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INTRODUCTION OF EXOTIC BIOCONTROLS INTO CALIFORNIA COTTON APHID POPULATIONS

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Abstract

The cotton aphid, *Aphis gossypii* is one of the most damaging pests of cotton and control efforts rely on chemical pesticides. To enhance the natural enemy complex of cotton aphid, two parasites and one entomopathogenic fungus were released into cotton aphid populations in California. One of the parasites, *Aphelinus* near *paramali* successfully overwintered at two nursery sites in the San Joaquin Valley. The fungus, *Neozygites fresenii*, although able to cause very limited infections in the field following release, failed to establish in populations during the season and apparently did not overwinter.

Introduction

The cotton aphid, *Aphis gossypii* is one of the most important pests affecting cotton in the San Joaquin Valley and elsewhere. It is responsible for physiological damage, thus reducing yield and can cause problems with sticky cotton late in the growing season. Estimated losses approach 40,000 - 60,000 bales per year in California alone. While properly applied chemical pesticides can be effective in controlling aphids, few non chemical alternatives exist. With the threat of pesticide use restrictions, it is important to consider biological control as an option for controlling aphids. However, native natural enemies do not effectively keep aphid populations below economic thresholds, especially in mid summer when naturally occurring predators, entomopathogenic fungi and parasitoids are not prevalent (Rosenheim et al. 1997). In an attempt to increase the amount of biological control on cotton aphid in mid to late season cotton, a cooperative project involving CDFA, USDA and University of Arkansas personnel began in 1996 to construct a natural enemy complex using natural enemies not currently found in California to compliment the existing natural enemy complex. The project has resulted in the release of two parasitoids and one entomopathogenic fungus.

Materials and Methods

Parasites

Two parasites were selected for release. *Aphelinus gossypii* (AG) was originally collected from China in 1997 and passed through quarantine in Florida. AG prefers cotton aphid and black citrus aphid while spirea aphid is less preferred (Yokomi and Tang 1995). *Aphelinus* near *paramali* (ANP) was thought to have been initially collected from crape myrtle in Florida in the spring of 1995 but host range studies demonstrated ANP would not attack Crape myrtle aphid. Instead ANP preferred cotton aphid, green peach aphid, black citrus aphid and spirea aphid (unpublished data from Tang et al.) Both parasite species were reared at CDFA and aphid mummies were released weekly into 10 unsprayed cotton nursery sites in the San Joaquin Valley starting in July 2000. Releases were made from the time aphids appeared (usually July) until defoliation. Weekly samples of aphids and parasite mummies were collected during the summer release times and monthly samples were obtained from surrounding weedy plants during the winter. All samples were held for parasite emergence and subsequently identified. Introduced parasites could be easily distinguished from native parasites.

Fungus

The entomopathogenic fungus *Neozygites fresenii* has played a major role in limiting cotton aphid populations in the Southeast US (Steinkraus et al. 1991) but has not been found occurring naturally in California cotton aphid populations (Godfrey et al. 2001). Attempts were made in the mid 1990's to introduce *N. fresenii* into California in efforts to establish a new natural enemy that might reduce populations. However, despite some horizontal transmission that occurred after release, the fungus did not establish nor affect cotton aphid populations (Steinkraus et al. 2002). In 2001 and 2002, efforts were renewed to attempt to introduce the fungus into California using several methods of release and several methods of irrigation. Infected cotton aphid cadavers were collected from an Arkansas field and shipped to California. When aphid populations reached 75 per leaf or more, cadavers were placed onto wetted cotton leaves and covered with a sleeve cage or left uncovered (2001; in 2002 all leaves were covered). All plots received irrigation (either by sprinkler or furrow in 2001 and furrow only in 2002) the day before release. Leaves were sampled 14 days after release in 2001 and at 7, 14, 21 and 30 days after release in 2002.

Live and dead aphids were counted on each leaf and then placed into alcohol. Aphids were mounted in lacto phenol for microscopic examination of *N. fresenii* infection. In 2003, no releases were made but the field from the 2002 release was planted into cotton again and aphid populations were monitored for *N. fresenii* presence.

Results and Discussion

Parasites

A total of 74,650 ANP mummies and 189,140 AG mummies were released at the 10 sites from 2000 through 2002 (Table 1). Both parasites were recovered during the time of parasite releases with more ANP recovered than AG (Table 2). A total of 813 ANP, 349 AG and 7,038 native aphidiid parasites were recovered from the nursery sites. The majority of the parasites were recovered during the cotton season when releases were made and aphid numbers were high. However, only ANP was recovered at 2 of the 10 nursery sites during the winter and early spring suggesting that it can over winter in the San Joaquin Valley.

Fungus

Although a few infections were observed in 2001 (Tables 3, 4), infections in 2002 (Table 5) were very minimal. Each year, the aphid population crashed but the reduction in population could not be attributed to *N. fresenii*. In 2001, one leaf had 20 infected aphids but no infections were observed on any leaf in close proximity suggesting the fungus did not spread beyond the initial release sites. In 2003, a careful search of the 2002 release field did not reveal any *N. fresenii* infections despite high aphid numbers. It is unclear why *N. fresenii* did not persist in these studies when, in Arkansas, it plays such a predominant role in aphid population dynamics. We suggest several hypotheses for this lack of success. First, it is possible not enough inoculum was released into the environment. Although more than 50% of the released cadavers sporulated each year, the limited number of cadavers may have reduced the opportunity for individual conidia to land on a suitable host. There is currently no mass production system for *N. fresenii* and the tests relied on field-collected cadavers. Second, the aphid population in California may be slightly different genetically, thus the strain of *N. fresenii* may not be as capable of infecting California populations as it is Arkansas populations. In laboratory tests with greenhouse reared California cotton aphids, infections were difficult to obtain using standard techniques. Third, the arid climate of California may be too severe for *N. fresenii* to rapidly spread and persist during the summer. Although irrigation was applied before cadavers were released, interruptions of sufficient humidity during critical phases of the life cycle of *N. fresenii* could end the infection cycle. It is interesting to note that previous surveys have revealed other fungi attacking California cotton aphid populations but these fungi are only observed in the cooler wetter months of the winter (Godfrey et al. 2001).

Conclusions and Future Work

Classical biological control of cotton aphids in California has proven to be a challenge. Although some overwintering of *Aphelinus nr paramli* has occurred, it is unlikely to play a role in cotton aphid population dynamics in the near future. However, efforts will continue to monitor parasite populations and their effect on aphid populations. In addition, another parasite, *Lipolexis oregmae* is being imported into California for study. Assuming host range tests are acceptable, this parasite will also be released following proper permitting. Insect fungi will also be pursued. The hot, dry conditions occurring in the central valley of California are generally not conducive to fungal epizootics and, while irrigation application may provide temporary periods of high humidity, there may be some problems with horizontal transmission if humidity required for sporulation is not present at critical times. However, it is very important to recognize the role *N. fresenii* plays in the Southeast US and it may be possible to find other strains of the fungus that are better adapted to cotton aphid populations in the Western US. Efforts will continue to identify these strains and the environmental parameters important to successful introduction of fungi.

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Table 1. The total number of ANP and AG mummies released at each parasite nursery site and the dates of parasite releases in the San Joaquin Valley from 2000 through 2002.

	2000 (Dates of Release)		2001 (Dates of Release)		2002 (Dates of Release)	
Nursery Site	**ANP**	**AG**	**ANP**	**AG**	**ANP**	**AG**
Madera 1	1,700 (7/19/00 – 10/11/00)	2,480	4,950 (7/11/01 – 10/11/01)	8,800	600 (7/18/02-9/10/02)	1,700
Madera 2	1,500 (8/1/00 – 10/11/00)	2,080	4,250 (8/8/01-10/11/01)	5,500	1,100 (7/18/02 – 9/10/02)	4,700
Madera 4	1,800 (7/19/00 – 10/11/00)	2,480	4,450 (8/1/00 – 10/11/00)	5,800	600 (8/21/02 – 9/10/02)	2,200
Merced 3	1,400 (7/27/00 – 10/11/00)	2,180	4,150 (7/24/01 – 10/3/01)	5,050	600 (8/21/02 – 9/10/02)	1,200
Kern 1	700 (7/27/00 – 9/21/00)	3,900	3,750 (8/1/01 – 10/10/01)	2,850	4,050 (7/10/02 – 10/23/02)	16,900
Kern 2	1,050 (7/17/00 – 9/28/00)	3,550	3,850 (7/25/01 – 10/3/01)	3,000	1,850 (7/10/02 – 10/9/02)	11,600
Kern 3	1,600 (7/27/00 – 10/12/00)	4,855	5,350 (7/18/01 – 10/10/01)	4,650	2,150 (7/10/02 – 10/30/02)	19,750
Kern 4	1,950 (7/27/00 – 10/12/00)	5,305	2,950 (8/8/01 – 9/27/01)	1,150	2,250 (7/10/02 – 10/30/02)	21,150
Kern 5	1,950 (8/4/00 – 10/12/00)	5,355	4,650 (7/18/01 – 10/3/01)	2,850	1,850 (7/10/02 – 10/2/02)	17,500
Kern 6	1,700 (7/27/00 – 10/12/00)	5,455	3,250 (8/1/01 – 9/27/01)	2,000	2,650 (7/10/02 – 10/30/02)	13,150

Table 2. The total number of primary parasites recovered from nursery sites in the San Joaquin Valley in 2000-2002. Parasite releases were conducted from July through November. Overwintering sampling was conducted from December through June.

	Time of	**ANP**			**AG**			**Native Aphidiidae**		
Site	**Season**	**2000**	**2001**	**2002**	**2000**	**2001**	**2002**	**2000**	**2001**	**2002**
Madera 1	Jul. - Nov.	4	6	0	0	0	0	28	7	0
	Dec. - Jun.	0	0	0	0	0	0	0	4	0
Madera 2	Jul. - Nov.	36	4	1	1	1	1	21	0	47
	Dec. - Jun.[a]	0	0	0	0	0	0	0	0	0
Madera 4	Jul. - Nov.	5	15	1	1	0	0	23	0	79
	Dec. - Jun.[a]	0	0	0	0	0	0	0	0	0
Merced 3	Jul. - Nov.	4	3	0	3	0	0	21	19	0
	Dec. - Jun.	1	3	0	0	0	0	0	5	0
Kern 1	Jul. - Nov.	0	14	83	0	1	123	198	359	76
	Dec. - Jun.	0	3	2	0	0	0	25	49	17
Kern 2	Jul. - Nov.	12	4	30	6	0	22	729	689	32
	Dec. - Jun.	0	0	0	0	0	0	9	102	1
Kern 3	Jul. – Nov.	19	103	235	6	3	77	1,094	117	64
	Dec. – Jun.	0	0	0	0	1[b]	0	0	236	7
Kern 4	Jul. – Nov.	24	33	31	10	0	27	657	244	129
	Dec. – Jun.	0	0	0	0	0	0	7	16	10
Kern 5	Jul. – Nov.	44	13	44	12	0	27	643	644	56
	Dec. – Jun.	0	0	0	0	0	0	0	4	58
Kern 6	Jul. – Nov.	13	4	19	2	0	25	155	202	140
	Dec. – Jun.	0	0	0	0	0	0	2	6	7

Table 3. Average number of aphids/leaf two weeks after fungus release (with sleeve cages) 2001.

Date	Water	Treatment (n)	Live	Dead	Infected
Aug 14	Furrow	Fungus (2)	15.5	17.5	3.5
		Control (4)	30.5	8.8	0
		F	0.18	10.90*	>100*
	Sprinkler	Fungus (3)	7.7	25.0	7.7
		Control (4)	52.2	16.3	0
		F	1.22	0.61	>100*
Aug 31	Furrow	Fungus (4)	9.0	4.8	0
		Control (5)	3.2	3.2	0
		F	0.50	0.34	
	Sprinkler	Fungus (4)	190.0	14.8	0
		Control (5)	31.8	3.8	0
		F	10.46*	6.47*	

* P<0.05

Table 4. Average number of aphids/leaf two weeks after fungus release (no sleeve cages) 2001.

Date	Water	Treatment (n)	Live	Dead	Infected
Aug 14	Furrow	Fungus (7)	1.3	13.7	7.6
		Control (9)	6.7	4.7	0
		F	11.63*	7.50*	8.59*
	Sprinkler	Fungus (8)	21.6	8.5	5.1
		Control (9)	41.2	7.1	0
		F	0.31	0.22	4.92*
Aug 31	Furrow	Fungus (9)	20.4	9.1	0.55
		Control (10)	2.0	15.4	0
		F	1.88	2.37	1.75
	Sprinkler	Fungus (7)	6.4	11.1	0.14
		Control (9)	19.7	7.9	0
		F	2.8	0.29	1.31

*P<0.05

Table 5. Results from release of *Neozygites fresenii*-infected cadavers on population dynamics of the cotton aphid in sleeve cages, 2002.

Days after Release	Treatment (n)[1]	Average Live Aphids	Average Dead Aphids	Average Percentage Mortality	# Leaves with Infected Aphids (n)[2]	Average Percentage Infection
7	Fungus (31)	400.6	20.1	5.7	3 (10)	0.6
	Control (9)	181.7	8.0	4.1	0 (4)	
14	Fungus (20)	72.7	90.2	66.1	2 (11)	1.0
	Control (8)	279.3	60.5	28.3	0 (3)	
21	Fungus (19)	215.1	43.5	54.2	1 (10)	1.0
	Control (6)	7.3	23.0	76.7	0 (3)	
30	Fungus (22)	159.1	73.2	62.3	0 (10)	0.0
	Control (7)	36.4	80.4	63.6	0 (3)	

[1] n= number of leaves examined. This includes the treated leaf and one leaf above and one leaf below for each sleeve cage. Some leaves were desiccated and were not included in the assay.
[2]Aphids from n treated leaves only were examined for presence of *N. fresenii*.