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Research Article

Functional response of Sunn pest egg parasitoid, *Trissolcus vassilievi* (Hym., Scelionidae) in response to thermal traits

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Abstract: The functional response of the egg parasitoid, *Trissolcus vassilievi* (Nees) (Hym., Scelionidae), on eggs of the sunn pest, *Eurygaster integriceps* Put. (Hem., Scutelleridae) was investigated at six constant temperatures (12, 15, 20, 25, 28 and 32 °C) for 8 hours. Using logistic regression, a type III functional response was determined for *T. vassilievi* at the two temperatures 15 and 32 °C. Any functional response was not detected at other temperatures. The Hassells' type III model was used for estimating searching efficiency (a) and handling time (T_h). At 32 °C the highest attack rate and the lowest handling time was recorded 0.2029 per hour and 0.1856 hour, respectively. Results show that the parasitoid can be used in the pest management programs at low and high temperatures.

Key words: Biological control, cereal bug, parasitoid behavior, temperature

Introduction

Sunn pest, *Eurygaster integriceps* Put. (Hemiptera, Scutelleridae), is the most important pest of wheat in Iran and The Middle East (Radjabi, 2000). The pest has one generation per year with an obligatory diapause. During early spring, overwintered adults arrive at cereal fields where they, after feeding, begin to lay eggs for about one month (Allahyari *et al.,* 2004). The pest can cause considerable damage to wheat and barley (Radjabi, 2000).

The sunn pest eggs are parasitized by various parasitoid species in the family Scelionidae (Hymenoptera) while the adults are parasitized by flies in the Tachinidae (Diptera). Among these parasitoids, *Trissolcus* spp. (Scelionidae) are the most effective and important species in all Sunn infected areas in Iran and that have received considerable attention (Kasraii, 2010; Allahyari *et al.,* 2004; Noori and Asgari 2003; Asgari, 2002; Asgari *et al.,* 2002; Amir-Maafi, 2000; Shahrokhi-Khangah, 1997; Safavi, 1973). Insect parasitoids are important subjects for behavioral and population studies, because they are remarkably common in nature and more importantly from an economic perspective, often being key species for biological control of many insect pests (Waage and Hassell, 1982; Godfray, 1994).

Among the species attributes related to parasitoid success, an important one is the behavior of an individual parasitoid in response to increasing prey density (Huffaker *et al.*,

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1971; Berryman, 1999); This is referred to as its functional response, defined as the relationship between prey (host) density and the number of prey (hosts) attacked by an individual predator or parasitoid (Solomon, 1949; Holling, 1959). Functional response studies have been used by evolutionary biologists and ecologists to clarify co-evolutionary relationships and basic mechanisms underlying the interactions of predator-prey behavior (Houck and Strauss, 1985). Holling (1959), in his classical paper, defined three types of functional responses. A type I response describes a linear relationship between prey density and the attack rate until a maximum value is reached due to, *e.g.*, satiation. The type II response is an increasing curve with a decelerating slope, which approaches an upper asymptote determined by the time it takes the predator to manipulate its prey (handling time). The type III response is a sigmoid curve. The accelerating part of the curve is often attributed to the parasitoid or predator becoming increasingly efficient at finding hosts or prey. It then levels off under influence of handling time or satiation (Berryman, 1999; Hassell, 2000).

Many factors influence the functional response of a parasitoid, especially the temperature range over which the parasitoid must find their hosts (Gitonga *et al*., 2002). Thus, general models of temperature effects on the functional response have been developed (Mack *et al*, 1981; Flinn, 1991). Some studies have been conducted on the functional response of Scelionid wasps (Allahyari *et al.,* 2004; Amir-Maafi, 2000; Asgari *et al*., 2002; Fathipour *et al*, 2001; Laumann *et al*., 2008), but no detailed study has been carried out to reveal the functional response of *T. vassilievi* (Mayr) (Hymenoptera, Scelionidae) at constant temperatures.

The present study describes the effect of temperature on the functional response of *T. vassilievi*. Knowledge of the functional response of a parasitoid species being used in biological control is a prerequisite for developing predictive models of the pest-enemy interactions (Mills, 2001).

Materials and Methods

Host and parasitoid preparing

Adults of *E. integriceps* were collected from Kermanshah fields, in March–April 2009. Sunn pests were reared in plastic cages (30 × 40 × 55 cm) on dry wheat. For water and humidity source, four tubes (10 × 2 cm) full of water with cotton caps were placed in the corners of each cage. Folded papers served as a site for egg laying. Cages were kept in a greenhouse chamber at 27 ± 2 °C and a photoperiod of 16:8 (L: D) hours and 65 ± 5% RH. The eggs of *E. integriceps* laid on the folded papers were collected in Petri dish (15 × 2cm) and transferred to a refrigerator (5 ± 1 °C) every day.

T. vassilievi specimens were collected from wheat fields in Kermanshah by egg trap cards. By this method, two or three batches of the pest eggs were glued onto trap cards and then placed in the fields. After seven days, egg traps were recovered and brought to the laboratory and placed in a growth chamber until parasitoid emergence.

Newly emerged parasitoids were kept at 25 ± 2 °C, 65 ± 5% RH and a photoperiod of 16:8 (L: D) hours and were fed on 10% honey. For increasing colony size, *ca* 500 eggs of *E. integriceps* were exposed to 10 female parasitoids. After 24 hours, the eggs were separated from parasitoids and placed in a growth chamber. Many parasitoids emerged after 11-12 days, enabling us to use the F2 generation for our experiments.

Experimental methods

The experimental set-up consisted of a glass vial (2 × 10 cm) with a food stripe of 10% honey water droplets and host eggs stripe (1 × 5 cm) in six densities of eggs (2, 4, 8, 16, 32 and 64). One mated female wasp (1-2 days old) was added to each vial which was then placed in a growth chamber for 8 hours. The experiments were carried out at six temperatures (12, 15, 20, 25, 28 and 32 ± 1 °C) with 65 ± 5% RH and a photoperiod 16:8 (L: D) hours. Each density was replicated six

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times and all density experiments exposed to the same temperature were set up simultaneously.

At the end of an experiment, eggs were removed from vials and maintained in growth chamber until emergence of adult parasitoids.

Statistical analysis

Data analysis of the functional response includes two steps (Messina and Hanks, 1998; De Clercq *et al*., 2000; Juliano, 2001; Mohaghegh *et al*., 2001). In the first step, the type of functional response, II or III, is determined by fitting the data in the logistic regression of the proportion of parasitized hosts to initial number of hosts as in eq. 1 (Juliano, 2001). eq. 1.

$$\frac{N_a}{N_0} = \frac{\exp(P_0 + P_1 N_0 + P_2 N_0^2 + P_3 N_0^3)}{1 + \exp(P_0 + P_1 N_0 + P_2 N_0^2 + P_3 N_0^3)}$$

Where *Na* is the number of host parasitized and N_0 is the initial number of hosts. P_0, P_1, P_2, and P_3 are the parameters associated with the intercept, linear, quadratic, and cubic terms respectively, estimated by means of maximum likelihood (Juliano, 2001). A significant negative or positive linear parameter (i.e. P_1) indicates a type II or III functional response, respectively (Juliano, 2001).

Because type III functional response was obtained, depending on results, the model suggested by Hassell *et al*. (1977) was used for estimating handling time (T_h) and attack rate (a), as follow: eq. 2.

$$N_a = N_0\{1 - \exp[-(bTN_0)/(1 + cN_0 + bT_h N_0^2)]\}$$

Where T is the total time of an experiment, b, c and d are constants expressing the relationship between N_0 and a, *i.e.* eq. 3.

$$a = \frac{d + bN_0}{1 + cN_0}$$

The equation is a general form of hyperbolic relationship wherein d is zero when intercept = 0. The parameters of the functional response model were estimated using SAS, PROC NLIN (SAS Institute Inc., 2002, Juliano, 2001).

Results

The data of response of *T. vassilievi* females to increasing host density at six different temperatures were fitted by equation (1). At 15 and 32 °C, the estimated values of parameters of linear, quadratic, and cubic terms (P_1, P_2 and P_3) were significantly different from zero and the sign of linear term parameter at the two temperatures was positive, indicating the type III functional responses (Table 1). At other temperatures (12, 20, 25 and 28 °C), the parameters or at least one of them were not significantly different from zero and therefore after omitting cubic term in equation 1, the data were fitted again by it, but the results did not change. Therefore type II or III functional responses could not be detected in these temperatures.

Hassells' type III model was fitted separately for the two temperatures (15 and 32 °C) in order to compare the search rates and handling times. The instantaneous attack rate is defined as the proportion of the experimental arena searched per time unit (Gitonga *et al*., 2002) and determines how steeply the curve approaches the upper asymptote. Handling time includes all time spent occupied by the prey and unable to attack other prey (Juliano, 2001). The inverse of handling time (*i.e.* handling rate) represents the potential number of hosts that could be parasitized during an experiment of duration T (Cave and Gaylor, 1987), corresponding to the upper asymptote of functional response curve.

The parameters of and the maximum attack were estimated 0.2029, 0.1856 and 43.10, respectively at 32 °C (Table 2). Efficiency of *T. vassilievi*, *i.e.* find and attack eggs of *E. integiceps*, tended to increase at higher temperature (Fig. 1).

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Table 1 Estimates (± SE) for coefficient P_1 in the polynomial function (eq. 1) at various temperatures.

Parameter	12 °C	15 °C	20 °C	25 °C	28 °C	32 °C
P_1	0.106 ± 0.094	0.244 ± 0.095	0.023 ± 0.120	0.108 ± 0.134	0.004 ± 0.141	0.473 ± 0.131
P_2	-0.0037 ± 0.0035	-0.0122 ± 0.0035	-0.0016 ± 0.0043	-0.0051 ± 0.0049	-0.0005 ± 0.0052	-0.0244 ± 0.0049
P_3	0.00003 ± 0.00003	0.00013 ± 0.00003	0.00002 ± 0.00004	0.00005 ± 0.00005	0.00001 ± 0.00005	0.00027 ± 0.00005

Table 2 Estimates (± SE) of searching parameters of *Trissolcus vassilievi* at two temperatures.

Table 2.1

Temperature (˚C)	Coefficient b (h^{-1})	Handling Time (T_h)1 (h)	Handling Rate (Maximum Attack Rate) ($^T/T_h$)2	r^2
15	0.0072 ± 0.00049 (0.0062 - 0.0082)	0.2031 ± 0.0059 (0.1912 - 0.2150)	39.39	0.9955
32	0.0097 ± 0.00074 (0.0082 - 0.0112)	0.1856 ± 0.0056 (0.1743 - 0.1969)	43.10	0.9948

The values in parenthesis are asymptotic 95% confidence interval.
1 T_h = The time that a parasitoid spends for handling a host.
2 $^T/T_h$ = Maximum number of hosts that a parasitoid can attack at time T.

Table 2.2

Instantaneous attack rate (a) 1 (h^{-1})	Temperature (˚C)	$N_0 = 2$	$N_0 = 4$	$N_0 = 8$	$N_0 = 16$	$N_0 = 32$	$N_0 = 64$	Mean
$a = b\ N_0$	15	0.0144	0.0288	0.0576	0.1152	0.2304	0.4608	0.1512
	32	0.0194	0.0388	0.0776	0.1552	0.3104	0.6208	0.2029

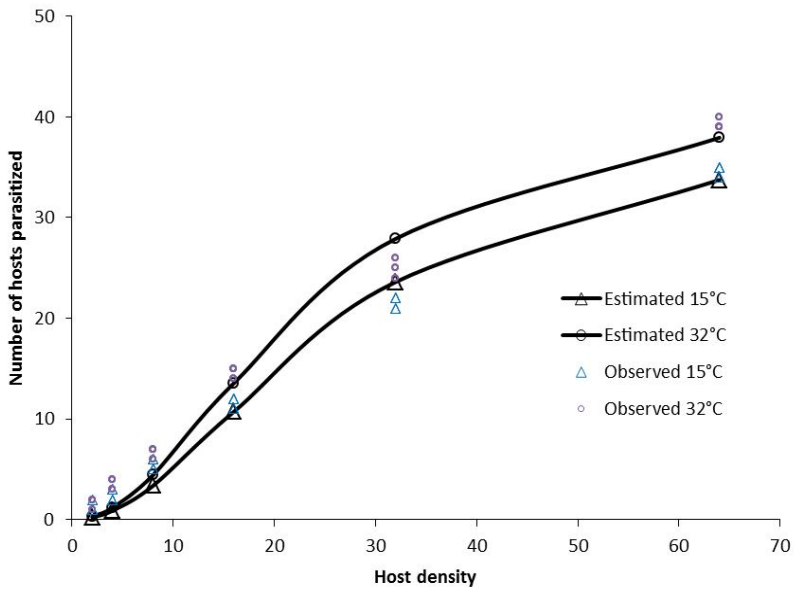

Figure 1 Type III functional response of *Trissolcus vassilievi* on different densities of host at two temperatures.

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Discussion

Parasitoids are common in nature and may influence the population dynamics of a large number of economically important phytophagous pest species. Therefore, detailed studies on the behavior of parasitoids leading to host mortality remain an important issue (Fernandez-Arhex and Corley, 2003). In this respect, studies of the functional response of natural enemies are widely used for understanding the interactions between hosts and parasitoids, as well as between prey and predators. Besides, functional response studies may guide researchers involved in biological control to select species of natural enemies (or age groups within such species) that are the most effective at killing a particular prey or host species (Allahyari *et al.,* 2004).

The positive values of the linear parameter (P_1) in the logistic regression model (Eqn 1) indicated a type III functional response for *T. vassilievi* at 15 °C and 32 °C. It means, as the host density rises, the response initially accelerates due to the parasitoid becoming increasingly efficient at finding hosts. The reason is attributed to an increasing attack rate and/or a decreasing handling time with host density, though Hassells' model assumes that only the attack rate is affected. The response curve then levels off under the influence of handling time or egg load limitation (Berryman, 1999; Hassell, 2000). The current study showed that temperature had significant effect on functional response of *T. vassilievi*, such that the parasitoid showed type III functional responses at 15 °C, the lowest temperature and 32 °C, the highest temperature but had no response to fluctuations of host population at other temperatures. Increased host density, activated parasitoid search at higher temperature (32 °C), more than at lower one (15 °C) so that the slope of response curve rapidly rose to asymptote at 32 °C (Fig. 1). The capacity of parasitoids to locate hosts occurring at low densities can be used as a criterion for selecting candidates for introduction in classical biological control programs (Van Lenteren, 1986; Van Alebeek *et al.*, 1996). Traditionally, parasitoids exhibiting a type III response have been considered more capable of regulating their hosts than type II species (Bernal *et al.,* 1994), though recent studies have questioned this view and instead focused on the role played by the spatio-temporal interactions between hosts (prey) and parasitoids (predators) (e.g., Ives and Settle, 1997, Lei and Hanski, 1997, Cronin and Reeve, 2005, Nachman, 2006).

Apart from temperature, several factors may affect the type of functional response model and its parameter values in an insect species, such as the host plant (both as a substrate for the animals and as food for the prey), and the type of prey or host used in an experiment (Juliano and Williams, 1985; Coll and Ridgway, 1995; Runjie *et al.*, 1996; Messina and Hanks, 1998; Mohaghegh *et al.*, 2001). Our results demonstrating a type III functional response of a scelionid wasp are quite similar to those previously reported (Laumann *et al.*, 2008; Asgari *et al.*, 2002; Allahyari *et al.,* 2004). The estimated search rate of *T. vassilievi* was higher at 32 °C than 15 °C. In general, *T. vassilievi* was more capable of parasitizing *E. integriceps* eggs at higher temperatures of the pests' egg laying period than the lower temperatures. It is therefore suggested that the wasps be released in areas where the temperature is above 25 °C during spring, or late in egg laying period of *E. integriceps* when it is warmer than the earlier of the period. Results of the present study agree with other findings for *Trissolcus grandis* (Thomson) and *Trissolcus semistriatus* (Nees), as both species exhibit a type III functional response when attacking their preferred hosts. The responses of the two species were influenced by plants and host species (Fathipour *et al.*, 2001; Asgari *et al.*, 2002; Allahyari *et al.*, 2004). Functional response studies in the laboratory have been criticized as being unnatural because of the differences in the size of the area that parasitoids have to search to find hosts (Kareiva, 1990; O`Neil, 1989). However, such studies are useful in providing the first step information for comparing the efficiency of different species/strains (Overholt and Smith,

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1990) and also provide a valid means of comparing host finding abilities of candidate natural enemies (Munyaneza and Obrycki, 1997; Zamani *et al*,. 2006).

In conclusion, temperature has a significant impact on the magnitude of parasitization capacity and thereby on the functional response of parasitoids as demonstrated by our study of *T. vassilievi* conducted under laboratory conditions. However, to test the usefulness of *Trissolcus* species as biological control agents against *E. integriceps*, studies should be carried out under natural conditions in the field. Hopefully, the findings of this study can stimulate further research within this important topic in order to reduce future problems with sunn pest in Iran and elsewhere.

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واکنش تابعی زنبور پارازیتوئید تخم سن گندم، *Trissolcus vassilievi* (Hym., Scelionidae) در واکنش به تیمارهای دمایی

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چکیده: واکنش تابعی زنبور *Trissolcus vassilievi* (Nees) (Hym., Scelionidae)، پارازیتوئید تخم سن گندم، *Erygaster integriceps* Put. (Hemiptera, Scutelleridae) در ۶ دمای ثابت (۱۲، ۱۵، ۲۰، ۲۵، ۲۸ و ۳۲ درجه سلسیوس) به‌مدت ۸ ساعت ارزیابی شد. با استفاده از رگرسیون لجستیک، واکنش تابعی زنبور در دماهای ۱۵ و ۳۲ درجه سلسیوس از نوع ۳ تعیین شد. در سایر دماها واکنش تابعی تشخیص داده نشد. برای تخمین کارآیی جستجو (a) و زمان دستیابی (T_h) از مدل نوع ۳ هسل استفاده شد. طبق نتایج، در ۳۲ درجه سلسیوس نرخ حمله آنی (a) با ۰/۲۰۲۹ در ساعت و زمان دستیابی ۰/۱۸۵۶ ساعت به‌ترتیب بیش‌تر و کم‌تر از ۱۵ درجه سلسیوس بودند. نتایج نشان می‌دهد که این پارازیتوئید می‌تواند در دماهای بالا و پایین در برنامه‌های مدیریت این آفت استفاده شود.

واژگان کلیدی: کنترل بیولوژیک، سن غلات، رفتار انگل، درجه حرارت

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