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OVERWINTERING OF *APHELINUS* NEAR *PARAMALI*
(HYMENOPTERA: APHELINIDAE), AN INTRODUCED PARASITE
OF THE COTTON APHID IN THE SAN JOAQUIN VALLEY, CALIFORNIA

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Natural biological control influencing cotton aphid population dynamics in the San Joaquin Valley does not maintain cotton aphid [*Aphis gossypii* Glover (Homoptera: Aphididae)] densities at low levels throughout the year. In the late fall through late spring, native insect parasites, generalist arthropod predators, and fungi typically maintain cotton aphid densities at non-pest status (Rosenheim et al. 1997, Godfrey et al. 2001, K. Godfrey and M. McGuire, unpublished data). However, in mid to late season cotton, the action of the natural enemies often appears to be minimal. The arthropod predator complex loses effectiveness due largely to hemipterous predators (reduviids and nabids) feeding upon lacewing larvae, another group of predators that feed heavily upon aphids (Rosenheim et al. 1993, Rosenheim and Cisneros 1994). Also, at this time, the native parasites and fungi are at their lowest levels (Godfrey et al. 2001, K. Godfrey and M. McGuire, unpublished data). Hot, dry climatic conditions that are not conducive for parasite survival and/or fungal growth exist at this time (Tang and Yokomi 1995, Godfrey et al. 2001).

In an attempt to increase the amount of biological control on cotton aphid in mid to late season cotton, a cooperative project began in 1996 to construct a natural enemy complex using natural enemies not currently found in California to compliment the existing natural enemy complex. After four years of research, two parasite species, *Aphelinus* near *paramali* and *Aphelinus gossypii* Timberlake (Hymenoptera: Aphelinidae), were identified as the first components of the introduced natural enemy complex. In field cage studies conducted in cotton over 4 years (1996-1999), these parasites reduced cotton aphid densities 10-38% as compared to the densities of cotton aphid in cages without parasites present (K. Godfrey, J. McLaughlin, and M. McGuire, unpublished data). Distribution of both parasites began at ten nursery sites in 2000. The nursery sites were monitored to determine if the parasites had begun to overwinter and establish at the sites.

Both parasites were obtained from researchers in Florida. *Aphelinus* near *paramali* (ANP) was thought to have been initially collected from crape myrtle aphid (*Tinocallis kahawaluokalani* (Kirkaldy)) on crape myrtle in Florida in the spring

of 1995. However, additional host range studies demonstrated that ANP would not attack crape myrtle aphid, but preferred cotton aphid, green peach aphid (*Myzus persicae* (Sulzer)), black citrus aphid (*Toxoptera aurantii* (Fonscolombe)), and spirea aphid (*Aphis spiraeicola* Patch; Homoptera: Aphididae) (Y. Tang, L. Osborne, and R. Yokomi, unpublished data). Female wasps preferred younger aphid instars for oviposition and would host feed on all aphid instars. The adult females survived 10-20 days, laid about 120 eggs, and fed upon about 25 aphids at 21°C (Y. Tang, L. Osborne, and R. Yokomi, unpublished data). *Aphelinus gossypii* (AG) was collected in southern China in July 1997 and passed through quarantine in Florida. Host preference studies have shown that cotton aphid and black citrus aphid are the preferred hosts, while spirea aphid is a less acceptable host (Yokomi and Tang 1995). The size of the adult parasite produced is greatest when AG is reared on cotton aphid (Yokomi and Tang 1995). Female AG host fed on all aphid instars. They survived from 5-17 days and had a mean fecundity of 57 eggs (range 20-115; Tokumaru and Takada 1996).

All parasites were reared at the California Department of Food and Agriculture Biological Control Program in Sacramento. Each species was maintained in a greenhouse at daytime temperatures (16 hours) of 24°C ($\pm 3^\circ\text{C}$) and nighttime temperatures (8 hours) of 20°C ($\pm 3^\circ\text{C}$) with supplemental lighting (600 watt high pressure sodium lights). ANP was reared on green peach aphid on potted green pepper plants, while AG was reared on cotton aphid on potted hibiscus plants. The parasites were reared on different host aphids and plants to insure that there was no cross contamination of parasite colonies. ANP was periodically tested to insure that it would accept cotton aphid as readily as green peach aphid. Given the above rearing conditions, parasite mummies (i.e., mummified aphids containing parasite pupae) could be found about 7 days after adult parasites were introduced into a cage, and new adult parasites began to emerge from the mummies in about 3-5 days. Approximately 56% (range 44-63%) of all ANP mummies produced adults within 7 days of the first adult emergence, and 60% (range 55-67%) of AG mummies produced adults. The sex ratio was approximately 1:1.

The nursery sites were established in Merced, Madera, and Kern Counties in and around cotton fields. Each nursery was located on a corner of a cotton field and was 8 rows wide by 10 m in length. The nursery was not sprayed with insecticides during the cotton-growing season. Most sites had other habitats that were favorable for cotton aphid throughout the year in close proximity to the nursery. Beginning in July of each year, each site was sampled weekly by examining 40 to 80 cotton plants for the presence of cotton aphid. Once cotton aphids were found, weekly releases of both parasites began and continued until the cotton at each site was defoliated. To make a release, brown paper bags containing leaves with parasite mummies that were about to emerge (i.e., adult emergence within 2-5 days) were opened and placed between cotton plants along a single row in the nursery prior to 10:00 am. No special accommodations were necessary to protect the mummies from predation. The densities of native arthropod predators were at their lowest levels at the time of the releases. In addition, ants were not abundant at any of the nursery sites.

A total of 74,650 ANP mummies and 189,140 AG mummies were released at the 10 sites from

2000 through 2002 (Table 1). Approximately 2 weeks after the parasite releases began, samples of cotton leaves with aphids and mummies were collected weekly. These aphids and mummies were held in the laboratory (24°C, 12L:12D) for adult parasites to emerge. The sites were sampled on a weekly basis until approximately 2 weeks after cotton harvest. After that time, the areas around the nursery site that harbored cotton aphid were sampled at approximately monthly intervals. Any aphids or parasite mummies found in these samples were returned to the laboratory and held for parasite emergence. The number and identity of the parasites was recorded for each sample site and date.

A total of 813 ANP, 349 AG, and 7,038 native aphidiid parasites were recovered from the nursery sites (Table 2). The introduced aphelinids could be distinguished from the native aphidiids using family characteristics. The aphidiids recovered belonged to the genera *Lysiphlebus*, *Aphidius*, and *Diaeretiella*. A small number (38) of native aphelinids were also recovered. These individuals could be distinguished from ANP and AG because they had more than 15 setae in the triangular area at the base of the forewing. ANP and

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.

| Nursery Site | 2000 (Dates of Release) | | 2001 (Dates of Release) | | 2002 (Dates of Release) | |
|--------------|-----------------------------|-------|-----------------------------|-------|-----------------------------|--------|
| | 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.

| Site | Time of Season | ANP | | | AG | | | Native Aphidiidae | | |
|----------|------------------------|------|------|------|------|----------------|------|-------------------|------|------|
| | | 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 ^b | 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 | 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 |

^aIn 2001 and 2002, overwintering sites for Madera 1, 2, and 4 were located at one site due to the close proximity of the three sites.

^bAG recovered approximately 1 month after an early season release at this site.

AG have 2-5 setae in this area of the forewing (Zehavi and Rosen 1988).

The majority of all parasites were recovered during the cotton season when releases were made (Table 2). This is not surprising considering that this is when cotton aphid densities are the largest. In general, the density of aphids in Madera and Merced Counties was less than in Kern County. In 2000, cotton aphid densities in cotton began to increase in mid-July and peaked in mid to late September with densities of 1.25-3.75 aphids per leaf in Madera and Merced Counties and 25-50 aphids per leaf in Kern County. In 2001, the density increase began later in the season and peaked in mid-August at 2-5 aphids per leaf in Madera and Merced, and 10-15 aphids per leaf in Kern. Numbers of aphids in all counties declined about 3-fold in mid-September. The cotton aphid populations in 2002 were much lower than in previous years. In Madera and Merced Counties, only 1 site had significant aphid numbers (5 aphids per leaf), and the other three sites had less than 1 aphid per leaf throughout the cotton season. In Kern County, cotton aphid densities began to increase in mid-July, but peaked in September at 3-5 aphids per leaf. Cotton aphid densities when cotton was not in the field (December to June) for all years remained at less than 1 aphid per leaf on alternate hosts.

Both parasites were recovered during the time of parasite releases with more ANP recovered than AG (Table 2). However, only ANP was recovered at 2 of the 10 nursery sites during the winter and early spring, suggesting that it could overwinter in the San Joaquin Valley. At the first nursery site, Merced 3, ANP was recovered in samples in February 2001 and 2002 (Table 2). A total of 6,150 ANP had been released in this nursery during the cotton seasons of 2000-2002 (Table 1). This site was adjacent to a river with winter vegetation that supported sufficient aphid densities to allow ANP to overwinter.

The second nursery site where ANP was found to overwinter was in Kern County (Kern 1). ANP was recovered in samples collected in April 2002 and March 2003 (Table 2). A total of 8,500 ANP had been released in this nursery during the cotton seasons of 2000-2002 (Table 1). This nursery

site was adjacent to a nectarine orchard with winter ground cover that provided habitat for the cotton aphid in the winter and spring.

SUMMARY

In an attempt to increase the amount of biological control exerted on the cotton aphid in the San Joaquin Valley, two aphelinids parasites were imported, evaluated for their ability to reduce cotton aphid densities, and released at ten nursery sites from 2000 through 2002. One of the parasites, ANP, was recovered for two consecutive years at two of the ten nursery sites. These recoveries suggest that ANP may be able to overwinter in the San Joaquin Valley and may have begun to establish.

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