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Demographic parameters of the predaceous ladybird, *Hippodamia variegata* (Goeze), on the aphid species, *Aphis craccivora* (Koch), reared on four host plants

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Abstract

The reproductive and demographic attributes of the aphidophagous ladybird, *Hippodamia variegata* (Goeze) (Coleoptera: Coccinellidae), when fed on the aphid species, *Aphis craccivora* (Koch), reared on four host plants, viz. *Dolichos lablab* L., *Vigna unguiculata* L. Walp, *Cajanus cajan* (L.) Millsp., and *Ranunculus sceleratus* L., were investigated in the laboratory to find host suitability for the production of the ladybird. Results indicated that there was a significant effect of host plant on both reproductive and demographic parameters of the predator. The host plant suitability was in the rank order *D. lablab* = *V. unguiculata* > *C. cajan* > *R. sceleratus*. This ladybird started reproducing very early and laid a high number of eggs for a longer period of time when the adults were fed on aphids raised on *D. lablab* and *V. unguiculata*. Both the fecundity and percent egg hatching increased significantly in the case of the abovementioned two host plants. Similar host plant effects were found on demographic parameters of *H. variegata*, with significantly high values of net reproductive rate (r_m), short generation (T_c), and doubling times (D.T.) on *D. lablab* and *V. unguiculata*. Age-specific fecundity of *H. variegata* at all host plant treatments was triangular in function with an initial increase in ovipositional rate with age, followed by attainment of peak and gradual decline till egg laying ceases. These peaks were higher on the host plant treatments, *D. lablab* and *V. unguiculata*. Sub-optimal values of both reproductive and demographic parameters were found on the host plant, *R. sceleratus*. Thus, it could be concluded that both *D. lablab* and *V. unguiculata* were the suitable host plants to raise aphid, *A. craccivora*, for the augmentative rearing of the ladybird, *H. variegata*.

Keywords: Coccinellidae, *Hippodamia variegata*, *Aphis craccivora*, Demographic analysis, Prey suitability, Host plants

Background

Predaceous coccinellids, popularly known as ladybirds (Coleoptera: Coccinellidae), are important biocontrol agents of numerous crop pests, viz. aphids, mealybugs, whiteflies, thrips, and other tiny soft-bodied insects (Dixon 2000 and Hodek et al. 2012). Their augmentative rearing in the laboratory depends largely on the prey food and the abiotic factors (Omkar and Pervez 2016). Hodek and Evans (2012) expanded the classical idea of food relationship in ladybirds in terms of essential food (that supports both development and reproduction), alternative food (that supports only survival), and rejected food (that is toxic and unpalatable). The degree of essential prey food varies among aphidophagous ladybirds, as they develop and reproduce optimally, when fed on certain aphids, viz. *Aphis craccivora* (Koch) and *Aphis gossypii* (Glover) (Pervez and Omkar 2004 and Omkar and Mishra 2005), whereas their development and reproduction are negatively affected, when fed on aphids containing toxic biochemical constituents. Among aphids, *A. craccivora* is one of the major crop pests infesting over 400 host plants, and the grain legumes are the major target crops (Blackman

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and Eastop 2000). *A. craccivora* is a major pest of Indian bean, [*Lablab purpureus* (L.) Sweet = *Dolichos lablab* L.], cowpea, *Vigna unguiculata* L. Walp (Karungi et al. 2000), and pigeon pea [*Cajanus cajan* (L.) Millsp.]. It also infests poison buttercup, *Ranunculus sceleratus* L. (Pervez and Kumar 2017).

Hippodamia variegata (Goeze) is a cosmopolitan, eurytopic, predaceous ladybird beetle with Palaearctic origin (Franzmann 2002 and Omkar and Pervez 2004) occurring abundantly in aphid prevalent hilly agro-ecosystems of North India. It is a polymorphic ladybird with an immense biocontrol potential (Honek et al. 2012). For the mass multiplication of this ladybird, a thorough understanding of its reproductive and demographic parameters is a prerequisite. These aspects have been investigated, using aphid prey cultured on only one host plant (Lanzoni et al. 2004; Farhadi et al. 2011 and Jafari 2011).

Aphid–host plant association also affects the prey suitability (Francis et al. 2000, 2004 and Pervez and Chandra 2018), as aphids reared on different host plants seem to bear different effects on the bio-attributes of *H. variegata* (Wu et al. 2010). Moghaddam et al. (2016) studied age-stage, two-sex life table of *H. variegata* feeding on the English grain aphid, *Sitobion avenae* (Fabricius), reared on four host plants. Rakhshan and Ahmad (2013) observed that *A. craccivora* reared on different host plants had significant effects on growth and development of predaceous ladybirds. In addition, consumption of *A. craccivora* was also varied when the ladybird was fed on prey raised from different host plants (Rakhshan and Ahmad 2015). The presence of allelochemicals and the morphology of different plants also affect the biocontrol potential of ladybirds.

Thus, the present study was designed to determine the demographic attributes of *H. variegata* on *A. craccivora* infesting four host plants, viz. *D. lablab*, *V. unguiculata*, *C. cajan*, and *R. sceleratus*, in the laboratory to determine host suitability for the mass multiplication of the ladybird.

Materials and methods

Collecting and rearing of *H. variegata* and aphid species

Adults of *H. variegata* were collected from the suburbs of a hilly region of Uttarakhand, India (30.2937° N, 79.5603° E), and brought to the laboratory. These were sexed and paired in Petri dishes (2.0 cm height × 9.0 cm diameter) in four sets (each containing 10 replicates for each host plant and each Petri dish had a pair of *H. variegata*) provided daily with a sufficient quantity of *A. craccivora* infested on (i) *D. lablab*, (ii) *V. unguiculata*, (iii) *C. cajan*, and (iv) *R. sceleratus*. The host plants were inoculated with the aphid, *A. craccivora*, and grown in a small crop-field, present in the college campus, Kashipur, India. The Petri dishes were kept in the Environmental Test Chamber (*REMI Instruments*, India), maintained at controlled abiotic conditions of 25 ± 1 °C, 65 ± 5% R.H, and 12L:12D. A preliminary experiment revealed that *H. variegata* reproduced best at 12L:12D photoperiod. Similarly, 27 °C was found to be the best temperature for laboratory rearing of the ladybird understudy (Maia et al. 2014). The F_1 eggs laid by the female ladybirds in each Petri dish were isolated in other dishes (size as above) to be used in the experiments.

Reproduction and demographic analysis

The F_1 eggs taken from the above stock were reared from egg-hatch to adult-emergence, using *A. craccivora* as prey obtained from the respective host plants (which were provided to the parents). The developmental duration and the number of adults that survived in each aphid–host plant combination were recorded ($n = 10$). After emergence, the adults were sexed and isolated in Petri dishes (space and diet as above). The adult male and female ladybirds were paired together in four sets and provided daily with a sufficient quantity of *A. craccivora* infested on (i) *D. lablab*, (ii) *V. unguiculata*, (iii) *C. cajan*, and (iv) *R. sceleratus* and observed daily for mating and oviposition for their lifetime until the death of female ladybird ($n = 10$). The data on pre-oviposition and oviposition periods, fecundity, and percent egg viability were recorded. The data for normality, using the Kolmogorov–Smirnoff test, and homogeneity of variance, using Bartlett's test via statistical software, SAS version 9.0, were tested, as well were the data of pre-oviposition and oviposition periods, daily oviposition, fecundity, and percent egg viability of *H. variegata*, when fed on *A. craccivora* raised on four host plants, to one-way ANOVA, using SAS, version 9.0. The age-specific fecundity of *H. variegata* was plotted by subjecting the data on mean reproductive age (i.e., mean values of ovipositional period) and daily oviposition to polynomial regression analysis using SAS, 9.0 (2002).

The demographic parameters, viz. net reproductive rate (R_o), intrinsic (r_m) and finite (λ) rates of increase, generation (T_c), and doubling times (D.T.) were estimated, using the jackknife method given by Maia et al. (2000, 2014) on SAS, 9.0 (2002). This method evaluates the pseudo-values of the demographic parameters, which helped to obtain their mean values and standard errors in each group. The box and whisker plots to illustrate the dispersion of pseudo-values within each group were constructed. This is directly related to the uncertainty of their estimates.

Results and discussion

The pre-ovipositional and ovipositional periods, fecundity, and percent egg viability of *H. variegata*, when fed on *A. craccivora* raised on four host plants, are presented (Table 1). All these parameters were significantly

Table 1 Reproductive parameters of *H. variegata* when fed on aphid, *A. craccivora*, infested on four host plants

Host plants	Pre-oviposition period (days ± S.E.)	Oviposition period (days ± S.E.)	Fecundity (eggs per female ± S.E.)	Egg viability (%)
Dolichos lablab	3.50 ± 0.22c (3–5)	45.70 ± 1.86a (35–54)	915.50 ± 23.16a (821–1064)	94.73 ± 0.55a (91.45–97.25)
Vigna unguiculata	3.40 ± 0.22c (3–5)	42.20 ± 1.50ab (39–50)	923.50 ± 22.57a (800–1062)	94.32 ± 0.77a (88.51–96.25)
Cajanus cajan	4.80 ± 0.29b (3–6)	39.00 ± 1.90b (30–49)	755.80 ± 22.42b (643–846)	83.54 ± 1.67b (75.62–91.13)
Renunculus sceleratus	8.40 ± 0.31a (7–10)	25.20 ± 1.53c (18–32)	337.30 ± 16.66c (229–440)	71.97 ± 1.86c (64.03–79.48)
F value	79.09*	165.50*	66.42*	27.54*

Different letters denote significance within the distribution. Range of each reproductive parameter has been provided in parentheses
*$P < 0.0001$; df = 3, 36; Tukey's range = 3.80880

affected, when the same aphid infested on different host plants was provided. Rank order of suitable host plants were *D. lablab* = *V. unguiculata* > *C. cajan* > *R. sceleratus.* The pre-ovipositional period of *H. variegata* was significantly ($F = 79.09$; $P < 0.0001$; df = 3, 36) shortened, when the aphid raised on *D. lablab* and *V. unguiculata* and delayed on *R. sceleratus*. Lanzoni et al. (2004) reported very short pre-ovipositional period of 2.1 ± 0.2 days, when this ladybird was fed on the aphid, *Myzus persicae* (Sulzer), that reared on *Pisum sativum* L. Farhadi et al. (2011) also found such short period (3.5 ± 0.16 days), when the female was fed on *Aphis fabae* Scopoli, reared on broad bean, *Faba vulgaris* L. Jafari (2011) recorded 6.20 ± 0.13 days for pre-ovipositional period of *H. variegata*, when fed on *A. fabae* infesting common bean, *Phaseolus vulgaris* L. Our findings closely agree with the data of Farhadi et al. (2011), due to the similarity of host plants.

Ovipositional period was significantly ($F = 165.50$; $P < 0.0001$; df = 3, 36) prolonged, when *H. variegata* was provided by the aphid infested on *D. lablab* and *V. unguiculata*, and shortened in the case of *R. sceleratus*. Lanzoni et al. (2004) found a shorter ovipositional period of 32.2 ± 3.2 days, while Jafari (2011) reported 43 ± 0.21 days. This period ranged from 24.37 ± 0.28 to 36.68 ± 0.19 days, when *H. variegata* was fed on *S. avenae*, infesting four host plants (Moghaddam et al. 2016). The ovipositional period extended to 45.70 ± 1.86 days, when the predator was fed on aphids reared on *D. lablab*, which was much longer than those periods reported earlier. Seemingly, nutrients from these host plants support oviposition for a longer time.

The fecundity of *H. variegata* was significantly ($F = 66.42$; $P < 0.0001$; df = 3, 36) higher when the host plants were *V. unguiculata* (923.50 ± 22.57 eggs) and *D. lablab* (915.50 ± 23.16 eggs), while it was minimal on *R. sceleratus* (337.30 ± 16.66 eggs). Previous studies recorded a fecundity ranging from 362.47 to 587.31 eggs on *S. avenae* (Moghaddam et al. 2016) and 599.29 to 667.12 eggs on *A. gossypii* (Wu et al. 2010), reared on multiple hosts. Comparing the fecundities, *A. craccivora* infesting *V. unguiculata* or *D. lablab* was recommended for the augmentative growth of the predator. Obtained data are in close agreement with the other reports on fecundity, i.e., 841.7 eggs on *M. persicae* (Lanzoni et al. 2004) and 943.90 eggs on *A. fabae* (Jafari 2011). Ladybirds fed on high-quality aphid diets had more ovarioles than those raised on inferior diets (Hodek et al. 2012). Egg viability in *H. variegata* was significantly ($F = 27.54$; $P < 0.0001$; df 3, 36) higher when the aphid was reared on *D. lablab* (94.73%) and *V. unguiculata* (94.32%) and lower in the case of *R. sceleratus.* The egg viability ranged from 72.11 to 86.5% on *A. gossypii* (Lanzoni et al. 2004).

A similar effect of host plants was noticed on the demographic parameters of *H. variegata* (Table 2). Significantly, the highest R_o values for *H. variegata* were estimated on *A. craccivora*, reared on *D. lablab* (457.93 ± 11.59 females/female) and *V. unguiculata* (450.21 ± 11.00 females/female), while the lowest was on *R. sceleratus* (82.16 ± 4.06 females/female). The same values ranged from 134.30 to 291.14 on *S. avenae* (Moghaddam et al. 2016) and from 153.98 to 199.03 on *A. gossypii* (Wu et al. 2010), when reared on multiple hosts, and only 52.75 on *M. persicae* (Lanzoni et al. 2004). Insignificant difference was found in r_m and λ values of *H. variegata* on *A. craccivora*, reared on multiple hosts. This was largely due to the short longevity of the predator on the host, *R. sceleratus*. This was further evident from the shortest generation time of the predator on *R. sceleratus* than on the other three hosts (Table 2). The r_m values of *H. variegata* ranged from 0.162 to 0.181 on *S. avenae* (Moghaddam et al. 2016) and from 0.1576 to 0.1681 on *A. gossypii* (Wu et al. 2010), when reared on multiple hosts, and 0.114 on *M. persicae* (Lanzoni et al. 2004). The present findings, regarding the intrinsic rate of increase, are in close agreement with those of Moghaddam et al. (2016) on aphids reared on multiple hosts. Previous studies had λ values of *H. variegata* that ranged from 1.176 to 1.199 on *S. avenae* (Moghaddam et al. 2016) and from 1.1708 to 1.1836 on *A. gossypii* (Wu

Table 2 Demographic attributes of *H. variegata* when fed on aphid, *A. craccivora*, infested on four host plants

Host plant	Demographic parameters				
	R_o	r_m	λ	T_c	D.T.
Dolichos lablab	457.93 ± 11.59a (431.72–484.15)	0.18 ± 0.0031a (0.1771–.1910)	1.20 ± 0.0037a (1.1938–1.2105)	33.27 ± 0.5259b (32.081–34.461)	3.76 ± 0.06c (3.6216–3.9060)
Vigna unguiculata	450.21 ± 11.00a (425.31–475.09)	0.19 ± 0.0035a (0.1778–.1937)	1.20 ± 0.0042a (1.1945–1.2136)	32.87 ± 0.7058b (31.288–34.482)	3.72 ± 0.07c (3.572–3.8895)
Cajanus cajan	261.20 ± 7.75b (243.68–278.73)	0.15 ± 0.0031a (0.1469–.1538)	1.16 ± 0.0018a (1.1583–1.1663)	37.00 ± 0.4450a (35.997–38.011)	4.60 ± 0.05a (4.5041–4.7128)
Renunculus sceleratus	82.16 ± 4.06c (72.99–91.35)	0.16 ± 0.0036a (0.1491–.1654)	1.17 ± 0.0042a (1.1607–1.1798)	28.00 ± 0.7849c (26.256–29.807)	4.40 ± 0.10b (4.1791–4.6333)

Data are Mean ± S.E. estimated from Jack-knife Method that includes Student t-test for pair-wise group comparison. Different letters denote significance at $P<0.01$ within the distribution estimated by Student's t-test. Data in parentheses are lower and upper range of the demographic parameter estimated from Jack-knife Method

R_o net reproductive rate (number of female population produce from one mother), r_m the intrinsic rate of increase for the population per individual, λ finite rates of increase for the population per individual, T_c generation time (time need to complete one generation), *D.T.* population doubling times (time needed to double the population)

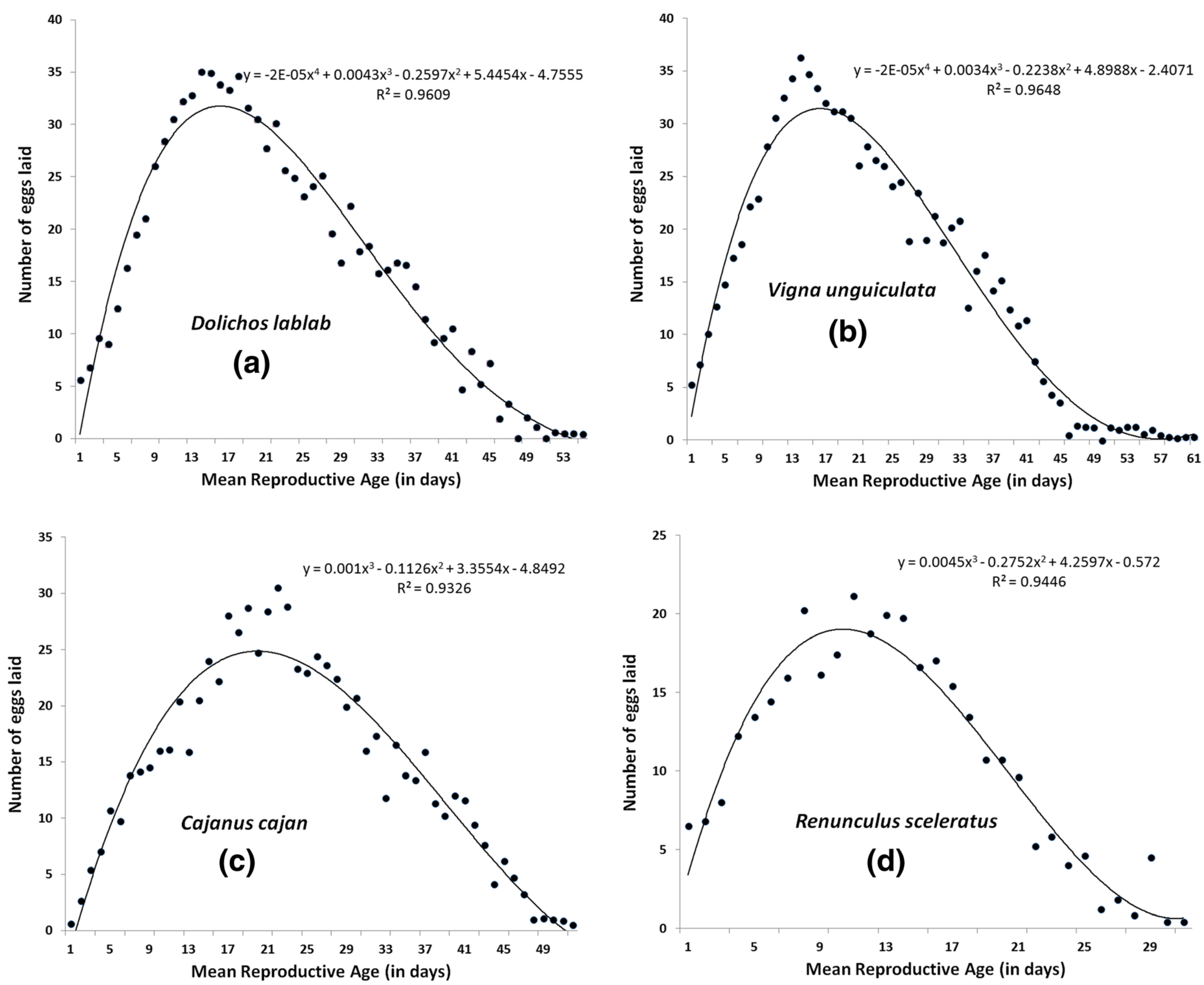

Fig. 1 Age-specific fecundity in terms of relationship between the number of eggs laid and the mean reproductive age of a ladybird, *H. variegata*, when fed on *A. craccivora* infesting **a** *Dolichos lablab*, **b** *Vigna unguiculata*, **c** *Cajanus cajan*, and **d** *Ranunculus sceleratus*

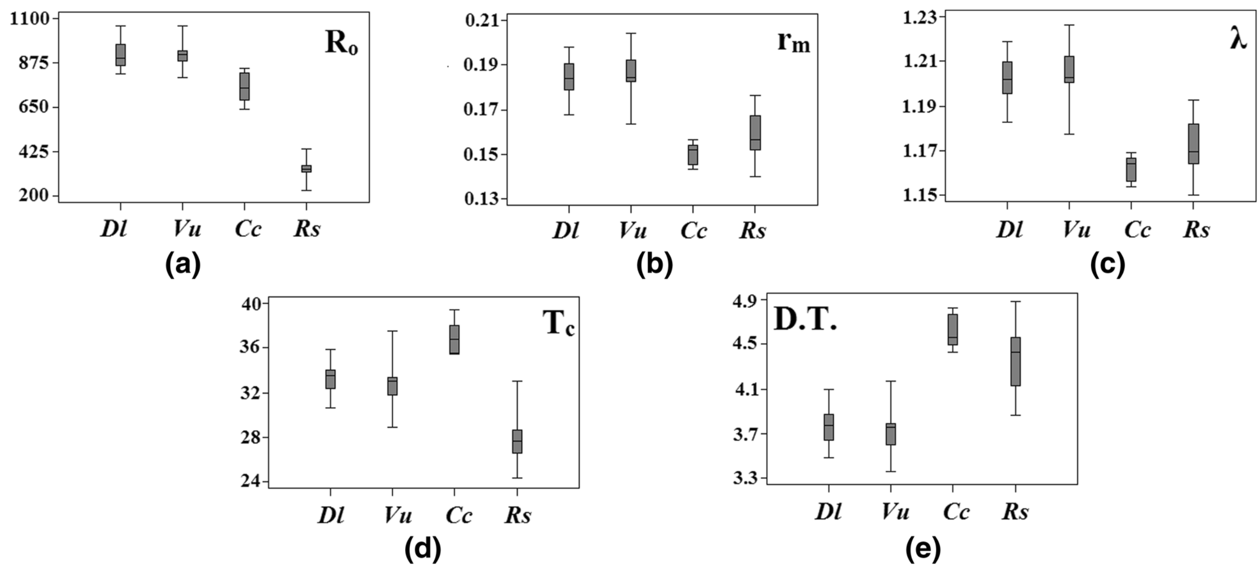

Fig. 2 a–e The box and whisker plots comprising of pseudo-values of the demographic parameters (R_o, r_m, λ, T_c, and D.T.) of *H. variegata*, when fed on *A. craccivora* reared on four host plants, viz. *D. lablab* (Dl), *V. unguiculata* (Vu), *C. cajan* (Cc), and *R. sceleratus* (Rs)

et al. 2010), when reared on multiple hosts, and 0.114 on *M. persicae* (Lanzoni et al. 2004). Doubling times (D.T.) of *H. variegata* on *A. craccivora*, reared on multiple hosts, varied significantly with the shortest D.T. on *D. lablab* and *V. unguiculata* and the longest on *R. sceleratus* (Table 1). The time taken to double the population of the predator ranged from 4.11 to 4.39 on *S. avenae* (Moghaddam et al. 2016), which was greater than that shown by obtained data. Thus, comparing the demographic analysis results, both *D. lablab* and *V. unguiculata* were the suitable hosts to rear *A. craccivora* for the augmentative rearing of *H. variegata*.

Age-specific fecundity of female *H. variegata* was triangular in function, i.e., there was an initial increase in ovipositional rate with age, reached a peak and then gradually declined (Fig. 1a–d). The maximum peak was recorded, when *A. craccivora* infested on *D. lablab* and *V. unguiculata* was provided. The fourth-order polynomial regression analysis gave the best fit to the age-specific curve of *H. variegata*, when fed on *A. craccivora* infested on *D. lablab* and *V. unguiculata*. However, the third-order polynomial regression analysis fits best to the age-specific curve, when host plants *C. cajan* and *R. sceleratus* were provided (Fig. 2).

Conclusion

It could be concluded that *A. craccivora* infesting *D. lablab* and *V. unguiculata* could be used as the best option for mass multiplication of *H. variegata*. In addition, its augmentative releases as a biocontrol agent against *A. craccivora*, infesting the plants above, are recommended as it can easily multiply faster than on other hosts.

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Availability of data and materials
Not applicable. Authors' cannot submit the raw data as they will further be used to developing suitable rearing techniques.

Authors' contributions
AP has planned the outline of this research and designed the methodology. He drafted the manuscript and did certain portion of statistical analysis. PA assisted in maintaining the laboratory culture and performing the experiments. HB helped in further drafting the manuscript and performing certain statistical analysis. All the authors read and approved the final manuscript.

Ethics approval and consent to participate
Not applicable

Consent for publication
Not applicable

Competing interests
The authors declare that they have no competing interests.

Publisher's Note

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