

Trichogrammatid egg parasitoids (Hymenoptera: Trichogrammatidae) of rice leaf folder and cabbage caterpillars in Sri Lanka

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Abstract: Use of egg parasitoids is an effective option available for managing lepidopteran pest species through augmentation and release of the parasitoids, particularly the locally available species. As the local egg parasitoids in Sri Lanka are poorly studied, an investigation was conducted to identify the egg parasitoids associated with rice leaf folder (*Cnaphalocrocis medinalis* Guenee) and cabbage semilooper (*Trichoplusia ni* Hubner). The host eggs were collected in different geographical locations for different durations in rice (*Oryza sativa* L.) and cabbage (*Brassica oleracea* L.) fields. The eggs were incubated until the emergence of host larvae or the adult parasitoid. Emerged parasitoids were preserved in 70% ethanol and subsequently dissected for male genitalia and the permanent slides were prepared using Canada balsam as the mounting media. Identification of specimens was done using taxonomic keys and related literature. Identities were confirmed by the Natural History Museum, London. Two species namely, *Trichogramma chilonis* Ishhi and *T. achaeae* Nagaraja and Nagarkatti were found parasitizing *T. ni* at Dodangolla and Thalatuoya regions in Sri Lanka. The record of *T. achaeae* is the first country report and *T. chilonis* had been recorded previously in Sri Lanka. The morphology of male genitalia was described and compared with the original description. *Trichogrammatoidea bactrae* Nagaraja and *T. nana* Zehnter were found parasitizing *C. medinalis* eggs at six sampling sites. The male genitalia of these two species were characterized and compared with the original descriptions.

Keywords: Egg parasitoids, *Trichogramma*, *Trichogrammatoidea*, Sri Lanka

Introduction

Biological control of insect pests is a viable alternative to insecticides, as insecticides cause many problems on the environment and human health (Holt and Hochberg, 1997). The aim of the biological control is to suppress pest populations below economic injury levels (Bale et al., 2008). *Trichogramma* spp. (Hymenoptera: Trichogrammatidae) is a well known group of egg parasitoids commonly used to control many insect pests in the world (Li, 1994; Smith, 1996; Doyen and Bovini, 2005). These parasitoids attack insect eggs, mainly those of Lepidoptera (Hassan, 1994) and thus, the control of pest is possible before the larvae damage the crops.

Availability and potential use of some *Trichogramma* species in Sri Lanka have been studied by several workers (Jardine, 1918; King, 1932; De Silva, 1960; Rajapakse

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and Kulasekara, 1980; Ketipearachchi and Paranagama, 1993; Galanihe *et al.*, 2003). Ten species of trichogrammatids belonging to five genera have been recorded (Noyes, 2012) in Sri Lanka however, their diversity could be much higher than the currently known. Biological control of insect pests is not a widely used tactic in Sri Lanka, except in certain crops through classical biological control.

Rice leaf folder and cabbage caterpillar complex causes severe damage to the rice and vegetable cultivations, respectively, and control of these pests is mainly by using insecticides. Biological control approaches are poorly used on these pests, as the required information is not available (Ahangama and Gilstrap, 2007). Therefore, field surveys and laboratory studies were conducted in rice (*Oryza sativa* L.) and cabbage (*Brassica oleracea* L.) growing areas of the mid country of Sri Lanka with the objective of investigating available species of egg parasitoids and their distribution to be used in augmentation and release programs.

Materials and Methods

This study was conducted during December, 2011 to August, 2013 based in the Department of Agricultural Biology of the Faculty of Agriculture, University of Peradeniya, Sri Lanka. The eggs of rice leaf folder *Cnaphalocrocis medinalis* (Guenee) (Lepidoptera: Pyralidae) were collected at weekly intervals over different durations in thirteen selected rice (*Oryza sativa*) fields in Kandy, Anuradhapura and Kurunegala districts of Sri Lanka. Simultaneously, the eggs of cabbage caterpillars were collected over four-five weeks at weekly intervals in two cabbage (*Brassica oleracea* L.) fields at Thalatuoya and Dodangolla in the Kandy district. The host eggs were removed from plants with a leaf piece using a cork borer (10 mm diameter). The eggs were reared individually in plastic vials (2.5 cm diameter, 5.5 cm height) at room temperature and incubated until the emergence of parasitoid adults or the host larvae.

The emerged parasitoid adults were preserved in 70% ethanol for further taxonomic studies. The parasitoid adults were dissected, and permanent slide mounts were prepared for identification as per the method proposed by Platner *et al.* (1999). Morphology of male genitalia was examined using compound microscope (400 x, Nikon, Japan) and compared with the original species description and taxonomic keys (Nagaraja and Nagarkatti, 1969; Nagarkatti and Nagaraja, 1971; Nagaraja, 1978). The specimens were compared with the type specimens located at the Natural History Museum, London and the identities were further confirmed by DNA analysis done at the Commercial Identification Service, California, USA.

Results and Discussion

Egg parasitoids of cabbage caterpillar *Trichoplusia ni* Hubner, 1800-1803 (Lepidoptera: Noctuidae)

Two species of egg parasitoids namely, *Trichogramma achaeae* and *T. chilonis*, were identified from the samples collected in the study area.

***Trichogramma achaeae* Nagaraja and Nagarkatti, 1969**

Materials examined: Sri Lanka, Kandy District, Thalatuoya (7° 14' 55.42" N, 80° 40' 53.52" E). 8 Males, 14 IX 2012, host: *Trichoplusia ni* (Hubner), host plant: cabbage; Sri Lanka, Kandy District, Dodangolla (7° 17' 32.35" N, 80° 42' 11.39" E), 5 Males, VII 2012, host: *T. ni*, host plant: cabbage.

Morphology: Adults are 0.5 mm long and 0.15 mm wide across the head. Males are yellow with blackish sides on pronotum, mesonotum, abdomen, hind coxa and femur. An antenna with long hairs, the longest is being nearly 2.5 x maximum width of flagellum. Dorsal expansion of gonobase is prominent and well chitinized, which is subtriangular and apically blunt (Figure 1). Adeagus is about 1.5 times longer as the apodermes. Chelate structures in level of tips of gonoforceps. Median ventral projection is minute and inconspicuous. Minute protrubences present at base of chelate structure, one on either side of median ventral projection.

Trichogramma achaeae is found in Asia, Europe, Africa and South America. In Asia, it was reported in China, India and Russia (Polaszek, 2010). This is probably the first record on the presence of *T. achaeae* in Sri Lanka. It parasitizes mostly the lepidopteran insect species. Polaszek (2010) reported 25 lepidopteran species, including *T. ni*, belonging to 23 genera in 11 families.

***Trichogramma chilonis* Ishhi, 1941**

Materials examined: Sri Lanka, Kandy District, Thalatuoya (7° 14' 55.42" N, 80° 40' 53.52" E) 10 Males, 14 IX 2012, host: *Trichoplusia ni*, host plant: cabbage. Sri Lanka, Kandy District, Dodangolla (7° 17' 32.35" N, 80° 42' 11.39" E) 7 Males, VII 2012, host: *Trichoplusia ni*, host plant: cabbage.

Morphology: Males yellow with blackish abdomen and mesoscutum. Antennal hairs sharply tapering and moderately long, longest being nearly 2.5 times the maximum width of flagellum. Fringe on tornus of forewing about one-sixth width of wing. Genitalia is triangular, with very prominent, lateral lobes; chelate structure markedly below the level of gonoforceps; median ventral projection prominent, broad at base; extending anteriorly to about two-thirds of length of genitalia (Figure 1); aedeagus as long as apodemes, both together slightly shorter than hind tibia.

Trichogramma chilonis is an Asian species found in Bangladesh, China, India, Indonesia, Japan, Malaysia, Nepal, Pakistan, Phillipines, Taiwan, Thailand, Vietnam (Polaszek, 2010), Sri Lanka and Papua New Guinea (Nagaraja and Nagarkatti (1971). It has been introduced to different places of Europe, Africa and North America as a biocontrol agent. *Trichogramma chilonis* widely uses lepidopteran eggs as its host including 127 species belonging to 92 genera and 22 families. In addition, two dipteran species from two families and one neuropteran species were reported as hosts (Polaszek, 2010).

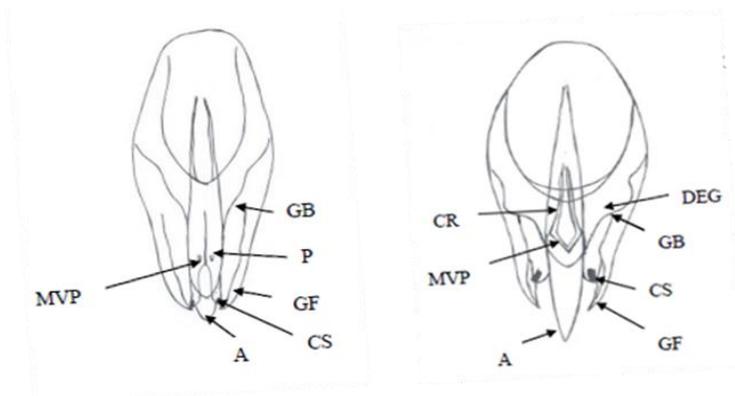


Figure 1. Male genitalia of *Trichogramma achaeae* (left), *T. chilonis* (Right); A - Aedeagus, CS - Chelate structure, GB - Gonobase, GF - Gonoforceps, MVP- Median ventral projection, P - Protuberances.

Egg parasitoids of Rice leaf folder *Cnaphalocrocis medinalis* (Guenee) (Lepidoptera: Pyralidae)

Two parasitoid species namely, *Trichogrammatoidea bactrae* Nagaraja, *T. nana* Zehntner, were identified parasitizing the eggs of rice leaf folder eggs. Both the species were recorded for the first time from Sri Lanka.

Trichogrammatoidea bactrae Nagaraja, 1979

Material examined: Sri Lanka, Kandy district, Megoda Kalugamuwa (7° 17' 4.05"N, 80° 38' 14.85"E), Hindagala (7° 14' 12.29"N, 80° 36' 12.78"E), Ihalawela (7° 17' 55.31"N, 80° 43' 13.27"E), Panideniya (7° 15' 15.97"N, 80° 35' 20.31"E), Ganegoda (6° 41' 45.03"N, 80° 23' 29.80"E) and Wathurakumbura (7° 17' 4.05"N, 80° 38' 14.85"E). Sri Lanka, Anuradhapura District, Mahalluppallama (8° 2' 31.57"N, 80° 35' 37.80"E).

Morphology: Head is light dull reddish yellow; eyes and ocelli are red; antenna is light dusky brownish grey colour. Pro- and mesothorax, scutellum and metanotum are dull yellowish brown to black. Legs yellowish brown, mid and hind femora greyish brown. Abdomen is with black tergites and ventral side is lighter. Antennal flagellum is about 1.75 × long as scape. Flagellum with 22-26 long hairs, the longest being nearly 3 × as wide as flagellum. The anterior rigid part of the forewing is narrow, its width is less than the length of wing with long setae; fringe setae long, almost uniformly long from apical angle to tornus, longest setae is about half the width of wing. Hind wing width with fringe setae as long as in fore-wing.

Male genitalia (Figure 2) is narrow, nearly 3 × as long as wide. Apex and base indistinctly are tapering, median cleft slightly less than one fifth of the length of genitalia. Median ventral projection is not conspicuous; lateral tubercles are distinct; central ridges are paired and short. Chelate structures are large, slightly below the level of gonoforceps. Aedeagus and apodemes are nearly equal length.

Trichogramma bactrae is reported to parasitize the eggs of major lepidopteran insect pests such as *Trichoplusia ni* (Hubner), *Chilo infuscatellus* Snellen, *C. partellus* Swinhoe, *Helicoverpa armigera* (Hubner).

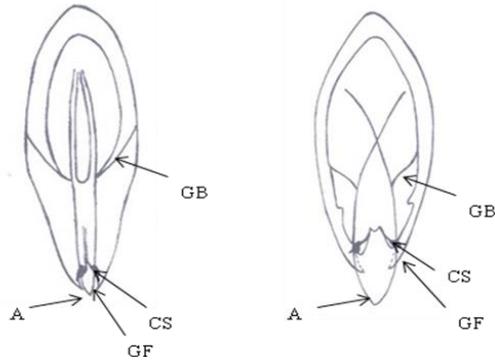


Figure 2. Male genitalia of *Trichogrammatoidea bactrae* (left), *T. nana* (Right); A - Aedeagus, CS - Chelate structure, GB - Gonobase, GF - Gonoforceps

***Trichogrammatoidea nana* Zehntner, 1896**

Material examined: *Trichogrammatoidea nana* was found only in Penideniya, Kandy district (7° 15' 15.97"N, 80° 35' 20.31"E) out of 13 sampling sites.

Morphology: General body colour is orange yellow. Eyes and ocelli are bright red. Pronotum, mesoscutum and anterior part of abdomen are light dusky brownish grey. Antenna with flagellum is slightly more than 1.5 × scape length; pedicel is 0.33 × as long as the scape; flagellum is with 22-26 long hairs; longest being 3.0 × the maximum width of flagellum; flageller segments are slender. Forewing is somewhat broad; width is slightly less than 0.5 × the length, outer margin is broadly rounded; setae of anterior rigid part are somewhat sparse and long; basal infuscation is in line with stigma. Fore-wing is with fringe setae; length 0.33 × of its width; the setae on tornus somewhat longer.

Male genitalia (Figure 2) width a little more than 0.33 × the length, with tapering base and perceptible dorsal connecting membrane. Aedeagus is longer than apodemes. Chelate structures is far below the gonoforceps. Median ventral projection is distinct but, short with indistinct tubercles (Figure 2). *Trichogrammatoidea nana* parasitizes on eggs of many micro-lepidopteran insects, such as *Corcyra cephalonica* and *Ephestia cautella* (Kazmi and Chauhan, 2003). The eggs of these stored pests thus, could be use as hosts in parasitoid mass-rearing programs.

Revised *Trichogrammatidae* species list in Sri Lanka

With the findings of this study, the total number of *Trichogrammatidae* in Sri Lanka increases to 13 under five genera. The revised list is given in Table 1. The egg parasitoids collected during this study belonged to two genera namely,

Trichogramma and *Trichogrammatoidea*. Of them, genus *Trichogramma* is the predominant group (250 spp. globally), which has been used in many bio-control programs worldwide. *Trichogrammatoidea* is less abundant (24 spp. globally) and only three species of this genera are found in Sri Lanka.

Table 1. The revised species list of *Trichogrammatidae* in Sri Lanka, based on the present study

Family: <i>Trichogrammatidae</i>
<i>Mirufens ceylonensis</i> Viggiani
<i>Oligosita staniforthii</i> Westwood
<i>Oligosita subfasciata</i> Westwood
<i>Trichogramma australicum</i> Girault
<i>Trichogramma achaeae</i> Nagaraja and Nagarkatti, 1969
<i>Trichogramma chilonis</i> Ishhi, 1941
<i>Trichogramma erosicorne</i> Westwood
<i>Trichogramma evanescens</i> Westwood
<i>Trichogramma minutum</i> Riley
<i>Trichogrammatoidea bactrae</i> Nagaraja, 1979
<i>Trichogrammatoidea nana</i> Zehntner
<i>Trichogrammatoidea nodicornis</i> Westwood
<i>Ufens mezentius</i> Owen

Conclusion

Four species of egg parasitoids belonging to family *Trichogrammatidae* were collected from Sri Lanka, of which three species are new country records. There is an immense potential for mass rearing in laboratories and subsequent field release of these species for effective biological control of the targeted pest species.

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