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Consumption Efficiency of Wolf and Lynx Spiders, *Pardosa pseudoannulata* and *Oxyopes javanus*, on Insect Pests of Asiatic Pennywort

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Absract Asiatic pennywort is a plant common to Central and Southeast Asia and is used for folk medicine. The most common predatory spiders found in the Asiatic pennywort growing areas were wolf and lynx spiders, *Pardosa pseudoannulata* Bösenberg and Strand and *Oxyopes javanus* Thorell. They were observed the onsuming two important insect pests of pennywort: young pennywort cutworm, *Zonoplusia ochreata* Walker, 1865 and a leafhopper, *Empoascanara alami* Ahmed. The consumption efficiency associated with spider sex and starvation condition of these two predatory spiders on the leafhopper and pennywort cutworm was assessed in the laboratory. The results showed that female spiders consumed more leafhoppers and pennywort cutworms than did the males. Both predatory spiders preferred the second larval instar of pennywort cutworm. The female and male of *O. javanus* consumed 18.07 and 17.11 insects/day, respectively, and *P. pseudoannulata* 14.39 and 12.18 insects/day, respectively. Starvation for a prolonged food of time did not increase hunger level of the spiders. The use of these two spiders combined, in comparison with *O. javanus* alone, against leafhoppers was not significant (P>0.05). However, *P. pseudoannulata* and *O. javanus* are important components of the natural enemy complex in pennywort and therefore they should be conserved to aid insect control in pennywort.

Keywords: Starvation, Asiatic pennywort, Insect pests, Predation

Introduction

Asiatic pennywort is an herbal plant long acknowledged for therapeutic value in folk medicine. Pennywort is native to India, Thailand, and Northern Australia as well as some other tropical and subtropical regions (Hashim *et al*., 2011) and grows well in swampy areas (Gohil *et al*., 2010). Pennywort is attacked by a leafhopper *Empoascanara alami* Ahmed, 1970 in early season and pennywort cutworm *Zonoplusia ochreata* Walker, 1865 throughout the

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growing season; their infestations can cause substantial yield loss. Heavy insecticide usage by growers to control these insect pests can disturb the natural ecosystem and eliminate the natural enemies along with the pest species. In addition, continuous use of insecticides commonly results in insect resistance and resurgence against various groups of pesticides (Butt and Sherawat, 2012; Khuhro *et al.*, 2012).

In Thailand, the wolf spider, *Pardosa pseudoannulata* Bösenberg and Strand, 1906 and lynx spider, *Oxyopes javanus* Thorell, 1887 are ubiquitous in pennywort farms (Yuphet and Bumroongsook, 2016). Wolf spiders are an important part of the natural enemy complex and recognized as a significant biological control agent (Matteson, 2000). These ground inhabiting hunting spiders are common in the agricultural ecosystem but little is known of their role in insect pest population control (Russell-Smith and Stork, 1995). About 3,500 species of spiders have been identified (Ghavami *et al*., 2007). They feed primarily on arthropods and are thought to play an important role in pest control. They are obligate predators in the ecosystem where they opportunistically kill and consume live prey; most adults can subsist for a week or more without prey and exhibit a functional response to congregate at nearby sites when prey becomes available. These characteristics suggest they may have a very important role to reduce and prevent the spread of insect pests in agriculture (Sunderland *et al*., 1986). Song *et al*. (1999) reported a correlation between the number of spiders and the number of plant hopper in rice fields in southern China for the first time. The spiders help to prevent epidemics of plant hoppers in rice fields. Sivasubramanian *et al*. (2009) reported that the wolf spider is effective against leafhoppers by consuming 3.34 adults, white flies 1.86, aphids 7.34 and plant hoppers 6.34, adults per day. Felix (1996) reported the adult spider would consume more adult insects than larvae (Foelix, 1996). *Schizocosa ocreata* Hentz is effective for cricket control (Stephanie and George, 2005). Female spiders devoured more prey compared to male spiders (Tahir and Butt, 2009). Nyffeler (1999) reported that the lynx spider is a polyphagous predator feeding on homopteran insects, flies and thrips. Thus, we examined two spider species, *P. pseudoannulata* and *O. javanus*, as potential predators in pennywort.

The objective of this research was to assess the consumption efficiency of both wolf and lynx spiders on pennywort cutworm (*Z. ochreata*) and leaf hoppers (*E. alami*).

Materials and methods

Spider rearing procedure

The study was focused on adult females and males of the hunting spiders, *P. pseudoannulata* and *O. javanus*. They were collected from the Asiatic pennywort farms at the university plantation of King Mongkut's Institute of Technology Ladkrabang, Bangkok in Thailand. Spiderlings of both species were collected from June to August 2015 and placed individually in a plastic cup 30 ml, and reared with the first larval instar of mealworms *Tenebrio molitor*. A small piece of cotton in a small amount of water was provided for spiders to access moisture (Whitecomb and Eason, 1965). After those spiderlings developed to adults, they were sexed and separated individually for consumption efficiency testing.

Rearing of pennywort cutworm

Two pairs of *Z. ochreata* adult moths were transferred to a Plexiglas cage 30×40×30 cm and provided a piece of cotton soaked with 10% honey solution as a food source. Inside each container, a pennywort plant was placed in the cage for egg-laying. Eggs were collected from the leaves and placed in a petri dish padded with a moist paper towel. Larvae hatched after 3–4 days. The newly emerged larvae were reared on pennywort leaves. To maintain the culture of *Z. ochreata* larvae, they were fed with fresh pennywort leaves and the adult had a honey solution as a food source. Different larval stages were used for the comparative consumption efficiency studies.

Comparative consumption efficiency of wolf and lynx spiders

In this study, we presented the five larval stages of *Z. ochreata* to *P. pseudoannulata* and *O. javanus* (total n = 60 of each species (30 male and 30 female) and measured consumption. Each spider was offered 20 pennywort cutworms of each instar on Asiatic pennywort plants, *Centella asiatica* L. cultivated in a Plexiglas cylinder (a diameter of 15 cm, height 40 cm) and allowed 24 h feeding time in the laboratory (27±2 ℃, 65±10% relative humidity and a photoperiod of 16L: 8D h). The number of larvae fed or killed by each predatory spider was recorded after 24 h.

Starvation effect on consumption efficiency of wolf and lynx spiders

The experiment was set up to test the starvation effect on consumption efficiency of *P. pseudoannulata* (total n = 60 of each species (30 male and 30 female) against the adult leafhopper, *E. alami*. There were two groups of treatments: starvation and no starvation of spiders by sex and species. For the starvation group, these spiders fully sated prior to being starved for five days, whereas the fed group was given worms every day. In the cage, 20 *E. alami* adults were transferred and placed on pennywort plants, with an individual of the spider. At 24 h after exposure, the number of leafhoppers killed by each predatory spider was recorded. Average temperature and relative humidity (%R.H.) during the whole experiment remained 27 ± 2^{o}C and $65 \pm 10\%$ relative humidity and a photoperiod of 16L: 8D h.

Feeding behavior of P. pseudoannulata and O. javanus

In these experiments, 20 adults of *E. alami* collected from the Asiatic pennywort farms were transferred to a pennywort plant in a plastic cylindrical cage (15 cm diameter, 40 cm high). The feeding behaviors of the predators placed in the cage were observed and recorded. The number of prey eaten or killed by each predatory spider was recorded at 24 h.

Data analyses

The collected data were analyzed by one-way ANOVA, and mean difference was compared with Tukey's HSD tests at the significance levels of $P = 0.01$.

Results

Comparative consumption efficiency of spider predators

Prey sizes might affect consumption capability of these spiders. They consumed the second instar larvae of *Z. ochreata* significantly more than the first, third and fourth instar larvae, respectively ($F = 2.68$, df = 3, 120, $P < 0.01$). They did not attack and consume the 5^{th} larval instar (Table 1). Starvation effect on consumption efficiency of both spiders was investigated at 24 h after treatment. No difference on amount of plant hopper consumed was found between the spiders with starvation and no starvation condition (Table 2).

Table 1. Consumption efficiency of predatory spiders on pennywort cutworms (mean ± SD).

Larval instar	*P. pseudoannulata*		*O. javanus*	
	Female	**Male**	**Female**	**Male**
First	3.20 ±1.5[b]	1.80 ±1.1[b]	3.20 ±1.2[b]	2.80 ±0.8[b]
Second	14.39 ±0.7[a]	12.18 ±1.6[a]	18.07 ±1.4[a]	17.11 ±1.2[a]
Third	1.90 ±1.9[bc]	0.80 ±1.2[c]	2.10 ±0.9[c]	1.50 ±1.2[c]
Fourth	0.50 ±1.2[c]	0.40 ±0.5[c]	0.51 ±0.7[d]	0.58 ±0.8[d]

Means followed by a different letter in a column are significantly different from each other (*P* < 0.01; Tukey's LSD test).

Table 2. Comparative consumption of leafhoppers (mean ±SD) by predatory spiders.

Treatment	*P. pseudoannulata*		*O. javanus*	
	Female[ns]	**Male[ns]**	**Female[ns]**	**Male[ns]**
No-starvation	17.82 ±0.28	10.50 ±0.34	19.38 ±0.41	16.25 ±0.65
Starvation	17.45 ±0.30	10.48 ±0.46	17.68 ±0.57	14.54 ±0.84

Means followed by a different letter in a column are significantly different from each other (P <0.01; Tukey's LSD test).

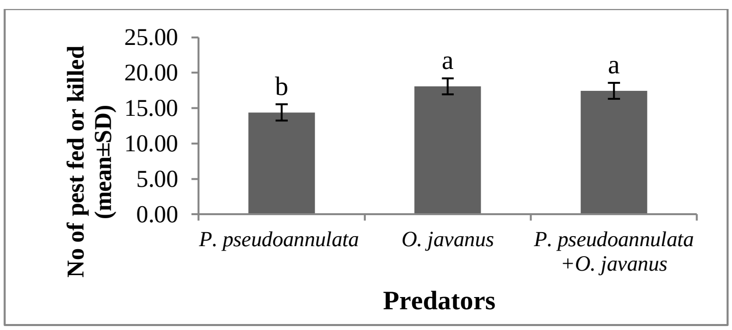

Figure. 1 The efficiency of predator species (solitary *P. pseudoannulata*, *O. javanus*, and together) in consuming the adult leafhopper, *E. alami*, observed over a 24-h period. Means ± SD with different letters are significantly different at P < 0.01.

O. javanus females could consume more leafhoppers than did *P. pseudoannulata* (19.38 ±0.41 and 17.82 ±0.28 insects/day, respectively). The same result was found with the male of *O. javanus* and *P. pseudoannulata*, which consumed leafhopper 16.25 ±0.65 and 10.50 ±0.34 insects/day, respectively (F=2.68, df = 3,120, P < 0.01) (Table 2). We found that both species of spiders readily feed on the leafhopper, *E. alami*, in Asiatic pennywort. Leafhopper predation was least with *P. pseudoannulata* by itself and higher with *O. javanus* by itself or in combination with *P. pseudoannulata* (F=3.15, df = 3, 120, P < 0.01) (Fig. 1).

Feeding behavior of P. pseudoannulata and O. javanus

P. pseudoannulata and *O. javanus* are hunting spiders; they feed on a wide range of insects and are generally considered to be polyphagous predators. Normally, the wolf spider waited for prey to ambush and relied on speed and vigor for target capture. Direct observation showed that wolf spider habitat was on the ground and the lower part areas of the pennywort stem. Wolf spiders are recognized as solitary ground dwellers that hunt alone. When prey was introduced into the cage, the spider approached the prey immediately and grasped it with its first pair of legs. If the prey struggled, the spider used its chelicerae and front legs to secure the prey. Once prey was immobilized, the spider pierced the prey exoskeleton and sucked out the liquid. A consumption period of 10-20 minutes was spent sucking the body fluid from a leafhopper. The lynx spider had bristles on its legs used to grasp prey, and this spider usually dwells in the upper canopy of pennywort plants. They were ambush predators and limited moving from one part of the plant to another. The lynx spider is attracted by prey movement; it attacked the prey with great speed and touches it with the first pair of legs. It hold the trapped prey with its first two pairs of legs, sucked the body fluids and finally discarded the exoskeleton as a dry ball. The feeding behavior of the spider showed that they consumed plant hoppers at night. The lynx spider took 10-30 minutes to suck the liquid from an adult plant hopper and for a cutworm larva.

Discussion

The predatory spiders prefer early stages of prey caterpillars. The 5^{th} larval size was a major determinant of a probability of predators encountering and attacking a prey, significantly shorter times to attack would be expected for larger larvae (Strand and Obrycki, 1996). A second possibility is that the defensive behavior of larvae repelled the attacking spider. Aggressive 5th instar behavior may repel parasitoid attacks. In our experiments, 4th instars tended to be more physically aggressive with the predator than first or second instars and were often observed biting at the approaching spider. The less aggressive behavior of first and second instar larvae could allow a higher proportion of successful attacks than for the more aggressive 5^{th} instars (Wagner and Wise, 1997). Prey within 50-110 percent of the lynx spider body length elicits feeding behavior (Huseynov, 2006). Studies by Pekar (2014) found that size of predator affects prey capture: adult spiders captured large prey while tiny juveniles captured smaller prey.

Starvation for short period of times had no effect on consumption efficiency of these two spider species. Inon (2016) observed spiders demonstrated a high starvation tolerance and low mortality rates even after 9 weeks of starvation. In particular, Walker *et al*. (1999) found that *Lycosa lenta* retained normal behavior over a 30 day starvation period. Also, we have found that hunger does not affect locomotor behavior. This is consistent with the experimental results of John (1974). Adult survival of the wolf spider, *Lycosa lenta* under starvation conditions averaged 208 days. Studies found that starvation did not seem to affect the feeding behavior of spiders (Persons and Uetz, 1996). Provencher and Reichert (1991) suggested that hunger is not an important factor influencing spider feeding behavior.

Female spiders showed a slightly higher percentage predation in comparison to the male spiders in feeding tests. Walker and Rypstra (2001); Mohsin *et al*. (2015) reported that females consumed more than males in similar conditions. This is presumed due to the female requiring more energy for egg laying, brood care and energy reservoir for reproduction and hibernation in the upcoming season consistent with what Muhammad *et al*. (2012) reported after female mating to have to fulfill their energy requirements to maximize their breeding. In addition, *R. rabida* females attack more prey than the males (Walker and Rypstra, 2001).

The studies showed that these spiders can be exploited for control of sucking pests. These studies of *P. pseudoannulata* and *O. javanus* indicated that they were effective against hemipteran sap feeders such as *Nilaparvata lugens* in rice fields (Sigsgaard *et al*., 2001). Sahito *et al*. (2010) found that spiders were biological control agents on all stages of sucking insect pests in *Brassica* and important in reducing pest densities. Spider predators play important roles in regulating the dynamics of prey populations and thereby indirectly benefit the resources eaten by prey species (Landis *et al*., 2000; Lang, 2003). According to Nyffeler *et al*. (1992) and Huseynov (2007), most spiders feed more at night than during daylight hours. Although laboratory conditions are quite different from field conditions, one factor in predation may be because they differ in hunting strategies and habitat preference in the agricultural field (Harwood *et al*., 2001; Maloney *et al*., 2003). Moreover, as the habitat and hunting of one predatory taxon decreases, the net effect could be a loss of the predator (Barton and Schmitz, 2009; Bhuyan and Basit, 1996).

Conclusion

Both *O. javanus* and *P. pseudoannulata* are hunting spiders. *O. javanus* consumed significantly more prey than did *P. pseudoannulata*. Females of wolf and lynx spiders consumed more prey than did their male counterparts. We noted that both spiders consumed *Z. ochreata*, which belong to the family Noctuidae that includes many other agricultural pests. The second larval instar of the pennywort cutworm was the preferred prey stage and subsequent instars were decreasingly preyed upon. This research suggested that *O. javanus* and *P. pseudoannulata* gave a predatory potential to mitigate damage from major insect pests of Asiatic pennywort.

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