

Research Article

Predation response of *Nabis pseudoferus* (Hemiptera: Nabidae) on untreated and *Metarhizium anisopliae*-treated larvae of *Tuta absoluta* (Lepidoptera: Gelechiidae)

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Abstract: Functional response is an important behavioral characteristic of prey-predator interactions that can be utilized for assessing impact of natural enemies. In this research, the functional response of *Nabis pseudoferus* Remane females was examined to the third-instar larvae of *Tuta absoluta* (Meyrick) which were exposed to LC₃₀ (2.03×10^4 conidia/ml) values of *Metarhizium anisopliae* (Metschnikoff) Sorokin isolate DEMI 001. Six densities of the prey (1, 2, 4, 8, 10 and 16) were exposed to the predator (0, 24, 48 and 72 h) after inoculation. *N. pseudoferus* exhibited a type II functional response to prey density in all treatments, indicating that predation increases asymptotically to a satiation level. The highest and the lowest attack rates (*a*) were 0.1052 ± 0.0440 and $0.0509 \pm 0.0133h^{-1}$ for 48h and 72h post-infection treatments, respectively. Maximum theoretical predation rate (*T/T_h*) was estimated 10.96 in control. Our results suggest that *M. anisopliae* and *N. pseudoferus*, can be a useful combination in pest management of tomato leaf miner, although it must be confirmed in field condition.

Keywords: Functional response, Attack rate, *N. pseudoferus*, *T. absoluta*, *M. anisopliae*

Introduction

The tomato leaf miner, *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) is one of the most important pests of tomatoes both in field and greenhouse (Barrientos *et al.*, 1998; Zappalà *et al.*, 2013). Originating from South America (Desneux *et al.*, 2010), this pest was recorded for the first time in Urmia, North West of Iran during November 2010 (Baniameri and Cheraghian, 2012). Subsequently, it was spread quickly and became one of the key pests of tomato in many regions in the country (Gharekhani and Salek-

Ebrahimi, 2014; Ghaderi *et al.*, 2017). Larvae of *T. absoluta* attack all aerial parts of the plants including the apical buds, leaves, stems, flowers and fruits, causing up to 100% losses when management methods are not efficiently implemented (Biondi *et al.*, 2018).

Recently, development of resistance by *T. absoluta* populations to traditional insecticides as well as other unfavorable side effects of the chemicals has encouraged safer methods of the pest control (Urbaneja *et al.*, 2012; Ingegno *et al.*, 2013). Thus, application of environmentally friendly tactics such as predators, parasitoids and entomopathogens is recommendable.

The damsel bug, *Nabis pseudoferus* Remane (Hemiptera: Nabidae) is a generalist and strong entomophagous predator that is commercially produced in Spain (Cabello *et al.*, 2009; Gámez *et*

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al., 2012). This predator is able to feed on different life stage of *T. absoluta* including egg, larvae (in spite of being hidden inside the mines) and pupa (Cabello *et al.*, 2009; Luna *et al.*, 2012; Zappala *et al.*, 2013; Mahdavi and Madadi, 2015; Mahdavi and Madadi, 2017), unlike the *Nesediocoris teneus* (Reuter) and *Macrolophus pygmeus* Rambur (Heteroptera: Miridae) that are predators of eggs and young larvae of tomato leaf miner. *N. pseudoferus* is a common species in Iran (Modarres Awal, 2008; Havaskary *et al.*, 2012; Arbab *et al.*, 2016) which may be used as a biological control agent in tomato crops (Cabello *et al.*, 2009; Ghoneim, 2014; Mahdavi and Madadi, 2017). Furthermore, the muscardine fungus, *Metarhizium anisopliae* (Metschnikoff) is a virulent entomopathogenic fungus that attacks the eggs (Pires *et al.*, 2009), larvae (Inanl and Oldarge, 2012; Tadele and Eman, 2017; Nozad-Bonab *et al.*, 2017), and pupae (Contreras *et al.*, 2014) of *T. absoluta*. The integration of *N. pseudoferus* and *M. anisopliae* can improve tomato leaf miner management; however, application of multiple biological control agents may act synergistically, additively or antagonistically (Roy and Pell, 2000). Accordingly, in order to make biocontrol programs more effective, multitrophic interactions among natural enemies, hosts and targeted plants need to be assessed. Alma *et al.* (2007) showed that utilization of entomopathogenic fungus, *Paecilomyces fumosoroseus* (Wize) to control *Trialeurodes vaporariorum* Westwood is compatible with simultaneous use of the whitefly predator *Dicyphus hesperus* Knight and these agents have additive effects. Similarly, Labbe' *et al.* (2009) indicated that *Beauveria bassiana* (Balsamo), *D. hesperus* and *Encarsia formosa* Gahan (Hymenoptera: Aphelinidae) can be successfully combined for biological control of *T. vaporariorum*, in short-term in greenhouse tomato crops.

Wekesa *et al.* (2007) demonstrated that the fungal pathogen *Neozygites floridana* (Weiser and Muma) could reduce egg predation rate of the predatory mite *Phytoseiulus longipes*, by increasing time and high energy invested grooming. In another study, population parameters of the predator *Eriopis connexa*

(Coleoptera: Coccinellidae) were influenced by entomopathogenic fungus, *B. bassiana* (Scorsetti *et al.*, 2017). Agboton *et al.* (2013) also reported a negative interaction between the predatory mite *Typhlodromalus aripo* and the entomopathogenic fungus, *N. tanajoae*.

The response of a predator to prey density is a key factor for the success of a biological control program (Barlow and Goldson, 1993; Lester *et al.*, 1999). This behavior may determine if a natural enemy is able to regulate, stabilize or destabilize the target pest's population (Dick *et al.*, 2013). The relationship between predation rate (i.e., number of prey consumption per predator in unit time) and prey density is termed "functional response" (Solomon, 1949). Holling (1959) categorized functional responses into three mathematical models, which he called types I, II, and III. These models are characterized by a hyperbolic curve: a linear rise in prey consumption with increasing prey densities to a plateau (type I), a decelerating curvilinear rise to a plateau (type II), or a sigmoidal shaped curve to a plateau (type III) (Holling, 1959).

Host-infection by an entomopathogen can affect natural enemies' performance and foraging behavior such as functional response (Wekesa *et al.*, 2007; Labbe' *et al.*, 2006; Pourian *et al.*, 2011; Seiedy *et al.*, 2012; Rännbäck *et al.*, 2015; Wu *et al.*, 2015; Jarrahi and Safavi, 2016 a; Jarrahi and Safavi, 2016b). Hence, in this research, we investigated effect of sub-lethal concentration (LC₃₀) of *M. anisopliae* isolate DEMI 001 on functional response of *N. pseudoferus* to infected larvae of *T. absoluta*. Taking into account that a pathogen needs some time for developing inside the host body, we also included time intervals from 0 to 72h from infection to exposure.

Materials and Methods

Plant cultivation and insect rearing

Tomato plants (cultivar Super Luna), were used for rearing of *T. absoluta* while they were approximately 45 days old. A stock culture of *T. absoluta* was initiated by collecting infected leaves from tomato fields in Urmia, West-Azerbaijan Province, Iran. The insects were reared on tomato

plants in wooden cages ($60 \times 60 \times 40$ cm) for two generations in a glasshouse at 25 ± 1 °C, 16: 8 (L: D) h and $65 \pm 5\%$ R. H. Adults of *N. pseudoferus* were collected by sweeping the alfalfa plants in Urmia fields, West - Azerbaijan Province, Iran (N $57^{\circ} 29' 53''$ E $45^{\circ} 3' 31''$). Ten to 20 adult *N. pseudoferus* of both sexes were introduced into rearing cups (14×22 cm), equipped with a cardboard panel as a shelter and covered by a fine mesh gauze at lid for ventilation. The predator was supplied by bean pods as an oviposition substrate, as well as moisture source. The bugs were fed by adults and nymphs of cotton aphid, *Aphis gossypii* Glover. A colony of the aphid was obtained from the culture collection of the Plant Protection Laboratory, Urmia University and reared on cucumber plants in a growth glasshouse at 25 ± 1 °C, $65 \pm 5\%$ RH, 16:8 (L: D) h. The cups were checked in 24-h intervals, and the bean pods involving the predator eggs were transferred to new cups (8×12 cm) in a growth chamber at 25 ± 1 °C, $65 \pm 10\%$ RH, and a photoperiod of 16: 8 (L: D) h. As soon as the eggs were hatched, the nymphs were transferred individually into Petri dishes (6 cm in diameter) containing fresh bean pods as well as nymphs of the prey, *A. gossypii*. The dishes were renewed every day until adulthood. The predator was reared for a generation, prior to experiments.

Fungal pathogen

Metarhizium anisopliae isolate DEMI 001 was obtained from the culture collection of the Plant Protection Laboratory, Urmia University. After passage of the fungus through *T. absoluta* larvae, it was cultured on Sabouraud's dextrose agar with yeast extract (SDAY) for two weeks at 25 ± 1 °C until sporulation. Fungal suspensions were prepared in distilled water containing 0.02% Tween-80 and spore concentration was determined using a Neubauer hemocytometer (Neubauer improved, Kavalier). The viability of the conidia was determined by inoculating plates of SDAY (four plates) with a conidial suspension ($100 \mu\text{l}$ of 10^7 dilutions) which was then incubated for 24h at 25 ± 1 °C. The conidia were considered viable when the germ tube lengths corresponded to the width (Inglis et al., 2012). The viability of conidia was assessed immediately before each experiment

and just those conidia which their viability was above 95% were used in experiments.

Virulence of *M. anisopliae* (DEMI 001) against *T. absoluta* larvae

Bioassays were carried out using third instar larvae (L3) of *T. absoluta* (high exposure to predator and fungus). Separate batches of L3 larvae were immersed in five spore concentrations including 10^3 , 10^4 , 10^5 , 10^6 , 10^7 conidia. mL^{-1} for 10s. Treated insects were transferred upon tomato leaves embedded in Petri dishes (10cm diameter) covered with a fine mesh gauze on lid for ventilation and fresh tomato leaves were provided daily. The control batch was treated by sterile distilled water plus 0.02% Tween-80. Mortality was monitored daily and dead larvae were removed. Larval cadavers were surface sterilized in 70% ethanol, followed by sterile distilled water and incubated on moist filter paper in Petri dishes (6cm diameter) to confirm infection by *M. anisopliae*. The experiment consisted of 4 replicates (15 insects per replicate) for each concentration.

Functional response experiment

Petri dishes (9cm diameter), with a meshed hole in the lid, were filled with a layer of 2% water agar. Excised tomato leaves were placed upside down onto the water agar. Six densities of *T. absoluta* L3 including 1, 2, 4, 8, 10 and 16 were exposed to sub-lethal (LC_{30}) concentration of 2.03×10^4 of *M. anisopliae* isolate DEMI 001 conidia/ml and then put into each Petri dish. Subsequently the treated larvae, exposed immediately or after 24, 48 or 72h incubation to experimental unit containing an individual predator. A 7-day-old inseminated female *N. pseudoferus* starved for 24h was used in each experimental unit. After 24h, the predators were removed and the number of consumed prey individuals were determined by counting number of survived larvae and subtracting it from initial prey number. Prey individuals were not replaced during the experiment and each treatment was carried out in 10 replications. The experiments were conducted at 25 ± 1 °C, $65 \pm 5\%$ RH and a photoperiod of 16:8h (L: D).

Data analysis

The logistic regression model was used to determine the type of functional response by taking into consideration the proportion of prey eaten (N_a/N_0) as a function of prey offered (N_0) (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 prey eaten, N_0 is the initial prey density, and P_0 , P_1 , P_2 and P_3 are the intercept, linear, quadratic, and cubic coefficients, respectively, estimated using the method of maximum likelihood (Juliano, 2001). The signs of the linear coefficients (i.e., P_1) from the regression can be used to distinguish the shape of the functional response (type II or III). A significant negative linear coefficient suggests a type II response, while a significant positive linear term indicates a type III response (Juliano, 2001).

The handling times and attack coefficients of a type II response were estimated as Royama (1971):

$$N_a = N_0 \{1 - \exp(a(T_h N_a - T))\} \quad (2)$$

where N_a and N_0 are described in equation (1), T is the time available for searching during the experiment (24h in this experiment), a is the instantaneous attack rate and T_h is per capita handling time. Parameters at various treatments were compared based on 95% confidence limits (CI). The coefficient of determination was calculated as $R^2 = 1 - \text{residual sum of squares/corrected total sum of squares}$. The functional response analyses were done using SAS PROC NLIN (SAS Institute, 2003). Curves were drawn by Excel (2016).

Results

Bioassays of *M. anisopliae* (isolate DEMI 001) on *T. absoluta* L3 resulted in LC_{30} value of 2.03×10^4 (CI 95% = $9.35 \times 10^3 - 3.81 \times 10^4$) conidia.ml⁻¹. In all treatments, the linear parameter of the polynomial regression of the proportion of prey consumed versus initial density was negative (Table 1).

Table 1 Maximum likelihood estimates of logistic regression parameters drawing of *Tuta absoluta* larvae eaten by female *Nabis pseudoferus* as a function of initial prey densities at 0, 24, 48 and 72h post-inoculation with LC_{30} of *Metarhizium anisopliae*.

Treatments	Parameters ¹	Estimate ± SE	χ^2	P-value
Control	Intercept	3.4309 ± 1.3005	6.96	0.0083
	N_0	-0.7787 ± 0.5817	1.79	0.1807
	N_0^2	0.0656 ± 0.0725	0.82	0.3660
	N_0^3	-0.0019 ± 0.0026	0.56	0.4545
0h	Intercept	2.9714 ± 1.1175	7.07	0.0078
	N_0	-0.8298 ± 0.5168	2.58	0.1084
	N_0^2	0.0826 ± 0.0655	1.59	0.2071
	N_0^3	-0.00275 ± 0.0023	1.37	0.2424
24h	Intercept	4.2802 ± 1.4740	8.43	0.0037
	N_0	-1.1128 ± 0.6372	3.05	0.0808
	N_0^2	0.0934 ± 0.0780	1.43	0.2311
	N_0^3	-0.0038 ± 0.0025	0.94	0.3332
48h	Intercept	3.7191 ± 1.2754	8.50	0.0035
	N_0	-1.0828 ± 0.5709	3.60	0.0579
	N_0^2	0.1117 ± 0.0711	2.46	0.1164
	N_0^3	-0.00380 ± 0.0025	2.26	0.1330
72h	Intercept	3.7456 ± 1.1413	10.77	0.0010
	N_0	-1.4324 ± 0.5244	7.46	0.0063
	N_0^2	0.1555 ± 0.0660	5.55	0.0185
	N_0^3	-0.00524 ± 0.2360	4.91	0.0267

¹ N_0 , N_0^2 and N_0^3 are linear, quadratic and cubic coefficients, respectively.

Fitting the polynomial logistic regression (equation 1) to the data suggested that adult females of *N. pseudoferus* exhibited type II functional response to *T. absoluta* L3 whether treated or untreated with *M. anisopliae* (DEMI 001) (Fig. 1). Moreover, in all post-inoculation intervals (0, 24, 48 and 72h treatments), the proportion of the consumed prey declined with increasing the prey densities (Fig. 2). Table 2 represents the coefficient of attack rates (a) and

handling times (T_h) of the predator. The attack rate coefficient and handling time of *N. pseudoferus* were significantly different from 0 as the asymptotic 95% confidence interval overlapped these values (Table 2). Maximum attack rate was estimated at 48h treatment ($0.1052 \pm 0.0440h^{-1}$). The lowest and highest handling time were observed in control (2.1899h) and 72h (3.3760h) post-inoculation treatments, respectively.

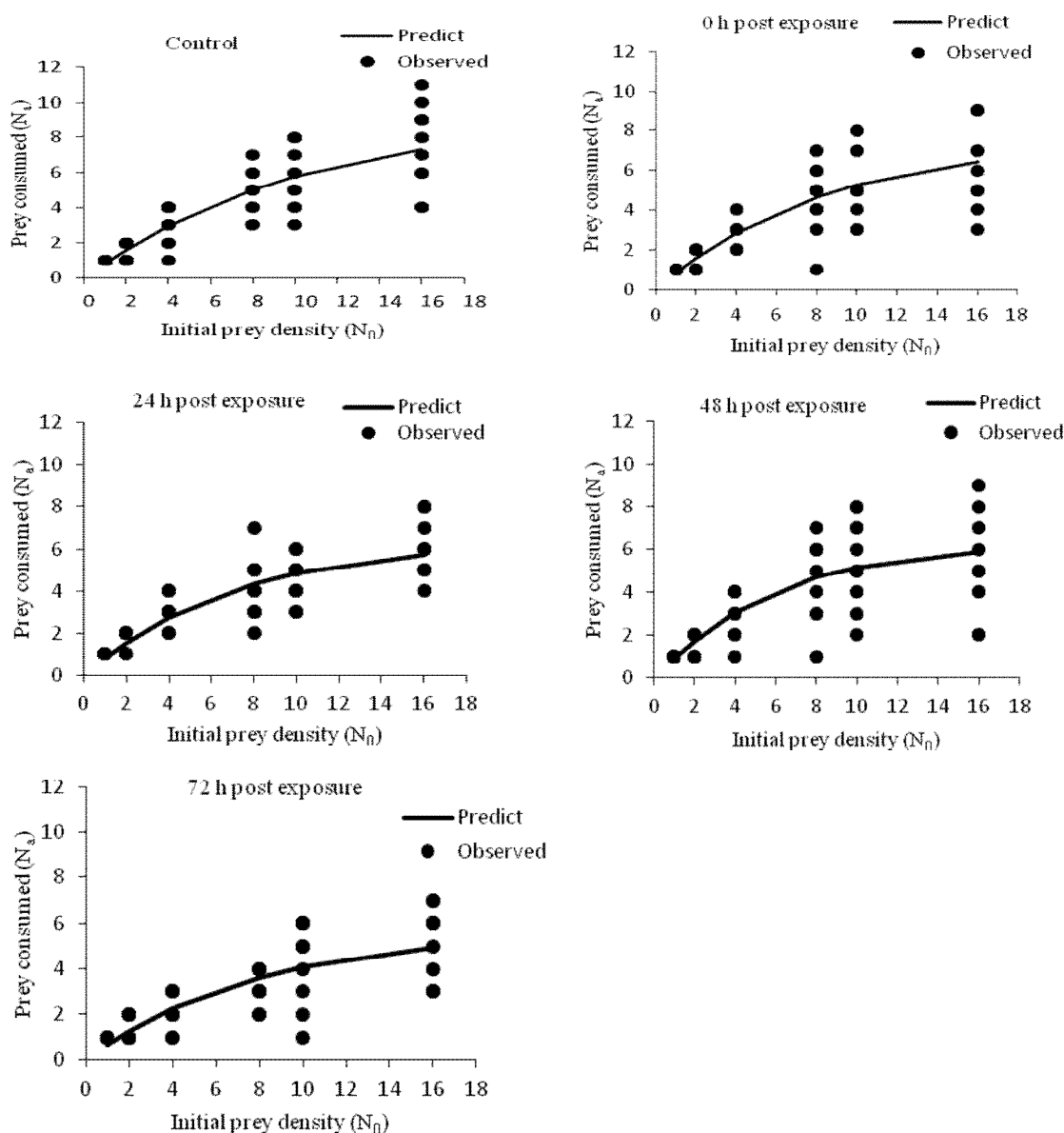


Figure 1 Functional response of *Nabis pseudoferus* to *Tuta absoluta* L3 at different post-exposure treatments to *Metarhizium anisopliae*.

Table 2 Parameters estimates (mean ± SE) of functional response of *Nabis pseudoferus* to *Tuta absoluta* larvae at different inoculation intervals to *Metarhizium anisopliae*.

Treatments	Functional response type	$a \pm SE (h^{-1})^1$ (95% CI) ²	$T_h \pm SE$	T/T_h	R ²
Control	II	0.0756 ± 0.0199 (0.0358 - 0.1153)	2.1899 ± 0.4293 (1.3306 - 3.0492)	10.96	0.92
0h	II	0.0744 ± 0.0215 (0.0315 - 0.1174)	2.6667 ± 0.4842 (1.6972 - 3.6359)	9.00	0.90
24h	II	0.0772 ± 0.0182 (0.0407 - 0.1137)	3.2005 ± 0.3942 (2.4114 - 3.9895)	7.50	0.94
48h	II	0.1052 ± 0.0440 (0.0170 - 0.1933)	3.3227 ± 0.5462 (2.2294 - 4.4160)	7.44	0.87
72h	II	0.0509 ± 0.0133 (0.0244 - 0.0774)	3.3760 ± 0.6102 (2.1547 - 4.5974)	7.11	0.90

¹ a , T_h and T/T_h are attack rate, handling time and maximum theoretical predation rate of predator, respectively.

² 95% confidence intervals.

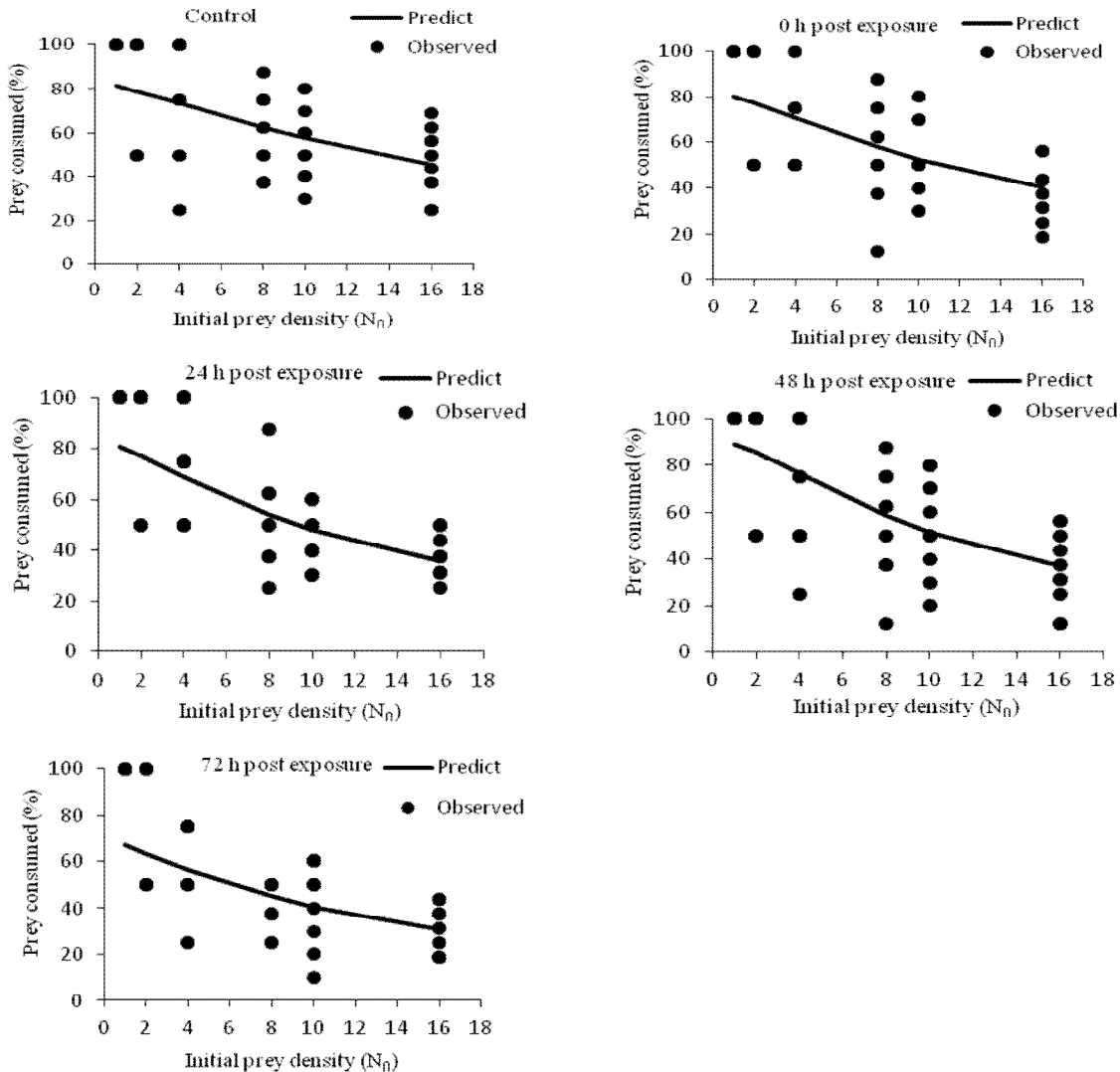


Figure 2 The percentage of consumed *Tuta absoluta* L3 by *Nabis pseudoferus* in different post-exposure treatments to *Metarhizium anisopliae*.

Discussion

The third instar larvae of *T. absoluta* tend to spread among tomato plant to avoid competition (Miranda, 1998; Cuthbertson, 2013). This behavior can increase exposure to external mortality factors such as natural enemies and insecticides (Miranda, 1998; Cuthbertson, 2013). As a result of this study, infection of *T. absoluta* third-instar larvae with a sub-lethal concentration (LC_{30}) of *M. anisopliae* (isolate DEMI 001) at different time intervals (0, 24, 48 and 72h) had no effect on the type of *N. pseudoferus* functional response. According to our results, the female predator exhibited type II functional response in all the treatments. A type II functional response indicates that the proportion of time a *N. pseudoferus* spends in handling prey increases as the density of *T. absoluta* larvae (treated or untreated) increases. These results agree with some studies in which fungus treatment of host has no effect on functional response type of some parasitoids (Jarrahi and Safavi, 2016a; Jarrahi and Safavi, 2016b) and predators (Seiedy et al., 2012; Wu et al., 2015). Furthermore, *Nabis* species commonly display type II functional responses (Fernandez-Maldonado et al., 2017). Similarly, Ma et al. (2005) demonstrated that functional response of adult damsel bug, *N. kinbergii* Reuter (Hemiptera: Nabidae) to *Plutella xylostella* (Linnaeus) (Lepidoptera: Plutellidae) was type II. Moreover, Propp (1982) illustrated that *N. americanoferus* displayed type II response to *Spodoptera exigua* (Hübner) and *Lygus hesperus* Knight. Furthermore, Fathipour and Jafari (2003) demonstrated that functional response of *N. capsiformis* to second instar nymphs of *Creontiades pallidus* (Rambur) was type II. Contrarywise, Fernandez-Maldonado et al. (2017) found that functional response of *N. pseudoferus* females was type I. These authors used dead larvae of *S. exigua*, as prey to remove fighting. This can be a reason for the difference in functional response patterns in the two studies.

Based on our data, the highest coefficient of predator successful attack rate (a) is observed at 48h treatment. Usually, host locomotion

declines at developed stages of infection (Roy et al., 1999). Faster movement lets a prey more frequently dose escape from predators (van den Meiracker and Sabelis, 1999). Hence, it can be concluded that lower mobility of larvae in 48h treatment might have been the cause for predator to overcome the prey faster both due to lower energy investment in fighting and higher speed of moving proportional to the prey. On the other hand, the slowest attack rate and the longest handling time both were observed in 72h treatment. Presence of hyphal bodies or fungal metabolites in the hemolymph of the diseased host, may act as an inhibitory factor against the predator which may in turn reduce intake rate of the predator. Reduced attack rate also may be due to ceased mobility of the prey that may cause visual detection by the predator became more difficult. These results are consistent with some results reported in the literature (Madurappulige, 2005; Wekesa et al., 2007; Seiedy et al., 2012; Wu et al., 2016).

Our results showed that the handling time of the predator increased on treated preys, resulting in a reduction in mean number of prey consumed. Seiedy et al. (2012) also obtained similar results in *P. persimilis*-*Tetranychus urticae* predator-prey system whether treated or untreated by *B. bassiana*. Moreover, handling time of predatory mite, *Neoseiulus barkeri* Hughes increased with longer incubation of *B. bassiana*-exposed adult *Frankliniella occidentalis* Pergande (Thysanoptera: Thripidae), while the feeding rate decreased (Wu et al., 2015). Also, predation rate of *Orius albidipennis* (Reuter) decreased on *Thrips tabaci* Lindeman (Thysanoptera: Thripidae) larvae treated by *M. anisopliae*, and predator was able to detect the treated larvae (Pourian et al., 2011).

Time interval between prey infection and subsequent access of predator affects acceptance or rejection of prey by predator (Labbe' et al., 2006). Likewise, in our study incubation time increased from 0 to 72h, which in turn, caused daily per capita prey consumption rate to decrease from 9 to 7.11.

Our laboratory results showed that long-time exposure to the entomopathogenic fungus

causes partial mal-effects on functional response parameters of the predator. However it is not an absolute effect and the predator can successfully attack and kill the treated hosts in high numbers enough to insure that the predator increases host mortality by the fungus. For example, 30% mortality by the fungus corresponds to ≈ 4.8 preys, plus 7.11 killed by the predator exceeds that of the control ($4.8 + 7.11 = 11.91 > 10.96$). However, studying other aspects of such an integration is necessary for example, effects of environmental factors (temperature and humidity) on prey, predator and fungus; application sequence of the predator and the pathogen, prey preference of the predator, numerical response, ovipositional behavior and inter-specific competition by the predator individuals.

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Statement of conflicting interest

The authors state that there is no conflict of interest.

Author contribution

All authors contribute equally in this research.

References

- Agboton, B. V., Hanna, R., Onzo, A., Vidal, S. and von Tiedemann, A. 2013. Interactions between the predatory mite *Typhlodromalus aripo* and the entomopathogenic fungus *Neozygites tanajoae* and consequences for the suppression of their shared prey/host *Mononychellus tanajoa*. *Experimental and Applied Acarology*, 60: 205-217.
- Alma, C. R., Goettel, M. S., Roitberg, B. D. and Gillespie, D. 2007. Combined effects of the entomopathogenic fungus *Paecilomyces fumosoroseus* Apoka-97 and the generalist predator *Dicyphus hesperus* on whitefly populations. *BioControl*, 52: 669-681.
- Arbab, A., Heydari, N. and Pakyari, H. 2016. Temperature-dependent development of pale damsel bug, *Nabis capsiformis* Geramer (hemiptera: nabidae) using linear and non-linear models. *Crop Protection*, 89: 248-254.
- Baniameri, V., and Cheraghian, A. 2012. The first report and control strategies of *Tuta absoluta* in Iran. *EPPO Bulletin*, 42: 322-324.
- Barlow, N. D. and Goldson, S. L. 1993. A modelling analysis of the successful biological control of *Sitona discoideus* (Coleoptera: Curculionidae) by *Microctonus aethiopoides* (Hymenoptera: Braconidae) in New Zealand. *Journal of Applied Ecology*, 30: 165-178.
- Barrientos, Z. R., Apablaza, H. J., Norero, S. A. and Estay, P. P. 1998. Threshold temperature and thermal constant for development of the South American tomato moth, *Tuta absoluta* (Lepidoptera, Gelechiidae). *Ciencia Investigacion Agraria*, 25: 133-137.
- Biondi, A., Guedes, R. N. C., Wan, F. H., Desneux, D. 2018. Ecology, worldwide spread, and management of the invasive South American tomato pinworm, *Tuta absoluta*: past, present, and future. *Annual Review of Entomology*, 63: 239-258.
- Cabello, T., Gallego, J. R., Fern'andez-Maldonado, F. J., Soler, A., Beltran, D., Parra, A. and Vila, f. 2009. The damsel bug *Nabis pseudoferus* (Hem.: Nabidae) as a new biological control agent of the South American tomato pinworm, *Tuta absoluta* (Lep.: Gelechiidae), in tomato crops of Spain. *IOBC/WPRS Bulletin*, 49: 219-223.
- Contreras, J., Mendoza, J. E., Martinez-Aguirre, M. R., Garcia-Vidal, L., Izquierdo, J. and Bielza, P. 2014. Efficacy of entomopathogenic fungus *Metarhizium anisopliae* against *Tuta absoluta* (Lepidoptera: Gelechiidae). *Journal of Economic Entomology*, 107, 121-124.
- Cuthbertson, A. G. S., Mathers, J. J., Blackburn, L. F., Korycinska, A., Luo, W., Jacobson, R. J. and Northing, P. 2013. Population development of *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) under simulated UK glasshouse conditions. *Insects*, 4: 185-197.

- Desneux, N., Wajnberg, E., Wyckhuys, K. A. G., Burgio, G., Arpai, S., Narvaez-Vasquez, C., Gonzalez-Cabrera, A. J., Ruescas, D. C., Tabone, E., Frandon, J., Pizzol, J., Poncet, C., Cabello, T. and Urbaneja, A. 2010. Biological invasion of European tomato crops by *Tuta absoluta*: ecology, geographic expansion and prospects for biological control. *Journal of Pest Science*, 83: 197-215.
- Dick, J. T. A., Gallagher, K., Avlijas, S., Clarke, H. C., Lewis, S. E., Leung, S., Minchin, D., Caffrey, J., Alexander, M. E., Maguire, C., Harrod, C., Reid, N., Haddaway, N. R., Farnsworth, K. D., Penk, M. and Ricciardi, A. 2013. Ecological impacts of an invasive predator explained and predicted by comparative functional responses. *Biological Invasions*, 15: 837-846.
- Fathipour, Y. and Jafari, A. A. F. 2003. Functional response of predators *Nabis capsiformis* and *Chrysoperla carnea* to different densities of *Creontiades pallidus* nymphs. *Journal of Agriculture Science and Natural Resource*, 10: 125-133.
- Fernandez-Maldonado, F. J., Gallego, J. R., Valencia, A., Gamez, M., Varga, Z., Garay, J. and Cabello, T. 2017. Cannibalism: Do risks of fighting and reprisal reduce predatory rates? *Community Ecology*, 18: 87-96.
- Gámez, M., Sebestyén, Z., Varga, Z., Garay, J., Gallego, J. R., Fernández, F. J., Cabello, T. 2012. Multi-stage dynamic model for prey-predator interaction: Application to *Spodoptera exigua* (Lep.: Noctuidae) and *Nabis pseudoferus* (Hem.: Nabidae) under greenhouse conditions. *Integrated Control in Protected Crops, Mediterranean Climate, IOBC-WPRS Bulletin*, Vol. 80, p. 184.
- Ghaderi, S., Fathipour, Y., Asgari, Sh. 2017. Susceptibility of Seven Selected Tomato Cultivars to *Tuta absoluta* (Lepidoptera: Gelechiidae): Implications for Its Management. *Journal of Economic Entomology*, 110: 421-429.
- Gharekhani, G. H., Salek-Ebrahimi, H. 2014. Life Table parameters of *Tuta absoluta* (Lepidoptera: Gelechiidae) on different varieties of tomato. *Journal of Economic Entomology*, 107: 1765-1770.
- Ghoneim, K. 2014. Predatory insects and arachnids as potential biological control agents against the invasive tomato leaf miner, *Tuta absoluta* Meyrick (Lepidoptera: Gelechiidae), in perspective and prospective. *Journal of Entomology and Zoology Studies*, 2: 52-71.
- Havaskary, M., Farshbaf Pour-Abad, R., Kazemi, M. H., Rafeii, A. and Havaskary, Sh. 2012. An investigation to the fauna of Cimicomorpha (Heteroptera) from Parsabad-E-Moghan and vicinity, NW Iran. *Munis Entomology and Zoology Journal*, 7: 1101-1107.
- Holling, C. S. 1959. Some characteristics of simple types of predation and parasitism. *The Canadian Entomologist*, 91: 385-399.
- Inanl, C. and Oldargc, A. K. 2012. Effects of entomopathogenic fungi, *Beauveria bassiana* (Bals.) and *Metarhizium anisopliae* (Metsch.) on larvae and egg stages of *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae). *Ege Üniversitesi Ziraat Fakültesi Dergisi*, 49: 239-242.
- Ingegno, B. L., Ferracini, C., Gallinotti, D., Alma, A. and Tavella, L. 2013. Evaluation of the effectiveness of *Dicyphus errans* (Wolff) as predator of *Tuta absoluta* (Meyrick). *Biological Control*, 67: 246-252.
- Inglis, G. D., Enkerli, J. and Goettel, M. S. 2012. Laboratory techniques used for entomopathogenic fungi: Hypocreales. In: Lacey, L. A. (Ed.), *Manual of Techniques in Invertebrate Pathology*, Academic Press, London, pp: 189-253.
- Jarrahi, A. and Safavi, S. A. 2016a. Effects of pupal treatment with Proteus® and *Metarhizium anisopliae* sensu lato on functional response of *Habrobracon hebetor* parasitizing *Helicoverpa armigera* in an enclosed experiment system. *Biocontrol Science and Technology*, 26: 206-216.
- Jarrahi, A. and Safavi, S. A. 2016b. Temperature-dependent functional response and host preference of *Habrobracon hebetor* between fungus-infected and uninfected

- Ephestia kuehniella* larvae. Journal of Stored Products Research, 67: 41-48.
- Juliano, S. A. 2001. Non-linear curve fitting: predation and functional response curves. In: Cheiner, S. M. and Gurven, J. (Eds.), Design and Analysis of Ecological Experiments. Oxford University Press, New York, pp: 178-196.
- Labbe', R. M., Cloutier, C. and Brodeur, J. 2006. Prey Selection by *Dicyphus hesperus* of Infected or Parasitized Greenhouse Whitefly. Biocontrol Science and Technology, 16: 485-494.
- Labbe', R. M., Gillespie, D. R., Cloutier, C. and Brodeur, J. 2009. Compatibility of an entomopathogenic fungus with a predator and a parasitoid in the biological control of greenhouse whitefly. Biocontrol Science and Technology, 19: 429-446.
- Lester, P. J., Pree, D. J., Thistlewood, H. M. A., Trevisan, L. M. and Harmsen, R. 1999. Pyrethroid encapsulation for the conservation of acarine predators and reduced spider mite (Acari: Tetranychidae) outbreaks in apple orchards. Environmental Entomology, 28: 72-80.
- Luna, M. G., Sa'nchez, N. E., Pereyra, P. C., Nieves, E., Savino, V., Luft, E., Virla, E. and Speranza, S. 2012. Biological control of *Tuta absoluta* in Argentina and Italy: evaluation of indigenous insects as natural enemies. Bulletin OEPP/EPPO, 42: 260-267.
- Ma, J., Li, Y. Z., Keller, M. and Ren, S. X. 2005. Functional response and predation of *Nabis kinbergii* (Hemiptera: Nabidae) to *Plutella xylostella* (Lepidoptera: Plutellidae). Insect Science, 12: 281-286.
- Madurappulige, D. 2005. Effect of *Beauveria bassiana* (Balsamo) Vuillemin (Ascomycota: Hypocreales) on *Diadegma Semiclausum* (Hellen) (Hymenoptera: Ichneumonidae), a Parasitoid of *Plutella Xylostella* (L.) (Lepidoptera: Yponomeutidae). PhD Thesis, Lincoln University, New Zealand.
- Mahdavi, T. S., and Madadi, H. 2015. Biology and life table parameters of *Nabis pseudoferus* by feeding on cotton aphid *Aphis gossypii*. Journal of Biological Control of Pests and Plant Diseases, 4: 121-129. (In Persian with English abstract).
- Mahdavi, T. S., and Madadi, H. 2017. Prey preference of *Nabis pseudoferus* Remane on *Aphis gossypii* Glover and *Tuta absoluta* Meyrick. Plant Protection (Scientific Journal of Agriculture), 40: 33-47.
- Miranda, M. M. M., Picanco, M., Zanoncio, J. C. and Guedes, R. N. C. 1998. Ecological life table of *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae). Biocontrol Science and Technology, 8: 597-606.
- Modarres Awal, M. 2008. Contribution to Heteroptera fauna of Khorasan Razavi province of Iran. Turkish Journal of Entomology, 32: 243-253.
- Nozad-Bonab, Z., Hejazi, M. J., Iranipour, S. h. and Arzanlou, M. 2017. Lethal and sublethal effects of some chemical and biological insecticides on *Tuta absoluta* (Lepidoptera: Gelechiidae) eggs and neonates. Journal of Economic Entomology, 110: 1138-1144.
- Pires, L., Marques, E., Wanderley-Teixeira, V., Teixeira, Á., Alves, L. and Alves, E. 2009. Ultrastructure of *Tuta absoluta* parasitized eggs and the reproductive potential of females after parasitism by *Metarhizium anisopliae*. Micron, 40: 255-261.
- Pourian, H. R., Talaei-Hassanloui, R., Kosari, A. A. and Ashouri, A. 2011. Effect of *Metarhizium anisopliae* on searching, feeding and predation by *Orius albidipennis* (Hem., Anthocoridae) on *Thrips tabaci* (Thy., Thripidae). Biocontrol Science and Technology, 21: 15-21.
- Propp, G. D. 1982. Functional response of *Nabis americanoferus* to two of its prey, *Spodoptera exigua* and *Lygus hesperus*. Environmental Entomology, 11: 670-674.
- Rännbäck, L. M., Cotes, B., Anderson, P., Räämert, B. and Meyling, N. Y. 2015. Mortality risk from entomopathogenic fungi affects oviposition behavior in the parasitoid wasp *Trybliographa rapae*. Journal of Invertebrate Pathology, 124: 78-86.
- Rogers, D. 1972. Random search and insect population models. Journal of Animal Ecology, 41: 369-383.

- Roy, H. R., Pell, J. K. and Alderson, P. G. 1999. Effects of Fungal Infection on the Alarm Response of Pea Aphids. *Journal of Invertebrate Pathology*, 74: 69-75.
- Roy, H. E. and Pell, J. K. 2000. Interactions between entomopathogenic fungi and other natural enemies: Implications for biological control. *Biocontrol Science and Technology*, 10: 737-752.
- Royama, T. 1971. A comparative study of models for predation and parasitism. *Researches on Population Ecology Supplement*, 1: 1-90.
- SAS Institute. 2003. A guide to statistical and data analysis, version 9.1. SAS Institute, Cary, USA.
- Scorsetti, A. C., Pelizza, S., Fogel, M.N., Vianna, F. and Schneider, M. I. 2017. Interactions between the entomopathogenic fungus *Beauveria bassiana* and the Neotropical predator *Eriopis connexa* (Coleoptera: Coccinellidae): Implications in biological control of pest. *Journal of Plant Protection Research*, 57: 205-211.
- Seiedy, M., Saboori, A., Allahyari, H., Talaei-Hassanlou, R. and Tork, M. 2012. Functional response of *Phytoseiulus persimilis* (Acari: Phytoseiidae) on untreated and *Beauveria bassiana*-treated adults of *Tetranychus urticae* (Acari: Tetranychidae). *Journal of Insect Behavior*, 25: 543-553.
- Solomon, M. E. 1949. The natural control of animal populations. *Journal of Animal Ecology*, 18: 1-35.
- Tadele, S. and Emana, G. 2017. Entomopathogenic effect of *Beauveria bassiana* (Bals.) and *Metarrhizium anisopliae* (Metschn.) on *Tuta absoluta* (Meyrick) (Lepidoptera: Gelechiidae) larvae under laboratory and glasshouse conditions in Ethiopia. *Journal of Plant Pathology and Microbiology*, 8: 411-414.
- Urbaneja, A., González-Cabrera, J., Arnó J. and Gabarra, R. 2012. Prospects for biological control of *Tuta absoluta* in tomatoes of the Mediterranean basin. *Pest Management Science*, 68: 1215-1222.
- van den Meiracker, R. A. F. and Sabelis, M. W. 1999. Do functional responses of predatory arthropods reach a plateau? A case study of *Orius insidiosus* with western flower thrips as prey. *Entomologia Experimentalis et Applicata*, 90: 323-329.
- Wekesa, V. W., Moraes, G. J., Knapp, M. and Delalibera- Jr, I. 2007. Interactions of two natural enemies of *Tetranychus evansi*, the fungal pathogen *Neozygites floridana* (Zygomycetes: Entomophthorales) and the predatory mite, *Phytoseiulus longipes* (Acari: Phytoseiidae). *Biological Control*, 41: 408-414.
- Wu, S. Y., Gao, Y. L., Xu, X. N., Goettel, M. S. and Lei, Z. R. 2015. Compatibility of *Beauveria bassiana* with *Neoseiulus barkeri* for control of *Frankliniella occidentalis*. *Journal of Integrative Agriculture*, 14: 98-105.
- Wu, S. Y., Gao, Y. L., Smagghe, G., Xu, X. N. and Lei, Z. R. 2016. Interactions between the entomopathogenic fungus *Beauveria bassiana* and the predatory mite *Neoseiulus barkeri* and biological control of their shared prey/host *Frankliniella occidentalis*. *Biological Control*, 98: 43-51.
- Zappala, L., Biondi, A., Alma, A., Al-Jboory, I. J., Arno, J., Bayram, A., Chailleux, A., El-Arnaouty, A., Gerling, D., Guenaoui, Y., Shaltiel-Harpaz, L., Siscaro, G., Stavriniades, M., Tavella, L., Aznar, R. V., Urbaneja, A. and Desneux, N. 2013. Natural enemies of the South American moth, *Tuta absoluta*, in Europe, North Africa and Middle East, and their potential use in pest control strategies. *Journal of Pest Science*, 86: 635-647.

واکنش شکارگری سن *Nabis pseudoferus* (Hemiptera: Nabidae) در تغذیه از لارو *Tuta absoluta* (Lepidoptera: Gelechiidae) تیمار شده با قارچ بیمارگر *Metarhizium anisopliae*

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چکیده: واکنش تابعی یک ویژگی مهم رفتاری در برهم‌کنش بین شکار و شکارگر می‌باشد که می‌تواند در ارزیابی تأثیر دشمنان طبیعی مورد استفاده قرار گیرد. در این پژوهش واکنش تابعی حشرات ماده سن شکارگر *Nabis pseudoferus* Remane نسبت به لارو سن سوم *Tuta absoluta* (Meyrick) تیمار شده با غلظت LC_{30} ($10^4 \times 2/0.3$ کنیدی در میلی‌لیتر) قارچ بیمارگر *Metarhizium anisopliae* (Metschnikoff) Sorokin، جدایه DEMI 001 مورد بررسی قرار گرفت. شش تراکم طعمه (۱، ۲، ۴، ۸، ۱۰ و ۱۶) در چهار بازه زمانی (صفر، ۲۴، ۴۸ و ۷۲ ساعت) پس از تیمار در معرض سن شکارگر قرار گرفت. واکنش تابعی سن *N. pseudoferus* نسبت به تراکم‌های طعمه در تمام تیمارها از نوع دوم به‌دست آمد که نشان‌دهنده افزایش نرخ شکارگری تا رسیدن به آستانه سیری است. بیش‌ترین و کم‌ترین مقدار نرخ حمله (a) به‌ترتیب 0.440 ± 0.1052 و 0.133 ± 0.0509 بر ساعت در ۴۸ و ۷۲ ساعت پس از تیمار به‌دست آمد. بیشینه نرخ حمله تئوریک (T/T_h) در شاهد و برابر با ۱۰/۹۶ بود. نتایج نشان داد ترکیب قارچ *M. anisopliae* و سن شکارگر *N. pseudoferus* می‌تواند یک ترکیب مفید در مدیریت تلفیقی مینوز گوجه‌فرنگی باشد، با این حال این نتایج باید در شرایط کاربردی نیز مورد تأیید قرار گیرد.

واژگان کلیدی: واکنش تابعی، نرخ حمله، *N. pseudoferus*، *T. absoluta*، *M. anisopliae*