Journal of Biological Control, 24 (4): 300–304, 2010

Seasonal occurrence of leaf miner, *Chromatomyia horticola* (Goureau) (Diptera: Agromyzidae) and its parasitoids, *Diglyphus* spp. on brown mustard, *Brassica campestris* in Kashmir valley

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ABSTRACT: *Chromatomyia horticola* (Goureau) was found infesting brown mustard (*Brassica campestris*) from third week of March till second week of May, during 2006 and 2007, at Srinagar, Kashmir. Per cent plant and leaf infestation by the leaf miners during March-May ranged from 4.16 to 100.0 and 0.46 to 92.73 per cent, with overall mean infestation of 64.13±40.66 and 44.64±39.3, respectively. Average number of miners / leaf ranged from 0.65 to 13.09 (mean = 8.0). *Diglyphus* spp. were found actively associated with the leaf miner and the extend of parasitism ranged from 4.14 to 97.26% (mean = 38.47±40.7). Correlation between number of leaf miners and per cent leaf damage (r=0.83**, d.f.=7) and between per cent parasitism by *Diglyphus* spp. and leaf miners (r=0.65*, d.f.=7) were found significant. The correlation analysis exhibited a positive correlation with temperature and negative with relative humidity with respect to infestation.

KEY WORDS: *Chromatomyia horticola*, mustard, *Diglyphus* spp., Kashmir, plant infestation, leaf infestation, parasitism

(Article chronicle: Received: 26.06.2010; Sent for revision: 30.08.2010; Accepted: 11.10.2010)

INTRODUCTION

The leaf miner, *Chromatomyia horticola* (Goureau) is known to attack about 35 families of host plants of crucifers, vegetables and ornamental crops throughout the world (Spencer, 1990). In Kashmir valley, this pest has been reported to infest pea, mustard, rape, kale, turnip, radish and some ornamental plants (Zakaur Rab, 1981; Bhagat *et al*., 1989; Ahmad and Zaki, 2004; Bhat and Bhagat, 2009). The world wide success and abundance of this pest is not only because of its wide host range but is also due to many generations per season (6-8), high fecundity (ca. 100-500 eggs @ 50 eggs per day), quick developmental rate at high temperature (approx. 9-12 days) and low rate of mortality at moderate temperature as indicated by earlier workers (Sasakawa *et al*., 1970; Molitas and Gabriel, 1975; Sharma *et al*., 1997; Mizukoshi and Togawa, 1999). Despite sufficient work on this pest, literature in relation to *Brassica campestris* is comparatively less in comparison to other attacked crops such as *Pisum* spp.

Ahmad and Zaki (2004) reported for the first time the useful association of *Diglyphus* sp. with *C. horticola* from Kashmir. Bio-regulatory potentials of *Diglyphus* spp., which are larval ecto-parasitoids of *Chromatomyia* spp. and *Liriomyza* spp. are reported from world over (Khan, 1985; Heinz and Parrella, 1990; Purwar *et al*., 2003; Gencer, 2005). Bhat and Bhagat (2009) presented a comprehensive scenario of the hymenopteran parasitoids of *C. horticola* on seven vegetable crops from Kashmir valley. The present work focuses on the seasonal occurrence pattern of *C. horticola* and the potential of its *Diglyphus* parasitoids on *B. campestris* (brown mustard) in Kashmir valley.

MATERIALS AND METHODS

The present study was conducted in Shalimar campus of Sher-e-Kashmir University of Agricultural Sciences & Technology–K, Srinagar, during Rabi 2006 and 2007 in three different pesticide free plots (ca. 2000 sq.m. size each) of brown mustard. Each plot was treated as one replication and isolated by a distance of approximately 200 meters from one another. Observations on the seasonal abundance of mustard leaf miner, *C. horticola* and *Diglyphus* spp. including parameters such as per cent plant and leaf infestation, average number of miners and *Diglyphus* per leaf and per cent host parasitism were

recorded from 2nd week of March (early vegetative phase) till 2nd week of May (early pod maturation) during both the years. Fifty randomly selected plants from each plot were observed every week for the incidence of leaf miners, and percentage of infested plants determined. Five hundred leaves from 50 plants (10 leaves / plant) from each replication were examined *in situ* every week and percentage of miner infested leaves determined. Average number of miners and *Diglyphus* parasitoids / leaf was based on the number of emergences per 100 leaves collected from 5 plants / replication / week. For this, individual infested leaf was kept in a translucent white envelope (15x10 cm), its top margin folded and kept in a thermostat BOD [26±1°C, 65±5% RH and photoperiodicity of 12: 12 (L: D)], for the emergence of adult miners and *Diglyphus* parasitoids. The emerged miners and parasitoids were counted after exposing them to subzero temperature for ten minutes. The number of emerged adult miners corresponded with the number of unparasitised miners, whereas, the number of emerged *Diglyphus* parasitoids indicated the number of parasitized miners. The sum of the number of adult miners and number of *Diglyphus* parasitoids reflected the actual number of leaf miners infesting a leaf at a given period. Per cent parasitism by *Diglyphus* parasitoids was determined by dividing the number of emerged parasitoids by the total number of miners per leaf. Means of these parameters were based on 60 observations recorded every week. The identity of *Diglyphus* was confirmed under a stereoscopic binocular micoscope on the basis of its 2-segmented antennal funicular segments, and other generic characters, whereas that of *C. horticola* was done at NBAII, Bangalore.

The data was statistically analyzed using Minitab after square root transformation ($\sqrt{n+0.5}$) of actual values. Mean, standard deviations and correlation coefficients were determined from the untransformed values. Student's *t-test* was done to compare the differences between the studied parameters of 2006 and 2007. Two-way ANOVA was done to analyze the interactions between various parameters.

RESULTS AND DISCUSSION

The incidence of *C. horticola* at Shalimar, Srinagar, was first noticed during the 3rd week of March, infesting an average of 4.16 per cent plants (mean of 2006 and 2007). A six-fold increase in plant infestation was recorded during the end of March, which reached up to one hundred per cent during the last week of April (Table 1). Average plant infestation during the entire crop season worked out to 62.7±40.11 (N=1350) and 65.55±41.91; (N=1350) per cent during 2006 and 2007, respectively. Similarly, a 12-fold increase was recorded during the fourth week of March which further increased up to 92.73 per cent during the second week of May (Table 1). Per cent leaf infestation during entire *rabi* season worked out to 40.54±36.65 (N=8100) and 48.74±41.38 (N=8100) during 2006 and 2007, respectively. When two year's data for per cent plant infestation and per cent leaf infestation where compared separately through Student's *t-test*, a non-significant difference was obtained for per cent plant infestation (t=1.03 NS; d.f.=25) whereas it was significant for per cent leaf infestation (t=9.18**; d.f.=1348). Further, per cent plant infestation and per cent leaf infestation were positively correlated (r=0.94**;d.f.=7) for the mean of two years.

A significant rise in the number of miners/ leaf was observed from March 3rd week to April 3rd week, followed by a decline thereafter (Table 2). Average number of miners per leaf during 2006 and 2007 was 7.26±6.16 (N=540) and 8.75±7.31 (N=540), respectively. *Diglyphus* parasitoids appeared during March 4th week and increased significantly until April 4th week, remaining statistically identical thereafter (Table 2). The number of *Diglyphus* parasitoids per leaf averaged 4.19±4.7 (N=540) and 5.0±5.91 (N= 540), during 2006 and 2007, respectively. Per cent parasitism by *Diglyphus* indicated a continuous rise from 4.14 to 97.26 during end of March to 2nd week of May. Mean parasitism by the parasitoid during 2006 and 2007 was 39.06±40.37 (N=540) and 37.88±40.9 (N=540) per cent, respectively. When two year's data of the individual parameters were compared through Student's *t-test*, statistically significant differences were obtained for average number of leaf miners/ leaf (t=7.42**; d.f.=538) and average number of *Diglyphus* parasitoids/ leaf (t=3.66**; d.f.=538), but non-significant for per cent parasitism (t=1.64 NS; d.f.=538). Correlation between various parameters and weather temperature and relative humidity indicated positive relation for temperature, whereas negative for relative humidity for all the parameters, *viz*. per cent plant infestation (r=0.96** and –0.84**), per cent leaf infestation (r=0.82** and –0.64*), average miners / leaf (r=0.82** and –0.76*), average *Diglyphus* / leaf (r=0.79** and –0.45 ns) and average per cent parasitism (r=0.81** and –0.39 ns).

Increase in the population density *C. horticola* on mustard during April onwards as exhibited by the severity of plant and leaf infestation (Table 1) was primarily because of gradual increase in day temperature (21.0 to 27°C) and simultaneous decline in relative humidity (74.6 to 61.62). Rise in temperature caused shortening of developmental periods of the miner and production of quick generations and the moderate climatic conditions in Kashmir valley favored the population of leaf miners through reduced mortality of puparia, as against in tropical areas, as indicated by Sasakawa *et al.* (1970). Rich abundance of

Table 1. Per cent plant and leaf infestation by *C. horticola* on mustard at Srinagar, during *Rabi* 2006 and 2007

Period of observations	% plant infestation			% leaf damage /10 leaves^{-1} plant		
	Rabi 2006	*Rabi* 2007	Mean*	*Rabi* 2006	*Rabi* 2007	Mean**
March 2nd week	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}
March 3rd week	5.0 (2.22)^{b}	3.33 (2.62)^{b}	4.16 (2.14)^{b}	0.66 (0.86)^{a}	0.27 (0.77)^{a}	0.46 (0.85)^{a}
March 4th week	28.0 (5.28)^{c}	26.0 (5.09)^{c}	27.0 (5.21)^{c}	6.6 (1.84)^{b}	4.48 (1.59)^{b}	5.54 (1.95)^{b}
April 1st week	58.66 (7.68)^{d}	68.66 (8.28)^{d}	63.66 (8.0)^{d}	18.06 (3.5)^{c}	23.13 (4.18)^{c}	20.6 (4.27)^{c}
April 2nd week	80.66 (9.0)^{e}	92.66 (9.62)^{e}	86.66 (9.33)^{e}	44.13 (6.05)^{d}	57.2 (7.2)^{d}	50.66 (7.02)^{d}
April 3rd week	92.0 (9.62)^{f}	99.33 (9.96)^{e}	95.66 (9.81)^{f}	55.2 (7.15)^{e}	79.6 (8.85)^{e}	67.23 (8.18)^{e}
April 4th week	100.0 (10.02)^{f}	100.00 (10.02)^{e}	100.0 (10.02)^{f}	68.46 (8.25)^{f}	85.86 (9.27)^{e}	77.16 (8.8)^{f}
May 1st week	100.0 (10.02)^{f}	100.00 (10.02)^{e}	100.0 (10.02)^{f}	81.86 (9.05)^{g}	92.93 (9.65)^{ef}	87.4 (9.36)^{g}
May 2nd week	100.0 (10.02)^{f}	100.00 (10.02)^{e}	100.0 (10.02)^{f}	89.86 (9.49)^{g}	95.6 (9.79)^{f}	92.73 (9.65)^{g}
CD (0.05)	0.56	0.52	0.38	0.54	0.45	0.31

*Figures in columns represent means of 300 observations; **each figure in the column represents mean of 3000 observations; values in parentheses are transformations of √n+0.5; similar alphabets in each column represent statistically par values

Table 2. Dynamics of *C. horticola* and its *Diglyphus* spp. at Srinagar during *Rabi* 2006 and 2007

Period of observations	Rabi- 2006			Rabi- 2007			Mean of two years		
	Av. no. of miners / leaf	Av. no. of *Diglyphus* / leaf	% parasitism	Av. no. of miners / leaf	Av. no. of *Diglyphus* / leaf	% parasitism	Av. no. of miners / leaf	Av. no. of *Diglyphus* / leaf	% parasitism
March 2nd week	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}
March 3rd week	0.36 (0.89)^{a}	0.0 (0.71)^{a}	0.00 (0.71)^{a}	0.95 (1.19)^{a}	0.0 (0.71)^{a}	0.0 (0.71)^{a}	0.65 (1.06)^{a}	0.00 (0.71)^{a}	0.00 (0.71)^{a}
March 4th week	2.68 (1.71)^{b}	0.2 (0.8)^{a}	3.31 (1.24)^{a}	1.9 (1.5)^{ab}	0.18 (0.79)^{a}	5.05 (1.49)^{a}	2.29 (1.63)^{b}	0.19 (0.81)^{a}	4.14 (1.57)^{a}
April 1st week	4.11 (2.09)^{b}	0.5 (0.93)^{a}	10.11 (2.31)^{a}	10.11 (2.31)^{a}	0.51 (0.95)^{a}	7.51 (2.1)^{a}	5.16 (2.35)^{c}	0.51 (0.97)^{a}	8.78 (2.54)^{ab}
April 2nd week	13.23 (3.67)^{c}	2.73 (1.72)^{b}	19. 69 (4.26)^{b}	11.58 (3.45)^{c}	1.72 (1.41)^{a}	14.28 (3.44)^{a}	12.41 (3.57)^{d}	2.22 (1.62)^{b}	16.99 (4.08)^{b}
April 3rd week	15.6 (3.97)^{c}	7.9 (2.86)^{c}	51.46 (7.14)^{c}	19.18 (4.37)^{d}	7.72 (2.78)^{b}	42.09 (6.31)^{b}	17.38 (4.2)^{e}	7.81 (2.84)^{c}	46.77 (6.82)^{c}
April 4th week	11.51 (3.29)^{c}	9.15 (2.96)^{c}	78.21 (8.67)^{c}	15.16 (3.92)^{c}	12.66 (3.58)^{c}	84.67 (9.19)^{c}	13.09 (3.65)^{d}	10.91 (3.34)^{d}	81.44 (9.01)^{d}
May 1st week	10.86 (3.34)^{c}	9.92 (3.19)^{c}	90.97 (9.55)^{cd}	13.51 (3.7)^{c}	12.4 (3.54)^{c}	91.73 (9.58)^{c}	12.19 (3.54)^{d}	11.12 (3.4)^{d}	91.35 (9.57)^{d}
May 2nd week	7.48 (2.8)^{c}	7.33 (2.77)^{c}	97.83 (9.91)^{cd}	10.28 (3.25)^{c}	9.83 (3.18)^{bc}	95.69 (9.8)^{c}	8.88 (3.05)^{cd}	8.58 (2.99)^{c}	97.26 (9.86)^{d}
C.D.(0.05)	0.64	0.61	1.7	0.63	0.58	1.67	0.44	0.38	1.23

Figures in columns represent mean of 60 observations; values in parentheses are transformations of √n+0.5; similar alphabets in each column represent the values statistically on par

Table 3. Two years' mean and two-way ANOVA of per cent plant and leaf infestation, number of miners and *Diglyphus* / leaf and per cent parasitism of *C. horticola*

	% plant infestation		% leaf infestation		Av. no. of Miners/leaf		Av. no. of *Diglyphus*/leaf		% miner parasitism	
	Mean ± SD(N)		Mean ± SD(N)		Mean ± SD(N)		Mean ± SD(N)		Mean ± SD(N)	
	64.13±40.66 (2700)		44.64±39.3 (27000)		8.00±6.8 (1080)		4.59±5.35 (1080)		38.47± 40.66 (1080)	
Parameters	F	P	F	P	F	P	F	P	F	P
Year	1.17 NS	0.29	95.19**	0.0	71.11**	0.00	17.68**	0.00	3.00 NS	0.08
Week	602.12**	0.00	1832.63**	0.0	860.01**	0.00	877.99**	0.00	1183.7**	0.00
Replications	0.6 NS	0.55	1.06NS	0.44	2.32 NS	0.44	3.35*	0.69	3.11*	0.34
Year * Week	1.59 NS	0.2	13.64**	0.00	12.96**	0.00	13.27**	0.00	3.89**	0.00
Week * reps	1.21 NS	0.34	1.08 NS	0.62	3.54**	0.95	3.46**	0.825	1.69*	0.37
Year * reps	0.21 NS	0.81	0.89 NS	0.33	0.43 NS	0.102	2.63 NS	0.269	1.3 NS	0.9

SD = standard deviation; N = total number of observations; * and ** = significant at *P* = 0.05 and 0.01 respectively; NS = non-significant; F = F-value; *P* = probability (0.05)

leaf miners on *Brassica campestris* during the month of May was also reported by Bhat and Bhagat (2009) from Kashmir valley. Poor population of *Diglyphus* parasitoids until mid-April and hence insufficient suppression of the leaf miners thereby caused the latter to assume a rich population density which resulted in considerable per cent plant and per cent leaf infestation. The observed decline in average number of leaf miners per leaf from the last week of April, however, is attributable to the active role of *Diglyphus* spp. which restrained the host population both through non-ovipositional host killing and larval parasitism which showed an increase from 46.77 to 97.26 per cent. Our finding on average larval parasitism (Table 3) was found comparatively higher than the observations made by Bhat and Bhagat (2009) who reported an average of 32.0 per cent parasitism during the entire *rabi* season. An upswing in parasitism after the second week of April is attributed to favorable temperature (25–27°C) and sufficient host density that helped increase the fecundity and population density of parasitoids as reported by Patel and Schuster (1991). Increased density of *Diglyphus* parasitoids / leaf in response to high host density might also be due to their behavioral attraction to more heavily infested mustard leaves as indicated by Heinz and Parrella (1990) and Patel *et al*. (2003). Decline in average number of *Diglyphus* during the last week of May, although statistically non-significant, was due to fall in host population (Table 3).

As the use of pesticides on *B. campestris* is almost negligible in Kashmir valley so far, a rich population of *Diglyphus* is therefore well associated with the leaf miners throughout the valley. In view of the immense potential of *Diglyphus*, if adult miners are managed during March to mid-April either through selective chemicals or the use of yellow sticky cards, subsequent generations of the miners might succumb under the pressure of high population density of *Diglyphus* spp.

ACKNOWLEDGEMENT

The present work was carried out under RCM project (Rabi) by the first author. Sincere thanks are due to the Head, Division of Entomology, SKUAST-K, for providing necessary facilities. Thanks are also due to Dr. J. Poorani, Principal Scientist, NBAII, Bangalore, for the identification of *Chromatomyia horticola.*

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