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Seasonal variation of stylopization on white leafhopper *Cofana spectra* (Distant) (Homoptera: Cicadellidae) by *Halictophagus australensis* Perkins (Strepsiptera: Halictophagidae)

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Abstract

White leafhopper *Cofana spectra* (Distant) is considered as a minor pest of paddy in West Bengal. *C. spectra* is an established host of obligate endoparasitoid Strepsiptera *Halictophagus australensis* Perkins. The sampling for seasonal variation of both parasitized and non-parasitized *C. spectra* was carried out 12 days per month for the period between March 2014 and February 2015. Stylopization of *C. spectra* by *H. australensis* Perkins was recorded from 1 to 15 % with seasonal variation. In post monsoon season higher percentage of parasitic infection was encountered relative to pre monsoon and monsoon seasons. Although percentage of stylopization varied with the relative abundance of *C. spectra*, ecological factors viz. temperature and relative humidity were found to augment it in optimum environmental conditions. Both the environmental factors have a negative correlation with the percentage of parasitization. The effect of parasitization on *C. spectra* by the *H. australensis* may be one of the factors employed for the bio-control of the host population.

Keywords: Strepsiptera, stylopization, *Cofana spectra*, *Halictophagus australensis*, endoparasitoid

1. Introduction

Strepsipteran parasitoid, *Halictophagus australensis* Perkins like other stylops (except Mengenellidae) exhibits extreme sexual dimorphism. Free living males have twisted hind wings, flabellate antennae, raspberry like compound eye, fore wing reduced to pseudohaltere while neotenic females lacking wings, antennae, and eyes are entomophagous endoparasitoid. It parasitizes white leafhopper *Cofana spectra* Distant (WLH) ^[1, 2].

Strepsiptera have an amazingly wide host range. They parasitize seven orders of insects viz. Thysanura, Blattodea, Mantodea, Orthoptera, Hemiptera, Diptera and Hymenoptera ^[3]. They are natural enemies of several insects i.e., particularly of leafhoppers (Cicadellidae) and plant hoppers (Delphacidae) that cause significant injury to paddy plants. Different paddy pests viz., *Nilaparvata lugens* Stål, *Nephotettix nigropictus* (Stål), *Sogatella furcifera* (Horváth) and *Cofana spectra* (Distant) are parasitized by strepsipteran parasitoids. Two species, *Cofana spectra* and *Cofana unimaculata* (Signoret), are considered as minor pests of rice in West Bengal ^[4] of which *C. spectra* (Homoptera: Cicadellidae) are abundant in West Bengal, Northeast and southern states of India ^[1, 2, 5]. They damage plant system by extensive feeding and ovipositing followed by secondary fungal and bacterial infection on the affected area. White leafhopper is also a vector of pathogenic virus like rice yellow mottle virus (RYMV) ^[6]. The RYMV pathogen is included under the Sobemovirus group and it is transmitted mechanically through rice plant injuries and by insect vectors ^[7]. Low infestation of WLHs results in tillering diminution; moderate infestation causes loss of ear production and leaves turn brown and curl; and severe infestation results in the death of paddy plants ^[8].

The aim of this study is to estimate the number of stylopized *C. spectra* with respect to total *C. spectra* collected from the field and the percentage of parasitization by *H. australensis*. Additionally evaluation of the seasonal variations of the percentage of parasitization and role of various environmental factors viz. temperature and relative humidity in parasitization has also been made.

2. Material and Methods

The study area is various rice fields of Burdwan district, which is one of the major rice growing areas in West Bengal, India.

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The collection sites are: Galsi (23.33°N, 87.69°E), Crop Seed Multiplication Centre, University of Burdwan (23.25°N, 87.84°E), Memari (23.20°N, 88.12°E), Shyamsundar (23.09°N, 87.86°E), Raina (23.06° N, 87.90°E), Jamalpur (23.06°N, 87.98°E), Kalna (23.22°N, 88.37°E) and Guskara (23.50°N, 87.75°E).

The host insects were collected using insect sweep nets (mesh size - 250 µm) in the rice agroecosystem and adjoining areas of grass weeds. The sampling was carried out for a total of 12 days per month for the period between March 2014 and February 2015. The hoppers were examined for parasitic infection under CARL ZEISS 2000-C Trinocular microscope in the laboratory. Data on the temperature and relative humidity of the sampling sites were recorded at the time of collection. The monthly data were taken from the meteorological office, Agricultural Farm, Burdwan. The relative abundance of the stylized *C. spectra* by *H. australensis* was compared with the environmental variables especially temperature and humidity.

Statistical analysis

The regression analyses were made following Zar [9] with the application Microsoft® office excel® 2007 [10]. The relationship between temperature and relative humidity with percentage of parasitization was presented through the regression equation using one way ANOVA.

3. Results

The stylization of the hopper hosts could easily be identified by the presence of protruded cephalotheca of parasitoid at their abdominal portion (Figs. 1-2). The infected insects were isolated and preserved in 90% ethanol. Some host insects harbouring strepsipteran parasitoids were sacrificed to observe the various stages of Strepsiptera (Figs. 3-4).



Fig 1: Stylized host white leafhopper *Cofana spectra*. Arrow indicating the location of strepsipteran endoparasitoid.

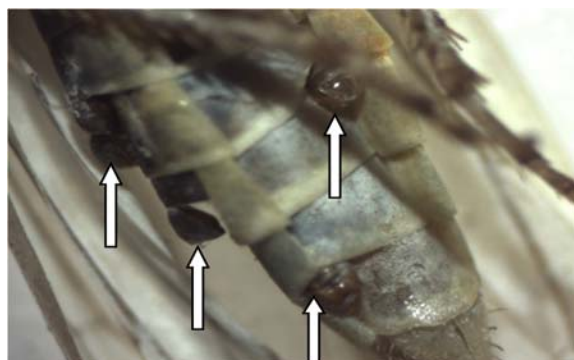


Fig 2: *C. spectra* stylized by four endoparasitoids, *Halictophagus australensis*: an example of superparasitism. Arrows indicating the location of strepsipteran endoparasitoids.

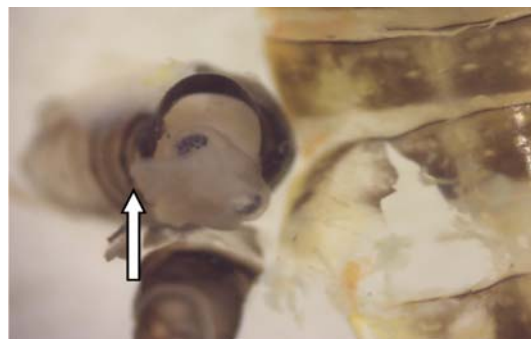


Fig 3: Pharate adult of male *H. australensis* (opening of cap of puparium) after removing from the abdominal portion of host *C. spectra*. Arrow indicating the head of pharate adult.



Fig 4: Adult male *H. australensis*.

During the study, it was observed that the amount of parasitization by *H. australensis* was greater in post monsoon period rather than in premonsoon and monsoon seasons (Fig 5). After observing the monthly data, number of parasitized *C. spectra* was mostly found in the month of December and less in the month of July and September (Fig 6). The percentage of parasitization was found to vary from 1% to 15% in the study period.

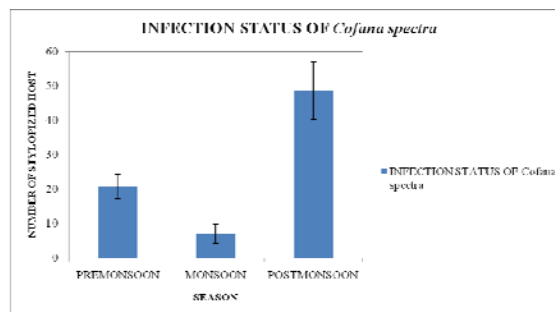


Fig 5: Figure showing the total number of infected *Cofana spectra* collected at different seasons.

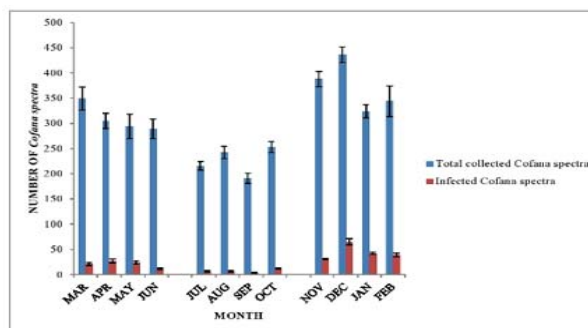


Fig 6: Monthly variation of total collected *Cofana spectra* and stylized ones.

The simple linear regression equation on the relationship between temperature and percentage of parasitization was evaluated.

$$Y = 49.10 - 1.38X$$

One way ANOVA has been performed using Microsoft Excel's statistical macro ANOVA (Table 1).

Table 1. ANOVA result (Temperature against percentage of parasitization).

Source of variance	Df	SS	MS	F-value	Significance F
Regression	1	152.50	152.50	28.27	0.000338
Residual	10	53.93	5.393		
Total	11	206.43			

Table 2. Correlation coefficient of independent variable considered in the regression analysis.

	Coefficient	Standard error	t Stat	P-value
Intercept	49.103	7.922	6.198	0.000101
X variable 1	-1.381	0.259	-5.317	0.000338

So, there was a significant correlation between temperature and percentage of parasitization ($P < 0.05$). From the above analysis it is found that the Coefficient of X (independent variable) is negative (-ve) (Table 2). The above analysis is represented through scatter plot (Fig. 7).

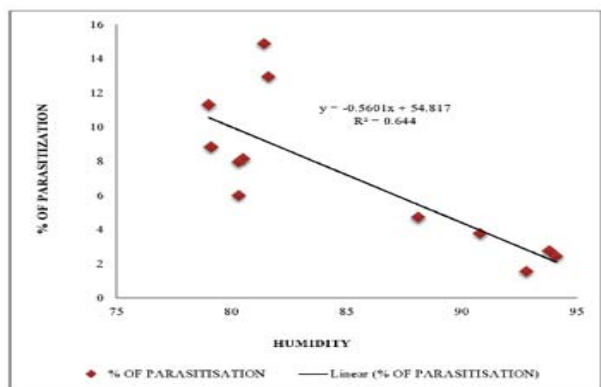


Fig 7: Scatter plot representation of temperature and percentage of parasitization by *H. australensis*.

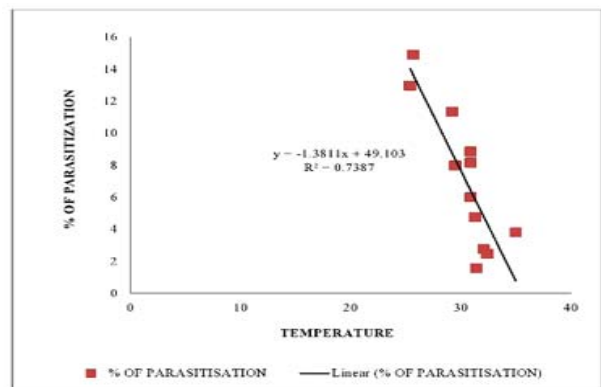


Fig 8: Scatter plot representation of relative humidity and percentage of parasitization by *H. australensis*.

The simple linear regression equation on the relationship between relative humidity and percentage of parasitization was also evaluated.

$$Y = 54.817 - 0.56X$$

One way ANOVA has been performed using Microsoft Excel's statistical macro ANOVA (Table 3).

Table 3. Anova result (Relative humidity against percentage of parasitization).

Source of variance	Df	SS	MS	F-value	Significance F
Regression	1	132.94	132.94	18.088	0.00168
Residual	10	73.49	7.349		
Total	11	206.43			

Table 4. Correlation coefficient of independent variable considered in the regression analysis.

	Coefficient	Standard error	t Stat	P-value
Intercept	54.817	11.24	4.876	0.000644
X variable 1	-0.56	0.131	-4.253	0.001681

So, there was a significant correlation between relative humidity and percentage of parasitization ($P < 0.05$). From the above analysis it is found that Coefficient of X (independent variable) is also negative (-ve) (Table 4). The above analysis is represented through scatter plot (Fig 8). Relationship between relative humidity and percentage of parasitization in *C. spectra* by *H. australensis* is presented in the figure 9. Relationship between temperature and percentage of stylization in *C. spectra* by *H. australensis* is shown in the figure 10.

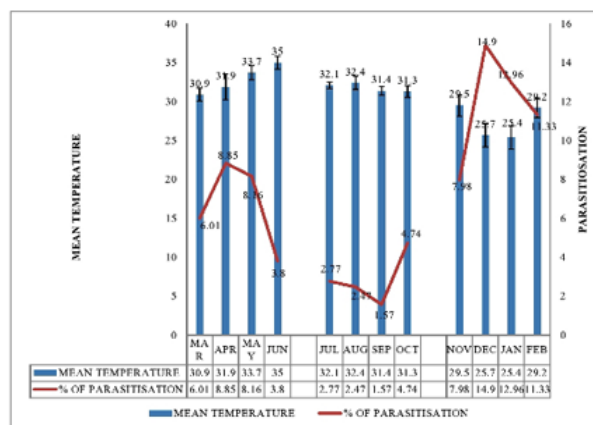


Fig 9: Relationship between humidity and percentage of parasitization in *Cofana spectra* by *Halictophagus australensis*.

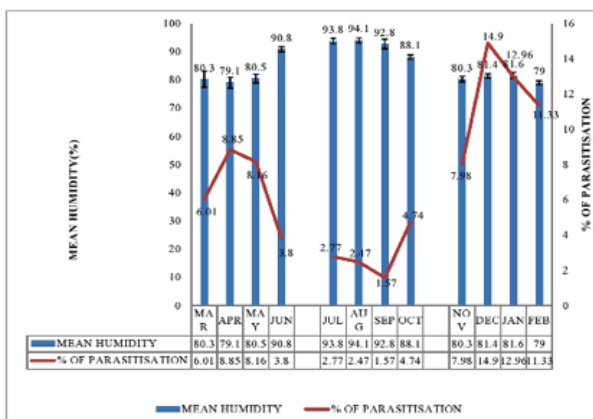


Fig 10: Relationship between temperature and percentage of parasitization in *Cofana spectra* by *Halictophagus australensis*.

4. Discussion

Infection of strepsipteran parasitoid to host *C. spectra* was observed to vary seasonally. In the post monsoon season, the rate of stylopization was greater relative to monsoon and premonsoon seasons. Percentage of stylopization varied from 1%-15%. It was previously suggested that the abundance of *C. spectra* increased the probability of parasitized WLHs ^[11]. Although the number of total collected *C. spectra* was high, the amount of parasitization did not exhibit proportionally increasing trend. Therefore it was evident that not only density of the host was important, but some environmental factors could also be responsible for this variation. Two environmental factors, i.e. temperature and relative humidity appeared the determining factors for parasitization of WLHs. In post monsoon season, the temperature (24 °C-26 °C) and humidity (79%-81%) were low relative to the premonsoon (31 °C-35 °C; 79%-90%) and the monsoon (31 °C-33 °C; 88%-95%). For this reason greater infection of strepsipteran parasitoids was detected in the post monsoon season while the lower number was noticed in the monsoon period due to high relative humidity. The result of the regression analysis shows the Coefficient of X (independent variable) for two ecological parameters (temperature and relative humidity) are found to be negative (-ve). That means both temperature and relative humidity have expressed negative correlation with the percentage of parasitization (Figs. 7-8). Lower rate of parasitization was encountered in some months when temperature was comparatively lower but with high relative humidity (September). Similarly low stylopization was observed in some months when temperature became high while relative humidity was low (June). Occurrence of stylopization was noticed higher in the post monsoon season when the temperature (24 °C-26 °C) and humidity (79%-81%) seem to be most favorable for the parasitoid. Although percentage of stylopization varied with the relative abundance of *C. spectra*, the optimal environmental conditions indicated significant effects on parasitization. Strepsiptera do not show a high percentage of stylopization due to their low searching ability ^[12] and very short lifespan of triungulins ^[13]. Limited control of host population is not only regulated by biotic factors, abiotic ones are also imperative.

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