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Research Note

Effect of host stage on parasitization and biological characteristics of *Aenasius bambawalei* Hayat (Hymenoptera: Encyrtidae), a parasitoid of *Phenacoccus solenopsis* Tinsley

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ABSTRACT: The effect of host stage on parasitisation and biological characteristics of *Aenasius bambawalei* Hayat (Hymenoptera: Encyrtidae), a parasitoid of *Phenacoccus solenopsis* Tinsley, was studied under laboratory conditions at 26-31°C and 46-73.6 per cent relative humidity. Mealybugs were reared on sprouted potato tubers and different stages of the pest were used in the host preference studies. Third instar and pre-reproductive adult females of the pest were most preferred by *A. bambawalei* for parasitization both under choice and no-choice conditions. First instars were not parasitized. Development of both male and female *A. bambawalei* was faster in large sized hosts (adult females). There was no significant difference in the adult emergence from different host stages. Emergence of adults from different stages ranged from 87.15 to 93.49 per cent. Maximum (73.39%) females emerged from reproductive adult females as compared to other stages of the pest. Parasitoid progeny emerging from second instar hosts consisted mostly (91.23%) of males.

KEY WORDS: *Aenasius bambawalei*, biology, host stage preference, solenopsis mealybug

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Solenopsis mealybug, *Phenacoccus solenopsis* Tinsley has emerged as a serious threat to cotton cultivation in India. The mealybugs suck the cell sap from leaves, twigs, stems, roots and fruiting bodies. Cotton plants infested during vegetative phase exhibit symptoms of distorted and bushy shoots, crinkled and/or twisted and bunchy leaves, and stunted plants that dry completely in severe conditions. In the Indian subcontinent, the pest has caused widespread and serious damage to cotton crop (Abbas *et al*., 2005; Dhawan *et al*., 2007; Jhala *et al*., 2008; Saini *et al*., 2009). *Aenasius bambawalei* Hayat is an important parasitoid of *P. solenopsis* (Ram *et al*., 2009). Surveys in cotton growing areas of Haryana showed 23.7 to 76.6 per cent parasitisation of *P. solenopsis* by *A. bambawalei* on cotton (Ram and Saini, 2010). Studies conducted on the biology of *A. bambawalei* revealed the potential of *A. bambawalei* for the biological control of *P. solenopsis* (Vijaya *et al*., 2011). However, there is little information available on the suitability of different host stages of the mealybug to the parasitoid. Information on the most suitable stage of the host is crucial in the development of a mass rearing programme of a parasitoid. Therefore, keeping this in view, the present study was undertaken to document the effect of different host stages of the pest on parasitisation and important biological characteristics of *A. bambawalei*.

Culture of *Phenacoccus solenopsis* was maintained in the laboratory on sprouted potato tubers. Small to medium sized potato tubers were procured from market. The tubers were washed and dipped in 5–10 ppm gibberellic acid (GA3) solution overnight, air dried and held at room temperature in the dark until sprouting. When the potato sprouts were 2.5–5cm in length, 1–2 field collected gravid females were released in glass jars (20 X 15 cm) with the help of brush at room temperature of 23.1– 33.3°C, and the mealybugs thus multiplied were used for further studies.

Adults of *A. bambawalei* were reared from field collected parasitized (mummies) *P. solenopsis*. The parasitoid adults were reared in the laboratory on mealybug colonies maintained on sprouted potato tubers in glass jars at room temperature (23.2-33.3°C). After 5-6 days of parasitisation, mealybugs transformed mummies. The mummies thus formed were collected and kept separately in glass vials (7.5 x 1cm) provided with honey streak on

a paper strip as a source of food for the emerging adult parasitoids. The freshly emerged adult parasitoids were used in host stage preference studies.

Host stage preference studies

Under free choice conditions, 20 mealybugs each of first, second and third instars and adults (pre-reproductive as well as reproductive females) were released on individual sprouted potato tubers kept in a glass jar. Under no-choice conditions, 50 mealybugs each of first, second, third instars and adults (pre-reproductive as well as reproductive females) were released on individual sprouted potato tubers kept in separate glass jars. Both under choice and no-choice conditions, a paper strip with a streak of honey served as a source of food for *A. bambawalei* adults was also placed in each jar. A pair of freshly emerged adults were released in the jar and removed after 24 hours and reared till all the parasitoid adults emerged from the parasitized mealybugs. The total number of mealybugs parasitized in each host stage was recorded and host stage preference was calculated both under choice and no-choice conditions. There were 15 replications both in choice and no-choice experiments.

In order to see the effect of host stage of the pest on biological characteristics of the parasitoid, observations were also recorded on the parasitoid emergence by observing exit holes in the mummies, proportion of females in the parasitoid progeny and parasitoid development period in all the host stages studied. All the experiments were conducted at room temperature ranging from 26–31°C and 46–73.6 per cent relative humidity.

The data obtained in the above experiments were subjected to the analysis of variance using single factor Completely Randomised Design (CRD).

Effect of host stage on parasitization under free choice and no-choice conditions

Both under free choice and no-choice conditions, *A. bambawalei* parasitized all the stages, except first instar of the host *P. solenopsis* (Table 1). Under choice conditions, number of *P. solenopsis* parasitized was significantly higher in third instar (4.33) and pre-reproductive adult females (3.46) as compared to second instar (2.53) and reproductive adult females (1.73). Under no-choice conditions, maximum number of mealybugs (7.80) was parasitized in the pre-reproductive adult stage and it was on a par with the number of mealybugs parasitized in the third instar stage (6.56). The number of mealybugs parasitized in second instar (5.66), third instar (6.86) and reproductive adult female (5.53) stages was on a par. Thus, it was concluded that third instar and pre-reproductive adult females were the most preferred host stages for parasitization by the parasitoid both under free choice and no-choice conditions. These results are consistent with the studies conducted by Cross and Moore (1992) and Bokonon-Ghanta *et al*. (1995) who found that certain encyrtids prefer to parasitize late instars and adult mealybugs. Jong and van Alphen (1989) reported that *Leptomastix dactylopii* Howard parasitized pre-reproductive and reproductive females of *Planococcus citri* (Risso) while first and second instars were never parasitized. Sagarra and Vincent (1999) also found that first instars of *Maconellicoccus hirsutus* Green were least preferred by *Anagyrus kamali* Moursi as compared to adult females and third instar host stages.

Effect of host stage on parasitoid development period

The development period of the parasitoid decreased as the age of the age of the host advanced from second instar to adult stage (Table 2). The development period of male parasitoids was longest (13.80 days) in second instar hosts while it was shortest in pre-reproductive

Table 1: Parasitization of different mealybug stages by *Aenasius bambawalei* under free choice and no-choice conditions

Host stage	Number of mealybugs parasitized (Mean±S.E.)	
	Free Choice	No-choice
1st Instar	No parasitization	No parasitization
2nd Instar	2.53±0.40	5.66±0.73
3rd Instar	4.33±0.52	6.86±0.69
Pre-reproductive adult females	3.46±0.42	7.80±0.46
Reproductive adult females	1.73±0.44	5.53±0.47
CD (P = 0.05)	1.27	1.72

adult females hosts (12.0 days). Similarly in case of female parasitoids development period was reduced as host grew in size. The development period of female parsitoids was longest (14.86 days) in third instar nymphs and shortest (13.53 days) in reproductive adult female hosts. The development period of the parasitoid in pre-reproductive (13.73 days) and reproductive adult females (13.53 days) was on a par. These results are in accordance with the findings of Nechols and Kikuchi (1985) that showed that development time for *Anagyrus indicus* (Subba Rao) decreased from 25 days to 18 days as the size of *Nipaecoccus vastator* (Maskell), increased from first instar to adult female stage. They also reported that male parasitoids developed faster than the females. Bertschy *et al*. (2000) also made similar observations for the parasitoid, *Aenasius vexans* Kerrich. They reported that in the latter development stages of *Phenacoccus herreni* Cox and Williams, parasitoids developed and emerged faster and in each host stage development time of male parasitoids was shorter than females.

Effect of host stage on parasitoid adult emergence

There was no significant difference in the percentage of parasitoid adults emerging from different stages of the host (Table 2). Parasitoid adult emergence from second instar, third instar, pre-reproductive and reproductive adult female host stages was 92.20, 92.26, 93.49 and 87.15 per cent, respectively. Similar observations were also recorded for parasitoids, *A. indicus* (Nechols and Kikuchi, 1985) and *Anagyrus pseudococci* (Girault) (Chandler *et al*., 1980) where parasitoid emergence from third instar and adult female host stages was consistently higher.

Effect of host stage on the proportion of females in the parasitoid progeny

Data on the proportion of female wasps emerging from the parasitoid progeny reared on different host stages revealed that small or young hosts yielded mainly male parasitoids (Table 2). Low percentage of female wasps (8.77%) emerged when parasitoid was reared from second instar mealybug hosts. Maximum female parasitoids (73.39%) emerged when parasitoid was reared from reproductive adult female hosts followed by pre-reproductive female hosts (63.31%) and third instar hosts (62.65%). The results of the present study are in conformity with those conducted by Nichols and Kikuchi (1985) who found that *N. vastator* parasitized in first and second nymphal stages of the host produced mostly males of *A. indicus* while third nymphal and adult female hosts yielded predominantly female parasitoids. Similar observations were also made by Cadée and van Alphen (1997) in *Leptomastidea abnormis* (Girault), a parasitoid of *P. citri*, who found that more female parasitoids were reared from latter instars (fourth instars and reproducing females) as compared to early instars (mainly third instars and smaller fourth instars). Gulec *et al*. (2007) found that sex-ratio of *A. pseudococci* was male biased in second instars of vine mealybug, *Planococcus ficus* (Signoret) but was equal in third instar hosts.

It was concluded from the present study that *A. bambawalei* parasitized all the nymphal and adult stages of *P. solenopsis* except the first instar. Third instar and pre-reproductive adult female mealybugs were

Table 2: Effect of mealybug stage on development period, adult emergence and proportion of female progeny of *Aenasius bambawalei*

Host stage	Total developmental period (Days)		Adult emergence (%)	Females in emerging progeny (%)
	Male	Female		
1st Instar		No parasitisation		
2nd Instar	13.80±0.32**	*	92.20 (77.08)	8.77 (10.01)
3rd Instar	12.80±0.20	14.86±0.25	92.26 (77.78)	62.65 (55.48)
Pre-reproductive adult females	12.0±0.20	13.73±0.34	93.49 (78.23)	63.31 (56.14)
Reproductive adult females	*	13.53±0.41	87.15 (72.57)	73.39 (68.12)
CD (P = 0.05)	0.63	0.98	(N.S.)	(10.16)

*Observations were not included as sample size was very small

**Mean±SE

Figures in the parenthesis are angular transformed values.

most preferred by *A. bambawaler* for parasitisation. Therefore, in mass rearing programmes of *A. bambawalei*, third instar and pre-reproductive adult female mealybugs must be used to obtain higher parasitism.

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