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Research Article

Natural parasitism by trichogrammatids (Hymenoptera: Trichogrammatidae) on lepidopteran eggs under diverse cropping system

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ABSTRACT: The present study was conducted to collect and identify the species of trichogrammatids from eggs of lepidopteran pests infesting the diverse cropping systems in the country. A total of 28 plant species were inspected for the collection of lepidopteran eggs either through collection of insect eggs or by placing sentinel egg cards. The species such as *Trichogramma chilonis* Ishii, *T. achaeae* Nagaraja and Nagarkatti*, T. danausicida* Nagaraja*, Trichogrammatoidea bactrae* Nagaraja and *Tr. armigera* Manjunath were recorded naturally occurring with 12 species of lepidopteran insect pests. In nature, the parasitism rate varied from 5.35 to 82.25% by the associated trichogrammatids. A total of 596 sentinel trap cards were placed in the agricultural, vegetables, fruits crops, and grasses on the bunds of fields to trap the egg parasitoids present in the different habitat. Through sentinel cards, three species, *viz., T. chilonis*, *T. achaeae* and *Tr. bactrae* were found inhabit the vegetables and ornamental crops. The percentage of adult emergence from the sentinel trap cards ranged from 70.59 to 100.0%. Natural parasitism of *Tr. bactrae* recorded for the first time on the eggs of *Lampides boeticus* L. infesting wild species of *Crotalaria*. In addition, the natural parasitism of *Tr. armigera* and *Tr. bactrae* on the eggs of *L. boeticus* laid on either cultivated or wild relatives of *Crotalaria* in undisturbed habitat had higher natural parasitism due to their bright yellow flower attracts parasitoids to parasitize the eggs and conserve these egg parasitoids. Recently, natural parasitism of *Trichogramma* sp. was recorded on invasive fall armyworm, *Spodoptera frugiperda* (J.E. Smith) infesting maize in Karnataka. Based on the present work, there could be choice of selecting right species which occur naturally on individual crops.

KEYWORDS: Biological control, collection, cropping System, egg parasitoid, identification

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INTRODUCTION

More than 1500 plant species are grown as edible or other multiple purposes under diverse cropping systems in India (Sivaraj *et al.,* 2016). In the intensified cropping system, and in the undisturbed habitat, several species of caterpillars of moths and butterflies are found feeding on the crops (Alfred *et al.,* 1998). The eggs and larvae of many species of lepidopteran are attacked by parasitic Hymenoptera in the diverse cropping system. The egg parasitoids belonging to genera *Trichogramma* Westwood and *Trichogrammatoidea* Girault (Hymenoptera: Trichogrammatidae) are extensively used for the management of lepidopteran pests in different parts of the world (Smith, 1996). The ease production of these egg parasitoids mainly trichogrammatids on the laboratory hosts, and their versatility to destroy egg stage before pests starts to damage the crops, added advantage over the other biocontrol agents (Ulrichs and Mewis, 2004; Gardner *et al.,* 2011). Further, the high likeness and belligerent parasitism towards the eggs of lepidopteran pests favours their use in biological control or their integration with other control measures (Jalali *et al.,* 2016). The success of biological control programs depends on the ample use of native species of parasitoids to reduce the pest population in the specific habitats as the native parasitoids are well adapted to natural environments (Hassan, 1994).Therefore, the documentation of local natural enemies in specific cropping system helps to use those parasitoids to control the pest population more effectively (Smith, 1996). Furthermore, conserving at least one potential parasitoid for a specific geographical area is a requirement for conservation biological control. For example under high pest population and low natural

occurring parasitoid density, the inundative release of potential parasitoids successfully manages the pests (Gurr *et al.,* 2000). Thus, the present work was aimed to identify the trichogrammatid egg parasitoids associated with the eggs of lepidopteran pests across number of locations in the country, which would help in biological control of lepidopteran pests after their identification from cropping systems.

MATERIALS AND METHODS

A random collections were made in seven states in the country, *viz*., Karnataka, Kerala, Tamil Nadu, Telangana, Punjab, Odisha, and Himachal Pradesh on the agricultural and horticultural crops. The collection of eggs and egg masses of lepidopteran pests was done by visual inspection. A total of 28 plant species were observed for the collection of lepidopteran eggs through visual inspection in 6 states, *viz.,* Karnataka, Telangana, Kerala, Odisha, Punjab and Tamil Nadu, and sentinel egg cards (trap cards) of *Corcyra cephalonica* (Stainton) were placed on 4 crops in 3 states, *viz*., Karnataka, Himachal Pradesh and Odisha. The plants that were observed for collections of eggs or used for keeping sentinel trap cards were banana, brinjal, cabbage, *Calotropis*, chilli, *Crotalaria* (wild), cassava, castor, cauliflower, curry leaves, *Cycas*, *Dolichos*, *Euonymus*, fenugreek, groundnut, jackfruit, jamun, mango, mustard, okra, pomegranate, red gram, sapota, sugarcane, sunflower, sunn hemp, *Tabernae montana*, and tomato. The sampling was done from October, 2016 to March, 2019. The plant species that were grown by the farmers in their field or were found on bunds were sampled in a particular region, and were sampled during the study period. A total of 596 sentinel cards were placed in the agricultural, vegetables, fruits crops and grasses on the bunds of fields to trap the parasitoids from different habitats.

The selected fields were sampled randomly and inspected for lepidopteran eggs. The eggs or egg masses on being observed were collected along with plant parts in the glass tubes (150×25 mm), labeled and brought to the laboratory. Collected egg masses and eggs of the insect pests were kept in individual glass tube, and each egg was considered as egg unit in observation. The tubes containing eggs and egg masses were kept under controlled climatic condition (Temperature: 27±2°C; Relative humidity: 65±5%; photoperiod 14L:10D) and observed on daily basis until the lepidopteran larvae or the adult parasitoid emerged. The percentage of parasitism of each host insect species was calculated by dividing the number of parasitized eggs by the total number of collected eggs or egg masses (number of parasitized eggs × 100/total number either of collected eggs or egg masses of collected).

The emerged adult parasitoids were counted as total and preserved in 80% ethanol for species identification. The males were used for species level identification based on their morphological characteristics and the females were identified, if they have emerged along with males from the same host eggs. If only females emerged from the host eggs, they were reared on *C. cephalonica* eggs to obtain males in the next generation and were used in identification in few cases. The identification was not performed, where only female emerged that were unable parasitize *C. cephalonica* eggs. The trichogrammatids thus emerged were mounted on microscope slides for identification by following procedure given by Noyes (1982). The species level identification was carried based on characteristics of the genitalia, antennae, and wings of males, using an illustrated identification key for species of *Trichogramma* (Nagaraja and Nagarkatti, 1969; Nagarkatti and Nagaraja, 1971; Nagaraja, 1973) and *Trichogrammatoidea* (Nagaraja, 1978). The collected specimens were deposited in the Division of Germplasm Collection and Characterization, ICAR-National Bureau of Agricultural Insect Resources, Bengaluru.

RESULTS AND DISCUSSION

The natural parasitism of trichogrammatid egg parasitoids was recorded from six states of India under different cropping system is listed in (Table 1). A total of 28 plant species were observed for lepidopteran eggs and/or were used for trapping the trichogrammatid egg parasitoids through sentinel trap cards. Among the 28 plant species, four vegetable crops (tomato, cabbage, eggplant and *Dolichos* bean), two ornamental plants (*Cycas* and *Euonymus*), two flowering plants (Jasmine and *Tabernae montana*), one plant each from fruits (mango), cereals (maize), fodder crop (sunhemp) and two plants from undisturbed habitat (wild species of *Crotalaria* and *Calotropis*) on which lepidopteran eggs were collected or trapped through sentinel cards had successful parasitism by trichogrammatids. A total of 11 species of trichogrammatids were recorded parasitizing different lepidopteran eggs in different cropping systems. The species composition consist of 3 species of *Trichogramma viz., T. chilonis* Ishii, *T. achaeae* Nagaraja and Nagarkatti*, T. danausicida* Nagaraja, two species of *Trichogrammatoidea viz., Tr. bactrae* Nagaraja and *Tr. armigera* Manjunath and 6 indeterminate species of belonging to *Trichogramma* were identified and their parasitism rate on the associated lepidopteran eggs was recorded.

The species, *T. chilonis* was obtained from the eggs of *Helicoverpa armigera* (Hübner) and *Plutella xylostella* (L.) infesting tomato and cabbage, respectively, with a natural parasitism of 16.21 and 7.69%, in Karnataka. In Telangana, the parasitism rate of *T. chilonis* on the eggs of *H. armigera* infesting tomato and Sphinx moth on jasmin was 36.0 and 26.92%, respectively. On the other hand, 50% parasitism of

H. armigera eggs laid on tomato was recorded from Odisha. In Punjab, the natural parasitism by *T. chilonis* was 35.71 and 60.0% on the eggs of *Lampides boeticus* (Linnaeus) and Sphinx moth eggs laid on *Cycas* and *Tabernae montana* plants. In Karnataka, the parasitism by *T. achaeae* was 5.35% on the eggs of invasive *Tuta absoluta* (Meyrick) and 28.57% eggs of *H. armigera* were also found parasitized by this parasitoid in tomato. Further, the eggs of *H. armigera* laid on tomato were also parasitized by *T. achaeae* (13.64%) in fields of tomato from Kerala.

The natural parasitism by *Tr. bactrae* was observed on the *T. absoulta* eggs collected from tomato along with *T. chilonis* in Karnataka. On wild *Crotalaria*, the eggs of *L. boeticus* were parasitized by *Tr. bactrae* with the parasitism of 21.05%. This is first report of parasitism by *Tr. bactrae* on the eggs of *L. boeticus* under natural condition from Kerala and Karnataka. The parasitism of *Tr. bactrae* and *Tr. armigera* together was 13.80% on eggs of *L. boeticus* collected from Karnataka on sunn hemp, *Crotalaria juncea* L. Moreover, natural parasitism of *Tr. armigera* was 25% on the eggs of *L.*

Table 1. Parasitism of trichogrammatids species on lepidopteran eggs inhabiting on plant species

Host plant	Host insects	No. of eggs		Percentage parasitism	Species
		Collection	Parasitism		
Karnataka					
Tomato	*Helicoverpa armigera*	21	6	28.57	*Trichogramma achaeae*
Tomato	*Tuta absoluta*	56	3	5.35	*T. achaeae* and *Trichogrammatoidea bactrae*
Tomato	*Helicoverpa armigera*	37	6	16.21	*T. chilonis*
Cabbage	*Plutella xylostella*	26	2	7.69	*T. chilonis*
Dolichos bean	Lepidopteran eggs mass	23	19	82.25	*Trichogramma* sp.
Mango	*Euthalia garuda*	13	4	30.77	*Trichogramma* sp.
Calotropis	*Danaus* sp.	66	38	57.48	*T. danausicida*
Sunn hemp	*Lampides boeticus*	123	17	13.80	*Trichogrammatoidea armigera* and *Tr. bactrae*
Maize	*Spodoptera frugiperda*	78	20	25.64	*Trichogramma* sp.
Telangana					
Tomato	*Helicoverpa armigera*	25	9	36.00	*T. chilonis*
Tomato	*Tuta absoluta*	7	3	8.82	*Trichogramma* sp.
Jasmin	Sphinx moth	26	7	26.92	*T. chilonis*
Kerala					
Tomato	*Helicoverpa armigera*	22	3	13.64	*T. achaeae*
Crotalaria (wild)	*Lampides boeticus*	19	4	21.05	*Tr. bactrae*
Odisha					
Tomato	*Helicoverpa armigera*	8	4	50.00	*T. chilonis*
Punjab					
Cycas	*Lampides boeticus*	14	5	35.71	*T. chilonis*
Tabernae montana	Sphinx moth	15	9	60.00	*T. chilonis*
Tamil Nadu					
Sunn hemp	*Lampides boeticus*	16	4	25.00	*Tr. armigera*

Table 2. Trichogrammatids trapped in sentinel cards placed in different habitat

Host plant	No. of parasitized eggs	No. of adult emerged	Percent Adult emergence	Species	State
Cabbage	8	6	75.00	*Trichogramma achaeae and Trichogrammatoidea bactrae*	Karnataka
Cabbage	7	6	85.71	*Trichogramma* sp.	
Tomato	29	29	100.0	*T. chilonis*	
Tomato	7	6	85.71	*T. achaeae*	
Tomato	35	30	85.71	*T. achaeae*	
Cabbage	3	3	100.0	*T. chilonis*	
Tomato	5	5	100.0	*T. chilonis*	
Tomato	94	73	77.66	*Trichogramma* sp.	
Egg plant	58	46	79.31	*T. chilonis*	
Egg plant	95	72	75.79	*T. chilonis*	
Cabbage	22	18	81.82	*T. chilonis*	
Tomato	13	11	84.61	*T. achaeae*	Himachal Pradesh
Euonymus	17	12	70.59	*T. achaeae*	
Tomato	3	3	100.0	*T. chilonis*	Odisha

boeticus laid on sunn hemp sampled from Tamil Nadu, while in Karnataka it was recorded along with *Tr. bactrae* on the same host. The indeterminate species of *Trichogramma* were also recorded under natural condition parasitizing the eggs of invasive, *Spodoptera frugiperda* (J.E. Smith) infesting maize in Karnataka. Furthermore, *Trichogramma* sp. were recorded on the eggs of mango butterfly, *Euthalia aconthea* Cramer with a 30.77% rate of parasitism in mango and on lepidopteran eggs on *Dolichos* bean with 82.25% parasitism. The species *T. danausicida* parasitized the eggs of *Danaus* sp. (57.48%) on milkweed, *Calotropis* sp.

The *Trichogramma* species, *viz*., *T. chilonis*, *T. achaeae* and *Tr. bactrae* were trapped in the sentinel trap card of *C. cephalonica* placed in different habitat from Karnataka, Himachal Pradesh and Odisha (Table 2). However, the successful parasitism of sentinel trap cards was observed in vegetable crops, *viz*., tomato, cabbage and eggplant. Besides, vegetable crops, parasitism of *Trichogramma* in the sentinel trap cards was observed in the *Euonymus*, an ornamental plant used for sampling. In cabbage field, 3-22 eggs on sentinel trap card were parasitized by *T. chilonis* and *Tr. bactrae*. *Trichogramma chilonis* appeared to be more frequently occurring species compared to *Tr. bactrae* under sampled conditions. The percentage of trichogrammatids that emerged from sentinel trap cards in cabbage was 75.0-100%. Further, sentinel trap cards placed in tomato were parasitized by *T. achaeae* and *T. chilonis* and both the species occurred twice during collection. The eggs parasitism of sentinel cards were in the range of 7-35 eggs by *T. achaeae* and 5-29 eggs by the *T. chilonis* recorded from Karnataka. The species of *T. achaeae* was trapped in sentinel cards placed in tomato and *Euonymus* plants from Himachal Pradesh with an 84.61 and 70.59% adult emergence. In organic tomato cultivation from Odisha, *T. chilonis* was collected through sentinel trap cards.

The natural parasitism of diverse trichogrammatids on the lepidopteran eggs has been observed under different cropping systems (Souza *et al.,* 2016). In the present study, survey to collect trichogrammatids was carried out in seven states, and natural parasitism consisted of 11 species, of which, five species were observed parasitizing the lepidopteran eggs infesting 12 plant species out of 28 plant species that were sampled for lepidopteran eggs or trapped in sentinel egg cards in the agricultural, vegetables and fruits crops grown under diverse cropping conditions. Previously, the natural parasitism of *Trichogramma* species was reported on lepidopteran eggs sampled from eight crops (Querino *et al.,* 2016; Jalali *et al*., 2018), and the seven crops through sentinel trap cards (Suroshe *et al.,* 2015) in the diversified cropping system. Besides, the lepidopteran eggs which were obtained from wild plants had a higher rate of parasitism by *Trichogramma* species in undisturbed habitats. The species, *T. chilonis* was recorded more frequently either though natural parasitism or trapped in sentinel cards in vegetable crops. In earlier studies,

dominant behaviour of *T. chilonis* (up to 80.0% parasitism) and *T. achaeae* has been recorded on eggs of *H. armigera* infesting tomato (Manjunath, 1970). Our survey revealed the natural parasitism of *T. achaeae* and *Tr. bactrae* on an invasive pest, *T. absoulta* in an open tomato fields. These species are mainly used in biological control of *T. absoluta* in the South American and European countries (Cabello *et al.,* 2009; Virgala and Botto, 2010). The emergence of *T. achaeae* and *Tr. bactrae* from the eggs of *T. absoluta* indicates that these parasitoids naturally inhabit the tomato crop. This further adds the importance of *Trichogramma* species in the natural biological control of lepidopteran pest populations under diverse cropping system. In addition, the natural parasitism of *Tr. armigera* and *Tr. bactrae* on the eggs of *L. boeticus* laid on either cultivated or wild relatives of genus *Crotalaria* collected from undisturbed habitat had higher natural parasitism by the trichogrammatids. The bright yellow flower of *Crotalaria* attracts these parasitoids to parasitize the eggs of *L. boeticus* and conserve these egg parasitoids. Furthermore, in the present study, about 25.0% eggs of *H. armigera* were naturally parasitized by *Tr. armigera* compared to earlier reports (11.8%) on tuberose from Karnataka (Manjunath, 1972). The natural parasitism of *Trichogramma* sp. was recorded on *Spodoptera frugiperda* (J.E. Smith) and *E. aconthea* infesting maize and mango, respectively and an occurrence of *T. chilonis* from the eggs of *L. boeticus* laid on *Cycas* indicate the ability of trichogrammatids to parasitize variety of lepidopteran eggs under different habitats. On milkweed *Calotropis*, the eggs laid by *Danaus* sp. were naturally parasitized by *T. danausicida* in undisturbed habitats. The existence of *Trichogramma* species has been reported from undisturbed forest habitats (Querino and Zucchi, 2003; Macedo-Reis *et al*., 2013).

The natural occurrence of indigenous trichogrammatid indicate that, these parasitoids are well adapted to local climatic conditions, and are able to survive in diverse cropping system. There activity helps to manage the build-up of lepidopteran pests through natural biological control in various economic agricultural and horticultural crops in different cropping conditions. This further provides information on the species inhabiting in specific environment which may be consider as potential parasitoid for biological control or inclusion of them in integrated pest management of major lepidopteran pests.

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