to successfully release *P. tricuspis* in Denton and in Red River counties. We plan to monitor the expansion of the phorid population at Ray Roberts Park in April and October 2009 using the passive "pizza" trap for 15 miles in each of the four cardinal directions. Also "pizza" traps will be placed in the *P. tricuspis* release area in Red River County in April and October 2009. The continued monitoring of both release sites will help in determining establishment and spread of the flies into new areas.

By introducing and establishing these two parasitic phorid fly, we are providing a natural biological stress on fire ant colonies. Hopefully, this will allow native ants to better compete with this introduced species and regain their territories. As the native ants reclaim their territories, it is hoped that the number of red imported fire ant colonies will decrease. The potential of sustainable fire ant population suppression will thereby reduce the $1.2 billion

annual impact of *S. invicta* in Texas and the need for insecticides for their control will be reduced.

Figure 1. Release site of *P. tricuspis* along the sidewalk below the dam of Lake Ray Roberts, Pilot Point, TX, with flags marking active fire ant mounds.

Figure 2. Passive "pizza" trap used to monitor the establishment and spread of both *P. curvatus* and *P. tricuspis* from the original release sites in Denton and Red River counties.

Figure 3. Release site of *P. curvatus* on a 100 acre site in Clarksville, Red River County, TX.

Table 1. Coordinates of each passive "pizza" trap placed 10 miles north, south, east and west of the original release site at Ray Roberts Park, Denton Co. in order to detect the spread of *P. tricuspis* used on April 15 and October 30, 2008.

Trap	North	South	East	West
1	33.3587N 97.332W	33.3494N 97.0355W	33.3545N 97.039W	33.347N 97.667W
2	33.3627N 97.032W	33.3452N 97.0318W	33.3579N 97.031W	33.344N 97.0625W
3	33.3725N 97.083W	33.3313N 97.0302W	33.3611N 97.0223W	33.3466N 97.644W
4	33.3793N 97.016W	33.3189N 97.0288W	33.3494N 97.0113W	33.3466N 97.0642W
5	33.383N 97.028W	33.3119N 97.0405W	33.3485N 97.0034W	33.3466N 97.0643W
6	33.3857N 97.028W	33.3069N 97.0427W	33.3490N 96.9932W	33.346N 97.06435W
7	In the middle of lake	33.3032N 97.042W	33.3486N 96.9834W	33.346N 97.0716W
8	33.406N 97.047W	33.2914N 97.035W	33.3464N 96.9737W	33.346N 97.08121
9	33.413N 97.046W	33.2815N 97.038W	33.3463N 96.963W	33.346N 97.0834W
10	33.421N 97.051W	33.2678N 97.0483W	33.3484N 96.954W	33.346N 97.0845W

Table 2. GPS coordinates of each active red imported fire ant mounds where *P. tricuspis* were released in Detroit, Red River County, TX beginning on May 12, 2008.

Mound Number	Number of *P. tricuspis* Released	LATITUDE	LONGITUDE
1	30	15SO300611	UTM3725978
2	30	15SO300603	UTM3725976
3	30	15SO300578	UTM3726055
4	30	15SO300638	UTM3725795
5	30	15SO300597	UTM3725772
6	40	15SO300575	UTM3725759
7	30	15SO300647	UTM3725841
8	25	15SO300643	UTM3725860
9	30	15SO300638	UTM3725878
10	30	15SO300628	UTM3725908
11	25	15SO300622	UTM3725930
12	25	15SO300556	UTM3726059
13	15	15SO300512	UTM3726093

Table 3. Number of *P. tricuspis* found on each passive "pizza" trap at each coordinate point from the original site at Ray Roberts Park in Denton County, in each of the four cardinal directions on April 15, 2008.

Trap Number	North	South	East	West
1	0	0	0	0
2	0	0	0	5
3	0	0	0	0
4	0	0	0	0
5	0	0	0	0
6	0	0	0	0
7	0	0	0	0
8	0	0	0	0
9	0	0	0	0
10	0	0	0	0

Table 4. Number of *P. tricuspis* and *P. curvatus* found on each passive "pizza" trap at each coordinate point from the original site at Ray Roberts Park in Denton County, set out on October 30, 2008.

Trap Number	North	South	East	West
1	0	5	5	0
2	2	2	0	1
3	1	4	3	2
4	10	32	0	Missing
5	0	9	4	0
6	2	1	1	6
7	0	2-*P. curvatus*	0	1-*P. curvatus*
8	2-*P. curvatus*	Missing	1	2-*P. curvatus*
9	4-*P. curvatus*	1-*P. curvatus*	1	1-*P. curvatus*
10	2-*P. curvatus*	2-*P. curvatus*	0	0

Table 5. Number of *P. tricuspis* found on each of the 8 passive "pizza" traps in Red River County set out on September 24, 2008 and retrieved September 25, 2008.

Trap Number	Number of Flies Recovered
1	0
2	0
3	1
4	0
5	0
6	0
7	Missing
8	0

Table 6. Number of *P. curvatus* found on each of the 10 passive "pizza" traps in Red River County set out on June 10, 2008 and retrieved June 11, 2008.

Trap Number	Number of Flies Recovered
1	0
2	0
3	0
4	0
5	0
6	1
7	0
8	1
9	2
10	0
11	Missing
12	0

Table 7. Number of *P. curvatus* found on each of the 10 passive "pizza" traps in Red River County set out on September 24, 2008 and retrieved September 25, 2008.

Trap Number	Number of Flies Recovered
1	1
2	9
3	10
4	1
5	0
6	0
7	0
8	0
9	0
10	0

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