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PARASITIZING POTENTIAL OF *Bracon hebetor* SAY AGAINST DIFFERENT LARVAL INSTARS OF *Earias vittella* (FABRICIUS) UNDER LABORATORY CONDITIONS

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ABSTRACT

The okra shoot and fruit borer, *Earias vittella* (Fabricius) (Lepidoptera: Nolidae), is an economically important pest of okra and other malvaceous crops. Its concealed feeding behaviour poses challenges for effective pest management, highlighting the need for alternative approaches, including biological control. The present study investigated the parasitizing potential of *Bracon hebetor* Say against different larval instars of *E. vittella* under laboratory conditions, with particular emphasis on the effects of host developmental stage and parasitoid-to-host ratio on parasitization. Significant variation in parasitization was observed across the tested parasitoid-to-host ratios and larval instars. The highest parasitization was recorded at a parasitoid-to-host ratio of 5:10, reaching 94.00% and 96.00% against third- and fourth-instar larvae, respectively. At the same ratio, parasitization of second- and fifth-instar larvae was 16.00% and 74.00%, respectively. The 4:10 ratio was the second most effective treatment, resulting in parasitization rates of 80.00%, 84.00% and 68.00% against third-, fourth-, and fifth-instar larvae, respectively, while only 6.00% parasitization was recorded against second-instar larvae. Lower parasitoid densities generally resulted in reduced parasitization. These findings indicate that parasitization by *B. hebetor* is strongly influenced by both parasitoid density and host developmental stage, with fourth-instar larvae exhibiting the highest susceptibility under the conditions tested. The results provide a foundation for further investigations into the potential use of *B. hebetor* in augmentative biological control and integrated pest management programmes targeting *E. vittella* in okra.

Key words : *Bracon hebetor*, *Earias vittella*, Larval instars, Parasitoid-to-host ratio, Parasitization, Biological control, Okra.

Introduction

Okra [*Abelmoschus esculentus* (L.) Moench] is an important vegetable crop widely cultivated in tropical and subtropical regions and valued for its nutritional and economic importance. The crop is attacked by several insect pests, among which the shoot and fruit borer, *Earias vittella* (Fabricius) (Lepidoptera: Nolidae), is one of the major constraints to profitable okra production. The larvae bore into tender shoots, flower buds and developing fruits, resulting in shoot wilting, fruit damage and deterioration in the marketable quality of fruits.

The management of *E. vittella* has traditionally relied heavily on synthetic insecticides. However, repeated and intensive use of insecticides can result in the development of insecticide resistance and may adversely affect beneficial arthropods and other components of the agroecosystem. In the case of *E. vittella*, the concealed feeding habit of larvae inside shoots and fruits makes chemical suppression more difficult once larvae have entered plant tissues. Consequently, biological control may provide an important complementary approach for reducing pest populations before or during vulnerable larval stages. Previous investigations on *E. vittella* have documented the potential of parasitoids and other biological approaches as components of integrated pest management (Reddy and Jalali, 2015; Das *et al*., 2017).

However, effective utilization of a parasitoid requires detailed information on the susceptibility and suitability of different developmental stages of the target pest.

Among parasitoids with potential for biological control of lepidopteran pests, *Bracon hebetor* Say (Hymenoptera: Braconidae) is a well-studied gregarious larval ectoparasitoid. Female *B. hebetor* typically paralyze host larvae by injecting venom through the ovipositor and subsequently deposit eggs on or near the surface of the immobilized host (Ghimire and Phillips, 2010). Its broad host range, high parasitization potential, reproductive capacity, and ability to rapidly suppress host populations have established *Bracon hebetor* as a promising candidate for augmentative biological control of economically important lepidopteran pests (Saadat *et al*., 2016). Kaur *et al.* (2015) also demonstrated the importance of interactions between *B. hebetor*, its lepidopteran host and the host plant, indicating that parasitoid performance can be influenced by the biological environment in which host–parasitoid interactions occur.

The effectiveness of *B. hebetor* as a biological control agent is influenced by several biological and ecological factors, including host species, host developmental stage, host density, parasitoid density, age and mating status of the parasitoid, and environmental conditions. Warsi *et al.* (2018) reported that the last larval instar of *Plodia interpunctella* was particularly important for development of *Habrobracon hebetor* and demonstrated that host physiological condition could substantially affect parasitoid reproductive performance. Raghavi *et al.* (2019), while studying host-recognition and acceptance behaviour of *B. hebetor*, demonstrated that host characteristics and larval stage influence parasitoid behaviour and host acceptance. Abou El-Ela *et al*. (2021) demonstrated that host density affected oviposition and reproductive performance of *B. hebetor*.

In particular, information on the response of *B. hebetor* to different larval ages of *E. vittella* is important because the parasitoid does not necessarily exhibit the same level of parasitization against all host instars. Differences in larval size, mobility, nutritional condition, defensive behaviour and suitability for parasitoid development may influence successful parasitization. Identification of the most suitable larval age can consequently improve laboratory mass multiplication and facilitate the development of effective release strategies.

Therefore, the present investigation was undertaken to determine the parasitizing potential of *Bracon hebetor* Say against different larval ages of *Earias vittella* (Fabricius) under laboratory conditions. The study was aimed at assessing the susceptibility of different larval stages of *E. vittella* to *B. hebetor* and determining the host stage that is most suitable for parasitization. The findings are expected to provide useful information for understanding the host–parasitoid interaction and for developing *B. hebetor*-based biological-control strategies for incorporation into integrated pest management of *E. vittella* in okra.

Materials and Methods

The experiment was conducted from October to December 2022 in the Biocontrol Laboratory, Department of Entomology, College of Agriculture, Indira Gandhi Krishi Vishwavidyalaya (IGKV), Raipur, Chhattisgarh, India. Laboratory cultures of *Bracon hebetor* Say and *Earias vittella* (Fabricius), as well as the parasitization experiments, were maintained under controlled environmental conditions of 25 ± 2°C temperature, 70 ± 10 % relative humidity (RH), and a 14:10 h light:dark (L:D) photoperiod.

Rearing of Parasitoid : For culture of *Bracon hebetor*, bracocards containing pupa of *B. hebetor* in varying number was used and maintained under above mentioned laboratory conditions until adult emergence. Newly emerged male and female parasitoids were allowed to mate and 48-h-old mated females were selected for the parasitization experiment.

Rearing of Host : The host, *Earias vittella* (Fabricius) was maintained on fresh okra fruit under laboratory conditions. Larvae were supplied with freshly cut pieces of okra fruit as food and were observed daily for moulting. Different larval instars were identified based on the occurrence of cast head capsules and exuviae. Healthy and active larvae of the second, third, fourth and fifth instars were selected for the parasitization experiment. Rearing *E. vittella* on fresh okra fruit is consistent with previously described laboratory-rearing procedures for this species (Pardeshi *et al.*, 2019).

Experimental Procedure

The experiment was designed to examine the effects of increasing parasitoid to host ratio within a confined space and also, to determine the most suitable age of the host larvae for maximum parasitization. The second-, third-, fourth- and fifth-instar larvae of *E. vittella* were exposed separately to five parasitoid-to-host ratios, *viz.* 1:10, 2:10, 3:10, 4:10 and 5:10. In each treatment, 10 host larvae were exposed to 1, 2, 3, 4 or 5 mated female parasitoids, respectively.

The parasitization assay was conducted using the sandwich method. The required number of mated female

B. hebetor was introduced into a plastic jar measuring 18 × 12 cm with the help of an aspirator. The experimental container was covered with muslin cloth to permit ventilation and prevent parasitoid escape. Fresh, healthy *E. vittella* larvae of the required instar were placed on a layer of muslin cloth and covered with a second layer of muslin cloth. The two layers were secured with a rubber band to restrict excessive movement of the host larvae while allowing access to the parasitoids. This arrangement facilitated close parasitoid–host contact during the exposure period.

The parasitoids were allowed to interact with the host larvae for 24 h. After completion of the exposure period, the host larvae were carefully removed and transferred individually to labelled Petri plates. Individual isolation was maintained to permit accurate assessment of parasitization and subsequent parasitoid development.

The number of parasitized host larvae was recorded for each treatment and replication. Parasitization was confirmed by subsequent observation of *B. hebetor* oviposition/development rather than host paralysis alone, because female *B. hebetor* may paralyze hosts without necessarily ovipositing on them. The host larvae were examined periodically under a digital stereo-trinocular microscope, and the number of *B. hebetor* eggs and developing parasitoid larvae associated with parasitized hosts was recorded.

Using the below formula, % parasitization was calculated:

$$\% \text{ Parasitization} = \frac{\text{Number of host larvae parasitized (containing eggs, grubs of } B.hebetor)}{\text{Number of host larvae exposed for parasitization}} \times 100$$

Statistical analysis : Each experiment was repeated 5 times for a particular instar. The data obtained from the laboratory studies were subjected to analysis of variance (ANOVA) under a completely randomized design (CRD). Percentage parasitization data were subjected to angular transformation using the arcsine square-root transformation before statistical analysis. The transformed data were used for analysis, whereas the original percentage values were used for presentation and biological interpretation. Treatment means were compared at the 5% level of significance following a significant F-test.

Results and Discussion

The results presented in Table 1 revealed significant differences in the parasitization of *Earias vittella* by *Bracon hebetor* across the five parasitoid-to-host ratios. Among the treatments evaluated, the 5:10 ratio was the most effective, recording parasitization rates of 16.00%, 94.00%, 96.00%, and 74.00% against second-, third-, fourth-, and fifth-instar larvae, respectively. The 4:10 ratio ranked second in effectiveness, with corresponding parasitization rates of 6.00%, 80.00%, 84.00%, and 68.00%. In contrast, the lowest parasitoid density (1:10) resulted in the minimum parasitization, with rates of 0.00%, 10.00%, 14.00% and 12.00% against second-, third-, fourth- and fifth-instar larvae, respectively. These findings indicate that parasitization efficiency was influenced by both parasitoid density and host developmental stage, with the 5:10 ratio consistently producing the highest parasitization across the tested larval instars.

Among the four larval instars, fourth-instar larvae exhibited the highest susceptibility to *B. hebetor*, followed by third- and fifth-instar larvae, whereas second-instar larvae were the least susceptible. The greater suitability of fourth-instar larvae was also reflected in the

Table 1 : Parasitizing potential of *Bracon hebetor* Say against larvae of *Earias vittella.*

Parasitoid : Host	Mean percentage of larval parasitization			
	II Instar	III Instar	IV Instar	V Instar
1 : 10	0.00 (00)	10.00 (18.42)	14.00 (21.67)	12.00 (20.05)
2 : 10	2.00 (3.68)	12.00 (20.05)	30.00 (33.07)	26.00 (30.54)
3 : 10	4.00 (7.37)	52.00 (46.13)	54.00 (47.28)	44.00 (41.52)
4 : 10	6.00 (11.05)	80.00 (64.70)	84.00 (66.95)	68.00 (55.68)
5 : 10	16.00 (23.30)	94.00 (78.92)	96.00 (82.61)	74.00 (59.42)
SE (m) ±	3.41	2.71	2.68	1.75
CD	10.14	8.05	7.69	5.22

Figure in the parenthesis are angular transformation.

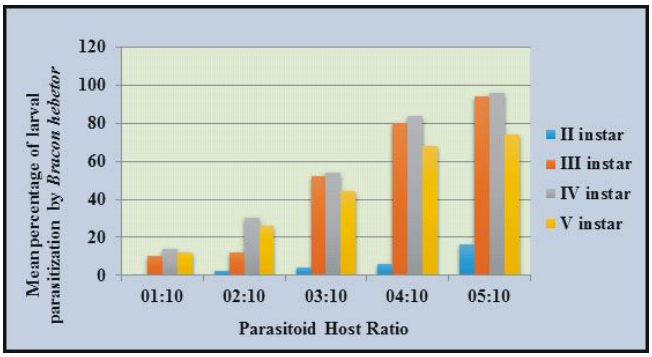


Fig. 1 : Parasitizing potential of *Bracon hebetor* Say against larvae of *Earias vittella.*

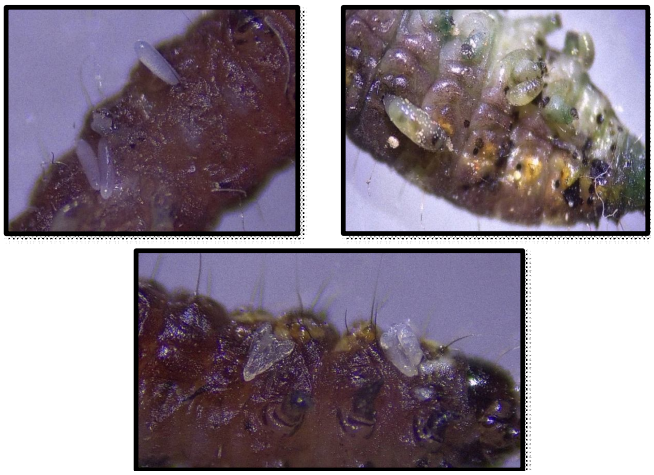


Plate 1 : Eggs and grubs of *Bracon hebetor* on parasitized larvae of *E. vittella.*

comparatively higher occurrence of parasitoid eggs and grubs on parasitized hosts. This pattern suggests that host developmental stage may influence parasitoid acceptance and subsequent progeny development. However, the presence of eggs and grubs should be interpreted as evidence of oviposition and immature parasitoid development, respectively, rather than as a direct measure of successful parasitoid emergence.

The present findings are consistent with those of Thanavendan and Jeyarani (2012), who reported significantly higher parasitization (97.50%) by *B. hebetor* at a 5:10 parasitoid-to-host ratio against fourth-instar larvae of *E. vittella*. Similarly, Hubballi *et al.* (2023) reported that the 5:10 ratio resulted in the highest parasitization (100%) of second-, third-, and fourth-instar larvae of *E. vittella*, followed by the 4:10 (96.83%), 3:10 (85.08%), and 2:10 (61.71%) ratios, while the lowest parasitization (52.71%) was recorded at the 1:10 ratio.

Overall, the findings demonstrate that a parasitoid-to-host ratio of 5:10 and the fourth-instar stage of *E. vittella* were the most favourable combination for parasitization under the conditions of the present study. These results highlight the importance of optimizing both parasitoid density and host developmental stage when evaluating *B. hebetor* for potential use in augmentative biological control programmes against *E. vittella*.

Conclusion

The present study demonstrated that the parasitization efficiency of *Bracon hebetor* Say against *Earias vittella* (Fabricius) was influenced by both parasitoid-to-host ratio and host developmental stage under laboratory conditions. Among the five ratios evaluated, the 5:10 parasitoid-to-host ratio was the most effective, achieving maximum parasitization of 96.00% and 94.00% against fourth- and third-instar larvae, respectively. In contrast, second-instar larvae exhibited comparatively low susceptibility, with parasitization rates of 16.00% and 6.00% at the 5:10 and 4:10 ratios, respectively. The consistently higher parasitization of fourth-instar larvae indicates that host developmental stage is an important consideration when assessing the biological control potential of this parasitoid. Overall, the findings establish the 5:10 parasitoid-to-host ratio and fourth-instar larvae as the most favourable treatment combination among those evaluated. These results provide a basis for further investigations into the efficacy of *B. hebetor* under semi-field and field conditions and its potential integration into augmentative biological control and integrated pest management programmes for the sustainable management of *E. vittella* in okra. Further studies should assess parasitoid establishment, progeny production, host suppression, and field-level effectiveness before practical recommendations are made.

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