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Influence of temperature on the functional response of the predatory bug, *Anthocoris minki pistaciae* (Hemiptera: Anthocoridae), a predator of *Agonoscena pistaciae* (Hemiptera: Psyllidae)

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ABSTRACT

The common pistachio psylla, *Agonoscena pistaciae* Burckhardt and Lauterer (Hemiptera: Psyllidae), is one of the most destructive pests of cultivated pistachio trees in Iran. A locally available anthocorid predator, *Anthocoris minki pistaciae* Wagner (Hemiptera: Anthocoridae) was evaluated as a biological agent candidate for the control of this pest. The functional response of this predatory bug was investigated at temperatures of 20, 25, 27.5, and 30 °C, relative humidity of 50–60%, photoperiod L:D of 16:8 h and at densities of 2, 4, 8, 20, 40, 60, 80, and 100 nymphs of *A. pistaciae* during a 24-h period. A type II functional response model was fitted at all tested temperatures. The results indicated that the instantaneous attack rate for the predatory bug ranged from 0.15 to 0.36 nymphs per hour and estimated handling times were 0.41, 0.35, 0.42, and 0.27 h at 20, 25, 27.5, and 30 °C, respectively. Based on asymptotic 95% confidence intervals, searching efficiency didn't differ significantly from 20 to 30 °C, but handling time at 30 °C was significantly different from other tested temperatures, except at 25 °C. This observation suggests that *A. minki pistaciae* can be more effective at 30 °C, compared with other experimental temperatures.

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Introduction

The common pistachio psylla, *Agonoscena pistaciae* Burckhardt and Lauterer (Hemiptera: Psyllidae), the key pest of pistachio trees (*Pistacia vera* Linnaeus) is distributed throughout pistachio producing regions in Iran. This pest is present from mid-March until early December on pistachio trees and has five to six generations per year. The presence of large populations of psyllid nymphs and adults causes severe problems in kernel development, causing bud drop and defoliation. The result has been a substantial reduction in pistachio yields for three consecutive years (Mehrnejad 2003). *A. pistaciae* also occurs in many pistachio growing regions in Asia Minor and the Middle East, as well as in Mediterranean regions (Burckhardt and Lauterer 1989, 1993; Mart et al. 1995; Lauterer et al. 1998). *A. pistaciae* has been controlled almost exclusively by pesticides; however, many populations of this pest have developed resistance to various insecticides (Mehrnejad 1998). Therefore, some alternative techniques to manage this economic pest, such as biological control are necessary.

Information on the status of most psyllids' natural enemies in pistachio growing areas of Iran has been

summarized (Mehrnejad and Emami 2005; Mehrnejad 2010). The predatory bug, *Anthocoris minki pistaciae* Wagner (Hemiptera: Anthocoridae) was reported as an egg and nymphal predator of *A. pistaciae* (Mehrnejad 2010). Thus, knowledge of this predator adaptation to climatic factors will play an important role in its rearing and implementation. Of all the ecological factors, the temperature is a major biotic factor influencing the biology and functional response of pests' natural enemies (Rosen and Huffaker 1983; Nechols, Tauber, and Masaki 1999). Knowledge of predatory potential of *A. minki pistaciae* would be useful for the development of IPM programs. Previously, the effect of temperature on its biology and prey consumption was studied (Pourali et al. 2010; Pourali, Mehrnejad, and Kheradmand 2010).

The objective of the present study was to determine the effect of temperature on the functional response of *A. minki pistaciae* at different prey densities under controlled laboratory conditions. The results of our investigation will be used to predict the attack rate as a function of host density and temperature for improving our understanding of prey-predator interactions and thus to develop a better strategy for the biological control of psylla in pistachio trees.

Materials and methods

Insect culture

Predatory bugs used in the present study were collected from pistachio trees in Rafsanjan (south-western part of Iran, 56°E, 30°N). *A. minki pistaciae* stock culture was maintained in a laboratory under the condition of $27.5 \pm 1^\circ\text{C}$, $60 \pm 10\%$ RH, and a photoperiod L:D of 16:8 h. The colony fed on nymphs of *A. pistaciae* and the pistachio leaf discs were used as an oviposition substrate. Leaf discs were placed into Petri dishes (52 mm diameter) that contained a layer of agar medium as a moisturizing source. Then the leaf-disc cages were placed into a plastic box ($25 \times 17 \times 10$ cm), having a hole (4 cm) covered by a nylon screen for ventilation. Relative humidity was regulated by placing saturated 'magnesium nitrate' salt inside the box (Mehrnejad 1998). Fresh nymphs of *A. pistaciae* used in our investigation were daily served as a food source.

Experimental design

To determine the functional response of *A. minki pistaciae* at various host densities, the predatory bugs were exposed to eight nymphal density levels (2, 4, 8, 20, 40, 60, 80, and 100) at four constant temperatures (20, 25, 27.5, and 30°C) at $60 \pm 10\%$ RH and a photoperiod L:D of 16:8 h. Selected temperatures in this study were based on the average monthly temperature during the pest activity in Rafsanjan (the main pistachio growing area of Iran). The experimental arena consisted of pistachio leaf discs as described above. For each density, nymphs were placed in a Petri dish and a single one-day-old starved adult female of *A. minki pistaciae* was introduced into each Petri dish. The tests were conducted for a 24-h period. There were 10 replicates per pest density. All Petri dishes were checked for predator activity after 24 h, and the number of nymphs killed by an adult predatory bug was determined using a binocular microscope (10 \times).

Statistical analyses

As it is difficult to discriminate between type II and III functional responses (Juliano 2001), the first step was to fit the data to determine the type of the functional response. A cubic logistic regression (1) between the proportion of host attacked (N_a) and initial host density (N_0) was used to determine the shape of the functional

response (Trexler, McColluch, and Travis 1988; Juliano 2001):

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

where N_a is the number of attacked prey, N_0 is the initial prey density, and P_0 , P_1 , P_2 , and P_3 are parameters to be estimated. These parameters were estimated using CATMOD procedures in SAS software (SAS Institute 2000). Significant negative or positive linear coefficients indicate type II or III, respectively, and a non-significant linear coefficient (P_1) indicates a type I response (Juliano 2001; Li, Tian, and Shen 2007).

As predators consistently displayed type II responses, the handling times (T_h) and searching efficiency (a) of type II responses were estimated using the weighted nonlinear least square regression (NLIN procedures with the DUD method in SAS). The Roger's random predator Equation (2) was used to fit data when host depletion occurred; this model is appropriate for describing a functional response (Juliano 2001):

$$N_a = N_0 \left[1 - \exp \left(- \frac{a T_t}{1 + a T_h N_0} \right) \right], \quad (2)$$

where a is the instantaneous attack rate, T_t is the time of exposure, T_h is the handling time. The differences in the attack rates and handling times at various constant temperatures were compared using approximate 95% confidence limits.

Results

A. minki pistaciae exhibited a decreasing curve type II functional response as determined by the logistic regression model at the different densities of hosts (Table 1).

According to the data, the logistic regression had a significant negative linear parameter $P_1 < 0$ at all tested temperatures. Therefore, the type of the functional response was not affected by temperature.

The Roger's random predator equation type II model was fitted at each temperature in order to compare the searching efficiency and handling time at the different tested temperatures. Using Roger's type II model, the approximate confidence limits ranged from 0.0906 to 0.6141 for searching efficiency and from 0.2187 to 0.4637 for handling time (Table 2).

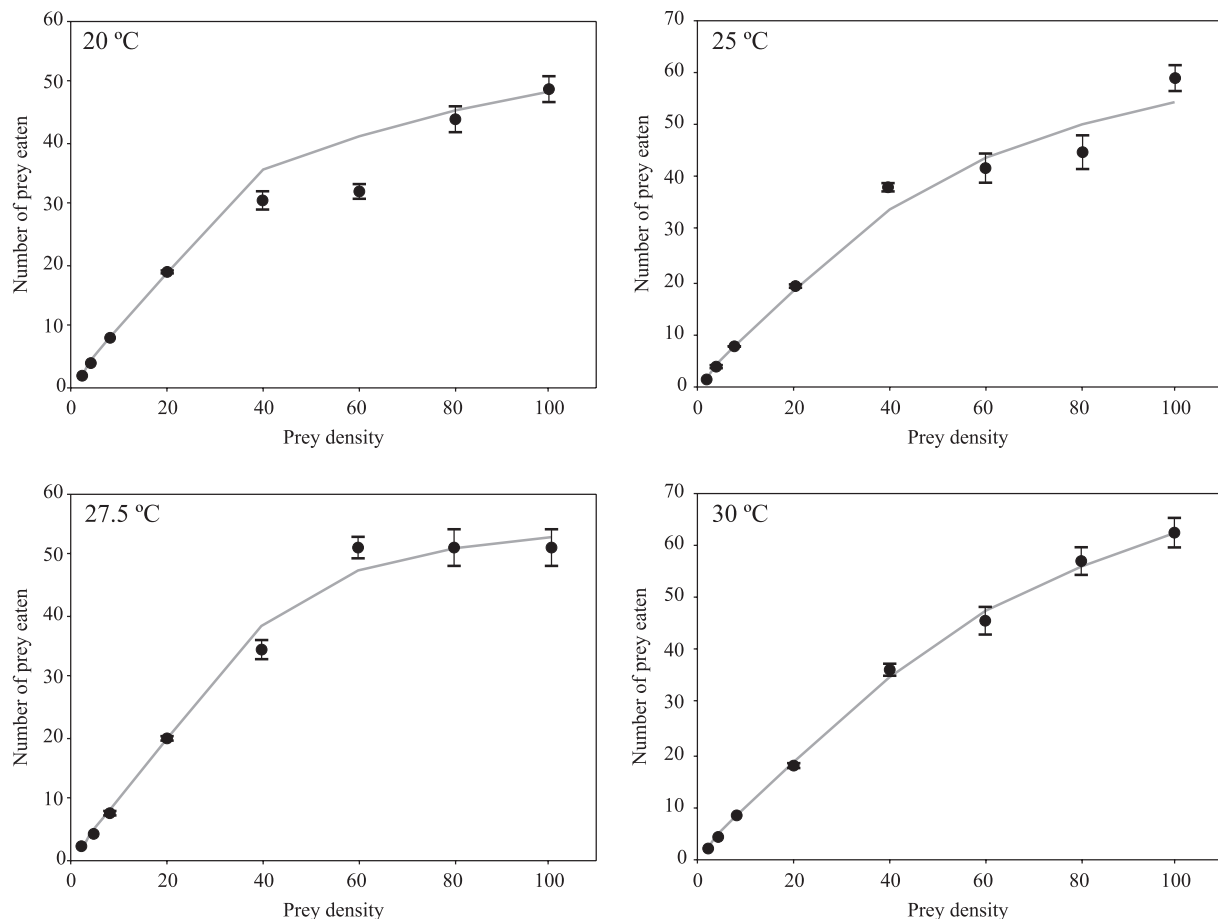
Table 1. Result of logistic regression analysis of the proportion of *A. pistaciae* killed by *A. minki pistaciae* to initial prey numbers at different constant temperatures.

Temperature ($^\circ\text{C}$)	Constant	Linear	Quadratic	Cubic
20	7.909 ± 1.023	$-0.298 \pm .0509$	0.00383 ± 0.000796	-0.00002 ± 3.928
25	6.827 ± 1.404	-0.1128 ± 0.0661	-0.00009 ± 0.00099	5.713 ± 4.804
27.5	4.19 ± 0.654	$-.0562 \pm 0.0358$	0.000181 ± 0.000609	-3.69 ± 3.177
30	3.922 ± 0.554	$-.0790 \pm 0.0315$	0.000772 ± 0.00055	-3.26 ± 2.921

Table 2. Estimates (\pm SEM) of searching efficiency and handling time of *A. minki pistaciae* at various constant temperatures.

Temperature ($^{\circ}\text{C}$)	Instantaneous attack rate (h^{-1})	Asymptotic 95% CI		Handling time (h)	Asymptotic 95% CI	
		Lower	Upper		Lower	Upper
20	0.1568 ± 0.0286	0.09991	0.2138	$0.4092 \pm 0.0247\text{a}$	0.3600	0.4583
25	0.1516 ± 0.0361	0.0796	0.2235	$0.3463 \pm 0.0332\text{ab}$	0.2803	0.4123
27.5	0.3587 ± 0.1283	0.1032	0.6141	$0.4175 \pm 0.0232\text{a}$	0.3714	0.4637
30	0.1355 ± 0.0226	0.0906	0.1804	$0.2705 \pm 0.0260\text{b}$	0.2187	0.3222

Means within a column followed by the same letter are not significantly different ($P > 0.05$).

**Figure 1.** Functional response of *A. minki pistaciae* to densities of *A. pistaciae* nymph at different temperatures (symbols – observed values (mean \pm SE), lines – values predicted by the model).

The number of *A. pistaciae* nymphs killed by *A. minki pistaciae* females increased at a decreasing rate until reaching an upper plateau as the number of *A. pistaciae* offered increased (Figure 1). The number of prey captured by the predator increased with temperature.

The relationship between temperature, handling times, and attack rates is shown in Table 2. Based on asymptotic 95% confidence interval (CI), a lower attack coefficient was found at 20 and 25 $^{\circ}\text{C}$ and the estimated instantaneous attack rate was the highest when the predatory bug was reared at 27.5 $^{\circ}\text{C}$.

The handling time ranged between 0.27 and 0.42 h and was significantly lower at 30 $^{\circ}\text{C}$ compared with 20 and 27.5 $^{\circ}\text{C}$.

Discussion

This is the first study that has investigated the functional response of *A. minki pistaciae* on the pistachio

psylla at a range of temperatures suitable for its development and survival. The selected temperatures should reflect thermal conditions frequently experienced by the predator in different protected and field crops in temperate areas. Our previous studies determined the effect of temperature on *A. minki pistaciae* development and population parameters (Pourali et al. 2010; Pourali, Kheradmand, and Mehrnejad 2012). The negative values for the linear parameters obtained in this study confirm the type II functional response at all experimental temperatures. This type of functional response has been reported for other close genera or species of predatory bugs by other researchers (Gitonga et al. 2002; Mahdian, Tirry, and Clercq 2007). The type of the functional response exhibited by a natural enemy can change with potential reasons due to changes in the foraging behaviour of the insects at different temperatures (Wang and Ferro 1998; Jamshidnia et al. 2010). In the present study, the tested temperatures apparently

did not influence the type of the functional response, which suggests this anthocorid can efficiently adapt itself to the environmental conditions in Iran. At all constant temperatures, the asymptote 95% CI for searching efficiency overlapped and thus did not demonstrate significant differences. By increasing the temperature from 20 to 27.5 °C, the searching rate was increased, although a decrease was calculated at 30 °C. Our result is in agreement with what was reported by Simonsen et al. (2008) for *A. nemorum*, but is not in agreement with Yanik and Unlu (2011), who believe that temperature has a significant effect on prey consumption by *A. minki*. Furthermore, the highest value for (*a*) was calculated at 27.5 °C, which was considerably higher than the attack rate reported by Gitonga et al. (2002) and Xu and Enkegaard (2009) for *Orius albidipennis* at 28 °C and *O. sauteri* when fed on *Tetranychus urticae* (0.029 and 0.042, respectively). According to our results, the handling time was decreased by increasing the temperature from 20 to 30 °C, the values indicated a significant difference at 20, 27.5, and 30 °C. The lower handling time for the adults at 30 °C indicates that predatory bugs can be present more efficiently at higher temperatures. Nesrin et al. (2012) and Zamani et al. (2009) report the handling times between 0.0122–0.0225 and 0.005–0.012 for *O. albidipennis*, which is considerably lower than our findings. To compare, Xu and Enkegaard (2009) showed T_h of 1.217 for *O. sauteri* when fed on *T. urticae*.

Our result shows that the maximum predation (T_t/T_h) by *A. minki pistaciae* is 58.7, 69.3, 57.5, and 88.7 at 20, 25, 27.5, and 30 °C, respectively. These data suggest this predator could be more effective at higher temperatures. Our results are in accordance with Yanik and Unlu (2011) who recommended 30 °C as an optimal temperature for the predator mass production.

To conclude, the present study has improved our understanding of the functional response of *A. minki pistaciae* to *A. pistaciae*. Also, our results show that *A. minki pistaciae* displays relatively high predation rates on *A. pistaciae* at a wide range of temperatures, indicating its potential for augmentative releases against the common pistachio psylla in the field. However, more accurate results of the prey-predator interactions in field-based studies will be needed to develop a reliable strategy for the biological control of psylla in pistachio trees.

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