



Effect of high temperature shocks on performance of *Trichogramma chilonis* Ishii (Hymenoptera: Trichogrammatidae)

RAVINDER SINGH and PALA RAM*

Department of Entomology, CCS Haryana Agricultural University

Hisar 125 004, Haryana, India

E-mail: pala_ram@rediffmail.com

ABSTRACT: Key biological characteristics of *Trichogramma chilonis* Ishii, which govern its performance in the field were studied by exposing pupae and the adults to different temperatures ($30 \pm 1^\circ\text{C}$, $33 \pm 1^\circ\text{C}$, $36 \pm 1^\circ\text{C}$, $39 \pm 1^\circ\text{C}$, $42 \pm 1^\circ\text{C}$ and $45 \pm 1^\circ\text{C}$) keeping 80 ± 5 per cent relative humidity daily for six hours. Temperature shocks at $\geq 33 \pm 1^\circ\text{C}$ resulted in decrease in adult emergence. Fecundity decreased when the parasitoid was exposed at $\geq 30 \pm 1^\circ\text{C}$ from day 1 and day 2; at $\geq 33 \pm 1^\circ\text{C}$ from day 3 and at $\geq 42 \pm 1^\circ\text{C}$ on day 4 of pupae formation. Female longevity decreased when the parasitoid was exposed at $\geq 36 \pm 1^\circ\text{C}$ from day 1 or day 2 and at $\geq 42 \pm 1^\circ\text{C}$ from day 3 or day 4 of pupae formation. Temperature shocks of $45 \pm 1^\circ\text{C}$ initiated from day 1 of pupae formation proved deleterious to the parasitoid resulting in low adult emergence (24.88%), low fecundity (26.50 egg hosts parasitized/female) and decreased female longevity (8.45 days). When females reared at $27 \pm 1^\circ\text{C}$ were exposed to different high temperatures, significant ($P = 0.05$) decrease in fecundity and female longevity was observed as compared to control. No effect of high temperature exposure on the sex-ratio in the progeny was observed. Results of the present study may help in developing augmentative release strategies of this parasitoid against lepidopterous pests in cotton by devising suitable release techniques.

KEY WORDS: Biological characteristics, high temperature exposure, *Trichogramma chilonis*

INTRODUCTION

In India, Trichogrammatid egg parasitoids are used as bioagents to control lepidopterous pests in different crops *viz.*, cotton, rice, sugarcane, tomato, castor, etc. (Singh, 2001; Ram *et al.*, 1997, 2002). The inundative releases of *Trichogramma* species against insect pests in the field are made in pupal or adult stage. Evaluation of the efficacy of augmentative releases as well as development of application techniques require a thorough

understanding of several factors that may affect the survival of released parasitoids. The most significant factors affecting the inundative releases are the number of parasitoids released, time of releases and local weather conditions (Smith *et al.*, 1986). For *Trichogramma* species, the influence of environmental factors has been most intensively studied. Researchers have been interested in the influence of high field temperatures on the parasitoid attributes such as longevity, fecundity, adult emergence, sex-ratio, host acceptance, etc.

*Part of M. Sc. thesis of the first author, submitted to the CCS Haryana Agricultural University, Hisar, Haryana

(Calvin *et al.*, 1984; Gross, 1988; Palvik, 1992; Bouchier *et al.*, 1993). In cotton, ambient air temperatures under Hisar (Haryana) agroclimatic conditions frequently exceed 41°C during parasitoid release periods (August-September), with a crop canopy temperature of 27.3-38.5°C (unpublished data). Since the parasitoid in the insectaries is reared at optimum temperatures its survival under field conditions will depend on its ability to resist inhospitable field conditions, particularly the high temperature. Keeping this in view, the present study was planned to examine the effect of high temperature shocks on key biological parameters of *T. chilonis*.

MATERIALS AND METHODS

Initiation of *T. chilonis* culture

The culture of *T. chilonis* was initiated by collecting and rearing parasitized eggs of sesame hawk moth, *Acherontia styx* Westwood, from sesame crop in Hisar and adjoining areas. The culture of the parasitoid was maintained in the laboratory on the eggs of *Corcyra cephalonica* Stainton at $27 \pm 1^\circ\text{C}$ temperature and 80 ± 5 per cent relative humidity (RH). Relative humidity was maintained in the desiccators using saturated sodium chloride solution.

Exposure to temperature shocks

Since the parasitoid is released in the field in pupal or adult stage, therefore, in order to simulate field conditions temperature shocks of $30 \pm 1^\circ\text{C}$, $33 \pm 1^\circ\text{C}$, $36 \pm 1^\circ\text{C}$, $39 \pm 1^\circ\text{C}$, $42 \pm 1^\circ\text{C}$ and $45 \pm 1^\circ\text{C}$ each at 80 ± 5 per cent relative humidity were given to different aged pupae of *T. chilonis* for six hours (10 AM to 4 PM). Blackening of host eggs parasitized by *Trichogramma* species indicated the initiation of pupal stage of the parasitoid (Dahlan and Gordh, 1996) and, hence, temperature shocks were applied daily to pupae beginning from day 1, 2, 3 or 4 of pupae formation. The adults emerged on 5th day. Similarly, females of *T. chilonis* from laboratory culture were subjected to the temperature shocks daily until death.

For this about 200 fresh, ultra-violet rays treated eggs of *C. cephalonica* were pasted on 25 x 6 mm paper strip of Post it[®] notes by dipping its 6 x 6 mm sticky portion in eggs. The host eggs were then provided to a large number of *T. chilonis* adults (about 500 females) in glass test tubes (25 x 150 mm) for three hours so as to achieve uniform and one time parasitization and it was replicated five times. Different aged parasitoid pupae, in the parasitized host eggs, were given high temperature shocks daily. Observations on adult emergence were recorded when all the adults had emerged. Immediately after the parasitoid adults emerged, 20 mated females from different replications were isolated per treatment in small glass vials (12 x 75 mm). Each female was provided with about 80-100 fresh, ultra-violet rays treated eggs of rice moth pasted on paper strips daily until death. A fine streak of honey on the back of paper strip was provided as food to the female. Similarly, 20 freshly emerged and mated females emerged at $27 \pm 1^\circ\text{C}$, 80 ± 5 per cent relative humidity regime were reared as mentioned above except that these were subjected to high temperature shocks daily until death.

Effect on biological parameters

Observations were recorded on fecundity (no. of host eggs parasitized/ female), female longevity and sex-ratio of progeny of the treated generation and that reared at $27 \pm 1^\circ\text{C}$ (control). To compare reproductive output under ideal ($27 \pm 1^\circ\text{C}$, $80 \pm 5\%$ RH) and simulated conditions (females subjected to high temperature shocks daily until death) in the laboratory, net reproductive rates (R_0), mean generation times (T), intrinsic rates of increase (r_m) and finite rates of increase (λ) were calculated according to the methods of Carey (1993) using mean development time from this study.

The data were subjected to Analysis of Variance (ANOVA) applying single factor Complete Randomized Design (CRD) and means were compared using critical difference (CD) at $p = 0.05$.

RESULTS AND DISCUSSION

A. Exposure of *T. chilonis* to temperature shocks in pupal stage

i. Effect of high temperature shocks on emergence of *T. chilonis*

Emergence of *T. chilonis* adults was noticed in all the treatments, though it was considerably reduced in treatments with high temperature shocks, there being maximum reduction at $45 \pm 1^\circ\text{C}$ (Table 1). Exposure of different age pupae to temperature shocks at $\geq 33 \pm 1^\circ\text{C}$ resulted in significant decrease in adult emergence as compared to control. Younger pupae were more vulnerable to high temperature shocks than the older ones, as there was higher decrease in adult emergence in the 1-day-old pupae as compared to 2, 3 or 4-day-old ones. Similarly, Lopez and Morrison (1980) reported more than 80

per cent reduction in emergence rate in the case of *T. pretiosum* when 5 to 7-days-old parasitized host eggs (parasitoid pupae) were exposed to heat shocks higher than 37°C for four hours. Likewise, Gross (1988) reported that when *T. pretiosum* was exposed to high temperatures between 38 and 43°C for several hours before emergence, emergence rate decreased by more than 70 per cent. Maissonhaute *et al.* (1999) observed 45 per cent pupal mortality of *T. brassicae* in the eggs of *Ostrinia nubilalis* when white pupal stage received two heat shocks at 44°C for six hours.

ii. Effect of high temperature shocks on *T. chilonis* fecundity

Females of *T. chilonis*, exposed to high temperature ($\geq 45 \pm 1^\circ\text{C}$) shocks were still able to parasitize host eggs. Mean lifetime fecundity decreased significantly over control when the

Table 1. Effect of high temperature shocks given to pupae on adult emergence and fecundity of *T. chilonis*

Temperature ($^\circ\text{C}$)	Adult emergence (%) of <i>T. chilonis</i> when heat shocks were initiated at different days of pupae formation					Fecundity of <i>T. chilonis</i> when heat shocks were initiated at different days of pupae formation				
	1	2	3	4	CD ($p=0.05$)	1	2	3	4	CD ($p=0.05$)
$27 \pm 1^*$	99.46 (86.76)	99.46 (86.76)	99.46 (86.76)	99.46 (86.76)	(NS)	207.65	207.65	207.65	207.65	NS
30 ± 1	98.83 (84.00)	99.44 (86.68)	99.37 (85.95)	99.49 (86.84)	(NS)	155.55	159.80	178.65	206.75	39.70
33 ± 1	96.39 (79.16)	97.42 (81.00)	97.83 (81.59)	98.48 (83.13)	(NS)	128.80	131.55	150.20	204.90	33.11
36 ± 1	95.98 (78.72)	97.08 (80.24)	97.78 (81.49)	97.73 (81.37)	(NS)	104.75	117.85	132.40	193.80	40.95
39 ± 1	85.71 (67.81)	93.36 (75.13)	93.39 (75.14)	96.15 (78.76)	(2.41)	82.90	89.35	123.55	168.50	27.30
42 ± 1	78.25 (62.22)	86.42 (68.41)	88.13 (69.98)	88.81 (70.48)	(3.12)	51.10	67.95	77.70	94.80	19.56
45 ± 1	24.88 (29.84)	65.16 (53.84)	74.17 (59.45)	80.32 (63.66)	(3.05)	26.50	48.95	63.20	78.10	17.88
CD ($p = 0.05$)	(3.22)	(3.23)	(2.85)	(2.79)	-	33.30	34.91	35.03	39.75	-

* Constant temperature as control

Figures in parentheses are angular transformed values.

parasitoid was exposed daily at $30 \pm 1^\circ\text{C}$ on day 1 and day 2; at $\geq 33 \pm 1^\circ\text{C}$ on day 3 and at $> 42 \pm 1^\circ\text{C}$ on day 4 of pupae formation (Table 1). Adverse effect of high temperature initiated in the younger pupae was more pronounced than in the older ones. There was a significant decrease in lifetime fecundity when high temperature exposure was initiated in the 1, 2 and 3 days old pupae than in the 4 days old pupae, being highest at $45 \pm 1^\circ\text{C}$ wherein only 26.50 egg hosts were found to be parasitized. Chihrane *et al.* (1993) reported that females of *T. brassicae* exposed to 44°C during pupal stage for six hours were less fecund and the parasitic efficiency was reduced to 50 per cent. Guine and Lauge (1997) also observed the reduction in fecundity of females of *T. brassicae* emerging from the pre-pupae exposed to single high temperature shock at $31\text{--}32^\circ\text{C}$. However, in the present study, no adverse effect on parasitoid fecundity was observed when pupae were given exposure to high

temperature ($\leq 39 \pm 1^\circ\text{C}$) one day before adult emergence.

iii. Effect of high temperature shocks on *T. chilonis* longevity

Females of *T. chilonis* given high temperature shocks at $45 \pm 1^\circ\text{C}$ on day 1 of pupation lived for 8.45 days as compared to 16.75 days in control (Table 2). High temperature shocks of $36 \pm 1^\circ\text{C}$ given daily to pupae starting from the day of pupae formation caused significant decrease in female longevity as compared to that given on 4th day of pupae formation. Temperature shocks at $45 \pm 1^\circ\text{C}$ given daily from the day of pupae formation proved harmful to the parasitoid as it resulted in about 50 per cent reduction in female longevity. Our results are in confirmation with findings of Chihrane *et al.* (1993) who reported that a heat shock of 44°C applied to pupae of *T. brassicae* resulted in reduced longevity of the adults.

Table 2. Effect of high temperature shocks given to pupae on female longevity and proportion of females in the progeny treated generation of *T. chilonis*

Temperature ($^\circ\text{C}$)	Female longevity (days) when heat shocks were initiated at different days of pupae formation					Per cent females progeny when heat shocks were initiated at different days of pupae formation				
	1	2	3	4	CD (p=0.05)	1	2	3	4	CD (p=0.05)
$27 \pm 1^*$	16.75	16.75	16.75	16.75	NS	77.33 (61.73)	77.33 (61.73)	77.33 (61.73)	77.33 (61.73)	(NS)
30 ± 1	14.35	16.70	16.95	17.50	NS	81.00 (64.15)	79.33 (62.95)	77.67 (61.80)	79.67 (63.19)	(NS)
33 ± 1	13.15	14.35	14.85	16.15	NS	75.67 (60.43)	83.33 (65.91)	86.00 (68.06)	74.00 (59.34)	(NS)
36 ± 1	12.25	12.25	14.00	16.05	3.12	74.33 (59.59)	83.67 (66.15)	83.00 (65.65)	73.00 (58.73)	(NS)
39 ± 1	12.15	12.35	14.30	14.90	NS	80.67 (63.89)	84.67 (66.99)	78.67 (62.50)	72.67 (58.47)	(NS)
42 ± 1	10.15	10.80	12.05	12.40	NS	81.00 (64.15)	81.67 (64.71)	77.33 (61.57)	80.00 (63.71)	(NS)
45 ± 1	8.45	8.75	10.60	11.15	NS	74.33 (59.57)	80.67 (63.91)	76.00 (60.72)	71.33 (57.62)	(NS)
CD (p = 0.05)	3.08	3.04	3.15	3.07	NS	(NS)	(NS)	(NS)	(NS)	-

* Constant temperature as control

Figures in parentheses are angular transformed values.

iv. Effect of high temperature shocks on sex-ratio of *T. chilonis*

As compared to control, the sex-ratio in the progeny of *T. chilonis* subjected to high temperature shocks during pupal stage was not adversely affected (Table 2). Females outnumbered males in all the treatments. Our observations are in line with the findings of other workers who reported greater proportion of females in the progeny of *Trichogramma* species exposed to high temperatures (Wang *et al.*, 1990; Hutchison *et al.*, 1990; Haile *et al.*, 2002). According to Bower and Stern (1996) the sex of the progeny is determined largely by the temperature to which their mothers were exposed during development.

B. Exposure of *T. chilonis* to temperature shocks in adult stage

The females of *T. chilonis* emerging at a constant temperature of $27 \pm 1^\circ\text{C}$ and exposed to different high temperatures for six hours daily until death were able to parasitize host eggs (Table 3). Mean lifetime fecundity at all the temperatures

above optimum decreased significantly, and maximum reduction was observed at $45 \pm 1^\circ\text{C}$. Similar trend was observed in the case of longevity. The adult longevity was as low as one day at $45 \pm 1^\circ\text{C}$. The results of the present study are in line with the results of several studies with different *Trichogramma* species (Calvin *et al.*, 1984; Harrison *et al.*, 1985; Ma *et al.*, 1988; Naranjo, 1993; Consoli and Parra, 1995).

Life table of *T. chilonis* showed a decrease in net reproductive rate (R_0) as temperature increased from $27 \pm 1^\circ\text{C}$ (control) to $45 \pm 1^\circ\text{C}$ with values of 162.51 and 7.56, respectively (Table 3). Similar relationship was observed between net generation time (T) and temperature. The intrinsic rate of increase (r_m) increased between $27 \pm 1^\circ\text{C}$ (control) and $33 \pm 1^\circ\text{C}$ and decreased at $36 \pm 1^\circ\text{C}$ indicating maximum capacity of population increase at $33 \pm 1^\circ\text{C}$. The finite rate of increase (λ) showed a direct relationship with the intrinsic rate of increase between $27 \pm 1^\circ\text{C}$ (control) and $33 \pm 1^\circ\text{C}$ and a decrease at $36 \pm 1^\circ\text{C}$. Life table of *T. chilonis* represents an important biological tool to evaluate

Table 3. Effect of high temperature shocks given daily to females on biological characteristics of *T. chilonis*

Temperature ($^\circ\text{C}$)	Biological characteristics			Life table parameters			
	Fecundity/ female	Female longevity (day)	Females in progeny (%)	Net reproductive rate (R_0)	Net generation time increase(T)	Intrinsic rate of (r_m)	Finite rate of increase (λ)
$27 \pm 1^*$	207.65	16.75	77.33 (61.57)	162.51	14.30	0.356	1.428
30 ± 1	148.10	11.90	80.33 (63.67)	118.37	12.54	0.381	1.463
33 ± 1	139.60	10.75	76.33 (60.89)	106.55	12.10	0.386	1.471
36 ± 1	87.75	9.05	74.00 (59.34)	64.94	12.10	0.345	1.412
39 ± 1	63.90	8.35	80.67 (63.91)	51.19	11.58	0.340	1.405
42 ± 1	30.55	3.80	81.33 (64.40)	24.99	10.01	0.321	1.379
45 ± 1	9.95	1.00	76.00 (60.67)	7.56	9.00	0.225	1.252
CD(p = 0.05)	36.44	2.67	(NS)	-	-	-	-

* Constant temperature as control

Figures in parentheses are angular transformed values.

its behaviour at different temperatures. High reproductive potential of the parasitoid at different temperatures increases the possibilities of using this species in integrated management programmes of cotton bollworms in cotton crop. The reproduction and multiplication of the local population of *T. chilonis* at high temperatures could be due to its adaptation to conditions of its habitat from where it was collected.

The results of these laboratory studies have implications for the utility of *T. chilonis* as a biocontrol agent and in developing augmentative release strategies of this parasitoid against lepidopterous pests in cotton. The ambient air temperatures under Hisar agroclimatic conditions sometimes exceeded 41°C during August-September *i. e.* the period during which inundative releases of the parasitoid are made in cotton crop, but the temperature in cotton crop canopy ranged from 27.3 - 38.5°C (unpublished data). Thus, under such conditions local population of *T. chilonis* used in the present study appeared to be a good candidate for use in augmentative biological control programmes against lepidopterous pests in cotton because it performed well under similar conditions in the laboratory especially at high temperatures ($\leq 39 \pm 1^\circ\text{C}$). In augmentative biological control programmes, the parasitoid pupae are released in the field just before adult emergence. For developing augmentative release strategies of this parasitoid against lepidopterous pests in cotton, the results of the present study suggest that different aged pupae may also be released in the field in suitable cages for continuous emergence of the parasitoid wasps over several days. The parasitoid could also be released in adult stage since in the laboratory the female wasps parasitized host eggs and survived for more than eight days at high temperatures ($\leq 39 \pm 1^\circ\text{C}$).

REFERENCES

- Bourchier, R. S., Smith, S. M. and Song, S. J. 1993. Host acceptance and parasitoid size as predictors of parasitoid quality for mass reared *Trichogramma minutum*. *Biocontrol Science and Technology*, **4**: 353-362.
- Bower, W. R. and Stern, V. M. 1996. Effect of temperature on the production of males and sexual mosaics in a uni-parental race *Trichogramma semifumatum* (Hymenoptera: Trichogrammatidae). *Annals of Entomological Society of America*, **59**: 823-824.
- Calvin, D. D., Knapp, M. C., Welch, S. M., Poston, F. L. and Elzinga, R. J. 1984. Impact of environmental factors on *Trichogramma pretiosum* reared on southwestern corn borer eggs. *Environmental Entomology*, **13**(3): 774-780.
- Carey, J. R. 1993. *Applied Demography for Biologists*. Oxford University Press, New York.
- Chihrane, J., Lauge, G. and Hawtlitzky, N. 1993. Effect of high temperature shocks on the development and biology of *Trichogramma brassicae*. *Entomophaga*, **38**(2): 193-198.
- Consoli, F. L. and Parra, J. R. P. 1995. Effects of constant and alternating temperatures on *Trichogramma galloi* Zucchi (Hymenoptera: Trichogrammatidae) biology, parasitism capacity and longevity. *Journal of Applied Entomology*, **119**(10): 667-670.
- Dahlan, A. N. and Gordh, G. 1996. Development of *Trichogramma australicum* Girault (Hymenoptera: Trichogrammatidae) on *Helicovera armigera* (Hübner) eggs (Lepidoptera: Noctuidae). *Australian Journal of Entomology*, **35**: 337-344.
- Gross, H. R. 1988. Effect of temperature, relative humidity and free water on the number and normalcy of *Trichogramma pretiosum* Riley (Hymenoptera: Trichogrammatidae). *Environmental Entomology*, **17**(3): 470-475.
- Guine, G. and Lauge, G. 1997. Effects of high temperatures recorded during diapause completion of *Trichogramma brassicae* prepupae (Hymenoptera: Trichogrammatidae), on the treated generation and its progeny. *Entomophaga*, **42**(3): 329-336.
- Haile, A. T., Hassan, S. A., Ogol, C. K. P. O., Baumgartner, J., Sithanatham, S., Monje, J. C. and Zebitz, C. P. W. 2002. Temperature dependent developments of four egg parasitoid *Trichogramma* species (Hymenoptera: Trichogrammatidae). *Biocontrol Science and Technology*, **12**(5): 555-567.

- Harrison, W. W., King, E. G. and Ouzts, J. D. 1985. Development of *Trichogramma exiguum* and *T. pretiosum* at five temperature regimes. *Environmental Entomology*, **14**: 118-121.
- Hutchison, W., Moratoria, M. and Martin, J. M. 1990. Morphology and biology of *Trichogrammatoidea bactrae* (Hymenoptera: Trichogrammatidae) imported from Australia as a parasitoid of pink bollworm (Lepidoptera: Gelechiidae). *Annals of Entomological Society of America*, **83**(1): 46-54.
- Lopez, J. D. Jr. and Morrison, R. K. 1980. Effects of high temperatures on *Trichogramma pretiosum* programmed for field release. *Journal of Economic Entomology*, **73**: 667-670.
- Ma, W. Y., Peng, J. W. and Zuo, Y. X. 1988. Studies on the biology of *Trichogramma dendrolimi* Matsumura. *Scientia Silvae Sinicae*, **24**(4): 488-495.
- Maisonhaute, C., Chihirane, J. and Lauge, G. 1999. Induction of thermo-tolerance in *Trichogramma brassicae* (Hymenoptera: Trichogrammatidae). *Environmental Entomology*, **28**(1): 116-122.
- Naranjo, S. E. 1993. Life history of *Trichogrammatoidea bactrae* (Hymenoptera: Trichogrammatidae), an egg parasitoid of pink bollworm (Lepidoptera: Gelechiidae), with emphasis on performance at high temperatures. *Environmental Entomology*, **22**(5): 1051-1059.
- Palvik, J. 1992. The effect of temperature on parasitization activity in *Trichogramma* spp. (Hymenoptera: Trichogrammatidae). *Zoologisch Jahrbuecher Abteilung für Allgemeine Zoologie and Physiologie der Tiere*, **96**: 417-425.
- Ram, P., Sharma, P. D. and Chaudhary, S. D. 1997. Evaluation of inundative releases of *Trichogramma chilonis* Ishii against bollworm pests of cotton. *Journal of Cotton Research and Development*, **11**(1): 116-119.
- Ram, P., Sharma, S. S. and Saini, R. K. 2002. Role of egg parasitism by *Trichogramma chilonis* Ishii to control *Helicoverpa armigera* (Hübner) in cotton-sesame intercropping. *Journal of Cotton Research and Development*, **16**(1): 109.
- Singh, S. P. 2001. Advances in biological control in India, pp. 12-30. *Proceedings of Symposium on Biocontrol Based Pest Management for Quality Crop Production in the Current Millennium*, July 18-19, 2001, PAU, Ludhiana.
- Smith, S. M., Hubbes, M. and Carrow, J. R. 1986. Factors affecting inundative releases of *Trichogramma minutum* Riley against the spruce budworm. *Journal of Applied Entomology*, **101**: 29-39.
- Wang, J. L., Yang, Z. C. and Zhang, J. 1990. Study on biological characters of *Trichogramma ostrinae* (Hymenoptera: Trichogrammatoidea). *Natural Enemies of Insects*, **12**(2): 56-61.