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Article

Diet dependent olfactory response and predation rate of *Neoseiulus californicus* (Acari: Phytoseiidae) in the presence of *Frankliniella occidentalis* and *Tetranychus urticae*

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Abstract

Responses of female predatory mite, *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae) with different diet experiences were examined in a Y-tube olfactometer. The odor sources were clean air vs. clean bean leaves, clean air vs. *Tetranychus urticae* (Koch) (Acari: Tetranychidae) infested bean leaves, and *T. urticae* infested bean leaves vs. clean bean leaves. Female predatory mites that fed on *T. urticae* as their previous diet for three generations, preferred clean leaves, spider mite infested leaves and again spider mite infested leaves over the alternative odor sources. When *N. californicus* was fed on *T. urticae* plus pollen, a significant movement was recorded towards clean leaves rather than clean air, but no significant preference was observed in two other olfactory experiments. Feeding on pollen alone, thrips first instar alone and thrips first instar plus pollen, the predators did not make a significant preference towards the arms in any of the olfactory experiments. Comparing the mean foraging time and predation rate of *N. californicus* when their prey (*T. urticae*) were experienced in a direct contact with *Frankliniella occidentalis* with predators whose prey had previousely received signals due to thrips (*F. occidentalis*) presence, we recorded a significantly higher foraging time and consumed prey in the patches where thrips were removed. According to our results, *N. californicus* is capable of detecting its prey and patches with heterospecific (*F. occidentalis*) and conspecific predators through its diet-based-understanding of chemical cues. Explanations for the olfactory response and foraging behavior of *N. californicus* are discussed.

Keywords: Experience, Predatory mite, Thrips, Tetranychidae, Two-spotted spider mite.

Introduction

Nutritional investigations are important for achieving successful culture of predators (Thompson & Hagen, 1999). Several studies have discussed the importance of ecological considerations in the diet of entomophagous insects (Hagen 1987; Vinson & Barbosa 1987). In order to enhancing the effectiveness of entomophagous insects and mites in the field and biological control programs, we need to manipulate their nutrition (Hagen 1987).The role of nutritional supplements for adult predators and parasitoids is well established. Anunciada & Voegele (1982) have reported the significant effect of adult feeding on the longevity and fecundity in *Tricogramma*. McEwen *et al.* (1996) demonstrated the value of adding honeydew for improving the performance of *Chrysopa carnea* (Stephens) on olive trees. Feeding attractants and deterrents produced by prey could also affect the foraging behavior of predatory mites (McMurtry & Rodriguez 1987). Pratt *et al.* (2003) studied the adult female survivorship, activity and oviposition in two populations of the predatory mite, *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae). They found that oviposition of female *P. persimilis* was significantly higher on *Tetranychus* species with which they were originally associated, in comparison with the predator populations that were reared on the congeneric prey species with which they were not originally associated.

Diet could be a key influence on the developmental time of predatory mites. *P. persimilis* performance on a diet of *Tetranychus evansi* Baker & Pritchard (Acari: Tetranychidae) was recorded as poor, but it was greatly improved when fed on a diet of *T. urticae*, *T. turkestani* Ugarov and Nikolski or *T. ludeni* Zacher (Galazzi & Nicoli, 1996). Dixon *et al.* (1997) studied the development of *P. persimilis* on diets of *T. urticae* and *T. lintearius* Dufour and showed that the mortality and development were similar on both diets. Dixon *et al.* (1997) suggested that the impact of *P. persimilis* on *T. lintearius* populations would be as great or greater than on *T. urticae* due to the latter's longer generation time.

Diet may also affect prey-predator interacations such as antipredator behavior. Magalhaes *et al.* (2005) found that intraguild [IG] prey can recognize its IG predator cues by means of the predator diet. They showed that the IG prey, *Neoseiulus cucumeris* (Oudemans) avoided its IG predator, *Orius laevigatus* (Fieber) only when the latter was fed on their shared food, Western Flower Thrips, *Frankliniella occidentalis* (Pergande). A previous study showed that thrips started anti-predator behavior (seeking refuge) when its predator, *Orius laevigatus* (Fieber), had previously fed on thrips conspecifics, while other diets of *Orius* did not trigger the thrips' escape response (Venzon *et al.* 2000). Thus, odours associated with diets, not only affect arthropod developmental periods, but also influence their behaviors.

Phytoseiidae includes the most important group of predatory mites that are successfully used for pest control on several plants and crops; e.g. of *Tetranychus urticae* (Nadeali *et al.* 2014) *Neoseiulus californicus* McGregor (Acari: Phytoseiidae), is a generalist predator which prefers spider mites, specially two-spotted spider mite, but can survive on alternative food sources such as pollen and other mites (McMurtry & Croft 1997). It has a broad range of tolerance in arid conditions and different temperatures in comparison with other phytoseiid predators (Castagnoli & Simoni 2003; van Baal *et al.* 2007). The homogeneous infection of *F. occidentalis* is usually observed in greenhouse treatments as one of the most important greenhouse pests beside spider mites and whiteflies. *F. occidentalis* is a major worldwide pest of various crops associated with serious losses of vegetables and ornamentals (Manners *et al.* 2013). There are commercially available predators (*Neoseiulus cucumeris* (Oudemans), /*Orius* spp.) that consume its different stages in the soil or on the foliage (Manners *et al.* 2013). *Frankliniella occidentalis* control needs an environmental friendly method to avoid the intensive use of insecticides and chemical resistance (Trdan *et al.* 2007). *Neoseiulus californicus* along with *Amblyseius swirskii* are suggested to establish a stable bio-control system by suppressing the pests mentioned above (Bolckmans *et al.* 2005; van Baal *et al.* 2007).

Here, we investigated whether the previous diet of the predatory mite *N. californicus* could affect its olfactory response and foraging behavior when it receives cues related to *F. occidentalis* and *T. urticae* on bean plants. We assessed whether *N. californicus* olfactory responses varied when it had previously fed on *T. urticae* or *F. occidentalis*. Second, we tested how the predator's previous diet affected its foraging behavior.

Material & methods

Mites and experimental units

Common bean plants (*Phaseolus vulgaris* L. var. Red Alamouti (Fabaceae)) were grown in plastic pots containing a mixture of perlite and soil (2:1). Plants were irrigated daily with tap water and fertilized (NPK: 20-20-20) and were kept in controlled conditions (23±1°C, 16L: 8D photoperiod, 65±5% RH) in growth chambers in Department of Plant Protection, Faculty of Agriculture, University of Tehran, Karaj, Iran. Spider mites, *T. urticae* and *F. occidentalis,* were reared on bean plants. Fresh bean plants were added to the rearing system regularly. Oilseed pollen was added to thrips colonies once every 3–4 days. Thrips pupae were allowed to develop on adjacent soil trays.

The predatory mite, *N. californicus* were reared on detached bean leaves put upside down on plastic sheet on a water saturated sponge. The plastic sheet was surrounded by napkin tapes which were put into the water from another side so that the predatory mites could drink water.

Olfactory experiments

We used an olfactometer to test whether the olfactory responses of female predatory mites would change when receiving different diets. The olfactometer consisted of a Y-shaped glass tube (4cm in diameter) with a Y-shaped metal wire in the middle of the tube, positioned parallel to the tube walls (Sabelis & van de Baan 1983). The lateral arms of the olfactometer through which the odors diffuesed towards the searching predator, were connected to the odor source boxes by means of plastic tubes. Clean air blown by a fan, passed through charcoal sacs just before odor source boxes. The hotwire air flow meters before the lateral arms of the olfactometer, provided a fixed 0.5 m/s air flow into each of the arms. The odor sources consisted of "spider mite infested plants" (5 same-aged bean plants (4 leaves), each infested by 150 female spider mites) as spider mite infested plants, 5 same-aged clean bean plants and clean air. Prior to the olfactory experiments, five separate cultures of *N. californicus* were provided on five different diets consisting of pollen (*Brassica napus*), *T. urticae*, *F. occidentalis* (first instar), *T. urticae* with pollen and *F. occidentalis* with pollen for three generations. The cultures were prepared on masses of detached bean leaves as mentioned previously. Three olfactory experiments were designed for each of the cultures as follows: *N. californicus* received: 1) odors related to clean bean plants from one arm vs. clean air from another arm: 2) odors related to spider mite infested bean plants from one arm vs. clean air from another arm; and 3) odors related to spider mite infested bean plants from one arm vs. odors related to clean bean plants from another arm. The same-aged predator females were kept starved for five hours prior to the experiments. Each treatment consisted of 20–25 replicates (predators). One predator was used in each replicate. The predator left for 5 minutes in the olfactometer. When the predator passed 9cm of the lateral railroad length, it was considered as a preference response to that stimulus. The odor sources were exchanged after five replicates and were renewed for each experiment. The olfactometer arms were cleaned after predator liberation (Zahedi-Golpayegani *et al*. 2007; Nadeali *et al*. 2014). Data analysis was performed by Goodness of fit test which includes a test for heterogeneity among replicate experiments (Sokal & Rohlf 1995).

Foraging and the predation rate of N. californicus *in the presence & absence of western flower thrips,* F. occidentalis

We tested whether the foraging time and predation rate of the predator would differ when *T. urticae* had previously experienced thrips (in the same patch) in comparison with when they had just exposed it. We put 10 spider mite females along with two female thrips in a patch (9 cm in diameter) consisting of a 6 cm bean leaf placed upside down on agar substrate. After 24 hours, the remaining spider mites (experienced the presence of thrips) were transferred to a new patch (considered as Control) with a female *N. californicus*. The treatment consisted of the same patch, including a female thrips (direct exposure). The Petri dishes were sealed by parafilm and kept in a growth chamber for 24 hours. Twenty five replicates were considered both for treatment and Control. After 24 hours, we monitored the Petri dishes for three continuous hours with a stereomicroscope to record the period of time the predator spent searching for prey. Foraging periods were recorded within three hours. Data analysis was performed by independent t-test using SPSS 16.

To find out whether the predator previous diet would affect its predation rate, two cultures of *N. californicus*, one feeding on *T. urticae* and the other feeding on *F. occidentalis* (1^{st} instar) were provided 10 days prior to the experiments. To start the experiment, same-aged *N. californicus* females (kept starved for five hours) related to either of the diets mentioned above, were put on leaf discs consisted of a bean leaf (2×2 cm^2) put upside down on saturated cotton wool in a Petri dish (9 cm in diameter). Five *T. urticae* protonymphs/*Five F. occidentalis* (1^{st} instar) were added to the leaf discs. The replicates which received similar prey with their previous culture were considered as Control. The number of consumed prey was recorded after 24 hours. The experiments replicated 30 times. Data analysis was performed by independent t-test in SPSS 16.

Results

Olfactory experiments

Tetranychus urticae *as diet*

When *N. californicus* received odors related to clean bean plants from one arm vs. clean air from another arm, 35 predators (from 60) moved towards each of the arms (so that 25 without any preference), 24 of which (68.5%) preferred odors related to clean bean leaves. There was a significant difference between the numbers of predators moved towards clean leaves in comparison with the ones attracted to clean air (11 predators, 31.4%) (P= 0.026) (Table 1a).

When the predatory mites received odors related to *T. urticae* infested bean plants from one arm vs. clean air from another arm, 36 predators (from 60) moved towards each of the arms, 26 of which (72%) preferred odors related to spider mite infested leaves. There was a significant difference between the numbers of predators moved towards infested leaves in comparison with the ones attracted to clean air (10 predators, 28%) (P = 0.006) (Table 1b).

When *N. californicus* received odors related to clean beans leaves from one arm vs. *T. urticae* infested leaves from another, 37 predators (from 60) moved towards each of the arms, 26 of which (70%) were attracted to infested bean leaves. A significant difference was observed between the number of predators that moved towards each of the arms (P= 0.012) (Table 1c).

Table 1. Results for replicated G-tests for the response of *N. californicus* to odours related to: a) clean bean plants from one arm (+) and clean air from another arm (-), b) *T. urticae* infested bean plants from one arm (+) and clean air from another arm (-), and c) *T. urticae* infested leaves from one arm (+) and clean beans leaves from another arm (-) when previous diet was consisted of spider mites

Odour source	G_h	d*f*	P value	G_p	d*f*	P value	G_T	d*f*	P value
a) clean bean plants vs. clean air	0.748	2	0.68 *n.s.*	4.946	1	0.026*	5.694	3	0.12 *n.s.*
b) *T. urticae* infested bean plants vs. clean air	1.290	2	0.52 *n.s.*	7.365	1	0.006**	8.656	3	0.034*
c) *T. urticae* infested leaves vs. clean beans leaves	0.114	2	0.94 *n.s.*	6.259	1	0.012*	6.374	3	0.094 *n.s.*

*: P < 0.05, **: P < 0.01, *n.s.*: not significant

Tetranychus urticae *and pollen as diet*

When *N. californicus* received odors related to clean bean plants from one arm vs. clean air from another arm, 44 predators (from 60) moved towards each of the arms, 32 of which (73%) preferred odors related to clean bean leaves. There was a significant difference between the numbers of predators moved towards clean leaves in comparison with the ones attracted to clean air (12 predators, 27%) (P = 0.002) (Table 2a).

When the predatory mites received odors related to *T. urticae* infested bean plants from one arm vs. clean air from another arm, 43 predators (from 60) moved towards each of the arms, 27 of which (63%) preferred odors related to spider mite infested leaves. No significant difference was observed between the number of predators moved towards infested leaves and the ones attracted to clean air (16 predators, 37%) (P = 0.09) (Table 2b).

Table 2. Results for replicated G-tests for the response of *N. californicus* to odours related to: a) clean bean plants from one arm (+) and clean air from another arm (-), b) *T. urticae* infested bean plants from one arm (+) and clean air from another arm (-), and c) *T. urticae* infested leaves from one arm (+) and clean beans leaves from another arm (-) when previous diet was consisted of spider mites and pollen

Odour source	G_h	d*f*	P value	G_p	d*f*	P value	G_T	d*f*	P value
a) clean bean plants vs. clean air	2.418	2	0.29 *n.s.*	9.433	1	0.002**	11.851	3	0.007**
b) *T. urticae* infested bean plants vs. clean air	0.701	2	0.7 *n.s.*	2.845	1	0.091 *n.s.*	3.547	3	0. 314
c) *T. urticae* infested leaves vs. clean beans leaves	0.925	2	0.62 *n.s.*	9.433	1	0.87 *n.s.*	0.948	3	0.813 *n.s.*

*: P<0.05, **: P<0.01, *n.s.*: not significant

When *N. californicus* received odors related to clean beans leaves from one arm vs. *T. urticae* infested leaves from another, 43 predators (from 60) moved towards each of the arms, 22 of which (51%) attracted to infested bean leaves. No significant difference was observed between the number of predators moved towards each of the arms (P = 0.87) (Table 2c).

Pollen as diet

When *N. californicus* received odors related to clean bean plants from one arm vs. clean air from another arm, 43 predators (from 60) moved towards each of the two arms, 17 of which (40%) preferred odors related to clean bean leaves. No significant difference was observed between the number of predators moved towards clean leaves and the ones attracted to clean air (26 predators, 60%) (P = 0.16) (Table 3a).

When the predatory mites received odors related to *T. urticae* infested bean plants from one arm vs. clean air from another arm, 43 predators (from 60) moved towards each of the arms, 20 of which (47%) preferred odors related to spider mite infested leaves. No significant difference was observed between the number of predators moved towards infested leaves and the ones attracted to clean air (23 predators, 53%) (P = 0.6) (Table 3b).

When *N. californicus* received odors related to clean beans leaves from one arm vs. *T. urticae* infested leaves from another, 44 predators (from 60) moved towards each of the arms, 21 of which (48%) attracted to infested bean leaves. No significant difference was observed between the number of predators moved towards each of the arms (P = 0.76) (Table 3c).

Table 3. Results for replicated G-tests for the response of *N. californicus* to odours related to: a) clean bean plants from one arm (+) and clean air from another arm (-), b) *T. urticae* infested bean plants from one arm (+) and clean air from another arm (-), and c) *T. urticae* infested leaves from one arm (+) and clean beans leaves from another arm (-) when previous diet was consisted of pollen

Odour source	G_h	d*f*	P value	G_p	d*f*	P value	G_T	d*f*	P value
a) clean bean plants vs. clean air	0.008	2	0.995 *n.s.*	1.897	1	0.168 *n.s.*	1.905	3	0.591 *n.s*
b) *T. urticae* infested bean plants vs. clean air	1.633	2	0.441 *n.s.*	0.209	1	0.647 *n.s*	1.841	3	0.605 *n.s*
c) *T. urticae* infested leaves vs. clean beans leaves	5.397	2	0.067 *n.s*	0.090	1	0.762 *n.s* .	5.487	3	0.139*n.s.*

n.s.: not significant

Thrips larvae as diet

When *N. californicus* received odors related to clean bean plants from one arm vs. clean air from another arm, 44 predators (from 60) moved towards each of the arms, 24 of which (55%) preferred odors related to clean bean leaves. No significant difference was observed between the number of predators moved towards clean leaves and the ones attracted to clean air (20 predators, 45%) (P = 0.54) (Table 4a).

When the predatory mites received odors related to *T. urticae* infested bean plants from one arm and clean air from another arm, 54 predators (from 60) moved towards each of the arms, 27 of which (50%) preferred odors related to spider mite infested leaves. No significant difference was observed between the number of predators moved towards infested leaves and the ones attracted to clean air (27 predators, 50%) (P = 1) (Table 4b).

When *N. californicus* received odors related to clean beans leaves from one arm vs. *T. urticae* infested leaves from another, 51 predators (from 60) moved towards each of the arms, 24 of which (47%) attracted to infested bean leaves. No significant difference was observed between the number of predators moved towards each of the arms (P = 0.67) (Table 4c).

Table 4. Results for replicated G-tests for the response of *N. californicus* to odours related to: a) clean bean plants from one arm (+) and clean air from another arm (-), b) *T. urticae* infested bean plants from one arm (+) and clean air from another arm (-), and c) *T. urticae* infested leaves from one arm (+) and clean beans leaves from another arm (-) when previous diet was consisted of thrips larvae

Odour source	G_h	d*f*	P value	G_p	d*f*	P value	G_T	d*f*	P value
a) clean bean plants vs. clean air	0.306	2	0.857 *n.s.*	0.364	1	0.546 *n.s.*	0.670	3	0.880 *n.s.*
b) *T. urticae* infested bean plants vs. clean air	0.807	2	0.667 *n.s.*	0	1	1	0.807	3	0.847 *n.s.*
c) *T. urticae* infested leaves vs. clean beans leaves	0.104	2	0.948 *n.s.*	0.176	1	0.674 *n.s.*	0.280	3	0.963 *n.s.*

n.s.: not significant

Thrips larvae and pollen as diet

When *N. californicus* received odors related to clean bean plants from one arm vs. clean air from another arm, 49 predators (from 60) moved towards each of the arms, 21 of which (43%) preferred odors related to clean bean leaves. No significant difference was observed between the number of predators moved towards clean leaves and the ones attracted to clean air (28 predators, 57%) (P = 0.31) (Table 5a).

When the predatory mites received odors related to *T. urticae* infested bean plants from one arm vs. clean air from another arm, 52 predators (from 60) moved towards each of the arms, 22 of which (42%) preferred odors related to spider mite infested leaves. No significant difference was observed between the number of predators moved towards infested leaves and the ones attracted to clean air (30 predators, 58%) (P = 0.26) (Table 5b).

Table 5. Results for replicated G-tests for the response of *N. californicus* to odours related to: a) clean bean plants from one arm (+) and clean air from another arm (-), b) *T. urticae* infested bean plants from one arm (+) and clean air from another arm (-), and c) *T. urticae* infested leaves from one arm (+) and clean beans leaves from another arm (-) when previous diet was consisted of thrips larvae and pollen

Odour source	G_h	d*f*	P value	G_p	d*f*	P value	G_T	d*f*	P value
a) clean bean plants vs. clean air	0.296	2	0.862 *n.s*	1.003	1	0.316 *n.s*	1.299	3	0.729 *n.s*
b) *T. urticae* infested bean plants vs. clean air	0.022	2	0.988 *n.s*	1.235	1	0.266 *n.s*	1.257	3	0.739 *n.s*
c) *T. urticae* infested leaves vs. clean beans leaves	0.125	2	0.939 *n.s*	0.183	1	0.668 *n.s*	0.308	3	0.958 *n.s*

n.s.: not significant

When *N. californicus* received odors related to clean beans leaves from one arm vs. *T. urticae* infested leaves from another, 49 predators (from 60) moved towards each of the arms, 23 of which (47%) attracted to infested bean leaves. No significant difference was observed between the number of predators moved towards each of the arms (P = 0.66) (Table 5c).

The effect of previous diet on the predation rate of N. californicus

When *N. californicus* had experienced thrips as its previous diet for 10 days, 2.86 ± 0.16 *T. urticae* protonymphs were eaten during the 24 h experimental period. The mean number of *T. urticae* protonymphs consumed by the predators with spider mites as their previous diet were recorded as 3.50 ± 0.16 which was significantly more than the mean number of *T. urticae* protonymphs consumed by *N. californicus* which had previously fed on thrips (P < 0.01) (Table 6a).

When *N. californicus* experienced *T. urticae* as its previous diet for 10 days, 1.86 ± 0.14 thrips 1st instar were eaten during the 24 h experimental period. The mean number of thrips consumed by the predators with thrips as their previous diet were recorded as 3.33 ± 0.12 which was significantly more than the mean number of thrips consumed by *N. californicus* which had previousely fed on *T. urticae* (P < 0.01) (Table 6b).

Table 6. Independent t test for the effect of previous diet on the predation rate of *N. californicus* when its previous diet was consisted of *T. urticae* (Control) and thrips (treatment)

		Mean number of *T. urticae* protonymphs consumed	Standard Error	P value
a)	Control (fed on *T. urticae*)	3.50	0.16	< 0.01
	Treatment (fed on thrips)	2.86	0.16	
b)		Mean number of thrips 1st instar consumed	Standard Error	P value
	Control (fed on thrips)	3.33	0.12	< 0.01
	Treatment (fed on *T. urticae*)	1.86	0.14	

Foraging and the predation rate of N. californicus *in the presence and absence of western flower thrips,* F. occidentalis

We compared the foraging time of *N. californicus* for *T. urticae* prey within three hours. The mean foraging time of predator in the presence of *F. occidentalis* (67.61 ± 2.57 min.) was significantly less than when thrips were removed (113.73 ± 2.31) (P < 0.01) (Table 7). Furthermore, the predators consumed significantly higher numbers of *T. urticae* (6.28 ± 0.22) in the patches from which thrips were removed, in comparison with those which thrips were present in (3.84 ± 0.19) (P > 0.01).

Table 7. Independent t test for the effect of thrips presence on the foraging time of *N. californicus*

	The mean of movement time	Standard Error	P value
Control (without thrips)	113.73	2.31	< 0.01
Treatment (with thrips)	67.61	2.57	

Discussion

In the olfactory experiments where *T. urticae* was used as prey, *N. californicus* was able to differentiate spider mite infested and clean leaves; they were attracted towards those rather than clean air. Dicke & Sabelis (1988) also showed that *Phytoseiulus persimilis* preferred infested bean leaves rather than clean air. Wijk *et al.* (2008) reported on the olfactory response of *P. persimilis* to *T. urticae* infested leaves in a similar way. It seems that as the predators in these experiments used the same diet as they used previously in the main culture, they got used to the odors related to *T. urticae* presence such as prey alarm pheromone and HIPVs (Herbivore induced plant volatiles). Drukker *et al*. (2000) examined *P. persimilis* females with the same feeding background as our predators through a set of similar olfactory experiments on potato leaves and observed that the predator preferred the volatiles related to spider mite infested leaves rather than both clean leaves and clean air. Their results contrasted with ours as *P. persimilis* did not differentiate between odors related to clean leaves and clean air. This could be due to the present volatiles with the ones the predator used to receive in the main culture previously. Koveos & Broufas (2009) studied the olfactory response of the predatory mite, *Amblyseius andersoni* Chant and pointed out that the similarity between the present and previous diet (volatiles) is of high importance for the predator which is searching for prey.

In the olfactory experiments which both pollen and prey were considered as predator diet, *N. californicus* exhibited a preference only between clean air and odors related to clean leaves, moving towards the latter. Unlike the first set of experiments, no attraction was observed towards infested leaves when the alternative choices were either clean leaves or clean air. One would wonder why adding pollen to the predator previous diet would change its olfactory response. van Rijn & Tanigoshi (1999) noted that pollen is utilized as an easy food source for phytoseiid predators for feeding. Cook *et al.* (2002) supported the hypothesis that pollen odor sources are important for insects in order to locate their food. Koveos & Broufas (1999) showed that *Typhlodromus kerkirae* did not respond to spider mite infested bean leaves when they had been reared on *Vicia fabae* pollen previously. Vantornhout (2006) reported that the predatory mite *Iphiseius degenerans* showed a similar olfactory response (attraction with no significant difference) towards spider mite induced plant volatiles and odors related to *Ricinus communis* pollen. Therefore, it seems that the predator formed an association between pollen odors and food presence so that pollen was used as a cue to signify its prey. Here, *N. californicus* was reared on both pollen and spider mites so that, after eliminating pollen from the food odor source, we observed no attraction towards prey.

In the olfactory experiments which *N. californicus* was fed on pollen alone, the predator did not differentiate between clean leaves vs. clean air, infested leaves vs. clean air and infested leaves vs. clean leaves. This means that *N. californicus* was not able to recognize odors related to clean leaves and odors related to spider mite infested leaves. The only difference between this experiment and the first olfactory ones refers to the predator previous diet. It seems that as the predator was reared on pollen (no prey was added) for three generations, no recognition ability of spider mite infested leaves could be expected. This is again similar to Vantornhout (2006) who reported that the predatory mites *I. degenerans*, which had previously been reared on *Ricinus communis* pollen could not differentiate odors related to spider mite infested bean leaves or whitefly infested ones rather than clean bean leaves. He concluded that, as *I. degenerans* had not experienced the prey species in its previous diet, it showed a naïve behavior toward them. Papaj & Prokopy (1989) proposed that repeated exposure to a stimulus would bring about an increase in response to that stimulus. Dicke *et al.* (1990) found that *P. persimilis* reared on spider mite-infested bean leaves preferred the odor from spider mite-infested bean leaves over the odor from infested cucumber; and that this preference changed gradually to a preference for infested cucumber during a period of 7 days in which the predators were reared on infested cucumber.

When *N. californicus* were fed on thrips larvae, they did not differentiate between clean leaves vs. clean air, infested leaves vs. clean air and infested leaves vs. clean leaves. This was to some extent similar to Zhang & Sanderson (1992) who reported that *P. persimilis* with previous experience on *T. urticae* infested rose leaves did not make a preference between rose and bean infested leaves. From another point of view, as stimuli from both prey and plant are involved in the predator foraging behavior (Zhang & Sanderson 1992), one may wonder why our predator with previous experience on *T. urticae* infested bean leaves, could not recognize the odors related to the same clean leaves rather than clean air.

Again in the experiments in which *N. californicus* was previously fed on both pollen and thrips larvae, the odors related to *T. urticae* induced bean volatiles were not so attracting for the predator to make it move towards *T. urticae* infested plants. The effect of each of the diets as previous experience was discussed above. The probable covering effect of odors related to the thrips larvae and pollen on odors related to clean leaves needs further investigations.

Foraging and the predation rate of N. californicus *in the presence and absence of western flower thrips,* F. occidentalis

Our results showed that the predator previous diet could affect its predation rate. The predators with similar present and previous diet consumed more prey regardless of the prey species (thrips or spider mite). Similarly, Castagnoli *et al.* (2002) noted that the diets used in predator mass rearing processes, not only influenced the predator predation rate through changing the rate of prey and predator encountering, but also changed their other biological characteristics. Vantornhout *et al.* (2006) demonstrated that diet had a significant effect on the life cycle parameters of the predatory mite, *Iphiseius degenerans* (Berlese). Mendel & Schausberger (2011) reported that in the mutual IGP (Intraguild predation) between *N. californicus* and *N. cucumeris*, the predation rate on larvae was not affected by previous diet but larvae from pollen-fed mothers were more profitable. Castagnoli *et al.* (2002) suggested that time since diet transfer should be added to the factors (i.e. feeding history) that may affect the predator functional and numerical response. Meng *et al.* (2006) offered whiteflies choice between plants with *Typhlodromips swirskii* (Athias-Henriot) with different previous diets and similar present diets. They showed that both present and previous diets of predators were important in prey anti-predator behavior (in addition to predator consumption rate) with present diet having a larger effect. Whereas it has been shown earlier that the diet of predators affects their life cycle parameters and anti-predator behavior, we show here that previous diet could also affect their predation rate.

Our expectation in the foraging experiment was that in the patches which thrips were present, *T. urticae* (as the predator diet) should reduce its movement (Grostal & Dicke, 1999) as it had experienced the risk of thrips presence (both in treatment and control), so that *N. californicus* might increase its foraging behavior through prey searching. In contrast the results showed a significant decrease in *N. californicus* mean movement time in the presence of thrips. It seems that the predatory mite has detected thrips as a competitor (van Baal *et al.* 2007) and avoided from a direct contact in order to decrease the IGP (intraguild predation) consequences. The predator less predation rate on *T. urticae* could also be due to both prey and predator less movements in the presence of thrips.

Predators are capable of detecting their prey and patches with heterospecific and conspecific predators through their diet based understanding of chemical cues. Diet based foraging behavior of predators can have ecological consequences at the individual and community levels. Many studies have focused on diet related predator cues which elicits anti-predator behavior in prey species but information about the effect of previous diet on the foraging behavior of predators themselves is rare. It is still an open question how long the predator foraging behavior could be influenced from its previous diet.

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پاسخ بویایی و نرخ شکارگری وابسته به رژیم غذایی در *Neoseiulus californicus* (Acari: Phytoseiidae) در حضور *Frankliniella occidentalis* و *Tetranychus urticae*

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چکیده

در این پژوهش پاسخ‌های کنهٔ ماده شکارگر *Neoseiulus californicus* (McGregor) (Acari: Phytoseiidae) تحت رژیم‌های غذایی متفاوت در لوله Y شکل دستگاه بوسنج بررسی شد. منابع مواد فرار بویایی عبارت بودند از: هوای پاک در مقابل برگ‌های تمیز لوبیا، هوای پاک در مقابل برگ‌های لوبیای آلوده به کنهٔ تارتن دولکه‌ای *Tetranychus urticae* (Koch) (Acari: Tetranychidae) و برگ‌های آلوده به کنهٔ تارتن دولکه‌ای *T. urticae* درمقابل برگ‌های تمیز لوبیا. کنه‌های ماده شکارگری که تا سه نسل پیشین خود از *T. urticae* تغذیه کرده بودند، به ترتیب برگ‌های تمیز، برگ‌های آلوده به کنهٔ تارتن و دوباره، برگ‌های آلوده به کنهٔ تارتن را در مقابل گزینه مخالف هر کدام انتخاب کردند. هنگامی که کنهٔ شکارگر ماده *N. californicus* از کنه تارتن *T. urticae* همراه با گرده تغذیه کرده بود، در مواجهه با برگ‌های تمیز در مقابل هوای پاک، به‌طور معنی‌داری به سمت برگ‌های تمیز حرکت کرد، این درحالی است که در دو آزمون بویایی‌سنجی بعدی، ترجیحی مشاهده نشد. کنه شکارگر با تغذیه از گرده تنها، تریپس تنها و تریپس به همراه گرده، گرایش معنی‌داری را به سمت هیچ‌یک از بازوها نشان نداد. میانگین زمان کاوشگری و میزان شکارگری *N. californicus* در شرایطی که شکارش-کنه تارتن دولکه‌ای، *T. urticae*- پیش‌تر در تماس مستقیم با تریپس *F. occidentalis* بوده، با زمان کاوشگری و میزان شکارگری کنه‌هایی که شکارشان پیش‌تر علایم شیمیایی مربوط به تریپس را دریافت کرده بود مقایسه شد. نتایج نشان داد که در حالت تماس مستقیم شکار با تریپس، کنه شکارگر به‌طور معنی‌داری مدت زمان بیشتری را به کاوشگری اختصاص داده و شکارهای بیشتری را تغذیه کرده بود. براساس این نتایج، کنه شکارگر *N. californicus* قادر است بر اساس اطلاعاتی که به‌واسطه رژیم غذایی پیشین خود به‌دست

می‌آورد، شکار و همین‌طور لکه‌های دربرگیرنده افراد هم‌گونه و غیرهم‌گونه (*F. occidentalis*) را شناسایی کند. پاسخ کنهٔ شکارگر در آزمون‌های بویایی و همین‌طور رفتار کاوشگری آن مورد بحث قرار گرفته است.

واژگان کلیدی: تجربه، کنه شکارگر، تریپس، Tetranychidae، کنه تارتن دولکه‌ای.

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