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Authors: Reddy, Gadi V. P., and Kikuchi, Rosalie

Source: Florida Entomologist, 94(4): 853-858

Published By: Florida Entomological Society

URL: https://doi.org/10.1653/024.094.0419

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LABORATORY HOST RANGE ASSESSMENT OF A PREDATORY PENTATOMID, *PODISUS MACULIVENTRIS* (HEMIPTERA: PENTATOMIDAE) FOR FIELD RELEASE ON GUAM

GADI V. P. REDDY¹ AND ROSALIE KIKUCHI Western Pacific Tropical Research Center, University of Guam, Mangilao, Guam 96923, USA

¹Corresponding author; e-mail: reddy@uguam.uog.edu

ABSTRACT

Predation by *Podisus maculiventris* (Say) (Hemiptera: Pentatomidae) was evaluated with *Erionota thrax* (L.) (Lepidoptera: Hesperidae), *Pericyma cruegeri* (Butler) (Lepidoptera: Noctuidae), *Pareuchaetes pseudoinsulata* Rego Barros (Lepidoptera: Arctiidae), *Papilio polytes* (L.) (Lepidoptera: Papilionidae) and *Eudocima phalonia* (L.) **comb. nov.** (Lepidoptera: Noctuidae). Both free-choice and no-choice experiments indicated that the *P. maculiventris* attacked and consumed all the larvae of the 5 species included in the tests. Although the larvae died at different intervals, most of them were dead within 24-120 h of the introduction of the predatory species. Since the *P. maculiventris* is polyphagous in nature and the present findings indicate that these predators will feed on the introduced biocontrol moth, *P. pseudoinsulata*, it is recommended not to take the predators out of the quarantine laboratory for the field release on Guam. Additionally, *P. maculiventris* will feed on some native species as they become available.

Key Words: biological control, Podisus maculiventris, risks, field release, Guam

RESUMEN

Se evaluó la depredación por *Podiscus maculiventris* (Say) (Hemiptera: Pentatomidae) utilizando *Erionota thrax* (L.) (Lepidoptera: Hesperidae), *Pericyma cruegeri* (Butler) (Lepidoptera: Noctuidae), *Pareuchaetes pseudoinsulata* Rego Barros (Lepidoptera: Arctiidae), *Papilio polytes* (L.) (Lepidoptera: Papilionidae) y *Eudocima phalonia* (L.) **comb. nov.** (Lepidoptera: Noctuidae). Los resultados de ambos experimentos de opción-libre y de no opción indicó que *P. maculiventris* ataca y consume todas las larvas de las cinco especies incluidas en las pruebas. Aunque las larvas murieron en diferentes intervalos, la mayoria de ellos murió entre las 24 y 120 hors después de la introdución de la especie depredadora. Debido a que *P. maculiventris* es polífago en la naturaleza y los resultados actuales indican que estos depredadores ea alimentan de *P. pseudoinsulata*, una polilla introducida como un agente de control biológico, se recomienda que no se dejan salir estos depredadores del laboratorio de cuarantena para liberarlos en el campo. Por otra parte, *P. maculiventris* se pueden alimentar de algunas especies nativas en cuanto estén disponsibles

In the Pacific Islands, agricultural crops such as banana, papaya, head cabbage, Chinese cabbage, cucumbers, eggplant, bell peppers, green onions, tomatoes, taro, yam and cassava are common crops grown in the region (Reddy 2011). Most of these have been uninterruptedly grown throughout the year on Guam and other regions in the Pacific. These crops are vigorously attacked by 2 polyphagous insect pests viz., cutworm Spodoptera litura (F.) (Lepidoptera: Noctuidae) and corn earworm Helicoverpa armigera (Hübner) (Lepidoptera: Noctuidae) (Nafus & Schreiner 1989). However, there are several specialist insect species that are also being attacked on different crops, but they remain a minor problem. The polyphagous insect species, particularly S. litura, are economically important caterpillars on almost all crops that are grown in the Pacific Islands. Since the Micronesian climate is tropical,

with little temperature variation between seasons and weather is typically hot and humid; the pest situation is very severe. If suitable control is not undertaken, especially during the dry season, the yield loss caused by this insect pest may be up to 100% (Reddy 2011). Guam and other Micronesian islands are therefore in the midst of a decline in agriculture production. However, the local government is promoting agriculture to cut down the imports and also avoid any possible introduction of pests associated with the food commodities.

Although chemical control is effective to some extent, it has been increasingly difficult due to the pesticide residue problem and restricted use in food crops (Reddy et al. 2011). While some farmers and growers continue to use chemicals at the recommended dosages for spraying, many insect pests have developed a level of resistance to pesticide sprays. Therefore, alternative control

methods such as biological control could be among the best options. Many parasitoids have been released on Guam for the control of insect pests on different crops (Nafus & Schreiner 1989). The recent few such examples are Euplectrus maternus Bhatnagar (Hymenoptera: Eulophidae) (Muniappan et al. 2004), Lixophaga sphenophori (Villeneuve) (Diptera: Tachinidae) (Reddy et al. 2011), Coccobius fulvus (Compere & Annecke) (Hymenoptera: Aphelinidae) (Moore et al. 2005). However, these parasitoids, even several years post release, have yet to be reported as established on Guam. However, some effective classical biological control programs have been achieved in case of Paracoccus marginatus Williams and Granara de Willink (Hemiptera: Pseudococcidae) (Meyerdirk et al. 2004), Coccinia grandis (L.) Voigt (Cucurbitaceae) (Reddy et al. 2009) and Chromolaena odorata (L.) King & Robinson (Asteraceae) (Cruz et al. 2006).

Of the various biocontrol agents evaluated by different workers, *Podisus maculiventris* (Say) (Hemiptera: Pentatomidae) has shown to be the highest potential biological control agent. The spined soldier bug is one of the generalist predators that help keep the pest populations low in crops such as cruciferous, alfalfa, soybeans, and fruit crops (De Clercq 2000; Wiedenmann & O'Neil 1992). These are medium-sized predatory stink bugs which prey on a wide variety of arthropods, especially larval forms of Lepidoptera and Coleoptera (De Clercq 2004). Reported prey include the larvae of *Pieris rapae* (Linnaeus) (Lepidoptera: Pieridae), Plutella xylostella (L.) (Lepidoptera: Plutellidae), Epilachna varivestis Mulsant (Coleoptera: Coccinellidae), Ostrinia nubilalis (Hübner) (Lepidoptera: Pyralidae), Helicoverpa spp. (Lepidoptera: Noctuidae), Spodoptera spp. (Lepidoptera: Noctuidae), Trichoplusia ni (Hübner) (Lepidoptera: Noctuidae), Leptinotarsa decemlineata (Say) (Coleoptera: Chrysomelidae), Anticarsia gemmatalis (Hübner) (Lepidoptera: Noctuidae), and Phyllotreta flea beetles (Coleoptera: Chrysomelidae) (Hoffmann & Frodsham 1993; McPherson 1980, 1982). There are many reports suggesting *P. maculiventris* could be useful in augmentation of biological control programs for many agricultural crops (Hough-Goldstein & Whalen 1993; Desurmont & Weston 2008). This should make for a positive outcome, as there will be an increase in the preying capacity of the biocontrol agent P. maculiventris on various insect pests. Because P. maculiventris is a polyphagous predator, this biocontrol agent was selected to see if its release, on several crops, on Guam can control the lepidopteran pests.

Several different elements of risks with *P. maculiventris* introduction into virgin territory must be considered. For example, intraguild predation between *P. maculiventris* and the coccinellid *Harmonia axyridis* (Pallas) in the absence or presence

of the extraguild prey *Spodoptera littoralis* (Boisduval) and *Myzus persicae* (Sulzer) was described by Declerq et al. (2003). However, the authors noticed that larger larvae and adults of *H. axyridis* will escape most attacks by *P. maculiventris*. The laboratory tests by Mallampalli et al (2002) indicated that even though ladybeetle adults (*Coleomegilla maculata* Lengi) were generally smaller than *P. maculiventris* adults, they were never preyed upon. However, the effectiveness of generalist predators in biological control may be reduced if increased availability of alternative prey causes individual predators to decrease their consumption of the target species (Alhmedi et al. 2010).

The aim of the study is to investigate whether *P. maculiventris* will affect the beneficial insects that have been released for the control of invasive plants and insect pests.

MATERIALS AND METHODS

Predatory Insects

Since P. maculiventris had not been introduced to Guam previously, the experiments were conducted at the Western Pacific Biocontrol Quarantine Laboratory (WPBQL) at the University of Guam during September to December 2010. All experiments were done at 24 ± 1 °C, relative humidity of 75 to 85% under a photoperiod of 16:8 h L:D. A shipment with eggs of *P. maculiventris* was obtained from Biocontrol Network, Brentwood, Tennessee and successful predator culture was established. P. maculiventris were fed with the cotton leafworm, Spodoptera litura (Lepidoptera: Noctuidae) but were also provided with larvae of the corn earworm, Helicoverpa armigera (Hübner) (Lepidoptera: Noctuidae). The rearing at the WPBQL confirmed the absence of parasitoids.

Prey Species

Prey species used for the experiments were early and later instars of the banana skipper, *Erionota thrax* (L.) (Lepidoptera: Hesperiidae), flame tree looper, *Pericyma cruegeri* (Butler) (Lepidoptera: Noctuidae), Arctiid moth, *Pareuchaetes pseudoinsulata* Rego Barros (Lepidoptera: Arctiidae), black citrus swallowtail, *Papilio polytes* L. (Lepidoptera: Papilionidae), and fruit-piercing moth, *Eudocima (fullonia) phalonia* (L.) **comb. nov.** (Lepidoptera: Noctuidae). These species were cultured in the laboratory on their respective host plants.

Host Preference Experiments

These studies were carried out with 5 prey species using free-choice and no-choice tests. Predator *P. maculiventris* nymphs used for the experi-

ments were second instars (5-7 d-old) and were starved for 24 h before testing. To investigate the prey preference of *P. maculiventris*, early instars (2-3 instars) and later instars (4-5 instars) were given a free-choice between caterpillars of E. thrax, P. cruegeri, P. pseudoinsulata, P. polytes, and E. phalonia. Fifty early or later instars of each prey species were introduced into the individual cages with few leaves of their host plants to provide food for the prey and moisture for the predators. Twenty P. maculiventris were placed singly in a cage (measuring $120 \times 90 \times 60$ cm). Thus, each cage contained 20 predator nymphs with 5×50 caterpillars (total 250). Slices of green beans were added to provide food for the prey and moisture for the predators. The number of larvae preyed or partial prey consumption (De Clercq & Degheele 1994) by P. maculiventris was counted at 24, 48, 72, 96, and 120 h after the start of the experiment. A separate cage each with 50 larvae of each prey species without a predator (control treatments) was maintained to evaluate natural mortality of the prey in the experiments. In addition, survival of the prey was recorded. This experiment was replicated 20 times. The data were analyzed by the SAS GLIMMIX procedure in SAS version 9.2 (SAS Institute 2009). The proportions of larvae preyed upon were analyzed using a generalized linear mixed model and the model was fit with a binomial distribution and logit link function. Treatment means were compared with the least squares means option (LSMEANS, SAS Institute 2009).

In no-choice tests, the prey preference was monitored using single species larvae of *E. thrax*, *P. cruegeri*, *P. pseudoinsulata*, *P. polytes*, and *E. phalonia*. Fifty early or later instars were introduced into the cage with their respective host plant. Ten *P. maculiventris* were placed in a cage. Thus, each cage contained 10 predator nymphs with 50 caterpillars. A separate cage each with 50 larvae of each prey species without a predator (control treatments) was maintained to evaluate natural mortality of the prey in the experiments. This experiment was replicated 20 times. The

number of larvae preyed by *P. maculiventris* was counted at 24, 48, 72, 96, and 120 h after the start of the experiment. Because of the binomial nature of the response data (i.e. each larva was either preyed on or not), the proportions of larvae preyed on relative to the total per replicate were analyzed using a generalized linear model (SAS Institute 2009). Treatment means were compared with the least squares means option (LSMEANS, SAS Institute 2009).

RESULTS

No prey mortality was observed in the control treatments of free-choice and no-choice experiments. Both early and later instar larvae of prey were readily attacked by nymphal P. maculiventris (Tables 1-4). In free-choice tests, P. maculiventris freely preyed among all the early instars and no significant difference was observed (Table 1; P > 0.05). In regard to the later instars under free-choice tests, *P. maculiventris* preyed significantly higher on E. thrax, P. cruegeri, and P. polytes than on P. pseudoinsulata at 24, 48, and 72 h (Table 2; P < 0.05). P. pseudoinsulata was least preferred by P. pseudoinsulata and took 120 h to consume all the larvae. Overall, *P. maculiventris* preyed significantly higher in early instars of all the species within 24 h compared to later instars (P < 0.05). Furthermore, *P. maculiventris* significantly (P < 0.05) took a longer time to prey on P. pseudoinsulata than other prey species particularly in cases of later instars.

In no-choice tests, *P. maculiventris* preyed 44-54% of early instars of all the species within 24 h (Table 3). No significant difference was observed among the species preyed by *P. maculiventris*. However, the significant preying difference (P < 0.05) was observed at 48 and 72 h and consumed 100% of *E. thrax*, *P. cruegeri*, *P. polytes*, and *E. phalonia*. *P. maculiventris* took 96 h to consume 100% of *P. pseudoinsulata*. In regard to the later instars, in no-choice tests, *P. maculiventris* preyed equally on all the species within 24 h and 48 h but preyed significantly higher (Table 4; P < 0.05) on

Table 1. Mean percentage (\pm SEM) early instars of different prey species preyed on by nymphs of *Podisus maculiventris* in free-choice tests at different intervals.

Prey species	Cumulative mean percentage preyed (hours after)			
	24	48	72	
Erionota thrax	11.5 ± 1.2 a	18.5 ± 2.4 a	19.2 ± 1.2 a	
Pericyma cruegeri	$13.5 \pm 2.4 \text{ a}$	20.5 ± 0.5 a	21.2 ± 2.1 a	
Papilio polytes	$10.8 \pm 0.8 a$	$19.5 \pm 1.4 a$	22.2 ± 0.4 a	
Eudocima phalonia	$12.2 \pm 1.6a$	$17.6 \pm 0.8 a$	20.4 ± 1.3 a	
Pareuchaetes pseudoinsulata	$9.8 \pm 2.1 \text{ a}$	12.1 ± 0.6 a	$17.0 \pm 2.6 a$	

Means within the column followed by same letters are not significantly different (P < 0.05; least squares means option (LS-MEANS); mean of 10 replications, each replicate consisted of 20 larvae of each species.

Table 2. Mean percentage (\pm SEM) later instars of different prey species preyed on by nymphs of *Podisus maculiventris* in free-choice tests at different intervals.

Prey species	Cumulative mean percentage preyed (hours after)				
	24	48	72	96	120
Erionota thrax	6.5 ± 1.2 a	15.5 ± 0.2 a	20 ± 0.0 a	_	_
Pericyma cruegeri	$7.2 \pm 2.1 \text{ a}$	16.4 ± 0.7 a	$20 \pm 0.0 \text{ a}$	_	_
Papilio polytes	5.2 ± 0.6 a	15.7 ± 1.2 a	$20 \pm 0.4 \text{ a}$	_	_
Eudocima phalonia	$7.0 \pm 0.8 \text{ a}$	$14.0 \pm 2.1 a$	$20 \pm 0.0 \text{ a}$	_	
Pareuchaetes pseudoinsulata	$0.5 \pm 1.6 \mathrm{\ b}$	$0.5 \pm 1.6 \mathrm{\ b}$	$8.5\pm1.4~\mathrm{b}$	14.5 ± 1.2	20.0 ± 0.4

Means within the column followed by same letters are not significantly different (P < 0.05; least squares means option (LS-MEANS); mean of 10 replications, each replicate consisted of 20 larvae of each species.

Table 3. Percentage (± SEM) early instars of different prey species preyed on by nymphs of *Podisus maculiventris* in no-choice tests at different intervals.

Prey species	Cumulative mean percentage preyed (hours after)				
	24	48	72	96	
Erionota thrax	48.2 ± 2.2 a	$98.0 \pm 0.4 \text{ a}$	100.0 ± 0.2 a	_	
Pericyma cruegeri	$54.3 \pm 1.8 a$	$96.6 \pm 1.2 \text{ a}$	100.0 ± 0.0 a	_	
Papilio polytes	$52.6 \pm 0.8 a$	$95.6 \pm 2.1 a$	100.0 ± 0.4 a	_	
Eudocima phalonia	$50.2 \pm 3.1 a$	$100.0 \pm 1.8 a$	_	_	
Pareuchaetes pseudoinsulata	$44.5 \pm 1.4 \text{ a}$	$58.4 \pm 1.6 \text{ b}$	$72.0 \pm 2.3 \text{ b}$	100.0 ± 0.0	

Means within the column followed by same letters are not significantly different (P < 0.05; least squares means option (LS-MEANS); mean of 10 replications, each replicate consisted of 20 larvae of each species.

Table 4. Percentage (± SEM) later instars of different prey species preyed on by nymphs of *Podisus ma-Culiventris* in no-choice tests at different intervals.

Prey species	Cumulative mean percentage preyed (hours after)				
	24	48	72	96	120
Erionota thrax	22.3 ± 2. 2a	37.3 ± 0.4 a	73.3 ± 0.2 a	100.0 ± 0.0 a	_
Pericyma cruegeri	$24.3 \pm 1.2 \text{ a}$	$41.6 \pm 1.2 a$	$78.3 \pm 0.8 \text{ a}$	$100.0 \pm 0.0 a$	_
Papilio polytes	$20.6 \pm 1.3 \text{ a}$	$40.5 \pm 1.1 a$	$69.3 \pm 1.3 \text{ a}$	$100.0 \pm 0.2 a$	_
Eudocima phalonia	21.3 ± 0.4 a	$42.6 \pm 2.2 \text{ a}$	$66.5 \pm 0.4 a$	$83.2 \pm 0.2 \text{ b}$	100.0 ± 0.2 a
Pareuchaetes pseudoinsulata	$18.2 \pm 2.1 \text{ a}$	$38.4 \pm 2.2 \; a$	$50.5 \pm 0.6~\mathrm{b}$	$63.5 \pm 0.5~\mathrm{c}$	$98.5 \pm 1.6 \text{ a}$

Means within the column followed by same letters are not significantly different (P < 0.05; least squares means option (LS-MEANS); mean of 10 replications, each replicate consisted of 20 larvae of each species.

E. thrax, P. cruegeri, P. polytes, and E. phalonia than on P. pseudoinsulata at 72 h. Similarly, P. maculiventris had consumed 100% of the larvae of E. thrax, P. cruegeri, and P. polytes at 96 h, while P. pseudoinsulata and E. phalonia were less preferred.

DISCUSSION

The prey species selected were based on the availability of the insect species which includes pest, common species, and beneficial insects. For examples, according to Muniappan (1974), *P.*

[—]No larvae survived.

⁻No larvae survived

⁻No larvae survived.

cruegeri is extremely abundant on Guam and defoliates most of the poincianas every year. Outbreaks of the *E. thrax* can occur at the beginning of the wet season and are severe enough to reduce yield of banana (Nafus & Schreiner 1989). P. polytes is an abundant butterfly on Guam and the caterpillars are quite harmless (Schreiner & Nafus 1997). E. phalonia is considered one of the top 10 pests in the Pacific region (Waterhouse & Norris 1987) and causes damage to fruit crops and vegetables (Reddy et al. 2007). In Africa, Asia, and Australia the larvae of fruit-piercing moths feed only on the vines of the family Menispermaceae, whereas in the Pacific and Papua New Guinea they feed on plants of the family Menispermaceae and the genus Erythrina variegata Linn. (Fabaceae) (Reddy et al. 2005). On the other hand, P. pseudoinsulata is a beneficial insect and a biological control agent for Chromolaena odorata (Asteraceae). This biological control agent is established on Guam and other neighboring islands. By 1987, due to the rapid spread, the moth had reached almost all areas on Guam. At this time, the release has resulted in successful suppression of the weed, but the moth is still in an invasion phase (Zachariades et al. 2009).

The results from the present study indicated that it took a longer time for *P. maculiventris* to prey on instars of *P. pseudoinsulata* than other prey species. This could be due to the presence of heavy hairs on the body of the later instars, which could lead to attacking and preying difficulties for *P. maculiventris*. Consequently, *P. maculiventris* has to spend more time in capturing the more agile late instars of *P. pseudoinsulata* than they did to kill larvae of early instars. However, the hairs on the early instars have not highly developed compared to later instars (Zachariades et al. 2009). Although *P. maculiventris* took a longer time, it was able to feed on almost all the larvae of *P. pseudoinsulata*.

Moreover, in our study, P. maculiventris preyed significantly higher in early instars of all the species within 24 h compared to later instars. These results corroborate with Herrick et al. (2008) and DeClerg et al. (2003) who reported that prey size can be vital in the selection of prey by P. maculiventris. The results by Herrick et al. (2008) indicated that predation by P. maculiventris on Plutella xylostella (Lepidoptera: Plutellidae) is far greater on young larvae than on older larvae because young, smaller P. xylostella larvae are less able to defend themselves and lack the ability to escape from *P. maculiventris*. Similar such results were reported by DeClerq et al. (2003) in cases of Harmonia axyridis Pallas (Coleoptera: Coccinellidae).

Although *P. maculiventris* is an abundant generalist predator that preferentially feeds on most of the lepidopteran larvae and can be found in a variety of agroecosystems and is common in many

agricultural crops in North America (McPherson 1980; Culliney 1986), it has not been recorded yet in Micronesia. Since many agricultural crops are being grown and are often attacked by many lepidopteran larvae in Micronesia, it would be worth the importing of *P. maculiventris* from North America to Guam as a biological control agent. Since the *P. maculiventris* is polyphagous in nature and the present findings indicate that these predators are feeding on the introduced biocontrol moth *P. pseudoinsulata*, it is recommended not to take the predators out of the quarantine laboratory for field release. Additionally, *P. maculiventris* will feed on some native species as they become available.

ACKNOWLEDGMENTS

This study was made possible, in part, by a Cooperative Agreement from the United States Department of Agriculture's Animal and Plant Health Inspection Service (APHIS). It may not necessarily express APHIS' views. This project was supported by FY 2010 United States Department of Agriculture (USDA), APHIS, Cooperative Agricultural Pest Survey (CAPS) Program Agreement#10-8510-1353-CA and Hatch Project W-2185 Biological Control in Pest Management Systems of Plants (Project# GUA0612). The authors thank Ms. Yolisa Ishibashi, Pest Survey Specialist, USDA, APHIS, PPQ, Honolulu, Hawaii 96850 for the encouragement and support during all phases of the project. In accordance with federal law and USDA policy, this institution is prohibited from discrimination on the basis of race, color, national origin, sex, age, or disability.

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