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Aswini R
Department of Agricultural Entomology, TNAU, Coimbatore, Tamil Nadu, India

S Jeyarajan Nelson
Department of Agricultural Entomology, TNAU, Coimbatore, Tamil Nadu, India

N Chitra
Department of Agricultural Entomology, TNAU, Coimbatore, Tamil Nadu, India

M Alagar
Department of Agricultural Entomology, TNAU, Coimbatore, Tamil Nadu, India

S Mohan Kumar
Department of Plant Biotechnology, TNAU, Coimbatore, Tamil Nadu, India

Corresponding Author:
S Jeyarajan Nelson
Department of Agricultural Entomology, TNAU, Coimbatore, Tamil Nadu, India

Fecundity of *Bracon hebetor* Say (Braconidae: Hymenoptera) on five lepidopterous hosts larvae

Aswini R, S Jeyarajan Nelson, N Chitra, M Alagar and S Mohan Kumar

Abstract
Bracon hebetor Say is a gregarious Lepidopteran larval ectoparasitoid that is used in the Integrated Pest Management programme as a biocontrol agent. Fecundity of *B. hebetor* on five lepidopteran larvae *viz.*, *Corcyra cephalonica* (Stainton), *Galleria mellonella* (Linnaeus), *Spodoptera frugiperda* (J.E. Smith), *Spodoptera litura* (Fabricius) and *Opisina arenosella* (Walker) was assessed. The results of this study showed that fecundity of *B. hebetor* was higher in 3rd instar larvae of *G. mellonella* followed by 5th instar larvae of *S. frugiperda, C. cephalonica*, *S. litura* and *O. arenosella.*

Keywords: *Bracon hebetor*, *Corcyra cephalonica*, *Galleria mellonella*, *Spodoptera frugiperda*, *Spodoptera litura*, *Opisina arenosella*

Introduction

Bracon hebetor Say is one of the most important biocontrol agents and cosmopolitan ectoparasitoid and it is reared on several Lepidopteran larvae (Dabhi *et al.*, 2012) [3]. *B. hebetor* is being used to control pests both in the field and in storage (Landge *et al.*, 2009) [11] due to its high reproductive rate and short generation time. In the current study, the aim is to identify the preferred host of *B. hebetor* for oviposition and evolve strategies for the mass production.

Materials and Methods

Experiments were conducted in the Biocontrol laboratory, Department of Agricultural Entomology, Tamil Nadu Agricultural University, Coimbatore, India. The five different hosts insects were mass cultured and used for the experiments. Mass culturing of *B. hebetor* was done continuously using *C. cephalonica* and used for conducting various experiments.

Mass culturing of different host insects

Corcyra cephalonica (Pyralidae: Lepidoptera) larvae were reared on cumbu medium, which consists of 2.5 kg of broken cumbu grains, 100 g of broken groundnut kernels, 5 g of dry yeast (Bakers), 5 g of wettable sulphur, and 10 ml of 0.01-0.05 percent streptomycin sulphate as per the method given by Kumar *et al.* (2017) [10].

An artificial diet consisting of wheat flour 200 g, maize flour 200 g, oats 200 g, Milk powder 200 g, yeast powder 20 g, honey 200 g, glycerine 250 ml, and vitamin E 2 capsules, streptomycin sulfate 0.5 g was used for rearing *Galleria mellonella* (Pyralidae: Lepidoptera) larvae as per the method suggested by Meyling (2007) [13].

Spodoptera frugiperda (Noctuidae: Lepidoptera) larvae were reared on an artificial diet which has three parts, *viz.*, Part A (Lab Lab (88 g), wheat germ (25.2 g), yeast (22.7 g), ascorbic acid (2.5 g), sorbic acid (1.3 g), methyl para hydroxyl benzoate (2 g), ethanol (10 ml), and distilled water (230 ml)). Part B (Agar agar (12.6 g), distilled water (250 ml)), and Part C (vitamin E (2 ml), formaldehyde (2 ml), and tetracycline (2.5 g)). Part B was added to Part A and Part C was added to the mixture of Part A and B following the method developed by Ashok *et al.* (2021) [2].

An artificial diet consisting of wheat germ (26.0 g), kidney bean flour (51.3 g), chickpea flour (56.0 g), yeast powder (31.6 g), L-ascorbic acid (3.2 g), two multivitamin multimineral capsules, one vitamin E capsule, castor oil (1 ml), methyl-p-hydroxybenzoate (1.8 g), sorbic acid (1.3 g), streptomycin sulphate (0.25 g), formaldehyde solution (2 ml), agar–agar (16.4 g), and 820 ml distilled water were used for mass rearing of the tobacco caterpillar *Spodoptera litura* (Noctuidae: Lepidoptera) from the neonate to the adult stage as per the protocol developed by Gupta *et al*. (2005) [6].

The black-headed caterpillar, *Opisina arenosella* (Xyloryctidae: Lepidoptera) larva was reared on an artificial diet 11 ingredients *viz.*, chickpea (50 g), yeast (15 g), ascorbic acid (1.5 g), sorbic acid (0.5 g), methyl-para hydroxybenzoate (1.0 g), multivitamin (1 capsule), vitamin E capsule (1 capsule), streptomycin sulphate (0.1 g), formaldehyde (1 ml), distilled water (240 ml) and agar-agar (5 g) (Murthy *et al.*, 2002) [14].

Mass culturing of *B. hebetor* (Braconidae: Hymenoptera)

A pair of *B. hebetor* adult was confined in a small container covered on both sides with muslin cloth and rubber bands. A cotton swab soaked in a 50 percent honey water solution was stuck to the side of the container to serve as food for *B. hebetor*. Larvae of *C. cephalonica* were placed between two muslin cloths and fastened with a pair of rubber bands. Females of *B. hebetor* were attracted to the host larvae, probed through the muslin, and paralysed the larvae, on each of which they deposited their eggs. After 24 hours, the parasitized larvae were removed and kept in Petri plates until the adult emergence. The parasitoid larvae were renewed daily and kept in separate Petri dishes until emergence of new adult parasitoids. The cycle was repeated to get a continuous supply of parasitoids for conducting laboratory experiments (Fig 3).

Fecundity of *B. hebetor* on different hosts

Fecundity of *B. hebetor* was studied on five different hosts *viz.*, *C. cephalonica*, *G. mellonella*, *S. frugiperda*, *S. litura* and *O. arenosella* under the controlled laboratory environment (70±5% R.H., 25±1 °C) in the Biological Control Laboratory, TNAU, Coimbatore, India. Plastic jars (12cm × 10cm) were used for conducting the experiments. Cotton swab soaked in a 50 percent honey solution was attached inside the jar wall for adult feeding.

A third instar larva of each host species was placed over the muslin cloth on the jar neck and then covered with the help of another muslin cloth piece and a rubber band was used to tighten the cloth pieces at the jar neck to keep the larvae in the same position for parasitization during the 24 hours exposition period. One pair of *B. hebetor* adults were released inside the jar. After 24 hours of parasitism, the parasitized larvae of the respective hosts were removed gently and kept in plastic Petri dishes (8.50 cm × 1.50 cm) for further observations. The larva was replaced by new ones for seven consecutive days at 24 hours interval to study the oviposition of a *B. hebetor*. Ten replications were maintained for each host species and five host species were considered as five treatments. Fecundity of *B. hebetor* was recorded. The same procedure was repeated for fourth and fifth instar larvae of five hosts (Fig 2).

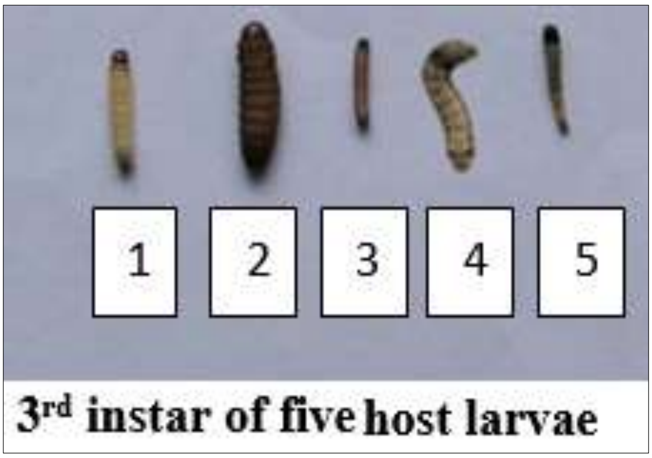
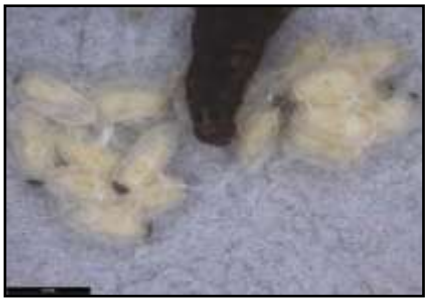


Fig 1: 1. *C. cephalonica*, 2. *G. mellonella*, 3. *S. frugiperda*, 4. *S. litura*, 5. *O. arenosella*

Eggs of *B. hebetor* on *G. mellonella* larva

Grubs of *B. hebetor* on *G. mellonella* larva



Pupae of *B. hebetor* on *G. mellonella* larva

Male and Female of *B. hebetor*

Fig 2: Egg, grub, pupa and adult of *B. hebetor*

Fig 3: Mass production of *Bracon hebetor* by sandwich method

Statistical Analysis

The data on fecundity was subjected to a one way ANOVA after square root transformation of the data. Means were compared using Tukey's test on SAS 9.4.

Results

Table 1: Number of eggs laid on 3rd instar larvae of 5 different hosts by *B. hebetor*

Host insect 3rd instar larvae	Number of eggs laid / female*							
	Age of *B. hebetor*							
	1 day old	2 days old	3 days old	4 days old	5 days old	6 days old	7 days old	Total
C. cephalonica	3.5±0.56b	3.0±0.54c	4.3±0.88bc	12.6±2.12b	3.2±0.59b	7.1±1.68b	5.5±0.87bc	39.2±0.46bc
G. mellonella	19.4±3.0a	17.3±2.61a	19.1±2.20a	24.6±3.30a	20.9±3.85a	34.2±4.62a	13.6±2.06a	149.1±1.03a
S. frugiperda	4.0±0.93a	4.6±1.20bc	6.0±0.97b	5.5±1.30c	5.7±1.33b	5.5±0.87bc	5.0±0.83bc	36.3±0.36c
S. litura	5.1±0.75a	8.2±1.38b	6.2±1.76b	6.5±2.09c	7.8±1.25b	8.6±1.28b	10.1±2.10ab	52.5±0.72b
O. arenosella	3.0±0.60a	2.8±0.70c	1.7±0.33c	1.7±0.33c	2.6±0.60b	1.8±0.29c	2.7±0.50c	16.3±0.18d
P=0.05	0.0004	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001

*Mean of ten replications for seven days
Means in the column followed by the same letters are not significantly different (P = 0.05) by Tukey's test

B. hebetor laid a maximum of 19.4 eggs on third-instar larvae of *G. mellonella* that shows the greater significant, followed by *S. litura* (5.1 eggs), *S. frugiperda* (4.0 eggs), *C. cephalonica* (3.5 eggs), and *O. arenosella* (3.0 eggs) on the first day. The same trend in egg laying was observed in two days and seven days old *B. hebetor*, except in four days old *B. hebetor*. Four days old *B. hebetor* laid maximum of 24.6 nos. of eggs on *G. mellonella* followed by *C. cephalonica* (12.6 eggs), *S. litura* (6.5 eggs), *S. frugiperda* (5.5 eggs) and *O. arenosella* (1.7 eggs). The maximum fecundity was recorded in *G. mellonella* (149.1 eggs) followed by *S. litura* (52.5 eggs), *C. cephalonica* (39.2 eggs), *S. frugiperda* (36.3 eggs) and *O. arenosella* (16.3 eggs) (Table 1 and Fig 4).

Table 2: Number of eggs laid on 4th instar larvae of 5 different hosts by *B. hebetor*

Host insect 4th instar larvae	Number of eggs laid / female*							
	Age of *B. hebetor*							
	1 day old	2 days old	3 days old	4 days old	5 days old	6 days old	7 days old	Total
C. cephalonica	11.8±2.25a	11.1±2.11a	16.4±3.54ab	9.1±1.98ab	16.4±2.57b	10.7±2.13b	11.7±2.61ab	87.2±1.07b
G. mellonella	15.9±5.32a	14.1±1.93a	25.4±4.78a	13.5±4.26a	27.8±3.56a	27.3±4.72a	14.7±3.38a	138.7±1.48a
S. frugiperda	12.9±2.11a	11.0±2.44ab	9.0±1.48bc	6.1±0.97ab	7.6±1.85cd	10.3±2.67b	12.3±2.53a	69.2±0.83bc
S. litura	7.0±1.16ab	6.2±1.33ab	10.6±2.10bc	6.6±1.61ab	10.5±1.85bc	8.8±1.47b	9.2±1.88ab	58.9±0.76c
O. arenosella	2.3±0.42b	3.5±0.87b	3.4±0.65c	2.4±0.43b	2.9±0.53d	2.2±0.39c	3.4±0.69b	20.1±0.24d
P=0.05	0.0024	0.0030	<0.0001	0.0106	<0.0001	<0.0001	0.0112	<0.0001

*Mean of ten replications for seven days
Means in the column followed by the same letters are not significantly different (P = 0.05) by Turkey's test

B. hebetor laid a maximum of 15.9 eggs on third-instar larvae of *G. mellonella* that shows the greater significant, followed by *S. frugiperda* (12.9 eggs), *C. cephalonica* (11.8 eggs), *S. litura* (7.0 eggs), and *O. arenosella* (2.3 eggs) on the first day. The same trend in egg laying was observed in seven days old *B. hebetor*. Two days old *B. hebetor* laid maximum of 14.1 nos. of eggs on *G. mellonella* followed by *C. cephalonica* (11.1 eggs), *S. frugiuperda* (11.0), *S. litura* (6.2 eggs), and *O. arenosella* (3.5 eggs). The same trend in egg laying was observed in 6 days old *B. hebetor*. Three days, four days, and five days old *B. hebetor* were followed a same trend in egg laying was in the order of *G. mellonella*, *C. cephalonica*, *S. litura*, *S. frugiperda*, and *O. arenosella*. The maximum fecundity was recorded in *G. mellonella* (138.7 eggs) followed by *C. cephalonica* (87.2 eggs), *S. frugiperda* (69.2 eggs), *S. litura* (58.9 eggs) and *O. arenosella* (20.1 eggs) (Table 2 and Fig 5).

Table 3: Number of eggs laid on 5th instar larvae of 5 different hosts by *B. hebetor*

Host insect 5th instar larvae	Number of eggs laid / female*							
	Age of *B. hebetor*							
	1 day old	2 days old	3 days old	4 days old	5 days old	6 days old	7 days old	Total
C. cephalonica	12.6±3.90a	12.2±1.83ab	8.6±1.37a	8.0±1.41b	10.6±2.15b	12.1±2.47ab	26.3±4.33a	90.4±1.03b
G. mellonella	15.3±4.36a	13.3±3.41ab	16.4±3.92a	20.6±3.32a	27.1±5.52a	21.5±2.71a	17.7±2.09ab	131.9±1.05a
S. frugiperda	9.0±1.45a	16.8±2.91a	17.3±3.23a	17.5±2.68a	14.7±3.02ab	21.7±2.93a	26.5±3.43a	123.5±1.10a
S. litura	11.1±2.12a	13.4±2.22ab	13.8±2.13a	15.4±3.33ab	13.8±2.37ab	13.0±2.49ab	12.7±2.56b	93.2±1.17b
O. arenosella	7.6±1.81a	5.9±1.19b	8.5±1.36a	6.3±1.00b	6.6±1.11b	9.5±1.69b	12.6±1.81b	57.0±0.55c
P=0.05	0.5814	0.0499	0.1051	0.0004	0.0039	0.0074	0.0037	<0.0001

*Mean of ten replications for seven days
Means in the column followed by the same letters are not significantly different (P = 0.05) by Tukey's test

B. hebetor laid a maximum of 15.3 eggs on third-instar larvae of *G. mellonella* that shows the greater significant, followed by *C. cephalonica* (12.6 eggs), *S. litura* (4.0 eggs), *S. frugiperda* (9.0 eggs), and *O. arenosella* (7.6 eggs) on the first day. Two days old *B. hebetor* laid maximum of 16.8 eggs on *S. frugiperda*, followed by *S. litura* (13.4 eggs), *G. mellonella* (13.3 eggs), *C. cephalonica* (12.2 eggs), and *O. arenosella* (5.9 eggs). Next to *S. frugiperda* (17.3 eggs), *S. litura* recorded as 13.4 eggs in three days old *B. hebetor*. The same trend in egg laying was observed in six days old *B. hebetor*. Four days old *B. hebetor* laid maximum of 20.6 nos. of eggs on *G. mellonella* followed by *S. frugiperda* (17.5 eggs), *S. litura* (15.4 eggs),C. *cephalonica* (8.0 eggs), and *O. arenosella* (6.3 eggs). The maximum fecundity was recorded in *G. mellonella* (131.9 eggs) followed by *S. frugiperda* (123.5 eggs), *S. litura* (93.2 eggs), *C. cephalonica* (90.4 eggs) and O. arenosella (57.0 eggs) (Table 3 and Fig 6).

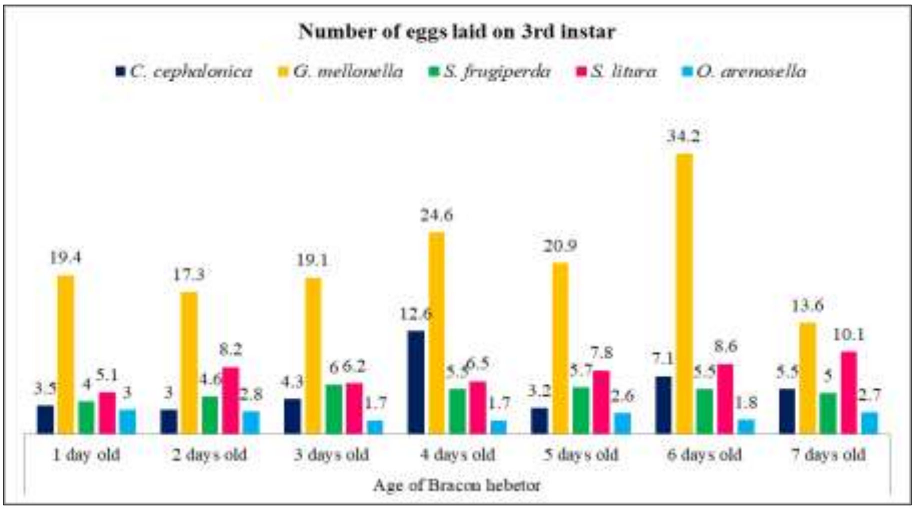


Fig 4: Number of eggs on 3rd instar of five host species

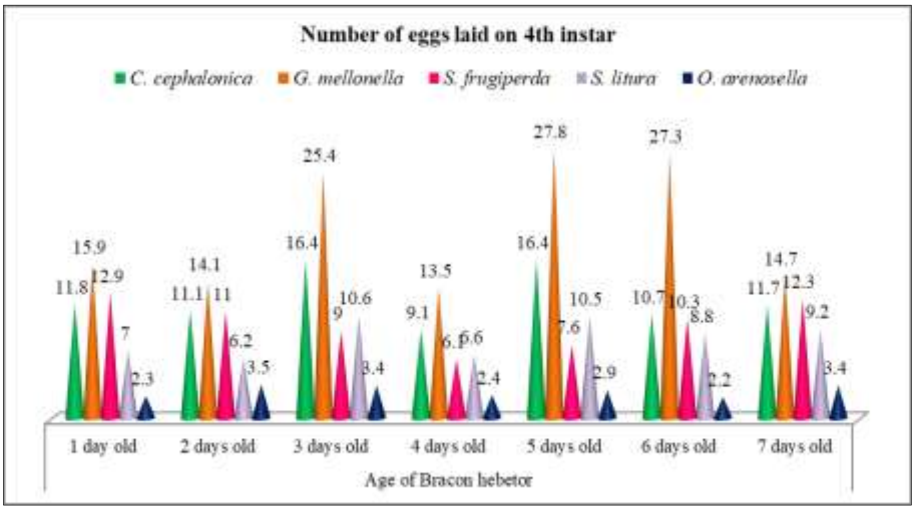


Fig 5: Number of eggs on 4th instar of five host species

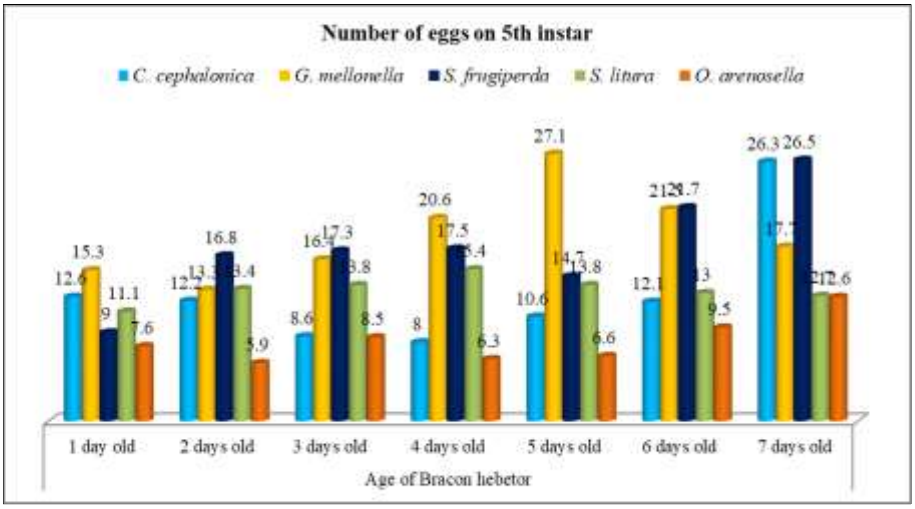


Fig 6: Number of eggs on 5th instar of five host species

Discussion

The oviposition rate of *B. hebetor* was the highest in 3rd instar larvae of *G. mellonella*, followed by 5th instar larvae of *S. frugiperda*, *C. cephalonica*, *S. litura*, and *O. arenosella*. Alam *et al.* (2016) [1] also found that *G. mellonella* was the most preferred host of *B. hebetor* among the several lepidopterous larvae. Ghimire and Phillips (2014) [5] reported that daily fecundity of *B. hebetor* was the highest on *G. mellonella*. Khalil *et al*. (2016) [9] revealed the highest biological activity in terms of paralysis, parasitization, and oviposition in *G. mellonella* larvae when compared to other host species. Muslim *et al.* (2017) [15] found that the paralysis and parasitization potential of *B. hebetor* were found to be greatest on *C. cephalonica*, followed by *G. mellonella*, *E. kuehniella*, and *P. interpunctella*, while they were found to be the lowest on *S. litura* and *Spodoptera littoralis*. In our study, *B. hebetor* laid 149.1 eggs on *G. mellonella* larvae but Imam (2013) [7] and Ghimire and Phillips (2014) [5] reported 343.83 and 808 eggs of *B. hebetor* respectively on *G. mellonella*. Magro and Parra (2001) [12], Jhansi and Babu (2003) [8], Dabhi *et al*. (2012) [3], Kumar *et al*. (2017) [10] reported that *C. cephalonica* was the better host for *B. hebetor* among the other lepidopterous hosts.

Contrary to the findings of Edpunganti and Kattula (2018) [4] who reported no oviposition by *B. hebetor* on S. *litura*, *B. hebetor* laid 93.2 eggs of fifth instar larvae of *S. litura* in our study. Fecundity of *B. hebetor* varied depending on the age of different host larvae. This might be due to the chemical compounds emenated from the host larvae and the cuticular structure.

Conclusion

The third instar larva of *G. mellonella* on six days old *B. hebetor* was found to be the best host for oviposition of *B. hebetor* followed by 5th instar of *S. frugiperda*, *C. cephalonica*, *S. litura*, and the least preferred host was *O. arenosella*. Further studies are needed to assess the role of kairomones on the oviposition behaviour of *B. hebetor*.

References

1. Alam MS, Alam MZ, Alam SN, Miah MRU, Mian MIH and Hossain MM. Host suitability of *Bracon hebetor* Say on different lepidopterous host larvae. Acad. J. Entomol. 2016;9:41-50.
2. Ashok K, Balasubramani V, Kennedy JS, Geethalakshmi V, Jeyakumar P and Sathiah N. Evaluating artificial diets for the fall armyworm, *Spodoptera frugiperda* (JE Smith) (Lepidoptera: Noctuidae) through nutritional indices and an age-stage, two-sex life table approach. African Entomology. 2021;29(2):620-634.
3. Dabhi MR, Korat DM and Vaishnav PR. Comparative biology of *Bracon hebetor* Say on seven lepidopteran hosts. Karnataka Journal of Agricultural Sciences. 2012;24(4).
4. Edpuganti SL and Kattula SY. Comparative biology of *Bracon hebetor* say on *Corcyra cephalonica* stainton, *Helicoverpa armigera* (hub.) and *Spodoptera litura* fabricius under no choice and dual choice conditions. IJCS. 2018;6(2):2264-2266.
5. Ghimire MN and Phillips TW. Oviposition and reproductive performance of *Habrobracon hebetor* (Hymenoptera: Braconidae) on six different pyralid host species. Annals of the Entomological Society of America. 2014;107(4):809-817.
6. Gupta GP, Rani S, Birah A and Raghuraman M. Improved artificial diet for mass rearing of the tobacco caterpillar, *Spodoptera litura* (Lepidoptera: Noctuidae). International Journal of Tropical Insect Science. 2005;25(1):55-58.
7. Imam AI. Life Tables of the Larval Parasitoid; Bracon brevicornis Wesm. (Hymenoptera: Braconidae) on Different Host Larvae. Egyptian Journal of Biological Pest Control. 2013;23(2):189.
8. Jhansi K and Babu PCS. Comparative biology of *Bracon hebetor* Say in two host insects. J Appl. Zool. Res. 2003;14:165-168.
9. Khalil MS, Raza ABM, Afzal M, Aqueel MA, Khalil H and Hance T. Effects of different host species on the life history of Bracon hebetor. Animal Biology. 2016;66(34):403-414.
10. Kumar S, Ghirtlahre RG and Ganguli J. Biology of *Bracon hebetor* Say (Braconidae: Hymenoptera) A Larval Ecto-Parasitoid on Rice meal moth, *Corcyra cephalonica* Stainton (Lepidoptera: Pyralidae). Journal of Plant Development Sciences. 2017;9(6):599-602.
11. Landge SA, Wankhede SM and Gangurde SM. Comparative biology of *Bracon hebetor* Say on *Corcyra cephalonica* Stainton and *Opisina arenosella*

Walker. International Journal of Plant Protection. 2009;2(2):278-280.
12. Magro SR and Parra JRP. Biologia do ectoparasitoide *Bracon hebetor* Say, 1857 (Hymenoptera: Braconidae) em sete especies de lepidopteros. Scientia Agricola. 2001;58:693-698.
13. Meyling NV. Methods for isolation of entomopathogenic fungi from the soil environment-laboratory manual.
14. Murthy SK, Venkatesan T and Jalali SK. Development of *Brachymeria nephantidis* Gahan (Hymenoptera: Chalcididae) on artificial diet reared *Opisina arenosella* Walker. In *Proc.* Symposium on Biological Control of Lepidopterous Pests. 2002;17-18.
15. Muslim M, Ansari MS and Hasan F. Paralysis and parasitisation potential of *Bracon hebetor* on various Lepidopteran hosts. Agricultural Science Digest-A Research Journal. 2017;37(4):327-328.
16. Yu SH, Ryoo MI, Na JH, Choi WI. Effect of host density on egg dispersion and the sex ratio of progeny of Bracon hebetor (Hymenoptera: Braconidae). Journal of Stored Products Research. 2003;39(4):385-93.