

Variations on prey preference and ability on functional response of a generalist insect predator, *Rhynocoris marginatus* (Hemiptera: Rduviidae) to three selected cucurbitaceae insect pests.

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Abstract:

In this investigation, the prey preference strategy, and functional response variation of entomophagous reduviid bug, *Rhynocoris marginatus* was observed. This reduviid bug feeds upon the insect pest belonging to Order Hemiptera, Lepidoptera, Arthropoda, Coleoptera, and Isoptera. The order of preference of the *R. marginatus* to Hadda beetle, Grasshopper, and Red Pumpkin Beetle was 59.09 ± 0.4 %, 22.72 ± 0.8 %, and 18.18 ± 1 % respectively. The functional response of *R. marginatus* against three selected insect pest was evaluated. It shows Type II functional response to kill three pests. Moreover, the negative correlation was observed between searching time and prey densities at each category. The maximum predation of *R. marginatus* were $k = 6.87$, $k = 2.25$, and $k = 2.12$ is a general predation towards hadda beetle, grasshopper, and red pumpkin beetle, respectively. This reduviid bug feeds upon all agricultural and some forest insect pest. Therefore can be utilized in integrated pest management as a biocontrol agents for reducing insecticide use on agricultural as well as forest ecosystem.

Key word: Biocontrol, Insect Pests, Hadda beetle, Red Pumpkin beetle, Grasshopper, Assassin bug, Pest choice, and Functional response.

Introduction:

Along with cereal grain plant Sorghum, *Sorghum bicolor* Linn., Moench (Poaceae), pumpkin fruit of certain varieties of squash such as varieties of *Cucubita pepo*, *C. moschata*, *C. maxima*, *Cucumis melo*, *C. sativum*, and *Citrullus lanatus* in the guard family (Cucurbitaceae) have been cultivated in winter and summer crop plant and they are served stable food of Indian population as they give balanced diet. Cucurbitaceae is a medium sized and specialized family of climbing plants and are mainly tropical or sub-tropical in distribution, with a few species extending into temperate climate (Subrahmanyam, 2004). These crops are attacked by variety of insect pests from seedling until harvest. Successful cultivation of cucurbits requires an effective and economical control of insect pests (Sharma et al., 2016). The present investigation has identified more than ten insects pests from cucurbitaceae crop which belong to the orders Hemiptera, Orthoptera, Lepidoptera, and Coleoptera are preferred by *R. marginatus*. Generally, three pests melon lady bird / hadda beetle, red pumpkin beetle, and grasshopper occurs in agricultural field of cucurbits in Gorakhpur district, Uttar Pradesh, India. Herbivorous insect pests are a serious threat to agriculture as they greatly affect plant reproduction and reduce the biomes and distribution of crops (Savary et al., 2019). Traditionally crop protection predominantly relies on chemical to prevent the damage caused by pests (Zhu et al., 2024). Insecticides have been playing a significant role in the field of agriculture but their debits have resulted in serious health implication to human beings, non-target organism and the environment (Ansari et al., 2014). A generalist predator, *R. marginatus* kill the pest having soft cuticle by releasing the venomous saliva into the body of prey followed by sucking inner body content (Nagarajan, 2010; Smith, 1966; Sahayaraj et al., 2020). Hence, these natural enemies can be utilized in IPM to reduced insect pests, crop yield loss, and harmful effect of insecticide on human being as well as environment. *Rhynocoris marginatus* is a natural predator, found into Agro forest ecosystem (Sahayaraj, 1999, 2002). Many literatures are available on the biocontrol potential of *R. marginatus* against several taxa of insects pests suggest that this predator primarily feed on early developmental stages of Lepidoptera, Hemipterans, Coleopterans, and Isopterans (Sahayaraj, 1999). Predatory reduviids feed on a variety of food sources and encounter several prey with different nutritional value and use defensive mechanisms while encountering prey and the predator has develop several attack strategies to exploit a variety of preys (Sahayaraj, 2018). According to Begon et al. (1996), a predator is classified as truly generalist when its prey selection is proportional to the relative abundance of the prey species in its environment. Thus, in the current study, we evaluated the prey preference, and functional response of *R. marginatus* against three pests' viz., melon or hadda beetle, red pumpkin beetle, and grasshopper. Prey preference of a predator depends upon nutritional requirements, predation capacity, and capturing ability of predator. Prey preference also elicited by size, softness – hardness of prey body, defensive ability, and volatile nature of the prey. Predator – prey interactions play a crucial role in shaping the structure and functioning of ecosystem and understanding these interactions and their effects is key to understanding animal communities (Chase et al., 2011). Prey preference of a polyphagous predator is determined by the size, shape, color, speed of the prey, texture of the prey, chemical produced by the prey (Saint – Cry and Cloutier., 1996). Functional response describe the ability of predator to attack ratio, consumption capacity, and minimize population dynamics of a selected prey species in particular period, and different climatic condition in prey and predatory Agro forest ecosystem. The functional response describes the rate at which a predator kills its prey at different prey densities and can thus determine the efficiency of a predator in regulating prey populations (Murdoch and Oaten., 1975). Ecologist has delimited functional response into three types (Holling 1959;

1966). The functional response may represent an increasing linear relationship (Type I), a decelerating curve (Type II), or a sigmoid relationship (Type III).

Material and Methods:

Predator Collection and rearing:

Gravid females of *Rhynocoris marginatus* were collected from the branch of a sheltering plant *Cleome viscos* Linn., Plant in Maha Yogi Krishi Vigyan Kendra Chaukhmafi (Lat: 26.93012N 26°55'48.42588'', Longi: 83.23686E 83°14'12.70212''), Pepeganj in Gorakhpur district, Uttar Pradesh, India. The *R. marginatus* were maintained in round, transparent, plastic container (25cm diameter, 8cm height) with twig, bark, green, and dry leaves to mimic microclimate of agro-ecosystem, and kept wet cotton bud to maintain humidity, grasshopper, *H. banyan* provided inside container as source of predator nutrition under laboratory condition at 30±2°C, 60-70% RH, and a photoperiod of 13h light (L): 11h dark (D). The stock culture of this predatory bug was raised inside laboratory.

Prey preference:

Choice experiment was carried out to study the prey preference of adult female of *R. marginatus* against melon or hadda beetle, red pumpkin beetle, and grasshopper in six arm olfactometer. The plant parts such as leaf, twig, and flower of host plant were kept inside each arm of olfactometer except central part to mimic agro-ecosystem. After the introduction of 48 hours starved predator in central arm and prey in other arm of olfactometer, the prey preference was assessed in terms of prey consumed by predator in 24 hours. Eight replicates were maintained in each species of insect pests.

Functional response studies:

The functional response of adult female *R. marginatus* was assessed at different prey densities viz., 1,2,4,8, and 16 prey / predator separately to above three insect pests in plastic container (diameter- 15.5 cm and height- 6.5cm). Adult female predator introduced into the plastic container where the prey with leaf, twig, and flower of host plant already placed. The selected adults of melon / hadda, red pumpkin beetle, and second instars of grasshopper were used on prey for functional response studies. After 24 hours of experimental period the number of prey killed by *R. marginatus* was recorded and the prey number was maintained constant by replacing them with fresh alive prey throughout the experimental days. Eight replications were maintained in 14 days observation.

“Disc” equation of Holling’s (1959) was used to calculate the functional response in each category of predators. “Disc” equation was derived from the following equation which signified the effect of prey density on attack

$$Y = a T_s \quad (1)$$

Where, a = rate of discovery per unit of searching time $[(y/x)/T_s]$

T_s = time spent by the predator in searching prey

Y = Total number of prey killed in a given period of time

x = Prey density

$$T_s = T_t - b y \quad (2)$$

T_t = total time in days when prey was exposed to the predator

$b = T_t/k$ = time spent for handling each prey by the predator

k = the maximum prey consumption

Substituting (2) in (1)

$$Y' = a (T_t - b y) x$$

The regression analysis was made to determine the relationship between the prey density and the number of prey consumed, searching time, attack ratio, handling and recovery time (Daniel, 1987).

Result and Discussion:

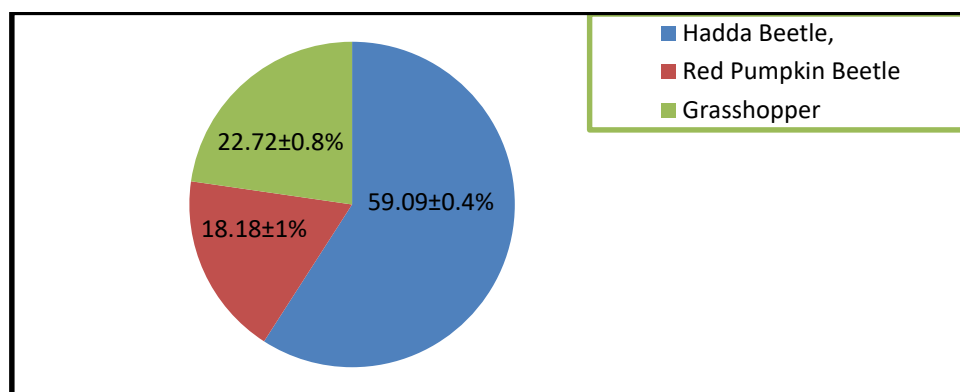
Prey preference:

The average prey preference estimated by choice tests is shown in table 1 and figure 1. Adult female of *R. marginatus* preferred adult of melon / hadda beetle (59.09%), grasshopper (22.72%), and red pumpkin beetle (18.18%) respectively. Balakrishana et al. (2011), reported that the adult of *A. pedestris* preferred the Slant-faced sp., 1 (25%), *T. varicoris* (16.38%), and cone-headed grasshopper (16.24%). Ambrose and Claver (2001), stated that reduviid predator, *R. marginatus* preferred red cotton bug, *Dysdercus cingulatus* (18.03%), flower beetle, *Mylabris pustulata* (4.26%). Reduviid predator, *R. kumarii* preferred Slant-faced grasshopper, *T. varicoris* Walker (21.57%), flower blister beetle, *Mylabris indica* Thunberg (13.45%), and *M. pustulata* Thunberg (8.03%). Reduviid predator generally preferred slow motion, soft cuticle, nymph, and defenseless prey. Reduviid predators generally prefer lepidopteran caterpillars due to their soft cuticle and slow movement (Ables, 1978). Prey selection reflects prey availability and, for each predators, this varies prey species, demographic and anti – predatory behaviors (Dreyer, 2024). Seasonal pulses of neonates will result in seasonal variations in prey availability; therefore prey selection is expected to vary seasonally and across prey demographic classes (Clements et al., 2016; Annear et al., 2023). The reduviid attacked hadda beetle, grasshopper were found to emit yellowish and greenish-black fluid in self defense. Ambrose and Claver (1996), reported that the reduviid attacked caterpillars were found to roll themselves and emit greenish fluid in self defense. Ambrose and Claver (1996) was reported size preference and functional response of the reduviid predator *R. marginatus* to its prey *Spodoptera litura*.

Table 1: Prey Preference % Mean ± SD of reduviid bug, *R. marginatus* against three different insect prey species.

Insect prey species	Preferred prey %
Hadda Beetle	59.09 ± 0.4
Grasshopper	22.72 ± 0.8
Red Pumpkin Beetle	18.18 ± 1

Figure 1: Prey Preference (%) Mean ± SD of *R. marginatus* on three chosen insect pests.



Functional response:

The evaluated functional response of *R. marginatus* to three selected insect pests viz., hadda beetle, red pumpkin beetle, and grasshopper were indicated in table 2 a, b, c. Curve linear functional response curves were obtained when contemplating the number of each prey species offered and killed in 24 hours by adult female *R. marginatus* (figure 2a, b, c). The predatory bug *R. fuscipes* exhibited curve linear functional response curve on Okra pest commonly known as red cotton bugs, *Dysdercus koengii* (Claver and Yadav., 2024). The results evidence type II functional response on hadda beetle, red pumpkin beetle, and grasshopper and it is possible in arthropod predators. The number of prey killed by predator gradually increased with the increased prey density and then stabilized on 16 prey density. Some time fluctuation was observed in case of red pumpkin beetle, and grasshopper (table 2a, b, c). Sahayaraj et al. (2014) reported similar type of functional response in the work on *R. kumarii* also exhibited on *Phenacoccus solenopsis*. The reduviid bug, *R. marginatus* was more predaceous on hadda beetle, followed by grasshopper, and red pumpkin beetle respectively. The number of hadda beetle, red pumpkin beetle, and grasshopper killed (y') by *R. marginatus* can be expressed in Holling's Disc equation [$y' = 0.17(14-8.12)x$, $y' = 0.15(14-13.2)x$, and $y' = 0.64(14-12.44)x$ respectively] in laboratory condition. The regression statistics and ANOVA indicated that the each insect prey species had significant impact on functional response of *R. marginatus* (variance in Column 1, 2 = 37.2, 7.34; 37.2, 0.27; 37.2, 0.22, $F = 0.47, 2.43, 2.53$, and P value = 0.51, 0.15, 0.15 for hadda beetle,

grasshopper, and red pumpkin beetle respectively). The maximum predation represented by k value was always found restricted to high prey density (k= 6.87, k= 2.25, and k= 2.12) for adult female predator of hadda beetle, grasshopper, and red pumpkin beetle. The searching day's (Ts = 11.97, 9.94, 5.88, 0.06, 0.06) of *Rhynocoris marginatus* decreased with the increasing melon / hadda beetle, grasshopper, and red pumpkin beetle density and this considered the *R. marginatus* as biological control potential. The type II functional response is typical of most heteropteran predators (Cohen and Tong, 1997; Cohen, 2000). Ambrose et al, 200. Reported that the *R. marginatus* responded to increasing density of two pests – *C. gibbosa*, and *H. banian* by killing more number of them than at lower densities; thus exhibiting type II functional response (Holling, 1959). Uniformly negative correlations were obtained between the prey densities and the searching times of the predator at all prey densities (Table 2 a,b,c). Ravichandran and Ambrose (2006), reported that the negative correlation obtain between prey densities and searching time in *Acanthaspis pedestris*. At high prey density, less time was spent in handling, whereas at low prey density the searching time always dominated the handling time (O' Neil, 1988). Similarly, Host stage preference, stage preference and functional response of assassin bug, *R. kumarii* to its most preferred prey tobacco cutworm, *Spodoptera litura* reported by Muniyandi et al., 2011. The naturally circumstance of *R. marginatus* is frequently not adequate to regulate insect pests. Hence, increment of the *R. marginatus* into the Agro forest ecosystem would be indispensable to achieve efficacious biological control.

Table 2 a: Functional response of *R. marginatus* to its preferred prey, *H. vigintiopunctata*.

Prey Density x	Prey Attacked y	Maxi. k	Days per y b = Tt/k	Days all Y's by	Days Searchi ng Ts=Tt-by	Attac k Ratio y/x	Rate of Discove ry y/x/Ts= a	Disc Equation Y'=a(Tt-by)x
1	1±0			2.03	11.97	1	0.08	
2	2±0			4.06	9.94	1	0.10	
4	4±0	6.87	2.03	8.12	5.88	1	0.17	Y'= 0.17(14-8.12)x
8	6.87±0.4			13.94	0.06	0.85	-	
16	6.87±0.4			13.94	0.06	0.42	-	

Table 2 b: Functional response of *R. marginatus* to its preferred prey, *H. banian*.

Prey Density x	Prey Attacked y	Maxi. k	Days per y b= Tt/k	Days all Y's by	Days Searchi ng Ts=Tt- by	Attac k Ratio y/x	Rate of Discove ry y/x/Ts= a	Disc Equation Y'= a(Tt-by)x
1	1±0			6.22	7.78	1	0.12	Y'= 0.64(14-12.4)x
2	2±0			12.44	1.56	1	0.64	
4	2.25±0 .4	2.25	6.22	13.99	0.01	0.56	-	
8	2.25±0 .4			13.99	0.01	0.28	-	
16	2.12±0 .3			13.18	0.82	0.13	0.15	

Table 2 c: Functional response of *R. marginatus* to its preferred prey, *A. foveicollis*.

Prey Density x	Prey Attac ked y	Maxi. k	Days per Y b=Tt/ k	Days all Y's by	Days Search ing Ts=Tt- by	Attac k Ratio y/x	Rate of Discov ery y/x/Ts= a	Disc Equation Y'= a(Tt-by)x
1	1±0			6.60	7.4	1	0.13	Y= 0.15(14-13.2)x
2	2±0			13.2	0.8	1	1.25	
4	2.12±0 .4	2.12	6.60	13.99	0.01	0.53	-	
8	2.12±0 .4			13.99	0.01	0.26	-	
16	2±0			13.2	0.8	0.12	0.15	

Figure 2a: Curve linear functional response of *R. marginatus* to its preferred prey, *H. vigintiopunctata*.

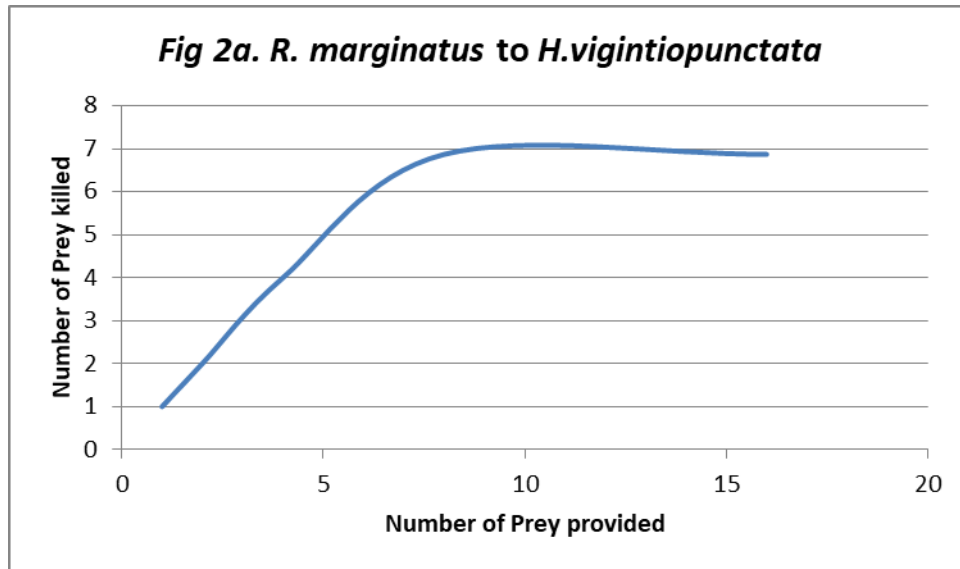


Figure 2b: Curve linear functional response of *R. marginatus* to its preferred prey, *H. banian*.

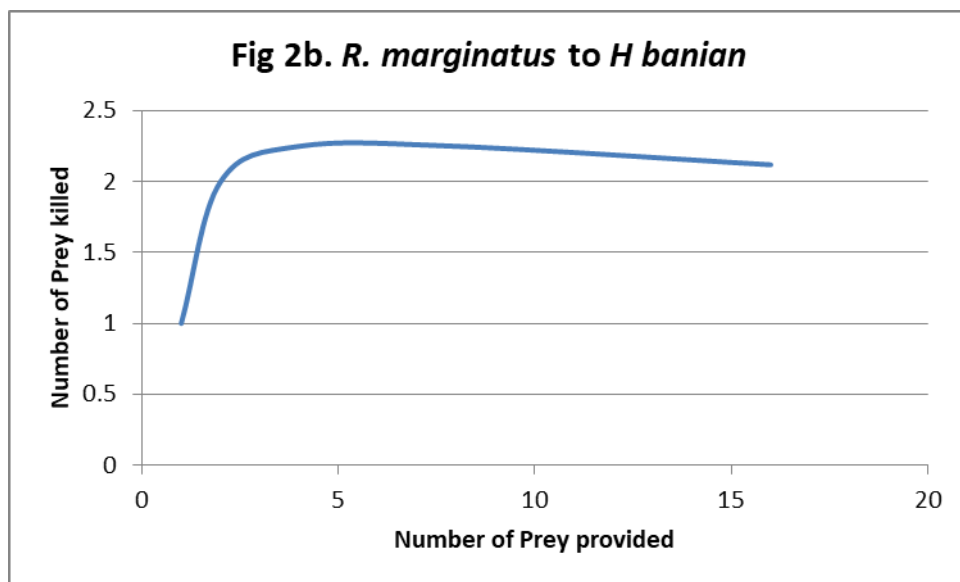
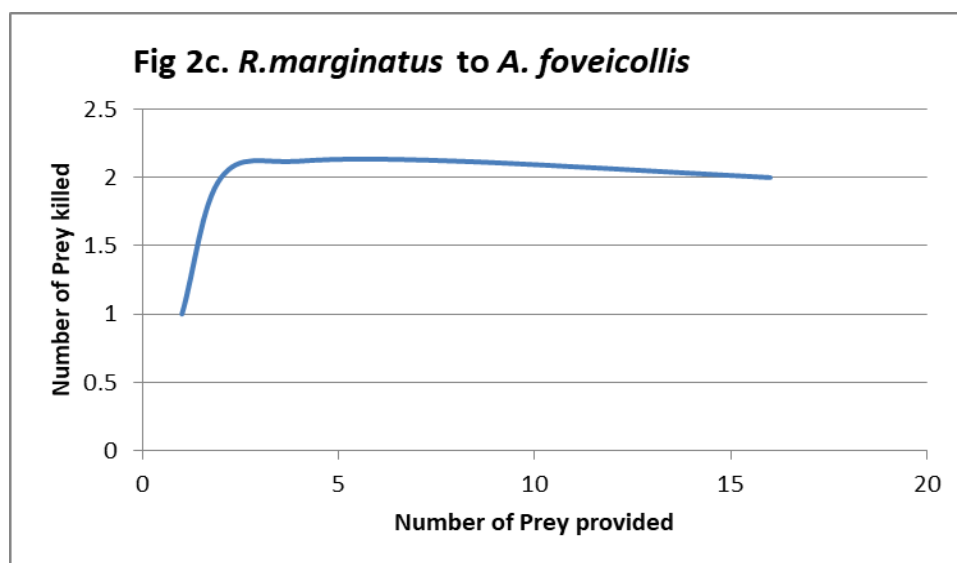


Figure 2c: Curve linear functional response of *R. marginatus* to its preferred prey *A. foveicollis*.



Conclusion

The above investigation finally revealed that the *Rhynocoris marginatus* can be applied in Integrated Pest Management. Because of *R. marginatus* was preferred melon/ hadda beetle followed by grasshopper and red pumpkin beetle. *R. marginatus* shows positive functional response towards above three insect pests under laboratory condition. Hence, application of this predator into IPM reduced demand of insecticide and its harmful impact on Agro forest ecosystem. Further studies are needed about these bugs on various another insect pests to gain more information.

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