Available online at www.elixirpublishers.com (Elixir International Journal)

Entomology

Elixir Entomology 92C (2016) 38778-38782

ISSN: 2229-712X

Choice based Olfactory Preference of two Predators, *Rhynocoris marginatus* and *Coranus spiniscutis* (Insecta: Heteroptera: Reduviidae) to different Insect Pests

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ARTICLE INFO

Article history:

Received: 27 January 2016;

Received in revised form: 1 March 2016;

Accepted: 4 March 2016;

Keywords

Y- Shaped Olfactometer,
Reduviids,
Agonoscelis tuberula,
Epilachna vigintioctopunctata,
Raphidopelpa foveicellis,
Chryrocoris stolli.

ABSTRACT

Both *Rhynocoris marginatus* (F.) and *Coranus spiniscutis* (Reuter) are polyphagus predator of agriculture important insect pests, collected from Deoria agroecosystem and under Electric light source, Gorakhpur respectively, during 2012-2013. To determine the olfactory preference of these polyphagus predators towards one prey over another, choice based experiments were conducted in the laboratory by using Y-shaped olfactometer. The distant ends of the first two tubes of the olfactometer was used for keeping two different prey types whereas the third tube end used for introducing the predator. *R. marginatus* was evaluated against the African cluster bug, *Agonoscelis tuberula* Stål (Hemiptera: Pentatomidae) and Hadda beetle, *Epilachna vigintioctopunctata* (Coleoptera: Coccinelidae), whereas *C. spiniscutis* against *Raphidopelpa foveicellis* (Coleoptera: Chrysomelidae) and *Chryrocoris stolli* (Hemiptera: Scutelleridae). The reduviid, *R. marginatus* was found to be more responsive to African cluster bug (60%) followed by Hadda beetle (50%) and *Coranus spiniscutis* strongly preferred *Raphidopelpa foveicellis* (91.67%) followed by *Chryrocoris stolli* (8.33%). In these observations, the preference of both preys during 24 hours by *C. spiniscutis* was converted into percentage value. The predator, *R. marginatus* showed higher rostral protruding activity with African cluster bug (8.29±0.159min) than that of Hadda beetle (1.65±0.049min). This may be due to the strength of the prey's chemical cues of one prey over another elicited a quicker approaching. Prey consumption time of *R. marginatus* was significantly longer with regard to African cluster bug (60±2.44 min) as compared to Hadda beetle (37±2.50 min).The results clearly showed that the prey chemical cues determine the prey selection pattern of these predators. The outcome of this result will be helpful to consider these predatory bugs as biological control agents against preferred insect species.

Introduction

Predators are regarded as 'keystone species' because of the great impact they promote in some ecosystems (Paine, 1966; Begon et al., 1996). These impacts of a generalist predator on any given prey species will depend not only on the abundance and susceptibility of that prey species but also on the abundance and susceptibility of other species that share the same enemy (Holt and Lawton, 1994).

Due to the problems posed by the use of synthetic insecticides for pest control, including environmental degradation and the pests' development of resistance to insecticides (Gets and Gutierres, 1982; Jaffé et al., 1993), there is a growing use of natural enemies and semiochemicals in integrated insect pest control program. Utilization of predatory insects such as assassin bugs helps in the regulation of insect pest population in integrated pest management (IPM). Evidence of assassin bugs ability as effective generalist predators in several agroecosystems is well documented (Cohen, 1990; James, 1994; Ambrose, 1995).

Prey selection by generalist insect predators could have important consequences for prey population. The choice of the best prey by a predator will depend on the energetic value of the prey, the handling time and the search time (Krebs and Davies, 1993).Simply observing a predator preying upon a prey species is not sufficient to conclude that the particular prey is the predators preferred prey or its normal prey (Huffaker and Messenger, 1964). Preference is evident when the predator selects certain species among others that are equally available (Saint-cry and Cloudier, 1996). Hence, it is imperative to understand the host specificity of predators before employing them in biological welfare. Generalist insect predators frequently are the most abundant natural enemies in annual agroecosystem (Ehler and Miller, 1978; Wiedenmann and O'Neil, 1990). Generalist predators may consume virtually and arthropod they are able to capture, which allows them to establish and maintain high population densities (Settle et al., 1996; Cisneros and Rosenheim 1997, 1998). Thus generalist predators may contribute to the suppression of herbivore population and could be used in biological control of insect pest (Murdoch, 1985; Settle et al., 1996). Hence the present study aims at investigating behaviour of two predators, *R. marginatus* and *C. spiniscutis*, to four different insect pests by using Y-shaped olfactometer under lab condition.

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Materials and Method

Environmental condition

The study was conducted in Entomology laboratory, St. Andrew's College campus, Gorakhpur. All insects rearing and experiments were carried out in an environmental controlled room at day temperature 35°C, night temperature 24°C with a photoperiod of 12:12 (L:D) hour and 50-70% relative humidity monitored with a hygrothermograph.

Insect culture

The insects used in the experiments taken from a research stock-rearing of predator *R. marginatus* adults originally collected from groundnut field near Bhujauli colony Deoria district and *C. spiniscutis* collected from electric light source, Gorakhpur. The predators were reared in plastic containers (15x7 cm) in the laboratory under optimal conditions on the head crushed larvae of rice meal moth *Corcyra cephalonica* (Stainton).

Pest collection and maintenance

Different life stages of *E. vigintioctopunctata* and *R. foveicellis* from potato and cucurbits field and the adult of *A. tuberula* were collected from thyme field near Bhalwani, Deoria and *C. stolli* were found in the campus of St. Andrew's College, Gorakhpur from *logan croton bonplandianum Bail* plant. The adults of *E. vigintioctopunctata* and *R. foveicellis* were maintained in the plastic troughs (34x9cm) on fresh leaves of cucurbits. The adults of *A. tuberula* and *C. stolli*were maintained on fresh thyme leaves and croton leaves, respectively. The rearing troughs or containers were examined daily and the fecal matter was removed to prevent the fungal contamination.

Prior to the experiments predators were held in separate cages and during 12-24 hours after their emergence, with water swabs and without food.

Bioassay for orientation/approaching behavior in Y-shaped olfactometer

A Y-shaped olfactometer made-up of glass (main stem 15cm length, two arms 12cm length and 4cm diameter, and 90° angle between them) was used for the bioassay studies (Plate1c),. Before starting the experiment the Y-shaped olfactometer with odor cells were cleaned with 70% alcohol followed by continuous blowing of air by air by an aerator for 15min to remove the unwanted odor from the odor cells. The 24h-starved predators were introduced through the main stem and their predatory behavior was observed for 15 min continuously. The predatory behavior was observed in terms of searching and consuming time.

This experiment aimed at understanding the host selection behavior of *R. marginatus* towards *A. tuberula* and *E. vigintioctopunctata* and *C. spiniscutis* towards *R. foveicellis* and *C. stolli* using a Y-shaped dual tube olfactometer at room temperature 30°C with twenty and twelve replications, respectively. All insect pests were maintain in laboratory and twenty adult female of *R. marginatus* and twelve adult female of *C. spiniscutis* were randomly selected from a rearing cage and staved for 24 hours.

Insect pests were placed in the alternative odor source tubes and a single predator was released in to the starting point of the olfactometer and collected in the observation arena. If predator did not move within 15 min we regarded this as no choice, predator than removed and replaced with another predator. We studied searching time (from starting point to observation arena) percentage of *R. marginatus* either selecting *A. tuberula or E. vigintioctopunctata* and *C. spiniscutis* either selecting *R. foveicellis* or *C. stolli.* The bioassay experiments were performed in the Y-shaped olfactometer and time spent by the predators provided a clear representation of behavioral response.

Statistical analysis

A student's 't' test was performed to determine the differences in the approaching and rostrum protrusion time and the approaching behavior of the predator between the prey and control chamber was determined by X^2 test and the significance was expressed at 5% level.

Result and discussion

The bioassay experiments were performed in the Y-shaped olfactometer and time spent by the predators to prefer insect pests provided a clear representation of behavioural responses (Table1 & 2).

When the predators, *R. marginatus* and *C. spiniscutis* were released into the main chamber of the Y-shaped olfactometer (Plate1c), it oriented towards the preferred pest cell with antennae directing towards the odor source. After getting perfect orientation, the reduviid palpated its antennae, followed by rubbing their legs, rostral cleaning and extended rostrum towards the odor source. Once the predator entered the preferred pest cell it exhibited quick walking and approaching with antennae, wings and legs cleaning and rostral protrusion. Behavioural chemicals associated with secondary plant metabolites and chemicals with the host prey that provides cues for orientation of reduviid predators in their prey finding sequence (Ambrose et al. 1983; Claver and Ambrose 2001, 2003; Claver et al. 2004).

The total number of visits of *R. marginatus* and *C. spiniscutis* to the positive and negative odor cells is shown in figure 1 and 2. Both the predators were strongly attracted to the chemicals produced by preferred prey and visit the positive chamber more frequently than the negative one. Similar positive approach response variedly exhibited by *Rhynocoris longifrons* Stål to the hexane fraction of *Helicoverpa armigera* (Hubner), *Spodoptera litura*,(F.) *Achaea janata* (L.), *Mylabris pustulata* (Thunberg) and *Dysdercus cingulatus* (F.) in 6-armed olfactometer (Kumar and Ambrose 2014)

R. marginatus was attracted by both prey African cluster bug and Hadda beetle 55% and 35% respectively wheras 10% of *R. marginatus* were did not leave the starting point. *C. spiniscutis* significantly (P=0.05) preferred leaf beetle 91.67% followed by jewel bug 8.33%. The response of *R. marginatus* towards the prey chemical cues can be visualized by exploration and probing behavior exhibited by the predators due to their chemosensory perception as observed by Bilgrami and Pervez (2001). The reduviid predators generally prefer lepidopteran caterpillars (Ables, 1978). McMahan (1983) noted that reduviids preference for one prey to another in choice test might be influenced by the noxious smell or unpleasant taste of the prey. But James (1994) and Fuseini and Kumar (1975) stated that, reduviid bugs apparently preferred heteropteran bugs by their better taste.

When *R. marginatus* and *C. spiniscutis* were released in to the starting point of the olfactometer 90% of them were able to search for and capture the prey. Once the predators approached the prey they clumped around it, extended their rostrum and started probing the prey. Sahayaraj and Paulraj (2001) also showed similar observations in reduviids. Several studies were also carried out in this regard in other pr edatory insects (Yasuda and Wakamura, 1996; Yasuda, 1997; Jhansilakshmi et al. 2000; Singh and Paul, 2002). They postulated that the chemical cues or kairomones of the preys stimulate the predators to respond towards them.

The response of *R.marginatus* to *A. tuberula* and *E. vigintioctopunctata* is shown in table 1. The response was higher for *A. tuberula*as compared to *E. vigintioctopunctata* (X^2=2.7; df=2; p< .005) The response of *C. spiniscutis* to *R. foveicellis* and *C. stolli* is shown in table 2. The response was higher for *R. foveicellis* as compared to *C. stolli* (X^2=5.83; df=2; p< 0.05).

The rostral protruding of *R. marginatus* and *C. spiniscutis* to the preferred pests is shown in the table 2. The predator *R.marginatus* exhibited the highest searching and rostral protruding time in African cluster bug (8.29±0.159min) followed by Hadda beetle (1.65±0.049min) and the predator *C. spiniscutis* showed highest time in leaf beetle (22.05±1.38 min) followed by jewel bug (0.01±0.00min).

Consuming time of *R. marginatus* exhibited the highest handling time in African cluster bug (60±2.44 min) than that of Hadda beetle (37±2.50 min).The handling time of *R. marginatus* on the two pests studied could be supported by the findings of Maran (1999) in different reduviids such as *Rhynocoris kumarii* Ambrose and Livingstone.

Preference of reduviids predator was determined by the activity of the prey, size of the prey and preying habits of the predator. Preference response elicited by a particular prey varied from one reduviid predator to another (Dolling, 1991; Ambrose, 1999). Such prey preference is intimately bound with prey choice. Although, assassin bugs are polyphagous predators, they exhibit a narrow range of host preference (Ambrose. 1999; Ambrose and Claver, 2001).

All these results and the present investigation reveal that the semiochemicals emitted from the prey species indeed elicit an intense host searching behavior in the predators. Although all the four prey tested in the present study elicited a positive response in *R. marginatus* and *C. spiniscutis*. The results tabulated show that the predator*, R. marginatus* preferred heteropteran pest whereas predator, *C. spiniscutis* preferred coleopteran pest. The chemical perception of these predators depends on the chemosensory system conveying the requisite quantity of information about the prey (Sujath et al 2012; Kumar and Dhiman, 2013). Moreover, the kairomones from the body of host prey probably act as sign stimuli, eliciting intensive searching behavior, rather than as a guiding cue attracting and directing the reduiids to the host (Sahayaraj and Delma 2004; Kumar et al 2011; Kumar and Ambrose 2014). The interaction between two factors should be reason for the preference of the different family of the prey by the predators. Thus this investigation offers new strategies for the chemical cues-mediated pest management in agroecosystem.

Table 1. Approaching behavior of *R. marginatus* to *A. tuberula* and *E. vigintioctopunctata* and *C. spiniscutis* to *R. foveicellis* and *C. stolli*

Response	*R.marginatus*	*C.spinscutis*
1st prey	11	11
2nd prey	7	1
No choice	2	0
X^2	2.7	5.83
Significance	N.S.	*

*=Significant at 5%level, N.S.= not significant

Table 2. Searching and rostral protruding time of *R. marginatus* and *C. spiniscutis* to the preferred insect prey (X±SE).

Predator	Insect preys	Searching and rostral protruding time (min.)
R. marginatus	*A.tuberula* *E. vigintioctopunctata*	8.29 ± 0.159 1.65 ± 0.049
C. spiniscutis	*R. foveicellis* *C. stolli*	22.05±1.38 0.01±0.00

*t-test significant at p<0.05

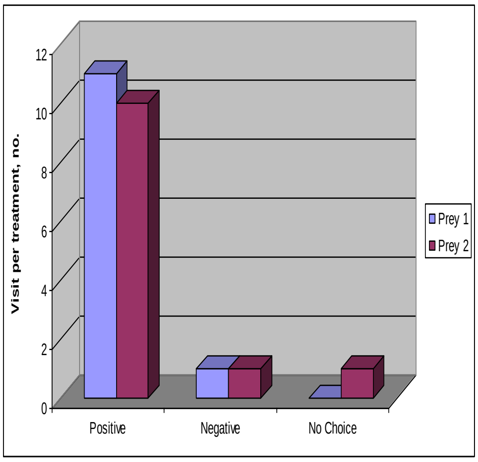

Figure 1. Effects of odors on *C. spiniscutis* adults as determined by the number of visits to chambers containing the following odor sources: *R. foveicellis* (Prey 1) and *C. stolli* (Prey 2).

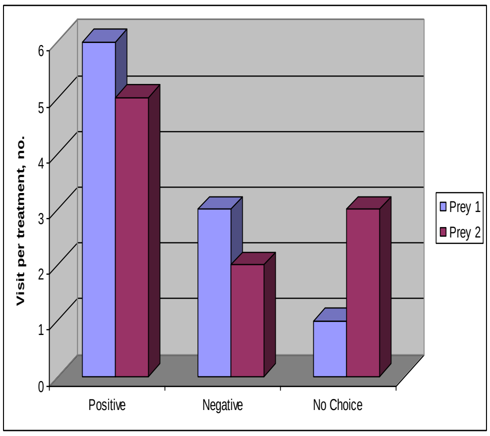

Figure 2. Effects of odors on *R.marginatus* adults as determined by the number of visits to chambers containing the following odor sources: *A. tuberula* (Prey 1) and *E. vigintioctopunctata* (Prey 2)

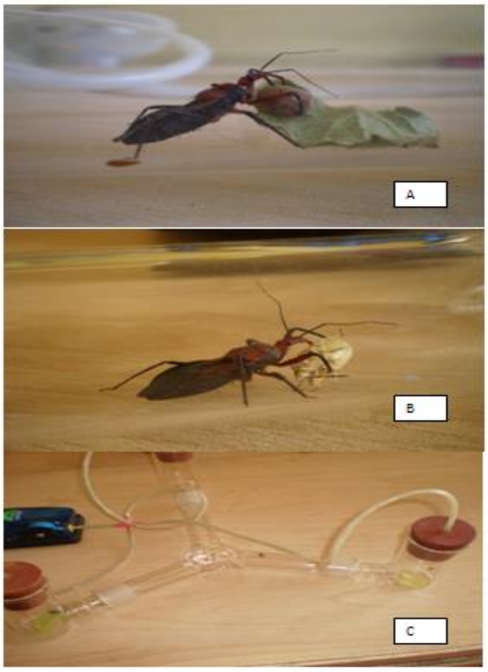

Plate 1. *R. marginatus* prey upon Hadda beetle (A), and African cluster bug (B), *Coranus spiniscutis* preferring leaf beetle (C) in Olfactometer.

Acknowledgements

We thank UGC, New Delhi for the financial support through a research project (F. 34-443/2008(SR) dated Dec 29, 2008). The authors are grateful to the authorities of St. Andrew's College especially its Principal Rev. Dr. J.K Lal for his kind words of encouragements and supports.

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