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Studies on diversity and abundance of parasitoids of *Chromatomyia horticola* (Goureau) (Agromyzidae: Diptera) in north-western Himalayas, India

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Abstract: Pea leafminer, *Chromatomyia horticola* (Goureau) is an important pest of many vegetable and ornamental crops. The present investigation was carried out to study the parasitoid diversity of this pest in different agroclimatic conditions of Himachal Pradesh, India. Sixteen species of parasitoids *viz. Diglyphus horticola* Khan, *Diglyphus isaea* (Walker), *Zagrammosoma* sp., *Pnigalio* sp., *Quadrastichus plaquoi* Reina and LaSalle, *Asecodes erxias* (Walker), *Closterocerus* sp., *Neochrysocharis formosa* (Westwood), *Chrysocharis* sp, *Chrysocharis indicus* Khan, *Pediobius indicus* Khan (Eulophidae), *Opius exiguus* (Wesmael), *Dacnusa* sp. (Braconidae), *Cyrtogaster* sp., *Sphegigaster* sp. (Pteromalidae), and *Gronotoma* sp. (Figitidae) were recorded parasitizing *C. horticola* in different agro-climatic zones of Himachal Pradesh. Agro-climatic zone II (sub-temperate mid-hills) was the richest in parasitoid diversity (14 species) followed by zone I (11 species), zone III (7 species) and zone IV (4 species) which are characterized by sub-tropical sub-montane, wet temperate high hills and dry temperate high hills, respectively. Shannon diversity index, species richness, species evenness and species dominance varied from 0.69-1.71, 1.39-2.64, 0.50-0.71 and 0.29-0.50, respectively. *D. isaea* and *D. horticola* were the dominant parasitoids of *C. horticola* contributing 41.46-80.15 and 9.16-50.65 per cent of the total parasitization, respectively, in different agro climatic zones. The study highlights the role of different parasitoids in natural control of the leaf miner and will be useful for designing the IPM strategies for the pest.

Keywords: Braconidae, *Chromatomyia horticola*, Diversity, Eulophidae, Pea leafminer

INTRODUCTION

Chromatomyia horticola (Goureau), commonly known as pea leafminer is an important pest of many economically important vegetable crops especially peas, crucifers, onion and some ornamental flowering plants (Spencer, 1973; Bhagat *et al.,* 1989). According to Shantibala and Singh (2008) amongst all the pests, *C. horticola* is the most serious and regular pest of pea crop. The first indication of damage is in the form of punctures made by the females on the leaves with their sharp and pointed ovipositor for oviposition and/ or feeding. The photosynthetic activity of the infested plants is considerably impaired which adversely affects the flowering and fruit bearing capacity of the infested plants. In Himachal Pradesh, leaf infestation up to 89.6 per cent in peas was recorded (Sharma *et al.,* 2014) which could cause heavy yield loss as more than 20 per cent avoidable yield losses in pea have been reported beyond 40 per cent leaf infestation by *C. horticola* (Mehta *et al*., 1994). For the management of this pest, farmers mainly rely on the application of synthetic insecticides. Nevertheless, indiscriminate use of these chemicals leads to many environmental consequences like pest resurgence, secondary pest outbreaks, insecticide resistance, elimination of beneficial organisms especially predators and parasitoids from the ecosystem and pesticide residues.

In nature leaf miners have been reported to be attacked by more than 300 species of parasitoids (Noyes 2013). Eulophid wasps are the most common parasitoids recorded on leaf miners worldwide (Minkenberg and Van Lenteren, 1986; Waterhouse and Norris, 1987; Konishi, 1998; Murphy and LaSalle, 1999; Mekhlif and Abdul, 2002; Reina and LaSalle, 2003; Chen *et al*., 2003 and Tran, 2009). From India there are some reports on the parasitoids of *C. horticola* (Kumar 1985a; Kumar 1985b; Kumar, 1990; Sureshan and Narendran, 2002; Purwar *et al*., 2003; Khan *et al*., 2005; Bhat and Bhagat, 2009; Bhat and Bhagat, 2011; Mahendran and Agnihotri, 2013), but, none of them were specifically aimed at to study the complete diversity and abundance of parasitoids of this leaf miner and their role in natural control of the pest. Further, the information on natural parasitoids of *C. horticola* from Himachal Pradesh which is situated in the North-western Himalayan region of India is lacking. The present study was therefore carried out to study the diversity and abundance of

parasitoids of *C. horticola* in different agro-climatic conditions of Himachal Pradesh along with natural parasitism of the pest.

MATERIALS AND METHODS

Himachal Pradesh is a mountainous state of India situated in western Himalayas between 350 and 7000 m above mean sea level 30° 22' 40'' to 33° 12' 40'' N latitude and 75° 45' 55'' to 79° 04' 20'' E longitude. Due to huge variations in altitude, the agro-climatic conditions of different parts of the state also vary greatly. Climate varies from hot and sub-humid-sub-tropical in the southern tracts to cold, alpine and glacial in the northern and eastern mountain ranges. Himachal Pradesh also varies greatly in rainfall and has areas like Dharamshala which receives as high annual rainfall as about 3400 mm, as well as Lauhal and Spiti that are cold deserts and almost rainless. Depending upon the agro-climatic conditions, Himachal Pradesh is divided into four zones. Zone-I is sub-tropical sub-montane region comprising of low hills and valley areas up to an elevation of 914 m amsl. Zone-II is sub-temperate sub- humid mid hills with an altitude ranging from 915 to 1523 m amsl. This zone is characterized by moderate to heavy monsoon rains. Zone-III of the state is a wet temperate zone with high hills spanning between altitudes of 1524 and 2472 m amsl. Zone -IV represents dry temperate high hills beyond 2472 m amsl. This zone is almost rainless and experience heavy snow fall (3-5 m) during winter. Keeping in view the variations in the agro-climatic conditions of the state, all the four agro-climatic zones of the state were covered to study the diversity of parasitoids of *C. horticola*.

Collection of parasitoids: For the collection of parasitoids, field surveys were conducted in all the four agro climatic zones of Himachal Pradesh. The details of the locations surveyed are given in table 1.

Leafminer infested leaves of peas (*Pisum sativum* L.), mustard (*Brassica compestris* L.), Chinese sarson (*B. chinensis* L.) and China aster (*Callistephus chinensis* L.) were collected periodically at random from three strata (top, middle and bottom) of the plant from 5-6 different sites from each location which were then pooled to get a representative sample. These leaves were brought to the laboratory and examined under stereo zoom microscope (SZ 61, Olympus make, Japan for live, empty, dead and parasitized mines. The numbers of these mines (except dead mines) were added to get total mines. Collected mines were kept in plastic jars or glass vials in the laboratory at 25±0.5°C temperature and 70±5 per cent relative humidity for the emergence of parasitoids and/or adult flies of the miner. Freshly emerged specimens of parasitoids were used to prepare permanent and temporary mounts for further identification.

Table 1. Details of the locations surveyed.

Zone	Location	Altitude (m amsl)	Latitude	Longitude
I	Nurpur	640	32.30° N	75.90° E
	Ghumarwin	670	31.45° N	76.68° E
II	Solan	1580	30.92° N	77.12° E
	Sarahan	1550	30.72° N	77.18° E
III	Rohru	1690	31.13° N	77.45° E
	Chopal	2190	30.95° N	77.58° E
IV	Sharbo	2290	31.54° N	78.27° E
	Pooh	2660	31.77° N	78.60° E

Identification of parasitoids: The specimens were first examined under stereo-zoom microscope to identify them up to genus or tribe level. For detailed microscopic observations on taxonomic characters, temporary and permanent mounts of specimens were prepared as per standard procedures given by Willoughby and Koszrtarab (1974), Noyes (1982) and Khan *et al*. (2005).

For identification up to species level the mounted specimens were examined under phase-contrast compound microscope and identified as per the keys and/ or morphological characters described by Mani (1971), Sureshan and Narendran (2002), Reina and LaSalle (2004), Khan *et al*. (2005) and Fisher *et al*. (2008). The specimens were also sent to National Bureau of Agricultural Insect Resources, Bangaluru for identification or confirmation of their identity.

Statistical analysis: The data obtained after identifying the parasitoids were used to calculate the percent parasitization by each species and the relative proportion of each species. Diversity indices were also calculated as per Shannon (1948).

RESULTS AND DISCUSSION

Diversity and abundance of parasitoids: During the present study 16 species of hymenopteran parasitoids *viz*. *Diglyphus horticola* Khan, *Diglyphus isaea* (Walker), *Zagrammosoma* sp., *Pnigalio* sp., *Quadrastichus plaquoi* Reina and La Salle, *Asecodes erxias* (Walker), *Closterocerus* sp., *Neochrysocharis formosa* (Westwood), *Chrysocharis* sp, *Chrysocharis indicus* Khan, *Pediobius indicus* Khan (Eulophidae), *Opius exiguus* (Wesmael), *Dacnusa* sp. (Braconidae), *Cyrtogaster* sp., *Sphegigaster* sp. (Pteromalidae), and *Gronotoma* sp. (Figitidae) were recorded from *Chromatomyia. horticola* (Goureau) from different agro-climatic zones of Himachal Pradesh (Table 2). The parasitoids of *C. horticola* reported in the present study is the first report from Himachal Pradesh, however, various workers have reported the parasitoid communities of *C. horticola* from other parts of the country. Among them are the reports of Kumar (1985a), Kumar (1985b), Singh and Kumar (1985), Hussain and Khan (1986), Kumar (1990), Khan (1995), Purwar *et al*. (2003), Bhat and Bhagat (2009), Bhat and Bhagat (2011) and Mahendran and Agnihotri (2013) who reported *N. formosa*, *D. isaea*, *Opius turcicus* Fischer, *O. exiguus*, *Sphegigaster* sp., *Tetrastichus* sp., *Eulophus* sp., *Pedi-*

Table 2. Diversity and abundance of parasitoids of *Chromatomyia horticola* in different agro-climatic zones of Himachal Pradesh.

Parasitoid species	Zone I		Zone II		Zone III		Zone IV	
	Parasitism (%)	Relative proportion	Parasitism (%)	Relative proportion	Parasitism (%)	Relative proportion	Parasitism (%)	Relative proportion
Diglyphus isaea	21.0 (2.2-44.6)	41.46	20.22 (0.2-38.6)	46.61	5.13 (2.1-7.1)	42.86	22.06 (17.0-26.4)	80.15
Diglyphus horticola	10.63 (1.8-31.4)	20.98	9.87 (0.5-23.7)	22.76	6.07 (1.9-8.2)	50.65	2.52 (1.2-3.2)	9.16
Quadrastichus plaquoi	5.77 (0.0-15.7)	11.38	4.45 (0.7-24.0)	10.26	0.16 (0.0-0.7)	1.30	-	-
Neochrysocharis formosa	0.74 (0.0-15.6)	1.46	2.97 (0.3-8.1)	6.84	0.16 (0.0-0.7)	1.30	-	-
Pediodius indicus	0.74 (0.0-2.7)	1.46	2.64 (0.0-25)	6.09	0.16 (0.0-0.3)	1.30	-	-
Chrysocharis sp	4.86 (1.4-30.6)	9.59	1.54 (0.0-21.5)	3.55	0.16 (0.0-0.6)	1.30	2.31 (1.1-3.5)	8.40
Chrysocharis indicus	0.74 (0.0-1.57)	1.46	0.23 (0.0-2.8)	0.53	0.16 (0.0-0.6)	1.30	-	-
Zagrommosoma sp	-	-	0.01 (0.0-0.2)	0.03	-	-	-	-
Closterocerus sp	-	-	0.07 (0.0-0.5)	0.16	-	-	-	-
Pnigalio sp	-	-	0.07 (0.0-0.6)	0.16	-	-	-	-
Asecodes erxias	0.41 (0.0-1.1)	0.81	-	-	-	-	-	-
Opius exiguus	4.53 (0.2-12.8)	8.94	0.86 (0.0-8.3)	1.98	-	-	-	-
Dacnusa sp	0.74 (0.0-2.4)	1.46	0.1 (0.0-2.0)	0.22	-	-	-	-
Gronotoma sp	-	-	0.03 (0.0-0.2)	0.06	-	-	-	-
Cyrtogaster sp	0.49 (0.0-0.8)	0.98	0.33 (0.0-3.7)	0.75	-	-	-	-
Sphegigaster sp	-	-	-	-	-	-	0.63 (0.0-1.1)	2.29
Total	50.66 (25.0-72.9)	100	43.39 (14.4-73.5)	100	11.98 (7.4-16.8)	100	27.31 (18.0-32.9)	100

obius acanthi (Walker), *O. phaseoli* Fischer, *Bracon* sp., *Sphegigaster stepicola* Boucek, *Hemiptarsenus indicus* Khan, *Chrysocharis funicularis* Khan, *Closterocerus agromyzae* Narayanan and Subba Rao, *Closterocerus phytomyzae* Mani, *P. indicus*, *P. thakerei*, *D. funicularis*, *D. mandibularis*, *C. horticola* Mani, *D. horticola*, *Euderus agromyzae* Gangrade, *Dacnusa* sp., *Diglyphus* sp. and *Opius* sp. to parasitize *C. horticola* infesting different host plants in different parts of the country. Besides, Khan *et al*. (2005) described 56 species of eulophid parasitoids parasitizing different agromyzids from different places of Uttrakhand. Parasitoids such as *N. formosa, H. varicornis*, *Q. plaquoi, Quadrastichus* sp., *Diglyphus* sp., *Asecodes* sp., *Chrysocharis* sp. and *O. exiguus*, however, have been reported from Himachal Pradesh attacking *L. trifolii* (Kaushik, 1999; Reina and LaSalle, 2004; Sharma *et al*., 2011). The parasitoid diversity varied with the agro -climatic conditions and zone II of the state which is sub-temperate sub- humid mid hills was more rich in parasitoid diversity having 14 species viz. *D. horticola, D. isaea*, *N. formosa*, *Chrysocharis* sp, *P. indicus C. indicus*, *Q. plaquoi*, *Closterocerus* sp*, Zagromosoma* sp, *O. exiguus*, *Dacnusa* sp., *Gronotoma* sp and *Cyrtogaster* sp (Table 1). Second most diverse zone was agroclimatic zone I (sub-tropical sub- montane region comprising of low hills and valley areas) of Himachal Pradesh where 11 species namely *D. horticola, D. isaea, N. formosa, Chrysocharis* sp, *P. indicus, Q. plaquoi, C. indicus, A. erxias, O. exiguus*, *Dacnusa* sp, and *Cyrtogaster* sp were collected from *C. horticola* (Table 2). Agroclimatic zone III (wet temperate high hills) and IV (dry temperate high hills) of the state had comparatively less diversity of the parasitoids as only 7 (*D. isaea*, *D. horticola*, *P. indicus*, *Chrysocharis* sp., *C. indicus*, *N. formosa* and *Q. plaquoi*) and 4 (*D. isaea*, *D. horticola*, *Chrysocharis* sp. and *Sphegigaster* sp, respectively, were reared from the leafminer (Table1). Among different parasitoids, *D. isaea* and *D. horticola* were the most common and abundant parasitoids of *C. horticola* throughout the state. *D. isaea* and *D. horticola* contributed 41.46 and 20.98, 46.61 and 22.76, 42.86 and 50.65; and 80.15 and 9.16 per cent of the total parasitization of *C. horticola* in agroclimatic zone I, II, III and IV of the state, respectively (Table 2). The present results find support from the finding of Ibrahim and Madge (1979) and Gencer (2004) who reported *D. isaea* as the dominant larval parasitoid of *Phytomyza syngenesiae* Griffiths and *C. horticola* resulting in 40 and 52.5 per cent mortality of the pest, respectively. The present results also agree with the findings of Bhat and Bhagat (2009) also reported *D. horticola* and *Diglyphus* sp as the dominant parasitoids contributing

Table 3. Indices of diversity of parasitoids of *Chromatomyia horticola* for different agro-climatic zones of Himachal Pradesh.

Parameter	Zone I	Zone II	Zon e III	Zone IV
Shannan index (H)	1.71	1.58	0.99	0.69
Species richness (Hmax)	2.40	2.64	1.95	1.39
Species evenness (J)	0.71	0.60	0.51	0.50
Species dominance (D)	0.29	0.40	0.49	0.50

Table 4. Effect of host plants on the diversity of parasitoids of *C. horticola.*

Parasitoid species	Host plants			
	Peas		Mustard	
	Parasitism (%)	Relative proportion (%)	Parasitism (%)	Relative proportion (%)
Diglyphus isaea	22.04 (1-33.9)	54.90	18.23 (0.5-44.6)	37.02
Diglyphus horticola	8.32 (1.9-18.4)	20.72	10.65 (1-18.9)	21.63
Neochrysocharis formosa	2.33 (1.9-5.7)	5.82	2.51 (0-5.1)	5.10
Quadrastichus plaquoi	2.33 (0-13.6)	5.82	10.56 (0-18.3)	21.44
Chrysocharis indicus	0.29 (0-1.2)	0.72	0.43 (0-1.6)	0.87
Chrysocharis sp.	0.84 (0.5-2.7)	2.10	1.99 (0.3-30.6)	4.04
Opius exiguus	1.83 (0.2-1.4)	4.56	0.14 (0.2-2.7)	0.29
Dacnusa sp.	0.25 (0-0.4)	0.63	0.05 (0-1.4)	0.10
Pediobius indicus	1.60 (0.5-11.3)	3.98	3.50 (1-25)	7.12
Cyrtogaster sp.	0.18 (0-0.8)	0.45	0.71 (0-1.6)	1.44
Closterocerus sp.	0.04 (0-0.2)	0.09	0.14 (0-0.5)	0.29
Pnigalio sp.	0.05 (0-0.7)	0.13	0.05 (0-0.7)	0.10
Asecodes erxias	-	-	0.24 (0-1.1)	0.48
Zagrammosoma sp.	-	-	0.05 (0-0.25)	0.10
Gronotoma sp.	0.04 (0-0.2)	0.09	-	-
Total	40.14 (29-59.8)	100	49.24 (8.1-72.9)	100
Shannon index (H)	1.45		1.68	
$H_{max.}$	2.56		2.64	
Evenness (J)	0.57		0.64	
Dominance (D)	0.43		0.36	

Figures in parentheses represent the range

35.66 and 27.44 per cent of the total parasitisation of *C. horticola*, respectively, in Kashmir valley of India. Shannon diversity indices (Table 3) also revealed the similar trend as the species richness was maximum (2.64) in agroclimatic zone II followed by zone I (2.40), zone III (1.95) and zone IV (1.39). Shannon index for zone I, II, III and IV was 1.71, 1.58, 0.99 and 0.69, respectively. As many as 71, 60, 51 and 50 per cent (J=0.71, 0.60, 0.51 and 0.50, respectively) of the species were evenly distributed in the respective zones and the species dominance in the respective zones was 0.29, 0.40, 0.49 and 0.50. The total per cent parasitization ranged from 25 to 72.9 (mean = 50.66) in zone-I, 14.4 to 73.5 (mean = 43.39) in zone II, 7.4 to 16.8 (mean = 11.98) in zone III and 18 to 32.9 (mean = 27.31) in zone IV. Pooling of data collected from different locations representing all the four agroclimatic zones of Himachal Pradesh reveal that the total parasitization of *C. horticola* ranged from 7.4 to 73.5 per cent with a seasonal mean of 40.62. The results obtained during the present investigation agree with the results of Kumar (1985a), Bhat and Bhagat (2009), Ahmad *et al*. (2010) and Mahendran and Agnihotri (2013) who reported 4.14 to 97.26 per cent parasitization of *C. horticola* by different parasitoids in different host plants like mustard (*B. campestris*), kale (*B. oleracea.* var. *acephala*), knoll-khol (*B. oleracea.* var. *gongylodes*), turnip (*B. rapa* L.), pea (*P. sativum*), onion (*Alium cepa* L.) and malva (*Malva sylvestris* L.).

Effect of host plant on the diversity and abundance of parasitoids of *C. horticola*: To study the effect of host plants on the diversity of parasitoids of *C. horticola* peas and mustard crops were selected. Both these crops are widely grown in the state and are heavily attacked by *C. horticola.* Study revealed that both peas and mustard had almost same parasitoids diversity as 13 and 14 species of parasitoids were active against *C. horticola* on peas and mustard, respectively. Out of the collected species, 12 namely *D. isaea*, *D. horticola*, *N. formosa*, *Q. plaquoi*, *Chrysocharis* sp., *C. indicus*, *O. exiguus*, *Dacnusa* sp., *P. indicus*, *Pnigalio* sp., *Cyrtogaster* sp.and *Closterocerus* sp. were common on both the host plants. Nevertheless, *Gronotoma* sp. was collected from *C. horticola* infesting peas only and *A. erxias* and *Zagrammosoma* sp. were found parasitizing *C. horticola* on mustard only (Table 4). The total parasitization (pooled data for all the locations) of the leafminer though varied on the two host plants. On peas it ranged from 29 to 59.8 per cent with on average of 40.14 per cent and was less than on mustard where it ranged from 8.1 to 72.9 per cent with a mean of 49.24 per cent. Earlier Bhat and Bhagat (2009), Ahmad *et al*. (2010) and Mahendran and Agnihotri (2013) also reported 4.14 to 97.26 and 19.96 to 71.69 per cent parasitization of *C. horticola* by different parasitoids in mustard (*B. compestris*) and peas (*P. sativum*), respectively.

Conclusion

The pea leaf miner, *C. horticola* is an important pest of many economically important crops in India. In nature 16 species of eulophid, pteromalid and braconid parasitoids are associated with this pest and play an important role in its natural control especially in peas and mustard. However, there is a need to conserve these parasitoids by discouraging insecticide applications during the peak period of their activity. The data generated during the study presents the variations in diversity and abundance of the leafminer parasitoids under different agroclimatic situations, and the extent of parasitization of the pest. The information generated in the present study can therefore be utilized in developing bio-intensive management strategies for this pest.

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