Reassessment of the Role of Predators and *Trissolcus basalis* in Biological Control of Southern Green Stink Bug (*Hemiptera: Pentatomidae*) in Hawaii

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The role of predators and parasitoids in biological control of the southern green stink bug, *Nezara viridula* (L.), in macadamia nut orchards was investigated in three field studies in Hawaii. Egg masses placed in the weeds at the orchard borders had significantly higher rates of parasitism by *Trissolcus basalis* (Wollaston) in 1990 and 1991 than eggs placed in the canopy of macadamia trees. Predators were more efficient at locating and destroying egg masses placed in trees than in weeds and were always more efficient than *T. basalis*, regardless of their location. An island-wide survey taken from July to December 1991 recovered no *T. basalis*, but predators were responsible for high levels of egg mortality at all locations. The importance of these findings in relationship to the commonly held view that *N. viridula* is under good biological control by *T. basalis* in Hawaii is discussed. © 1995 Academic Press, Inc.

**Key words:** biological control; *Trissolcus basalis*; *Nezara viridula*; *Macadamia integrifolia*.

Biological control of the southern green stink bug, *Nezara viridula* (L.), has been considered a landmark example of classical biological control of an introduced pest (Caltagirone, 1981). Although several authors (Clausen et al., 1977; DeBach, 1974; Waterhouse and Norris, 1987) claimed that substantial control of the southern green stink bug was achieved in Australia, New Zealand, and Hawaii, their information was based on the review of limited postrelease evaluations performed by other researchers. The success of the biological control programs in these three areas is generally considered to be the result of multiple introductions of different geographical strains of the egg parasitoid *Trissolcus basalis* (Wollaston) (Hymenoptera: Scelionidae) originally into Australia and later into Hawaii (Caltagirone, 1981). However, recent studies by Clarke and his co-workers have shown that percentage parasitism of *N. viridula* eggs by *T. basalis* in South East Queensland is commonly low (0 to 30%) and little evidence exists that the “success” of the Australian biological control project is complete or even substantial (Clarke, 1990, 1992a,b).

The southern green stink bug was first discovered in Hawaii in October 1961 feeding on macadamia nuts and vegetables on the island of Oahu (Davis, 1967). Subsequent collections of stink bugs occurred on all major Hawaiian islands by 1963. Upon its discovery in Hawaii, the Hawaii Department of Agriculture (HDOA) initiated a biological control program aimed at reducing *N. viridula* to subeconomic status (Davis, 1964). A total of seven species of egg parasitoids were introduced along with the adult parasitoid *Trichopoda pennipes* (F.) (Diptera: Tachinidae). The Hawaiian project was considered a success, and statewide releases of parasites were discontinued by 1966 (Davis, 1967). However, periodic small scale releases of *T. basalis* were performed on the island of Oahu in 1976, 1977, and 1983 for control of stink bugs using *T. basalis* colonies initiated from field collections (P. Conant, HDOA, personal communication). No further assessments of the effectiveness of *T. basalis* were regularly performed in Hawaii. In 1989, macadamia growers on the southwest (Kona) side of the island of Hawaii complained of extensive losses from the southern green stink bug. Jones and Caprio (1992) found that losses on certain cultivars over the 3 months of peak harvest exceeded 60%. Their data suggested that complete biological control of stink bug was not occurring. The studies reported herein were initiated to determine the effect of *T. basalis* and other parasitoids and predators on southern green stink bug eggs in both weeds at the edge of macadamia orchards and within macadamia trees. The comparison of the predation and parasitism in the trees and weeds was performed because other species of *Trissolcus* have been shown to partition the environment and preferentially occur in either low lying vegetation or within the tree canopy (Okuda and Yeargan, 1988).
MATERIALS AND METHODS

Monitoring method. Sentinel egg masses were used as described by Powell and Shepard (1982). Egg masses were collected from a colony of the southern green stink bug maintained in the laboratory at the University of Hawaii at Manoa and frozen at -20°C. Stink bug adults in laboratory culture occasionally prey on their own eggs. Therefore, each egg mass was examined under a dissecting microscope and eggs fed upon were marked with a fine-tipped permanent marker before they were placed in the field.

Egg masses were placed in the field for a period of no more than 1 week, retrieved and placed individually in 5-cm-diameter tight-fitting petri dishes, and held at ambient temperature (≈22–27°C) for parasitoid emergence for a minimum of 1 month. After the waiting period, each egg mass was examined under a dissecting microscope to determine whether eggs had been parasitized (as indicated by emergence hole or unemerged parasitoid) or eaten by predators. For each egg mass, the total number of eggs per mass, the number of eggs available (total eggs minus those eaten by stink bug adults), the number from which parasitoids had emerged, the number of parasitoids that had not emerged, and the number of eggs in a mass destroyed by predators were recorded. Parasitoids emerging were mounted and sent to Anthony Clarke (University of Tasmania, Australia) for identification of Trissolcus spp. and to Gary Gibson (Agriculture Canada, Ottawa) for identification of Anastatus sp.

Egg mortality experiments. Two field studies were designed to determine egg mortality caused by natural enemies of southern green stink bug at the UH Kona Experiment Station (Hawaii County). In 1990, 10 egg masses were placed in the weeds at the edge of a 0.6-ha macadamia orchard and 10 egg masses (one per tree) were placed inside the orchard between 1 and 2.5 m high at the edge of the tree canopy. Weeds consisted of a mixture of Crotalaria juncea (Desv.), Emilia sonchifolia L., Bidens pilosa L., Desmodium uncinatum (Jacq.), and Amaranthus spinosus L.). Locations for egg mass placement were randomly chosen each time. Egg masses were placed in the field between August 20 and December 13. Between September 4 and September 24, a 3- to 5-mm-wide ring of Tanglefoot was placed around each egg mass to determine if predation was obscuring parasitism by T. basalis or other parasitoids. Egg masses were changed at 3- to 7-day intervals over the entire period, depending on their availability.

In 1991, the study was repeated using the same methods and site, except the study was started on May 21 and ended December 20. Egg masses were replaced weekly, and at no time were egg masses ringed with Tanglefoot. Insecticides were not applied to the orchard during either year.

Island-wide survey. To determine parasitism and predation trends throughout the macadamia growing regions of the island of Hawaii, eight orchards were monitored using sentinel egg masses between July 17 and December 20, 1991. Because of logistical reasons, 10 egg masses were placed in each orchard once a month and removed 1 week later. Mortality of N. viridula eggs in weeds was assessed in only one of the orchards because of limited availability of egg masses. Insecticides were not applied in any of the study orchards.

Analysis. Mortality imposed on stink bug egg masses by natural enemies is composed of two major components. The first is the ability of a natural enemy to find an egg mass, and the second is the proportion of the eggs destroyed when an egg mass is discovered. It is important to consider the two components separately because a higher searching ability of a natural enemy can be offset by destruction of only a portion of the eggs within a mass and vice versa. The combination of these two factors is the proportion of eggs placed in the field that were destroyed and is a measure of the overall mortality imposed by a natural enemy.

The ability of natural enemies to find and destroy egg masses in trees and weeds was compared at the UH Experimental Farm in Kona. The ability to find egg masses in trees compared to weeds was analyzed using 2 x 2 G tests performed separately on data from 1990 and 1991. The data were pooled for the entire season for each experiment so that overall ability to find egg masses in the different habitats and by the different natural enemies could be determined.

The percentage of an egg mass destroyed once it was found by a natural enemy and the proportion of eggs placed in the field destroyed (overall egg mortality) were tested using ANOVA to determine if egg destruction was significantly different in the weeds and trees. ANOVA was run separately for parasitoids and predators for all experiments. A sin^-1 sqrt(x) transform was used before an ANOVA. The data from September 4 to 24, 1990 (ringed with Tanglefoot), were not included in the 1990 analysis, but were analyzed separately using the same methods.

RESULTS

During the study, only T. basalis (both sexes) and Anastatus sp. (Hymenoptera: Eupelmidae) (males only) were reared from eggs of N. viridula. Because only male Anastatus were recovered, their identification to species was not possible. The ants Monomorium floricola (Jerdon) and Pheidole megacephala (F.) were regularly seen preying on egg masses. M. floricola nests in trees in sticktight nuts (nuts whose abscession layer dies and which remain on trees from the previous season) while P. megacephala is a ground nesting species.
TABLE 1
Season-Long Destruction of N. viridula Sentinel Egg Masses in Kainaliu, Hawaii

<table>
<thead>
<tr>
<th>Sentinel egg location</th>
<th>% Egg masses found</th>
<th>When found by a natural enemy, % egg mass destroyed</th>
<th>% Eggs placed in field destroyed</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Parasitoids&lt;sup&gt;*&lt;/sup&gt;</td>
<td>T. basalis</td>
<td>Anatusus sp.</td>
</tr>
<tr>
<td>1990 Trees</td>
<td>4.6</td>
<td>2.8</td>
<td>3.4</td>
</tr>
<tr>
<td></td>
<td>16.9</td>
<td>13.5</td>
<td>6.9</td>
</tr>
<tr>
<td>Weeds</td>
<td>3.5</td>
<td>1.2</td>
<td>3.1</td>
</tr>
<tr>
<td></td>
<td>8.3</td>
<td>4.7</td>
<td>3.6</td>
</tr>
</tbody>
</table>

<sup>*</sup> Percentage destroyed calculated by using only those egg masses discovered by the given natural enemy. Mean with interquartile range in parentheses.
<sup>1</sup> Data for T. basalis and Anatusus sp. pooled.
Detection of *N. viridula* egg masses by predators was high at all locations. A minimum of 36.7 and a maximum of 92.6% of the egg masses were found by predators at the different orchards during the experiment (Table 2). As in the Kainaliu orchard, across all orchards the highest numbers of egg masses were found from August to October, and they decreased slightly during the other months (Fig. 3).

Considering the proportion of *N. viridula* eggs eaten during the entire experiment, variation between orchards was low except for two orchards in Pahala (Table 2). In the majority of the orchards, predation varied between approximately 45 and 77%. However, in both Pahala orchards <33% of the eggs were eaten (20.9 and 32.3%) over the course of the season. Examination of the data by month shows that at the first Pahala orchard egg predation was low during the first and last 2 months and was >35% only once. In the second orchard, predation of *N. viridula* eggs was >35% twice (Fig. 4).

**DISCUSSION**

Results from these experiments cast strong doubt on the prominent role attributed to *T. basalis* in the biological control of *N. viridula* in Hawaii. In the macadamia agroecosystem, my results clearly indicate that predation is much more important than *T. basalis* in limiting stink bug survival through the egg stage. Predation is probably attributable primarily to ants, but other insects may also be important. *P. megacephala*

parasitized *N. viridula* egg masses placed in trees, but was most common in eggs placed in weeds ($F = 19.5$, $df = 1, 334, P < 0.0001$), particularly in August and early October. Parasitism of eggs by *Anastatus* was not significantly different in the weeds and trees ($F = 0.58$, $df = 1, 334, P < 0.45$).

In 1991, parasitism of *N. viridula* eggs was rare throughout the season (Fig. 2). Egg predation was highest in the trees from August through December, and as in 1990, egg predation was lower ($F = 5.6$, $df = 1, 448, P < 0.02$) and more variable in weeds (Fig. 2). Peak egg predation in the weeds occurred at approximately the same time as in the trees, but was low in early May and in late October.

When destruction of *N. viridula* eggs was calculated over the entire sampling period in 1990, only 1.2 and 8.6% of the eggs were destroyed by parasitoids in the trees and weeds, respectively. During the same period, predators destroyed 26.0 and 14.5% in trees and weeds, respectively (Table 1). In 1991, parasitism of eggs dropped to 0.2 and 1.7% in the trees and weeds, respectively, while predation increased to 47.7 and 36.9%, respectively.

**Island-wide survey.** Parasitism of *N. viridula* eggs by *T. basalis* was not recorded from any of the orchards in the survey, although parasitoids were collected in the egg mortality studies (reported above) which were run at the same time. During the month of October, one egg mass was parasitized by *Anastatus* spp., from which only males were collected.
TABLE 2

<table>
<thead>
<tr>
<th>Location</th>
<th>N(^a)</th>
<th>% Egg masses found by predators</th>
<th>When found by predators, % egg mass eaten(^b)</th>
<th>% Eggs placed in orchard eaten by predators(^b)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hilo weed</td>
<td>35</td>
<td>65.7</td>
<td>76.7 (64.7–100)</td>
<td>46.8 (0–100)</td>
</tr>
<tr>
<td>Hilo tree</td>
<td>55</td>
<td>80.0</td>
<td>72.5 (39.6–100)</td>
<td>59.3 (13.4–100)</td>
</tr>
<tr>
<td>Kapulena</td>
<td>59</td>
<td>83.5</td>
<td>81.4 (70.7–100)</td>
<td>66.7 (20.8–100)</td>
</tr>
<tr>
<td>Honomalino 1</td>
<td>46</td>
<td>73.9</td>
<td>76.0 (57.9–100)</td>
<td>50.7 (0–100)</td>
</tr>
<tr>
<td>Honomalino 2</td>
<td>54</td>
<td>92.6</td>
<td>81.4 (70.2–100)</td>
<td>76.4 (48.4–100)</td>
</tr>
<tr>
<td>Pahala 1</td>
<td>60</td>
<td>36.7</td>
<td>52.7 (27.9–80.4)</td>
<td>20.9 (0–38.9)</td>
</tr>
<tr>
<td>Pahala 2</td>
<td>56</td>
<td>51.8</td>
<td>65.7 (41.8–100)</td>
<td>32.4 (0–68.8)</td>
</tr>
<tr>
<td>Keahou</td>
<td>55</td>
<td>65.4</td>
<td>74.0 (55.5–100)</td>
<td>48.4 (0–100)</td>
</tr>
</tbody>
</table>

\(^a\) Number of egg masses recovered from orchard over the entire season. Sixty egg masses placed at each location.

\(^b\) Overall mean with interquartile range in parentheses.

has been reported as being an important source of N. viridula egg mortality in other studies (Nishida, 1966; Seymour and Sands, 1993), and M. floricola has also been reported as predators of insect eggs (Way and Khoo, 1992). High populations of the long-legged ant Anoplolepis longipes (Jerdon) were also present in some of the orchards, but they were never observed attacking N. viridula egg masses. However, A. longipes is considered an efficient predator in coconut and cocoa (Way and Khoo, 1992) and may have been a contributing factor in the high egg predation in some orchards.

Nishida (1966) found that the big-headed ant caused mortality of N. viridula eggs on the island of Oahu (Hawaii) similar to what is reported in this study. He also found comparatively low levels of egg parasitism by T. basalis during April to January 1964, from 14 to 55% parasitism during the period of February to June 1965, and no parasitism during July and August 1965. Nishida (1966) concluded “These two parasites (T. basalis and T. pennipes) no doubt hastened the decrease and stabilization of N. viridula. However, there are cases of other insects where increase, decrease and stabilization occurred without the purposeful introduction of natural enemies. Thus, it seems other factors are involved. . . . The results of studies at Ewa, Oahu, indicate that the increase in ant predation could be involved in the decrease and stabilization of the population of N. viridula.”

My studies were performed during the summer and fall and this may provide a clue as to why T. basalis was rarer than expected. Velasco and Walter (1993) have shown that N. viridula does not continually produce egg masses in Queensland, but does so only when host plants are suitable (spring and fall). Clarke and Walter (1993) took this information to the next trophic level and showed that T. basalis can survive egg-free periods in the winter, but not during the summer. Their studies showed that under a winter temperature regime (in Brisbane, SE Queensland) no significant mortality of T. basalis occurs between 50 and 124 days after emergence if parasitoids are provided with honey and water. Further, if egg masses were placed after \( \approx 150 \) days, surviving females were able to successfully parasitize them. However, studies on T. basalis survival during the summer egg-free period suggest that summer temperatures may be a limiting factor in T. basalis population dynamics. They found that 90% of the T. basalis population exposed to summer tempera-

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**FIG. 3.** Percentage eggs eaten by predators in a survey of the island of Hawaii, 1991.

**FIG. 4.** Percentage eggs eaten in the two Pahala macadamia orchards during a survey of the island of Hawaii, 1991.
tures (average high temperature >25°C) died by Day 55, with 80% of the mortality occurring in the first 30 days (Clarke and Walter, 1993). If the same seasonality in stink bug reproduction and \textit{T. basalis} parasitism occurs in Hawaii, then \textit{T. basalis} may be of importance as a \textit{N. viridula} mortality factor only early in the winter.

Previous studies on the role of \textit{T. basalis} in controlling \textit{N. viridula} populations in Hawaii suggest a higher success than observed in this study. Davis (1967) reported 10 field-collected \textit{N. viridula} egg clusters found on the island of Oahu during the last 6 months of 1963 had an average of 94.9% of the eggs parasitized. He states that from 1964 to 1966, “only an occasional egg cluster which was 90 to 97% parasitized was found.” However, on the islands of Hawaii and Maui, where sentinel egg masses were employed, parasitism was much lower. A total of 13,230 eggs in clusters were placed in 27 locations on Hawaii and parasitism averaged only 34.4%. On Maui, 16,015 eggs were placed in 11 widely separated localities and parasitism averaged 58% (Davis, 1967). Although there is no mention of it in the literature, it is possible that \textit{T. basalis} is more effective when stink bug populations are higher, and a low level of parasitism is a reflection of lower stink bug populations presently found in Hawaii.

The role of \textit{T. basalis} in regulation of \textit{N. viridula} population levels has recently been questioned in Australia (Clarke, 1990) for the same reason it is being questioned in Hawaii. That is, on a relatively unsprayed crop, damage from the southern green stink bug is unacceptable. Many of our results are similar to information presented by Clarke and Walter (1993). However, there are differences which need to be closely explored. For example, when egg masses were found in our studies, comparatively few of the eggs within the mass were parasitized. Whereas, the Australian studies typically report ∼100% parasitism (Seymour and Sands, 1993). In California, Hoffmann et al. (1991) found the percentage of parasitized \textit{N. viridula} eggs to average between 21 and 85% at most locations and two collections where parasitism was >90%. Foraging theory (Stevens and Krebs, 1986) suggests that it makes little sense for \textit{T. basalis} to parasitize every egg in a mass, particularly when \textit{T. basalis} egg production is only approximately 140 eggs (∼2–3 stink bug egg masses) (Powell and Shepard, 1982) and predation on eggs is so high. However, studies need to be performed before differences in the proportion of the egg mass used by \textit{T. basalis} in different areas can be ascribed to differing predation pressure.

To clearly understand the role of \textit{Trissolcus} in biological control of \textit{N. viridula} in Hawaii, we need a better understanding of \textit{Nezara}'s seasonality and the ability of \textit{T. basalis} to survive during climatic extremes. As pointed out by Clarke and Walter (1993), the importance of understanding the \textit{Nezara}–\textit{Trissolcus} interaction is of local as well as international interest. In Hawaii, understanding this interaction is important because the assumption that \textit{T. basalis} is an important biological control agent causes growers (and researchers) to automatically assume the answer to their problem is to release more \textit{T. basalis} in their orchards. Following the release of \textit{T. basalis}, changes in stink bug population levels are not quantified, and the placebo effect makes growers (and researchers) feel as if something beneficial has happened. The assumption of high \textit{T. basalis} effectiveness therefore leads to misallocation of research and implementation resources. For private companies in Hawaii, this misallocation is not merely an academic concern; two of the largest macadamia growers established insectaries for the production of \textit{T. basalis}, but discontinued releases when no detectable declines in stink bug damage occurred after a period of several years. Internationally, both Hawaii and Australia are frequently contacted for source colonies of \textit{T. basalis}. As stated earlier, \textit{T. basalis} may be more effective during the winter, but even so, at least in Hawaii and Australia, there is strong evidence that a search for new natural enemies of \textit{N. viridula} other than \textit{T. basalis} should be performed (Clarke, 1992b).

Finally, the story of \textit{Nezara} clearly points out that biological control programs need to have a strong evaluation component from the start. Such an evaluation component is critical for biological control to continue as a useful pest management strategy. The specter of “Pandora’s Box” has been brought up with regards to biological control (Howarth, 1991) and needs to be strongly considered in planning natural enemy introductions into ecologically sensitive areas. However, it is implicit that good evaluations must be performed both by the proponents of biological control and the proponents of conservation so that rational decisions can be made about the desirability, effectiveness, and risk involved with the introduction of a particular biological control agent. Weakness in evaluation prevents us from learning from our mistakes and our successes. Ultimately, failure to perform proper evaluations of a biological control introduction will likely result in biological control remaining more of an art rather than becoming a science.

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