

Effect of host plants on the life-history traits of *Trichogramma chilonis* (Ishii) at different constant temperature

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Abstract: The pod borer, *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae), is considered to be the most important pest of several crops. In northeastern Uttar Pradesh, it is a major problem on chick pea (*Cicer arietinum*), Pigeon pea (*Cajanus cajan*), and Tomato. *Trichogramma chilonis* Ishii (Hymenoptera: Trichogrammatidae) and *Campoletis chlorideae* Uchida (Hymenoptera: Ichneumonidae) are tiny wasp, and considered as egg and larval parasitoid respectively, of several lepidopteran host including *H. armigera*, and are widely used in biological control. The aim of the present study was to control the pest population of *H. armigera* by using these parasitoids. Five districts of northeastern Uttar Pradesh, namely Gorakhpur, Kushinagar, Deoria, Mahrajganj and Sidharthnagar, was surveyed for the incidence of *H. armigera* on different crops such as chick pea, Pigeon pea and corn. The results revealed that pod borer was a major pest of chick pea and pigeon pea in northeastern Uttar Pradesh. Two parasitic wasp, *T. chilonis* and *C. chlorideae* were naturally occurring, also noted the high rate of parasitism in the field on crops under study. These findings can be used in the control of *H. armigera* on crops where natural parasitism seemed to be high.

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Introduction

The American bollworm, *Helicoverpa armigera* (Hübner) (Lepidoptera: Noctuidae) is a pest of major importance in most areas wherever it occurs, damaging a wide variety of horticultural agricultural and crops. This pest is regarded as number one among the ten worst pests all over the world and causes economic losses (Pandey *et al.*, 2000). In Uttar Pradesh, India, it is one among major biotic constraints for the cultivation of pulse crops (Pandey *et al.*, 2004; Pandey and Tripathi, 2008; Pandey *et al.*, 2009). The ecological and physiological features like direct attack on fruiting structures, voracious feeding, high fecundity, multi-voltinism, occurrence of overlapping generations and ability to diapause during unfavorable conditions has made this pest a 'bugbear', particularly in Eastern regions of this state. On average a 30% crop loss is reported. This can be witnessed from progressive decline in area and production of chickpea during the past three decades. While area has gone down from 17.26 lakh ha in 1975-76 to 7.40 lakh ha in 2005-06, the production has decreased from 12.50 lakh tonnes to 6.61 lakh tonnes (DACNET).

Chemical pesticide application is the most commonly used method against *Helicoverpa* in this region, but it cannot wipe *Helicoverpa* out since, it easily develops resistance to applied chemicals including pyrethroids (Patel and Koshiya, 1999). The

potential of applied chemicals is also screened off as the grown up *Helicoverpa* larvae mostly feed upon developing grains inside the pods. In addition, large quantities of persistent insecticides are raising concerns about applicator safety, environmental contamination and possible deleterious effects on non-target animals and humans.

As an alternative, biological control is generally perceived as providing both long-lasting insect control and having less potential for damage to the environment and non-target organisms than chemical interventions. The use of bacterial (e.g., *Bacillus thuringiensis*) and viral insecticides (e.g., NPV) for the control of *H. armigera* is the most recent achievement in biological plants protection (Grzywacz, 2001). However, at present the application of microbial pesticide in India is negligible and worldwide still present around 1% of crop protection. Undoubtedly, they are potential biocontrol tools for managing *Helicoverpa* population on chickpea; resistance to biorationals is likely to evolve rapidly unless they are used as part of coherent resistance management strategy.

The use of genetically modified (GM) crops that express insecticidal genes, such as those derived from the soil bacterium *Bacillus thuringiensis* (Bt), have opened new endeavors to control insect pests of agricultural crops. As with cotton, the expression of *B. thuringiensis* cry genes is an option to protect

chickpeas from damage by *H. armigera* (Romeis *et al.*, 2004). However, transgenic chickpea varieties that express either Cry₁Ac or Cry₂Aa, or both proteins, are currently under development and could become commercially available in the future (Sanyal *et al.*, 2005; McPhee *et al.*, 2007). It is therefore, an urgent need to devise alternative control measures, which can be applied to control this dreaded pest on chickpea in an economically and ecologically agreeable manner.

Biological control of insect pests utilizing parasitoids has been advocated as one of the most recent techniques and unlike other control measures its effect is permanent, ecologically non-disruptive, self-sustaining and after the initial costs involving investigations and release, the recurrent costs are nominal.

For successful biological control program of insect pests, the parasitoids should be synchronized with its host (Weeden and Hoffman, 2001). For example, if it is an endolarval parasitoid, sufficient number of hosts must be present in the management area at the time of its best reproductive fitness. Any delay in the availability of hosts may strongly affect their reproduction and survival. Host shortage is likely to occur in nature, reducing the efficiency of parasitoid species. None of the parasitoids can manage the fecundity and progeny sex ratio, if it is deprived of host for a long time. However, a parasitoid which can tolerate longer host deprivation times would be considered good. These aspects are highly important for the implementation of an efficient mass rearing program as well as in the inundative release of the parasitoid species.

The ichneumonid *Campoletis chlorideae* Uchida is a common parasitoid of the pod borer, *Helicoverpa armigera* (Hübner) on chickpea crop in India (Thakur *et al.*, 1995; Durairaj, 1999). It is an arrhenotokous, idiobiont parasitoid species, which effectively parasitises the second instar larvae of *H. armigera*, both at vegetative and fruiting stages of the chickpea crop. Previous studies revealed that *C. chlorideae* may be considered as a promising alternative to the exploitative and disruptive chemical control measures against *H. armigera* on chickpea in Eastern Uttar Pradesh, India (Pandey *et al.*, 2004; Pandey and Tripathi, 2008; Pandey *et al.*, 2009). However, before any attempt is made to mass rear and release this parasitoid, the factors that may affect their reproduction and survival must be understood. In this regard one important factor to be considered is host deprivation.

Materials and Methods

The age specific life table statistics of parasitoid *T.chilonis* was determined at five

parasitoid densities (1, 2, 4, 8 parasitoid/days). The newly emerged mated and well fed female parasitoids of the same cohort were introduced into marked wooden cage (30×30×45 cm) having young potted pigeon pea, chick pea and tomato with almost 200 *H.armigera* eggs. The female were introduced singly into the cages for 24 h.

A small sponge piece soaked in 30% honey solution was available as food for parasitoid. After every 24 h, the exposed eggs along with the host plant were replaced by flesh ones throughout the life of female parasitoid. The cages were illuminated by two 40 w flourosent lamps for 14 hour. The cages were sprayed with water from an atomiser at least once a day to maintain the proper female parasitoid (150 for all the four parasitoid density x three host plant) was used. The parasitized eggs when turned black that picked off together with part of leaf and were put into marked tube (1×5 cm) each having a moist filter paper at their bottoms. The emerged parasitoids were sexed and counted.

Culture of the host

The field collected larvae were transferred singly with help of a small camel hair brush into glass vials (10 x 3.35 cm) having moistened filter paper at their bottoms. The mouth of glass vials was plugged with absorbent cotton. Fresh and green leaves and pods of chickpea were provided as food for the host larvae and were reared until pupation. After pupation, the pupae were transferred to the fresh sterilized glass vials having moistened filter paper at their bottoms. Emerging adults were provided a 30% honey solution as food.

For the culture of *H. armigera*, a couple of adults were kept together in a beaker (1000 ml) until mating was observed. Moistened filter paper was kept at the bottom of the beaker to provide humidity inside it. A strip of muslin cloth was hung inside to provide rest to the flying moth when needed. The mouth of the beaker was covered by a muslin cloth. The mated females were then removed from the beaker and introduced into the small marked wooden cages (45x50x60 cm) containing potted young plants of chickpea. A piece of sponge soaked in 30% honey solution was kept in each cage as food and was changed daily. The eggs deposited each day on the leaves and pods of host plant, were transferred to the marked beakers (250 ml) and kept until hatching. The larvae were then collected in glass tubes (10x 3.25 cm). For the culture of hosts of a known age, only newly hatched first instar larvae were allowed to remain in the beaker and the rest were removed. Second instar larvae, which are most preferred by the parasitoid were collected from the maintained culture and were utilized as hosts for the experiments.

Culture of the parasitoid

The field collected cocoons of the parasitoid were transferred singly with a small camel hair brush into glass vials (10x 3.35 cm), each having moistened filter paper at their bottoms. Adults emerging from the cocoons were then fed 30% honey solution *ad libitum* for 2-4 hrs. Thereafter, the female and male parasitoids were put together in a glass tube (10 x 3.25 cm) until mating was observed (2-6 hours). The males then were removed from glass tubes. The mated females were introduced into the small marked wooden cages (45 x 50 x 60 cm) having potted young plants of chickpea and about 100 healthy second instar host larvae. A small piece of sponge soaked in 30% honey solution, was placed into the each wooden cage as food for the parasitoids. After parasitisation, the parasitoids were removed and the host plants were placed into cages (30 x 30 x 40 cm) for further development. The potted plants were examined daily for cocoon formation. The cocoons were then collected and transferred singly into the marked sterilized glass vials. After adult emergence, the number of each sex was determined.

To observe the effects of host deprivation times on the reproduction and survival, a couple of adults virgin male (M) and female (F) *C. chloridae* were obtained from culture and kept together into separate tubes (Ca. 1x10 cm) until the mating was observed. Now, mated females, deprived of hosts for 0, 1, 3 and 5 days were introduced into four separate wooden cages (Ca. 45 x 50 x 60 cm), each having potted young plants of chickpea and about 100 healthy second instar host larvae for parasitisation. After every 24 h the females were removed from their respective cages and after proper feeding and rest, re-introduced in other similar cages having potted young plants of chickpea and about 100 healthy second instar host larvae throughout their life. The exposed host larvae were placed in other cages and examined daily. As soon as the parasitised larvae transformed into cocoons, they were counted and transferred separately with a part of the leaf to glass vials (1 x 5 cm) having moist filter paper at bottom. Upon adult emergence, the number of each sex was determined (Pandey *et al.*, 2009).

Calculation of Life Table Statistics

Under constant environment conditions the growth rate of a population can be used to demonstrate the relative measure of exponential growth and to conceptualise the relationship between demographic variables and population growth, which is more or less constant and than the population assumes a stable age composition (Birch, 1948; Pressat, 1985). For such a situation, the growth rate of the population can be calculated directly from the

vital statistics of the age specific survival and net fecundity rates under natural condition which is fault as “intrinsic rate of increase (rm)”. the value of rm under optimum condition indicates the maximum biological potential of the population and growth in that situation.

Results

Progeny sex ratio

The data related with the influence of parasitoid density on the number of progeny yield as well as on the progeny sex ratio.

The progeny sex ratio was calculated as proportion of males in the progeny population. They showed significant effect of both the parasitoid density and host plants food (chickpea, pigeonpea, tomato) on the progeny sex ratio of the parasitoid *T. chilonis*. The regression analysis of the progeny sex ratio on the parasitoid density yielded significant relationship coefficients are significant (PSR_(Pigeon pea) = 0.444+0.014 X, r = 0.989, P < 0.001; PSR_(Chick pea) = 0.474+0.020 X, r = 0.981, P < 0.001; PSR_(Tomato) = 0.538+0.21 X, r = 0.972, P < 0.001) (Fig-1). We observe that alimited supply of the hosts with increasing number of parasitoids always increases the production of male progeny in the population irrespective of host plant used. It shows that to increase the proportion of female progeny in the population, sufficient number of hosts should be made available for the parasitoid.

The data of progeny sex ratio obtain from the eggs laid during subsequent days after emergence revealed are progressive increase in the sons population in progeny, irrespective of host density variation. It implies that the probability of deposition of daughter producing eggs (diploid eggs) decreased on the successive days of oviposition.

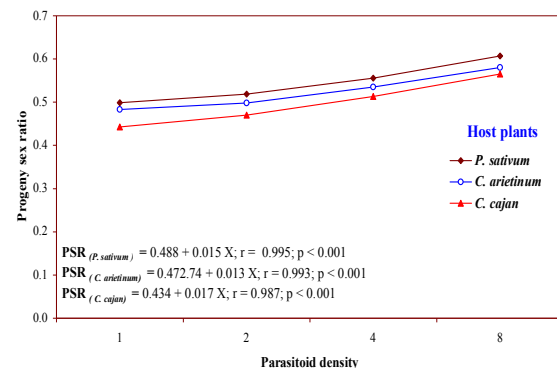


Figure 1. Effect of host plants on progeny sex ratio traits of *Trichogramma chilonis* (Ishii) at different constant temperature

Discussion

Figure shows that sex ratio decreased with increase of parasitoid density, however, it is still significantly more than the Fisherian ratio ($P < 0.001$) (Fisher, 1930). The Fisher's model predicts that, in panmictic population, investment in the production of male and female should be equal, however selection for sex ratio other than 0.5 may arise if the assumptions underlying Fisher's model do not apply.

In arrhenotokous wasps like *T. chilonis* (where males develop from unfertilised eggs and females from fertilised eggs), regulation of the release of sperms from the spermatheca may, therefore, control sex ratio (Flanders, 1939). The sperm release from the spermatheca is influenced by several extrinsic and intrinsic factors (Sinha and Singh, 1979; Islam and Copland, 1997). The decrease of sex ratio with increase of parasitoid density may be explained on the following accounts: (1) increase of parasitoid density increase the rate of superparasitism (van Alphen and Nell, 1982; Hofsvang and Hågvar, 1983; Dhiman and Kumar, 1987) in that situation the second female lays more male eggs (Wylie, 1976; Werren, 1980); (2) parasitised hosts provide less sources for larval development than healthy hosts (Waage and Lane, 1984; Tripathi and Singh, 1991b; Pandey and Singh, 1997; Honek *et al.*, 1998) and males because of their lower nutritional requirements (Charnov, 1982) fare relatively better (Narayanan and Subba Rao, 1955; Wilkes, 1963; Waage and Ng, 1984; Hardy, 1992) and (3) parasitising females, and change their sequence of sex allocation (Waage, 1986). Stimulus for this change may have been contact with traces of their female, by physical jostling of female by other individuals (Sinha and Singh, 1980a) or encounter of parasitised hosts (Singh and Sinha, 1981; Islam and Copland 1997;). Frequent contacts with conspecific females lead to male-biased sex ratio (Singh *et al.*, 2001c; Singh *et al.*, 2002). The physical encounters with conspecific females and/or their odour was observed to induce haploid oviposition producing male progeny (Decker *et al.*, 1993; Biswas and Singh, 1995b). Biswas and Singh (1995c) opined that by having feminine stimuli (female odour) the females somehow 'estimate' the density of conspecific females in her vicinity and respond for optimal progeny sex ratio by increasing the male progeny in the population. The concentration of pheromones may help in estimating the number of males in the ambient environment of the ovipositing females. However, King (1989) and Wylie (1976) could not observe such effect of female odour on the progeny sex ratio in other groups of the parasitoids such as in case of *Spalangia cameroni* and

Nasonia vitripennis (Hymenoptera: Chalcidoidea: Pteromalidae).

The results discussed so far demonstrate that the parasitoid densities influence the progeny yield as well as cause significant variation in the progeny sex ratio. The inversely female-density dependent progeny sex ratio indicates that for procuring maximum female progeny in the population for laboratory work or for mass culture, the ratio of female parasitoids to the hosts should not be limited. There should be about 100 hosts per female parasitoid in the mass culture program.

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