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# International Journal of Insect Science

### Estimation of Fitness of Normal and Stylopized Paddy Pest, White Leafhopper *Cofana spectra* (Distant) (Hemiptera: Cicadellidae), in West Bengal, India through Correlation of Life History Traits

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**ABSTRACT:** The assessment of the morphological and reproductive features of white rice leafhopper *Cofana spectra* (Distant) was carried out using selected characters that bear importance in determining the fitness at the individual and population levels. Morphometric measurements of the individuals reared in the laboratory as normal and stylopized with the strepsipteran parasitoid, *Halictophagus australensis* Perkins, were recorded and analyzed. A *t*-test was performed to justify whether parasitization by *H. australensis* affected the traits. Correlations and regression analyses were carried out to deduce the difference in relative importance of the morphological features in the life history of *C. spectra* and their variation because of stylopization by *H. australensis*. A principal component analysis (PCA) was applied on the morphometric data to further substantiate the difference observed in the traits. In case of stylopized white leafhopper (WLH), fecundity was inhibited almost completely irrespective of seasons. The effect of stylopization on the life history traits of *C. spectra* has been noted that supports its possible use in biocontrol.

KEYWORDS: Cofana spectra, leafhopper, rice pest, parasitoid, Strepsiptera, Halictophagus australensis

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#### Introduction

The white leafhopper (WLH) *Cofana spectra* (Distant) is a minor rice pest that rarely occurs at huge populations to cause yield loss.<sup>1</sup> WLH is widely distributed in the tropics from Africa; south and southeast Asian countries such as India, Indonesia, Malaysia, Philippines, Sri Lanka, and Taiwan; Pacific; and Australia.<sup>2</sup> It is very common in West Bengal and other regions of India where paddy cultivation is practiced.<sup>3,4</sup> Both nymphs and adults are xylem feeders and suck sap from leaves.<sup>5</sup> The paddy plants experience damage as a result of sap sucking and oviposition by *C. spectra* followed by secondary fungal and bacterial infection on the affected part. *C. spectra* is also a vector of pathogenic virus like rice yellow mottle virus (RYMV) in West Africa.<sup>6</sup> The RYMV pathogen is included under the Sobemovirus group and is transmitted mechanically through rice plant injuries and by insect vectors.<sup>7</sup> 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.<sup>4</sup> In Philippines, C. spectra co-occur with pest insects such as Nephotettix virescens (Distant), Nephotettix nigropictus (Stål), Recilia dorsalis (Motschulsky), Sogatella furcifera Horváth, and Nilaparvata lugens Stål, and the predatory Cyrtorhinus sp., coccinellids and spiders.8 Similar observations are made in Sri Lanka where among paddy pests, Cicadellidae (81.3%) dominates over Delphacidae (18.3%), and among Cicadellidae, C. spectra accounts for 14.2% in Yala-minor cropping season (April-August). A minor variation occurs in the Maha-major cropping season (October-February) with a higher percentage of C. spectra.9 The host of Halictophagus australensis Perkins has been confirmed as C. spectra. This endoparasitoid (Plates 1 and 2) is distributed in south-east Asia, Japan, Australia, and





India.<sup>10,11</sup> The percentage of stylopization of hoppers varies between 1 and 63% depending on the seasons.<sup>12,13</sup> Stylopization of *C. spectra* by *Halictophagus spectrus* Yang in Malaysia was first reported by Hirashima and Kifune<sup>14</sup> and Barrion and Litsinger.<sup>15</sup> They listed several natural enemies attacking the hopper showing 11–50% variation of stylopization depending on different biological and environmental factors such as the host population, weather conditions, rice cultivars, age of the paddy plants, and time of occurrence.

Several morphological and reproductive features are viewed as life history traits and are useful parameters to gage the fitness of the individual insect.<sup>16,17</sup> Life history traits are those that are associated with the survival and reproduction,<sup>18</sup> which include growth rate; size at birth; age; and size at maturity, fecundity, and longevity; and frequency of reproduction. Variations in life history traits may be because of genotypes and their interactions with the environmental factors.<sup>19</sup> Even if the genotypic variations are absent or minimal, the life history traits may be different quantitatively because of environmental impacts that are substantiated through observations in the insects of the orders Lepidoptera,<sup>20</sup> Hemiptera,<sup>21</sup> and Diptera.<sup>22</sup> Subsequently, life history traits differ quantitatively in a population bearing similar genotype but under different environmental regulations. Adjustment in the life history traits of an individual insect is obvious in response to varied abiotic factors for its survival. Alternations in environmental factors such as food, temperature (abiotic), and biotic factors such as crowding, predators, and parasitoids can provide useful insights to understand the importance of the life history traits in a pest species. For instance, the parasitoid Elenchus tenuicornis Kirby causes an effect on the morphology, anatomy, ethology, and mortality of its host Javesella pellucida (Fabricius).23 Similarly, external genitalia are lost in the delphacid hosts, S. furcifera and N. lugens, because of stylopization by Elenchids and Drynids, respectively.<sup>2</sup>

In insects, fecundity is a positive function of the body size and longevity.<sup>24</sup> Longevity, on the other hand, depends on the feeding pattern and calorie content in the food.<sup>22</sup> Variations in the life history traits are adaptive providing maximal fitness to the species that can be explained in terms of the tradeoffs in the life history traits.<sup>25</sup> The trade-offs are significant determinants for individual survival and maintenance of the population. Agricultural pests and their vectors face a continuous threat from environmental stochastic factors and the synthetic chemicals. Similarly, a parasitoid may induce a significant variation in these important life history traits in the host species.<sup>26,27</sup> This is evident from the effects of parasitoid on the internal and external reproductive structures, as a manifestation of parasitoid infection in delphacid J. pellucida,23 cicadellid Ulopa reticulata (Fabricius),28,29 and various other plants and leafhoppers.<sup>30,31</sup> According to Kathirithamby,<sup>32</sup> stylopization lengthens the life span of the host (macrynobionts), sex characters of stylopized male delphacid are lost, and the male resembles superficially a normal female but all other homopteran host families appear normal from outside but internal genitalia may be affected.<sup>33</sup>

In course of development, value of one life history trait is compensated by the other, or the cost of one phenotypic expression is mitigated through the benefits of the other in terms of fitness value. In the present study, an attempt has been made to quantify the life history traits such as longevity, body size, and fecundity of the host, *C. spectra*. The effects of the strepsipteran parasitoid, *H. australensis*, on these life history traits have been compared. Such variations may predict the effect of the parasitoid on the host *C. spectra* parasitized by Strepsiptera and may be used to determine the potentiality for regulation of population of *C. spectra*.

#### **Materials and Methods**

Paddy seedlings of *Oryza sativa* Linnaeus were transplanted in earthen pots containing dried loamy soil and water up to 3 cm from the soil level with holes plugged with cotton to prevent drainage of water. The pots were placed on plastic plates containing water, and 5% 1,2,3,4,5,6-hexachlorocyclohexane (HCH) insecticide was sprinkled as dust around the plates to repel the predators such as ants and spiders. NPK was used as a fertilizer for luxuriant foliage growth of the paddy plants. As the plants grew about 30 cm, those were covered with mylar





cages of PVC sheet, for protection from other insects. These cages were cylindrical and covered at one end with a nylon net of mesh size 0.47 mm<sup>2</sup> for aeration. Five to six seedlings were planted in the center of the tub, which was used for stock culture. Single plant was used for mating, oviposition, and emergence of nymph of *C. spectra*. WLHs of both sexes collected from the field were reared and multiplied successfully in the cages kept in the net house of the Department of Zoology, University of Burdwan. The nymphs hatched out from the stock culture were collected and released again in another plant. The nymph was transferred to different cages singly. In this way, WLH was raised in the net house and maintained during the experiment. Population of WLH became evident when air temperature was 26–32°C and humidity 55–75%.

The C. spectra infected by endoparasitoid H. australensis (Plates 3 and 4) were released in a cage for getting triungulins. Sometimes, infected C. spectra kept singly in a glass cylinder with a bulbous region before the wider region ends. Both ends of the cylinders were covered with nylon net of mesh size 0.47 mm<sup>2</sup> for aeration. A small hole was made in the center of the net at the narrow end of each cylinder for insertion of rooted seedling of height 20 cm keeping the root outside, and the hole was then plugged with non-absorbent cotton. The experimental cylinders were then placed in an inclined position on a rack in water-filled tray in such a way as roots of seedlings were kept immersed in water. The seedling remained alive for four to five days, and was then replaced by a fresh one. The hoppers harboring strepsipteran female ready for emerging of triungulins (Plate 5) from eggs were released to the experimental cage for inoculation of the first and second instar nymphs of the captive host. Inoculation was found more in live hopper for its mobility. Morbid hoppers harboring female parasitoids kept in moist condition were noticed to release triungulins. As a result, a large number of triungulins came out for consecutive three days from live or morbid hoppers. As soon as free living and host-seeking triungulin comes in contact with a host, it normally enters the body via the abdominal cuticle, though

entry via the host's tarsi has recently been recorded.<sup>34</sup> Entry is gained through a combination of enzymatic (eg chitinase) and physical activity. Fluid is secreted from the mouth and appears to partially dissolve the host's cuticle. In this way, inoculation was carried out to obtain stylopized hoppers.

Both normal and stylopized adult hoppers after death were kept in 70% ethanol. The hoppers were processed and mounted on microslides following the method of DasGupta and Wirth.<sup>35</sup> Measurements of different parts of WLH were taken using microscope and micrometer, and are expressed in centimeter (cm).

The assessment of the morphological features of *C. spectra* was made using selected characters that bear importance in



Plate 5. Triungulin (First instar larva) of Strepsiptera *Halictophagus australensis*.

determining the fitness at the individual and population levels. Morphometric measurements of the individuals reared in the laboratory as normal and infected with the strepsipteran parasitoid, *H. australensis*, were recorded and analyzed. Correlations and regression analyses were performed to infer the difference in relative importance of the morphological traits in the life history of WLHs and their variation attributable to stylopization. A principal component analysis (PCA)<sup>36</sup> was carried out on the morphometric data to further validate the difference observed in the traits. The morphological features considered were as follows:

- For both male and female: wing length (WL), body length (BL), width of seventh sternum (WS), femur length (FL), leg length (LL)
- For male: aedeagus length (AL)
- For female: ovipositor length (OL).

An assessment of degree of dimorphism was made following Sharmila Bharathi et al<sup>37</sup> to deduce about the difference in value of the morphological feature toward a particular sex. A comparison was made between normal and infected individuals for each of these traits. A t-test38 was applied to validate whether parasitization by H. australensis affected the traits. The null hypothesis was that the values for the normal and infected individuals for any trait are not significantly different from zero (H0: value for a trait expressed as a ratio of normal/infected = 1). As morphological traits are highly correlated,<sup>37,39</sup> the data on all the morphological features were applied for PCA following Gotelli and Ellison.<sup>40</sup> The basis for using PCA is to reduce the number of initial observed variables and redundancy in the relationship. Because PCA is a variable reduction procedure, it is useful when the data are obtained on a number of variables (life history traits or morphological parameters) and there is some redundancy in these variables. In this case, redundancy means that some of the variables are correlated with one another, possibly because they are measuring the same construct (like the relationship between FL and WL might be similar to BL and WL, expressed in terms of correlation coefficient as indicator). Because of this redundancy, it should be possible to reduce the observed variables into a smaller number of principal components (artificial variables) that will account for most of the variance in the observed variables. Thus, the method was applied to infer about the relationship between life history traits. The initial observed data are transformed into eigenvalues of the components/factors that indicate how much of the variance is explained by the components/factors using XLSTAT 2009 version.<sup>41</sup> The relative importance of the variables toward their representation as a component is explained through the factor loadings. As the life history traits are highly correlated and add to fitness of organisms as a whole, an analysis to reduce the redundancy of the variables is essential to infer about the relative importance of the life history of the organisms.<sup>33,39,42</sup>

#### Results

The morphometric features of both sexes of normal and infected C. spectra are presented in Table 1. The degree of dimorphism was prominent in the following traits: WL, BL, FL, LL, and WS, having a trend toward females. The observed values for each trait were higher in females than males, and thus the females are larger in size (Fig. 1). These differences were statistically significant at P < 0.05 level (Table 2). Significant difference in these morphological traits was observed between normal and infected individuals of C. spectra (Fig. 2, Table 3). In all instances, the ratio between normal and infected (N/I)traits was less than 1, indicating that the infected individuals had a higher measurement for these traits. These suggest that the parasitization of C. spectra favored positively all traits except LL in males and OL in females. The PCA was appropriate as revealed by the measure of sampling adequacy, which was >0.5, and Bartlett's sphericity test for the justification of the PCA was significant in all cases (Tables 4-7). The results are presented in a tabular form in sequence of correlation matrix, eigenvalues, and factor loadings. For the normal individuals, a single factor was accountable for the observed variances in the data, whereas for the infected individuals, two factors

WL	BL	WS	FL	LL	AL
Normal male					
$6-6.6,  5.62 \pm 0.49$	$6.4{-}8,7.29\pm0.42$	1.04–1.28, 1.11 $\pm$ 0.06	1.4–1.8, 1.49 ± 0.11	$5.87.86,6.65\pm0.58$	$0.24{-}0.32,0.28\pm0.03$
Infected male					
5.4–7, 6.06 ± 0.45	7.2–9.2, 7.91 $\pm$ 0.56	$1.04{-}1.6,1.29\pm0.2$	$1.44{-}2,1.67\pm0.15$	$5.87.4,6.52\pm0.42$	$0.28{-}0.44,0.35\pm0.06$
WL	BL	ws	FL	LL	OL
Normal female					
$5.47.8,6.61\pm0.58$	$6.8{-}9,7.93\pm0.5$	$1.28{-}1.84,1.55\pm0.13$	$1.4{-}1.92,1.68\pm0.14$	$6.087.52,6.93\pm0.35$	$2.24{-}3.04,2.64\pm0.21$
Infected female					
$5.67.8,7.01\pm0.49$	$7.4{-}10,8.68\pm0.7$	1.44–2.12, 1.71 $\pm$ 0.18	$1.6{-}2.2,1.86\pm0.16$	$6.67.92,7.36\pm0.38$	$2.23,2.57\pm0.22$

**Table 1.** The values (range, mean  $\pm$  SE) of different morphological traits of *C. spectra* for normal males: n = 21 and normal females: n = 31, and for infected males: n = 12 and infected females: n = 21.

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**Figure 1.** The box plot representations of the degree of dimorphisms in the morphological traits of *C. spectra*.

**Notes:** When the values are greater than 1, it signifies that the trait is biased toward female. The **blue marks** are outliers, the red cross is the mean value, and the box represents the lower (25%) and upper (75%) quartiles with the mean values marked as line.

Abbreviations: WLDD, WL degree of dimorphism; BWDD, body weight degree of dimorphism; WSDD, WS degree of dimorphism; FLDD, FL degree of dimorphism; LLDD, LL degree of dimorphism.

explained the observed variation. It might be because of the low sample size for the infected males and females or the effect of parasitization that yielded two components. The ordination of variables along the component values is shown in Figures 3 and 4. For the normal individuals, a single factor was accountable for the observed variances in the data, whereas for the infected individuals, two factors explained the observed variation. It might possibly be because of the low sample size of the infected males and females or the effect of parasitization that yielded two components. From the PCA tables and the ordination of the variables, it can be deduced that the life history features are altered for both infected males and females of *C. spectra*. The factor loadings for the traits were different

**Table 2.** The results of *t*-test indicating that the degree of dimorphism is inclined toward females, because all values have a mean of >1.

MORPHOLOGICAL TRAIT	WLDD	BLDD	WSDD	FLDD	LLDD
Normal C. spectra	5.059	5.378	12.673	5.935	2.989
Infected C. spectra	3.958	2.579	7.366	2.769	3.266

All *t*-values are significant at *P* < 0.05.

Abbreviations: WLDD, WL degree of dimorphism; BLDD, BL degree of dimorphism; WSDD, WS degree of dimorphism; FLDD, FL degree of dimorphism; LLDD, LL degree of dimorphism.



**Figure 2.** The difference between the morphological traits of *C. spectra* expressed as a ratio of the normal to infected individuals (normal males: n = 21; normal females: n = 31; infected males: n = 12; infected females: n = 21).

from normal and suggestive of the effect of parasitization by Strepsiptera. The relationship between the selected morphological traits is represented as regression equations (Table 8). The relationship was similar to that observed in other insects like mosquito *Culex pipiens quinquefasciatus*.<sup>16</sup> It was noted that the OL and WL relationship, and LL and WL relationship were asymmetrical in normal compared to infected *C. spectra*. This is further suggestive of the effects of parasitization by *H. australensis*. The observation of the differences in these life history traits is indicative of differential fitness and thus reproductive success of *C. spectra*. As life history traits are indicators of the reproductive features of the pest *C. spectra* to infer about the relative contribution of these traits toward the event of reproduction and fecundity.



**Table 3.** Results of *t*-test on the deviation of the morphological traits of infected individuals from the mean values of normal individuals for respective sexes of *C. spectra*.

FEMALE TRAITS	WL	BL	ws	FL	LL	OL
<i>t</i> -value	2.623	4.377	3.299	3.872	4.121	1.442
MALE TRAITS	WL	BW	ws	FL	LL	AL
<i>t</i> -value	1.314	2.387	2.892	2.304	0.048	3.658

Values that are significant at P < 0.05 level are marked as bold. **Table values:** for infected males,  $t_{0.05(1),12} = 1.782$ ; for infected females,  $t_{0.05(1),21} = 1.714$ .

**Table 4.** The results of PCA of the morphological traits of normal male *C. spectra*. **A.** Bartlett sphericity test  $\chi^2 = 84.246$ , df = 15, P < 0.001. **B.** Eigenvalues and factor loadings for the morphometric variables. The highest factor loadings are marked in bold.

Α					
VARIABLES	WL	BL	ws	FL	LL
BL	0.824				
WS	0.898	0.701			
FL	0.752	0.658	0.547		
LL	0.554	0.545	0.417	0.660	
AL	0.532	0.410	0.490	0.234	0.163
В					
		FACT	OR 1	FA	ACTOR 2
WL	0.964			0.097	
BL	0.878			-0.017	
WS		0.860		0.235	
FL		0.816		-0.375	
LL		0.692		-0.528	
AL	0.553		0.	705	
Eigenvalue	3.891		<b>891</b> 0.981		981
Variability (%)	64.847			16	6.349
Cumulative (%)		64.84	7	81	.196

#### Discussion

Analysis of morphological attributes indicates existence of degree of dimorphism in *C. spectra* having a tendency toward females as shown in Figure 1. This means that the females are larger in size and live for longer period of time than the males (Table 1). This will have important consequence as pest, where the females can be expected to inflict more damage than the males. Sexual size dimorphism is also an indicator of genetic variability.<sup>37,43</sup> The difference in the morphological traits was also prominent between normal and infected individuals of *C. spectra*. For all these traits, the parasitized individuals had a higher value than the normal ones (Fig. 2). Morphometric studies on the external features of pest insects allow interpretation of the importance of morphological features as life history traits. Many of the morphological

**Table 5.** Results of PCA of the morphological traits of normal female *C. spectra.* Bartlett sphericity test  $\chi^2 = 94.019$ , df = 15, *P* < 0.001. **A.** Correlation matrix (values in bold indicate significance at *P* < 0.05). **B.** Eigenvalues and factor loadings for the morphometric variables. The highest factor loadings are marked in bold.

Α					
VARIABLES	WL	BL	ws	FL	LL
BL	0.485				
WS	0.665	0.349			
FL	0.823	0.631	0.549		
LL	0.534	0.156	0.103	0.529	
OL	0.495	0.501	0.222	0.592	0.164
В					

	FACTOR 1	FACTOR 2
WL	0.912	0.163
BW	0.707	-0.408
WS	0.660	-0.160
FL	0.939	0.063
LL	0.534	0.793
OL	0.670	-0.354
Eigenvalues	3.382	0.977
Variability (%)	56.374	16.289
Cumulative (%)	56.374	72.664

**Table 6.** Results of PCA of the morphological traits of infected male *C. spectra*. Bartlett sphericity test  $\chi^2 = 42.09$ , df = 15, *P* < 0.001. **A.** Correlation matrix (values in bold indicate significance at *P* < 0.05. **B.** Eigenvalues and factor loadings for the morphometric variables. The highest factor loadings are marked in bold.

Α					
VARIABLES	WL	BL	WS	FL	LL
BL	0.407				
WS	0.392	0.417			
FL	0.178	0.545	-0.086		
LL	0.158	0.714	0.219	0.892	
LA	-0.043	0.414	-0.334	0.628	0.658
В					
	FACTOR 1		R 1	FACTOR 2	
WL		0.337		0.664	
BL	0.824			0.3	841
WS	0.182			0.872	
FL		0.884		-0	.245
LL		0.956		-0.059	
LA	0.714		-0	.547	
Eigenvalue	3.030		1.6	80	
Variability (%)	50.494		27.997		
Cumulative (%)		50.494		78	.491

**Table 7.** Results of PCA of the morphological traits of infected female *C. spectra.* Bartlett sphericity test  $\chi^2 = 60.21$ , df = 15, *P* < 0.001. **A.** Correlation matrix (values in bold indicate significance at *P* < 0.05. **B.** Eigenvalues and factor loadings for the morphometric variables. The highest factor loadings are marked in bold.

А							
VARIABLES	WL	BL	ws	FL	LL		
BL	0.839						
WS	0.608	0.657					
FL	0.506	0.398	0.364				
LL	0.049	0.075	0.113	0.368			
OL	0.624	0.638	0.566	0.389	-0.061		
В							
	FACTOR 1			l	FACTOR 2		
WL		0.896	5		-0.103		
BL		0.891	I	-	-0.140		
WS		0.797	7	-	-0.070		
FL		0.639	)	(	0.510		
LL		0.169	)		0.913		
OL	0.79		l		-0.261		
Eigenvalue	3.296				1.197		
Variability (%)	54.936			19.955			
Cumulative (%)		54.93	36		74.891		

features are considered as life history traits because of their contribution to the reproductive success of the species.<sup>16,22</sup> Such morphometric studies are important from the perspective of assessing the importance of the pest and its damage potential.44 Using multivariate analyses, the significance of morphological features has been further evaluated in Heteroptera,<sup>33</sup> Hymenoptera,<sup>45</sup> and Orthoptera.<sup>46</sup> In the context of the present study, the consequences of parasitization can be deduced from the deviation in the morphological features and life history traits of the hosts. In case of C. spectra parasitized by H. australensis, the changes in the morphological features were prominent. The WL, BL, and LL of stylopized individuals were increased compared to the normal ones. The OL in females and AL in males were also affected suggesting that parasitization affects the traits, which contribute to the reproductive success of WLH. The multivariate analyses revealed that the morphological traits are highly correlated and that the changes in one or the other character have correlated changes in rest of the features. For instance, regression analysis supports that a change in the OL in female C. spectra infected by H. australensis leads to a change in the relationship with the corresponding WL. Such changes can be deduced as an effect of induced response because of the parasitoid and can be viewed as phenotypically plastic trait.<sup>17,19,47,48</sup> A tradeoff in such characters<sup>23</sup> is prominent because the features like WL and LL are higher in infected C. spectra compared to OL. Thus, the morphological features that can be considered as life history traits are affected by endoparasitoid.

In the present study, the morphometric traits of WLHs were significantly affected by stylopization. The OL is reduced but the AL (Fig. 2) is incremented in stylopized C. spectra. In addition, the styles are also significantly different between normal and parasitized males. The morphological characters of nymphs and adults remain unaffected in case of stylopized Nilaparvata sp., but the sexual characters are affected because of parasitization.<sup>26</sup> The PCA analysis substantiates the differences in the normal and parasitized C. spectra at the level of morphological traits. It is reflected in the correlation coefficient and ordination along the component axis. This possibly suggests that the morphological variations occurred as a whole though individual differences remain obscured. Alternately, this difference may have been pronounced if larger samples were considered. The population level is expected to vary under field conditions as the seasonal variation in the fecundity of C. spectra is prominent. Although fecundity exhibited a positive relationship with the oviposition events and period, it varied with seasons, suggesting that the population level varied between the months.49,50 Thus, this observation supports that seasonal variation occurs in the relative abundance of C. spectra. Such variation in abundance of C. spectra is also observed in other parts of the world.<sup>8,51</sup>

Life history theory depicts the pattern of longevity, reproduction, and development in the light of natural selection. It is also a theory of life cycle and fitness as shaped by natural selection.<sup>20</sup> In this view, the study of life history and related traits of pest insects is important to describe the population characteristics and possible trade-off between traits to infer about the possible damage potential.<sup>44</sup> In the present study, the effect of parasitization by H. australensis on the life history features of C. spectra has been noted that supports its use in biological control. The stylopized females rarely mate and oviposit. If they lay eggs, the clutch size is smaller than the normal one but the size and morphology of the eggs do not vary significantly. The average length of eggs of normal and stylopized females measures 1.08 mm (n = 35) and 1.12 mm (n = 12), and the hatchability is less than normal, which is in accordance with Cronin and Strong.<sup>52</sup> Naturally, fecundity varies with environmental factors.<sup>53</sup> This is evident for fecundity of C. spectra, which remains high in February to March and October to November, and low in April to June. The number of offspring produced can be used as a surrogate to measure the fitness of an ovipositing female and the population as a whole.<sup>18,25,41,44</sup> In case of stylopized C. spectra, fecundity is almost repressed irrespective of seasons demonstrating that the reproductive success is reduced by the strepsipteran parasitoid.

Stylopization reduces the pest status of the host.<sup>31</sup> The biological control of leafhoppers using predators and parasitoids can be a viable alternative.<sup>52</sup> For instance, Strepsiptera damage the sex organ, cause sterilization (reproductive death), and suppress the population of *C. spectra* in West Africa<sup>49</sup> and thus can serve as an agent to suppress pest population, along with predators like spiders.<sup>8</sup> Thus, Strepsiptera can act as a biocontrol





Figure 3. A biplot showing the ordination of morphological traits of male *C. spectra*.

agent for the hopper acting as paddy pest. As reflected in the findings of the present study, *H. australensis* can affect the life history of *C. spectra* detrimentally and impart regulatory effect on the population growth. However, to prove the viability of *H. australensis* as potential biological control agents, further studies are required on the environmental factors and population interactions of *H. australensis* and *C. spectra*. Nonetheless, keeping in view the effects of synthetic pesticides on non-target species and costs associated with the use of pesticides, the Strepsiptera *H. australensis* is a better alternative because it is host specific and does not interfere with the functioning of the non-target organisms. Further, the effect of parasitoid can be retrospective because reproduction of pest is affected, indicating that it has a long-term effect on the population dynamics of the pest.

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#### **Author Contributions**

Conceived and designed the experiments: AM, SM. Analyzed the data: SM. Wrote the first draft of the manuscript: NH. Contributed to the writing of the manuscript: RH. Agree with manuscript results and conclusions: SM, RH, NH, AM. Jointly developed the structure and arguments for the paper: AM, RH. Made critical revisions and approved final version: NH. All authors reviewed and approved of the final manuscript.





**Figure 4.** A biplot showing the ordination of morphological traits of female *C. spectra*.

**Table 8.** The regression equations using WL as explanatory variable for LL and AL of males, and LL and OL of females of normal and infected *C. spectra*.

NORMAL	INFECTED
Male	
LL(y) = 3.795 + 0.484WL(x) r <sup>2</sup> = 0.307; df = 1, 19; F = 8.401; P < 0.009	LL(y) = $5.424 + 0.202WL(x)$ $r^2 = 0.025$ ; df = 1, 10; F = 0.254; not significant
$\begin{array}{l} AL(y) = 0.125 + 0.03WL(x) \\ r^2 = 0.283;  \mathrm{df} = 1,  19; \\ F = 7.494;  P < 0.013 \end{array}$	AL(y) = 0.380 - 0.005WL(x) $r^2 = 0.002$ ; df = 1, 10; F = 0.02; not significant
Female	
LL( <i>y</i> ) = 4.773 + 0.327WL( <i>x</i> ) <i>r</i> <sup>2</sup> = 0.285; df = 1, 29; <i>F</i> = 11.588; <i>P</i> < 0.001	LL(y) = 7.09 + 0.04WL(x) $r^2 = 0.002$ ; df = 1, 22; F = 0.052; not significant.
OL(y) = 1.458 + 0.179WL(x) r <sup>2</sup> = 0.245; df = 1, 29; F = 9.404; P < 0.005	OL(y) = 0.598 + 0.0282WL(x) $r^2 = 0.389$ ; df = 1, 22; F = 14.06; $P < 0.001$

#### DISCLOSURES AND ETHICS

As a requirement of publication the authors have provided signed confirmation of their compliance with ethical and legal obligations including but not limited to compliance with ICMJE authorship and competing interests guidelines, that the article is neither under consideration for publication nor published elsewhere, of their compliance with legal and ethical guidelines concerning human and animal research participants (if applicable), and that permission has been obtained for reproduction of any copy-righted material. This article was subject to blind, independent, expert peer review. The reviewers reported no competing interests.

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