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Feeding and oviposition preference of anthocorid predator, *Blaptostethus pallescens* Poppius to different prey species

A. NASREEN[1], S. SRIDHARAN[1], M. MUTHUKUMAR[2]* and K. ASHOK[1]

[1]Department of Agricultural Entomology, Tamil Nadu Agricultural University, Coimbatore-641003, Tamil Nadu, India

[2]College of Agricultural Sciences, SRM Institute of Science and Technology, Kattankulathur, Chengalpattu - 603203, Tamil Nadu, India

***E-mail:** muthukumar.tnau@gmail.com

ABSTRACT: Laboratory studies were conducted on the feeding and oviposition preference of anthocorid predator, *Blaptostethus pallescens* Poppius through behavioural bioassay to different prey species. The orientation and predation of *B. pallescens* was maximum on *Corcyra* eggs followed by spider mite, *Tetranychus urticae* Koch. The least preferred prey was whitefly, *Aleurodicus dispersus*. Among host plants tested, the nymphs and adults of *B. pallescens* preferred to orient and predate more preys on brinjal followed by bhendi. The least preferred host was cluster bean. Even though French bean pod was commonly used in mass culturing of *B. pallescens*, green pods of cluster bean were the most preferred substrate for oviposition. And also, the eggs laid on cluster bean pods showed the maximum hatching. The adults of *B. pallescens* provided with honey and water along with the normal diet of *Corcyra* eggs showed increased fecundity.

Keywords: *Blaptostethus pallescens,* biocontrol, feeding, oviposition, alternate host

INTRODUCTION

The use of indigenous natural enemies in biological control of crop pest is an alternative way to enhance the use of exotic natural enemies (Bonte and de Clercq, 2011). Indigenous natural enemies provide an advantage of their ability to readily exploit native or invasive pests as their prey and also persist on alternate prey when the target pests are rare or absent (Symondson *et al.*, 2002; Castane *et al.*, 2014). Anthocorid predators, commonly known as minute flower bugs or minute pirate bugs are recognized as a potential biocontrol agents of crop pests (Barber, 1936). They feed on small lepidopteran larvae, small grubs, psocids, mites, thrips, aphids and storage pests (Tawfik and El-Husseini, 1971). Natural population of anthocorid predators has been successful in maintaining pest populations at low levels (Muraleedharan and Ananthakrishnan, 1974). The majority of anthocorids are predaceous at nymphal and adult stages and few are phytophagous (Oku and Kobayashi, 1966). Anthocorids possess many of the characteristics of an ideal biocontrol agent, such as high searching efficiency and feeding rate, shorter duration of development, density dependent response to the pest population and synchronization of predator and prey population (Ballal *et al.*, 2009).

In countries like France, United Kingdom, Nether lands, Germany etc., several species of anthocorid predators are available commercially and are released in green houses and fields for the management of insect pests, especially sucking pests such as thrips and mites (Ballal *et al.*, 2003). In India, research has generally focused only on identifying indigenous anthocorids on different pests infesting different crops (Ballal and Yamada, 2016). Therefore, it is necessary to test the feeding and oviposition behaviour of anthocorids, before employing it as a general predator in any ecosystems. With this context, the present study has been taken up to identify the alternate natural ovipositional substrate, feeding choice of one such anthocorid predator *Blaptostethus pallescens* Poppius (Heteroptera: Anthocoridae) and also tested with different adult nutrition to enhance the fecundity.

MATERIALS AND METHODS

Mass culturing of predator *B. pallescens*

Mass culturing of *B. pallescens* was carried out as per the method developed by Ballal *et al.* (2003). UV irradiated *Corcyra cephalonica* eggs were sprinkled on cotton pad placed at the bottom of the transparent plastic container (500ml). Nymphs were released into the container along with bean pods which supply the required water for the nymphs. Fresh eggs of *Corcyra* were provided on alternate days till the adults emerge. Freshly emerged adults were shifted to plastic containers with green bean pods for oviposition. The pods with the eggs were removed daily and fresh pods were replaced as an oviposition substrate.

Mass culturing of red spider mite, *Tetranychus urticae* Koch

One month old host plants, brinjal raised in pots was infested with *T. urticae* by releasing the mite stages using camel hair brush and by placing the infested leaves collected from the field. Further, the uninfested potted plants were placed along the infested potted plants for uniform infestation of mites. The newly uninfested potted plants maintained in the insectary were periodically replaced after the removal of dried plants. Likewise, the mass culturing of *T. urticae* is maintained in the Insectary.

Prey preference of fourth stage nymph of *B. pallescens*

Sucking pests like whitefly (*Aleurodicus dispersus*), mite (*T. urticae*), aphids (*Aphis gossypii*), mealybug (*Phenacoccus solenopsis*) and factitious host *Corcyra* eggs were the prey tested for the preference of fourth instar nymph of *B. pallescens*. Twenty life stages of different prey were kept in moist filter paper placed in iron trough of 28 cm dia. at equidistant and ten fourth stage nymphs of the predator pre starved for two hours were released in the centre. The experiment was carried out with five treatments and four replications. Two hours after the release, the congregation of predator nymphs in different prey arena was recorded. The number of preys fed by the nymphs was also recorded at six and 24 hours after release. The preference of the prey by the fourth instar nymph of *B. pallescens* was studied using six arm olfactometer also. Twenty insects of five different preys were placed in each arm and ten fourth instar nymphs were placed in the centre. The settlement of nymphs in individual prey species two hours after the release of predator was recorded. Besides, the observation on the number of preys fed six and 24 hours after the release of predator also recorded.

Prey preference of adult *B. pallescens*

The preference of different prey species by the adult of *B. pallescens* was studied in two separate experiments using the multiple-choice test in open circular basin and in closed six arm olfactometer as mentioned previously for the fourth instar nymph of the predator.

Influence of prey host plant on feeding of fourth instar nymph of *B. pallescens*

The volatiles of the plant housing the prey shall act as allomones or kairomones for the predators feeding on the prey. The effect of plant hosts housing *T. urticae* on the orientation and feeding of *B.pallescens* was assessed through laboratory experiment with four treatments (brinjal, bhendi, cluster bean and cucurbit) and five replications. Ten nymphs and adults of *T. urticae* were released on the host leaf arena with the camel hair brush. The leaf discs of different host plants *viz*., brinjal, bhendi, cluster bean and cucurbit infested with mites were kept at equidistant in circular zinc tray of 28cm diameter lined with moist filter paper. Later ten fourth instar nymphs of the predator were released in the centre. The preference of predator to mite as influenced by the host was assessed two hours after the release by recording the assemblage of nymphs on different host arena housing the mite. In addition, the number of preys fed by the nymphs was also recorded at six and 24 hrs after the release.

Similar study to assess the influence of host plant on prey feeding was also conducted using six arm olfactometer. The four host plant discs infested with twenty mites kept in the arm of olfactometer were released in the centre with ten fourth instar nymphs of the predator. The number of predator nymphs occupying the different host plants housing mites two hours after release and the number of mites fed by the predator six and 24 hours after release were recorded.

Influence of host plant on prey feeding of adult *B. pallescens*

The effect of plant hosts housing *T. urticae*on the orientation and feeding of *B. pallescens* was assessed through laboratory experiment with four treatments (brinjal, bhendi, cluster bean and cucurbit) and five replications. Ten nymphs and adults of *T. urticae* were released on the host leaf arena with the camel hair brush. After settling of mites, the predator adults (10) were released in circular test arena which contains all the host plant leaves kept at equidistant around the inner periphery of the tray. Observation on the orientation of the predator adults two hours after the release was noted. Further, the number of mites preyed by the adult at six and 24 hours after the release was also recorded. The laboratory experiments were conducted as per the methodology mentioned previously for the fourth instar nymph of the predator.

Preference of plant host for oviposition

Endophytic insect predators generally prefer a host to lay eggs in a substrate which provide the essential nourishment to the newly hatched young ones. In the case of *B. pallescens,* it is mass cultured in the laboratory with the help of green French bean pods as ovipositional substrate which has sufficient moisture retention to support the requirement of hatched out nymphs. In order to find an alternate and cost-effective ovipositional substrate, a laboratory experiment was conducted with six plant substrates which were replicated four times. Each substrate (as fresh green pod) was taken in a plastic container (500ml) which was provided with *Corcyra* eggs and cotton pad at the bottom. Each container with

Table 1. Influence of prey on orientation and predation of fourth instar nymphs of *B. pallescens*

Prey	Orientation of predators* 2HAR Test arena		Number of prey predated*	
	Circular basin	Olfactometer	6HAR	24 HAR
Aphis gossypii	1.75 (1.32) [ab]	1.8 (1.32) [ab]	1.50 (1.22) [b]	1.50 (1.22)[b]
Tetranychus urticae	2.50 (1.58) [a]	2.5 (1.58) [a]	3.25 (1.80) [a]	4.50 (2.12)[a]
Aleurodicus disperses	0.50 (0.71) [c]	0.5 (0.71) [c]	0.25 (0.50) [c]	0.50 (0.71)[b]
Phenacoccus solenopsis	0.75 (0.87)[bc]	0.8 (0.87) [c]	1.00 (1.00)[bc]	1.50 (1.22)[b]
Corcyra eggs	2.00 (1.41) [a]	3.0 (1.73) [a]	4.00 (2.00) [a]	6.50 (1.55)[a]
SE(d)	0.20	0.18	0.23	0.22
CD(0.05)	0.43	0.39	0.48	0.46

*Mean of four replications; HAR- hours after release ; Values in the parentheses are square root transformed values

Table 2. Influence of prey on orientation and predation of adult *B. pallescens*

Prey	Orientation of predators* 2HAR Test arena		Number of preys predated*	
	Circular basin	Olfactometer	6HAR	24 HAR
Aphis gossypii	2.00(1.41)[b]	2.00(1.41)[ab]	1.75(1.32)[b]	1.8(1.32)[b]
Tetranychus urticae	3.50 (1.87)[ab]	3.00 (1.73)[a]	4.00 (2.00)[a]	5.0 (2.24)[a]
Aleurodicus disperses	0.50 (0.71)[c]	0.80 (0.87)[b]	0.25 (0.50)[c]	0.5 (0.71)[c]
Phenacoccus solenopsis	0.80(0.87)[c]	1.30(1.12)[b]	1.25(1.12)[b]	1.8(1.32)[b]
Corcyra eggs	3.80(1.94)[a]	3.80(1.94)[a]	5.00(2.24)[a]	7.0(2.65)[a]
SE(d)	0.20	0.23	0.20	0.22
CD(0.05)	0.42	0.49	0.43	0.47

*Mean of four replications; HAR- hours after release; Values in the parentheses are square root transformed values

substrate was released with mated female and allowed for oviposition upto seven days. Every 24 hours of release, green pod laden with eggs was removed and fresh green pod was supplied to the container. The number of eggs laid on the removed green pod was counted and kept in a separate container for observation on hatching.

Adult nutrition to enhance fecundity

The ongoing protocol of mass culturing of *B. pallescens* under the laboratory conditions uses green French bean pods and *Corcyra* eggs as adult diet. In order to enhance the fecundity of the adult, the mated female can be supplied with sugar rich adult diet. In this regard, the specialized adult diet followed in the culturing of other predators like *Chrysoperla* was tested along with treatment of honey + water. There were three treatments and seven replications. The treatments were *Corcyra* eggs + green French bean pod(T1), honey+ water+ *Corcyra* eggs+ green French bean pod(T2) and specialized diet (yeast + fructose + honey + Proteinex + water @ 1:1:1:1:1) + water + *Corcyra* eggs + green French bean pod (T3). The liquid diet was provided separately through soaked cotton pads stuck to the inner wall of the plastic container along with green French bean pod and cotton pads containing Corcyra eggs at the bottom. Individual mated female was released into the container and observed for seven days to record the number of eggs on the pods.

Statistical analysis

The results are expressed in the form of means ± S.D. Data analysis was done with significance ($p < 0.05$) of treatment effects using one-way ANOVA, followed by *post hoc* comparisons. The significance of the results was determined by Duncan's multiple range test (DMRT) using the Statistical Package for the Social Sciences (SPSS) software (version 20, IBM).

RESULTS AND DISCUSSION

Influence of prey on orientation and predation of fourth instar nymph

The orientation of predator two hours after the release observed on various preys like aphid (*A. gossypii*), mite (*T.urticae*),whitefly (*A. disperses)*, mealybug (*P.solenopsis)* and *Corcyra* eggs indicated maximum aggregation of predator nymph in mite (2.50) followed by *Corcyra* eggs (2.00). The least preferred prey was whitefly (0.50). The orientation of fourth instar nymph was also observed in olfactometer. The results indicated maximum aggregation of nymph in *Corcyra* eggs (3.00) followed by mite (2.50). The least preferred prey was whitefly (0.80). The prey predation six and 24 hours after the release was high in *Corcyra* eggs (4.00 and 6.50) followed by mite (3.25 and 4.50) respectively. Least predation was noted with whitefly (Table 1).

Influence of prey on orientation and predation of adult *B. pallescens*

The orientation of adult predator two hours after the release on various prey species *viz.,* aphids (*A. gossypii*), mite (*T.urticae*), whitefly (*A. disperses)*, mealybug (*P. solenopsis)* and *Corcyra* eggs was observed. The congregation of adult predator on *Corcyra* eggs was maximum (3.80) followed by mite (3.50). The least preferred prey was whitefly (0.25). The orientation of adult was also observed in olfactometer. The results indicated maximum aggregation of adult in *Corcyra* eggs (3.80) followed by mite (3.00). The least preferred prey was whitefly (0.80). The predation was maximum in *Corcyra* eggs (5.00 and 7.00) followed by mite (4.00 and 5.00) at six and 24 hours after the release respectively (Table 2).

Influence of prey host plants on the orientation and predation of fourth instar nymph

The aggregation of predator nymphs on mite prey provided in the host plant arena of bhendi, cluster bean, brinjal and bitter gourd was observed 2hrs after the release. The assemblage of predator nymphs to mites on host bhendi was maximum (4.8) followed by brinjal (3.0).Similar result was obtained on the orientation of nymph observed with olfactometer. The consumption of mites by the predator nymph six and 24 hours after the release noted in brinjal was high with 4.4 and 7.0 mites respectively. This was followed by predation of mites in bhendi host which recorded 2.6 and 4.0 mites at six and

Table 3. Influence of host plants of prey species on the orientation and predation of fourth instar nymph of *B. pallescens*

Prey host plants	Orientation of predators* 2 HAR Test arena		Number of mites fed*	
	Circular basin	Olfactometer	6 HAR	24 HAR
Bhendi	4.8 (2.19) [a]	3.6 (1.90) [a]	2.6 (1.61) [a]	4.0 (2.00) [b]
Cluster bean	0.8 (0.89) [c]	0.6 (0.77) [b]	0.4 (0.63) [b]	0.6 (0.77) [c]
Brinjal	3.0 (1.73) [b]	3.2 (1.79) [a]	4.4 (2.10) [a]	7.0 (2.65) [a]
Cucurbit	1.4 (1.18) [c]	0.8 (0.89) [b]	1.2 (1.10) [b]	3.2 (1.79) [b]
SE(d)	0.16	0.18	0.22	0.19
CD(0.05)	0.34	0.37	0.46	0.40

*Mean of five replications; HAR- hours after release; Values in the parentheses are square root transformed values

Table 4. Influence of host plants of prey species on the orientation and predation of adult *B. pallescens*

Prey host plants	Orientation of predators* 2 HAR Test arena		Number of mites fed*	
	Circular basin	Olfactometer	6 HAR	24 HAR
Bhendi	5.0 (2.24) [a]	3.8 (1.95) [a]	3.0 (1.73) [a]	6.0 (2.45) [a]
Cluster bean	0.8 (0.89) [c]	1.0 (1.00) [b]	0.4 (0.63) [b]	0.8 (0.89) [c]
Brinjal	2.6 (1.61) [b]	3.4 (1.84) [a]	4.0 (2.00) [a]	7.2 (2.68) [a]
Cucurbit	1.0 (1.00) [c]	1.2 (1.10) [b]	1.4 (1.18) [b]	3.6 (1.90) [b]
SE(d)	0.17	0.20	0.22	0.20
CD(0.05)	0.36	0.43	0.47	0.41

*Mean of five replications; HAR- hours after release; Values in the parentheses are square root transformed values.

24 hours after release respectively. The feeding of mites 0.4 and 0.6 noted in cluster bean was least respectively in six and 24 hours after the release (Table 3).

Influence of prey host plants on the orientation and predation of adult

The orientation of adult predator two hours after the release on various hosts like bhendi, cluster bean, brinjal and bitter gourd showed maximum congregation to bhendi (5.0) followed by brinjal (3.0). Similar result was obtained on the orientation of adult predator observed with olfactometer. The consumption of mites by the predator adult respectively in six and 24 hours after the release was significantly high in brinjal (4.0 and 7.2) and found on par with bhendi (3.0 and 6.0). The feeding of mites respectively in six and 24 hours after the release on cluster bean was least (0.4 and 0.8) (Table 4).

Preference of host plant as substrate for oviposition

The results revealed that the predator oviposited readily in all the substrates provided. The mean number of eggs laid per day on substrates like lablab, beans, cowpea, cluster bean, bhendi and green peas were 4.79, 4.46, 4.21, 8.43, 5.25 and 4.54 respectively. The mean number of eggs laid was high (8.43) in cluster bean as compared to beans (4.46). The hatching of eggs laid on different substrate was also observed. The per cent hatching was high in green peas (96.21) followed by cluster bean (94.64).The hatching of eggs in cowpea and beans were low 86.77 and 89.24 per cent respectively (Table 5).

Adult nutrition on fecundity

The data on egg laying of adult *B. pallescens* provided with different adult nutrition to increase the fecundity indicates that the adult fed on honey +water +*Corcyra* eggs laid a greater number of eggs (10.46) followed by the specialized diet mix (8.37). The number of eggs laid was low (7.20) when normal diet of *Corcyra* eggs was given (Table 6).

The variation of prey preference can be attributed to the nutritional quality of the prey besides the aggregation of prey in the feeding arena. The least preferred whitefly prey noted in the present study can attributed to the succulence of the prey. The nymphs of whitefly are less succulent and scaly which might have contributed to the lesser preference and predation of *B. pallescens.* In addition, the nature of exoskeleton of the prey and its external structure also plays a role in the preference of the prey. Among the prey host plants tested, brinjal followed

Table 5. Preference of *B.pallescens* to different plant substrates for oviposition

Ovipositional substrate	No. of eggs/pods* at different days after the release of mated female							Mean number of eggs/days	Egg hatching (%)
	1	2	3	4	5	6	7		
Lab lab	0.50 (0.71)	2.00 (1.41)	4.0 (2.00) b	5.50 (2.35)	7.50 (2.74) b	6.50 (2.55) bc	7.5 (2.74) ab	4.79	90.62
Beans	0.75 (0.87)	5.00 (2.24)	3.5 (1.87) b	6.00 (2.45)	5.75 (2.40) bc	6.00 (2.45) bc	4.25 (2.06) c	4.46	89.24
Cowpea	0.50 (0.71)	4.00 (2.00)	5.00 (2.24) ab	6.00 (2.45)	3.75 (1.94) c	4.25 (2.06) c	6.00 (2.45) bc	4.21	86.77
Cluster bean	1.25 (1.12)	6.25 (2.50)	7.75 (2.78) a	7.50 (2.74)	11.75 (3.43) a	13.75 (3.71) a	10.75 (3.28) a	8.43	94.64
Bhendi	0.50 (0.71)	3.75 (1.94)	5.25 (2.29) ab	5.25 (2.29)	6.25 (2.50) bc	7.75 (2.78) bc	8.00 (2.83) ab	5.25	93.48
Green peas	0.25 (0.50)	1.75 (1.32)	4.50 (2.12) ab	4.50 (2.12)	6.50 (2.55) b	9.75 (3.12) ab	4.50 (2.12) c	4.54	96.21
SE(d)	0.30	0.49	0.27	0.30	0.28	0.43	0.27	-	-
CD (0.05)	0.63	1.03	0.56	0.64	0.59	0.91	0.56	-	-

*Mean of four replications ; Values in the parentheses are square root transformed values.

Table 6. Effect of adult nutrition on fecundity

Treatment	No. of eggs laid*					Mean
	Day 1	Day 2	Day 3	Day 4	Day 5	
Honey + Water + *Corcyra* eggs	3.00 (1.73)	6.71 (2.59) [a]	9.86 (3.14) [a]	15.00 (3.87) [a]	17.71 (4.21) [a]	10.46
Specialized diet + water + *Corcyra* eggs	1.29 (1.13)	4.86 (2.20) [b]	8.57 (2.93) [ab]	12.00 (3.46) [b]	15.14 (3.89) [b]	8.37
Corcyra eggs + water	1.14 (1.07)	3.86 (1.96) [b]	6.57 (2.56) [b]	11.43 (3.38) [b]	13.00 (3.61) [b]	7.20
SE(d)	0.29	0.14	0.20	0.16	0.15	-
CD(0.05)	0.60	0.29	0.43	0.34	0.31	-

*Mean of seven replications ; Values in the parentheses are square root transformed values

by bhendi showed positive influence on the orientation and predation of *B. pallescens* on mites. Yarahmadi and Rajabpour (2017) also reported similar findings on the variation of predation of *Orius albidipennis* on *Tetranychus turkestani* and *Bemisia tabaci* in sweet pepper and cucumber. They opined those morphological characters like hairiness of leaves of cucumber might have negative impact on the predation as compared to sweet pepper. The volatiles of host plants harbouring the mite prey could also influence the orientation and predation of *B.pallescens.* The results obtained in the present study on preference of predator to brinjal and bhendi hosted mites shall also have attributed to the above factors.

The predator *B. pallescens* preferred to lay more eggs on cluster bean pods and least preference on vegetable cowpea and French bean pod. Regarding the hatching of laid eggs, maximum hatching was noted in green peas followed by cluster bean. The findings of Sobhy *et al.* (2005)who reported the variation among the oviposition substrates (bean pods, geranium leaves, sweet pepper seedling) in the egg laying of *Orius albidipennis* was in line with the present finding. Regarding the hatchability of eggs, Sobhy *et al.* (2005) showed variation among the ovipositional substrates with maximum hatchability in bean pods. The higher preference of cluster bean as an ovipositional substrate by *B. pallescens* might be due to the absence of dense trichomes and hairiness in cluster beanas compared to other substrates besides optimum moisture content retained for longer period. In addition, fungal moulds present on the pods of ovipositional substrate grow faster in other host pods as compared to cluster bean which slow down the mould growth. In addition, the plant volatiles also influence the ovipositional response in predators. The above reasons can also be attributed to more preference of cluster bean as anovi positional substrate to *B. pallescens*.

The reports of Lundgren and Fergen (2006) explains that the predatory bug *Oriusin sidiosus* preferred pole bean for oviposition to green foxtail, orchard grass and soybean well supported to the present results. Coll (1995) also found that *O.insidiosus* does not prefer to lay its eggs on the vegetative structures of *Zea mays* when given a choice among *Phaseolus lunatus*, *Capsicum anuum*, *Lycopersicum esculentum*. This observation was akin to the results obtained in the present study. The results of the study indicated that the mean number of eggs laid by the predator adult was maximum (10.46) when honey is provided along with *Corcyra* eggs and water followed by specialized diet mix (8.37). Heimpel and Jervis (2004) reported that provision of carbohydrates and water as adult food to predators and parasitoids enhance the longevity and fecundity. This report supports the present findings on the enhanced egg laying of predator with the adult food of honey +*Corcyra* eggs + water.The findings of Kiman and Yeargan (1985) who reported that the nymphs reared on pollen alone and along with arthropod preys successfully completed its development and when beans and water alone were given no nymphs completed its development into adults. They also found that the fecundity was significantly higher on diets containing *Heliothis virescens* eggs. This finding was in line with the present finding.

In conclusion, the nymph and adult predator preferred to orient and predate more preys when present on host plant brinjal followed by bhendi. Among the host plants *viz*., brinjal, bhendi, cluster bean and cucurbit, the least preferred host plant was cluster bean. Among the five preys offered to anthocorid nymph, the orientation and predation was maximum on *Corcyra* eggs followed by mite. The least preferred prey was noted as whitefly. The green pod of cluster bean was the most preferred substrate for egg laying as against the green French bean pod commonly used in mass culturing. The eggs laid on preferred cluster bean pod also showed maximum hatching (94.64%) next to green peas (96.21%).The adults of anthocorid bugs provided with additional nutrition of honey and water along with the normal diet of *Corcyra* eggs laid more eggs.

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