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Author: Honda, Jeffrey Y.

Source: Florida Entomologist, 88(3): 325-326

Published By: Florida Entomological Society

URL: https://doi.org/10.1653/0015-4040(2005)088[0325:PNAATP]2.0.CO;2

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PARTITIONING NATIVE AND AUGMENTATIVE *TRICHOGRAMMA PLATNERI* (HYMENOPTERA: TRICHOGRAMMATIDAE) PARASITISM OF *AMORBIA CUNEANA* (LEPIDOPTERA: TORTRICIDAE) EGG MASSES IN SOUTHERN CALIFORNIA AVOCADO ORCHARDS

JEFFREY Y. HONDA

Department of Biological Sciences, San Jose State University, One Washington Square, San Jose, CA 95192-0100

The objective of this experiment was to test the efficacy of augmentative releases of Trichogramma platneri Nagarkatti against the avocado pest, Amorbia cuneana Walsingham, and to determine natural parasitism rates concurrently in the field by examining parasitism of sentinel A. cuneana egg masses placed in avocado orchards. The released wasps used in the experiments were marked with a unique phosphoglucose mutase (PGM) allele from T. minutum Riley that is absent in the coastal, native populations of the closely related T. platneri (Pinto et al. 1992). Wasps were produced by introducing the unique PGM marker into native T. platneri parasitoids by repeated backcrossing (>20 generations), to ensure that released lines were different only at the PGM locus. Wasps from each generation were electrophoresed for the unique PGM locus by the methods of Kazmer & Luck (1995) to ensure culture purity and quality assurance.

Studies were done in three blocks of 'Haas' avocado on ranches located at Temecula (Riverside Co.), Vista (San Diego Co.), and Moorpark (Ventura Co.), California. Orchard characteristics were quite variable between the Moorpark site and the Temecula/Vista sites. Temecula and Vista orchards were categorized as 'mature' and possessed trees that were at least 25 years in age, over 12.0 m in height and spaced between 7.5 and 10.5 m apart. In contrast the Moorpark orchard was categorized as 'immature' and possessed trees that were approximately 10 years old, less than 4.5 m in height, and spaced between 4.5 and 6.0 m apart. Parasitism and dispersal was evaluated by monitoring sentinel A. cuneana egg mass traps placed in 10 trees per orchard. Egg-masses were placed in nine avocado trees (30 egg masses per tree) arranged in the shape of a cross. Thus, there were two trees per arm of the cross and one tree at the center beneath which wasps were released. An additional tree located 10 rows east of the release tree containing egg masses served as a control. Within each tree, 15 egg-masses were arranged in the tree canopy 0.5-1.5 m above the ground and 15 egg-masses were placed 3.0-6.0 m above the ground in the tree canopy.

Egg-masses were placed in the ten experimental trees on day one. These egg masses were used to determine the amount of parasitism by resident *T. platneri* prior to release of the marked line. These egg-masses were replaced with fresh egg-masses on day four and followed immediately by the release of marked *T. platneri* in the center tree of each plot. The second egg-mass group was collected on day seven of the experiment and replaced with fresh egg-masses that were then collected on day 10 of the experiment. Thus, a total of 900 egg masses were placed in each orchard over a 10 d sampling period.

Approximately 40,522 wasps were released under the centrally located release tree. Eggmass cards were collected and blackened eggs indicating parasitism were separated to collect parasitoids. Egg parasitism was calculated as the percentage of egg masses with at least one egg parasitized. Emerging parasitoids were snap frozen in liquid nitrogen and electrophoresed for PGM to determine if they were from the resident or released populations. Differences in the number of egg-masses parasitized between canopy treatments within trees, parasitism rate differences between adjacent and non-adjacent trees within an orchard, and comparisons between plot sites (mature vs. immature orchards) were analyzed by ANOVA (SAS Institute Inc. 1988).

Egg-masses placed in the experimental groves prior to release of the marked wasps remained unparasitized. Released wasps constituted almost all of the recorded parasitism as indicated by the presence of the unique PGM allele collected from wasps emerging from the sentinel eggmasses. A total of three sentinel egg-masses from a single tree at the Moorpark site were collected which contained wasps that had PGM alleles differing from those of the released T. platneri, however, they were similar to those of resident coastal T. platneri (Pinto et al. 1992), indicating that these wasps were the progeny of resident T. plat*neri*. This represents only a small percentage (0.33%) of the observed parasitism in this plot. None of the 1,800 sentinel egg-masses placed in 10 trees over the three monitoring periods in either the Temecula or Vista plots showed T. platneri allozyme patterns typical of resident wasp populations.

Augmentative parasitism rates of the sentinel egg-masses in the upper and lower portions of the trees for all three locations were not significant ($F_{1.108} = 3.31 \ P > 0.05$). Parasitism rates of egg masses collected on day seven were significantly higher than those collected on day 10 for all three orchards studied with 78%, 89%, and 94% of the total parasitism observed occurring within three days of release for the Temecula, Vista, and Moorpark sites, respectively (Table 1). At each site,

Treatment Pre Release	Release (eggs collected three days after release)	Post Release (eggs collected six days after release)
0/270 (0%)	71/270 (26.3%)	$9/270^{*}$ (3.3%)
0/270 (0%)	64/270 (23.7%)	$18/270^{*}$ (6.6%)
0/270 (0%)	150/270 (55.6%)	10/270* (3.7%)
	Treatment Pre Release 0/270 (0%) 0/270 (0%) 0/270 (0%)	Treatment Pre Release Release (eggs collected three days after release) 0/270 71/270 (0%) 0/270 64/270 (0%) 0/270 64/270 (0%) 0/270 150/270 (0%)

 TABLE 1. NUMBERS OF EGG MASSES PARASITIZED BY TRICHOGRAMMA PLATNERI BEFORE, DURING, AND AFTER RELEASE

 IN AVOCADO TREES IN THREE ORCHARDS.

*Each row indicates significance at the 0.05 level for chi-square tests of independence between release and post release treatments for each of the three orchards.

parasitism rates between the outermost trees and trees adjacent to the release tree decreased significantly ($F_{1,108} = 41.02 \ P < 0.05$). Thus, parasitoid searching efficiency appeared to be limited to the few trees in close proximity to the release tree and only for a brief period (<3 days) after release.

Parasitism rates appear to be affected by plant complexity and interplant distance based on this experiment as the Moorpark plot with smaller and closely spaced trees had higher levels of parasitism ($F_{2,108} = 11.79 P < 0.05$). Larger, more complex trees may cause parasitoids to search more area per host encounter than in smaller less complex trees.

In conclusion, although indigenous T. platneri were extremely scarce, the use of unique allozymes incorporated into wasps released in the field may be an effective tool to accurately determine indigenous and augmentative parasitism rates concurrently. Moreover, augmentative releases are most effective three days after release and only in those trees immediately adjacent to parasitoid release points. Thus, point releases of T. platneri should not be spaced more than a few trees apart and should be performed every 3-4 days against A. cuneana in avocado.

SUMMARY

The use of a unique PGM allozyme marker was introduced into a culture of *Trichogramma platneri* used for augmentative field releases in an effort to distinguish between native and augmentative parasitism against *Amorbia cuneana* in avocado orchards. Although native parasitism rates were extremely low, the marker was useful in distinguishing native parasitoids from those released in the field. Augmentative releases were most effective up to three days post release and in those trees adjacent to the release trees. Orchard composition appears to affect parasitoid efficiency.

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