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Article

The effects of two essential oils on the functional response of *Amblyseius swirskii* (Acari: Phytoseiidae) fed on *Frankliniella occidentalis* (Thysanoptera: Thripidae)

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ABSTRACT

Amblyseius swirskii Athias-Henriot (Acari: Phytoseiidae) is an important generalist predator used to control several important crops' pests such as spider mites, whiteflies, and thrips. In this study, the lethal concentrations of *Mentha piperita* L. (Lamiaceae) and *Laurus nobilis* L. (Lauraceae) essential oils (EOs) were calculated on the adult stage of the *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae). The side effects of EOs were investigated on the functional response of *A. swirskii* on *F. occidentalis* 1st instar larvae. The females of *A. swirskii* were exposed to sub-lethal concentrations (LC₃₀ and LC₅₀) of EOs for 12 h with the fumigant exposure method. In the control, the treatment was performed by using Triton X-100 (0.02). Treated predatory mites were selected randomly, and transferred to the experimental arenas containing 2, 4, 8, 16, 32, and 64 thrips. The predators were kept in the experimental arenas for 24 h at 25 ± 1 °C, 60 ± 10% RH, and a photoperiod of 16: 8 h (L: D). Results indicated that *A. swirskii* exhibited a type II functional response, regardless of EOs applications. The shortest (1.62 ± 0.046 h) and the longest (1.91 ± 0.049 h) handling times, were recorded in the control and the LC₅₀ treatment of *M. piperita* EO, respectively. There was not a significant difference between attack rate coefficients (α) under sub-lethal concentrations. Based on the results, the two EOs have the potential to be used in *F. occidentalis* management, but their side effects on *A. swirskii* should be considered in IPM programs.

KEYWORDS: Attack rate, botanical pesticide, handling time, integrated pest management, predatory mite.

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INTRODUCTION

Western flower thrips, *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae), is one of the most destructive plant pests due to its cosmopolitan occurrence as well as its wide range of host plants (He *et al.* 2020; Reitz *et al.* 2020). This pest causes serious damage to plants directly by piercing and sucking sap excessively from leaves, flowers, and fruits, and indirectly by transmitting tospoviruses including impatiens necrotic spot virus and tomato spotted wilt virus (Gao *et al.* 2012). Management of this pest by conventional pesticides is challenging due to its high reproductive potential and relatively short life cycle, which can facilitate the rapid development of resistance to various groups of insecticides (Jensen 2000; Mouden *et al.* 2017). Despite being aware of this issue, the main method

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for controlling this pest has been through conventional insecticides, which causes different problems such as environmental pollution, and toxicity to humans and non-target organisms such as natural enemies (Gill and Garg 2014). In this regard, there is a pressing need for the development of safe alternatives to establish an effective and environmental friendly pest management strategy. Natural enemies including predators and parasitoids are one of the groups that have received the most attention due to their values in integrated pest management (IPM) programs (Silva *et al.* 2019; Duso *et al.* 2020). Among them, the members of the family Phytoseiidae (Acari: Mesositimata) received a special interest in augmentative control of *F. occidentalis*. The phytoseiid mites are categorized into four basic lifestyle types based on their feeding habits; type I (specialized), type II (selective), type III (generalist), and type IV (pollen-feeding generalist predators) (McMurtry *et al.* 2013). *Amblyseius swirskii* Athias-Henriot belonging to the type III group is widely utilized as a biocontrol agent for several important crop pests including thrips. In addition, it has been available in the commercial market in more than 50 countries since 2005 (Messelink *et al.* 2005, 2006; Wimmer *et al.* 2008; Calvo *et al.* 2015; Fathipour and Maleknia 2016).

Essential oils (EOs), which are commonly extracted from various parts of plants using steam distillation or hydrodistillation methods are perceived by the general public as safer alternatives to synthetic insecticides (Isman 2006; Pavela 2016). Plant EOs have a high potential for development as an alternative to insecticides, particularly in small-scale cultivated crops or greenhouse production in organic farming (Isman 2006). Among EOs, those produced by species of the Lamiaceae family, have the potential to be used in pest management programs (Ebadollahi *et al.* 2020; da Silva 2021).

Some studies suggested that EOs could be used as an alternative to insecticides for the management of *F. occidentalis* in greenhouse conditions (Stepanycheva *et al.* 2018, 2019; Durr *et al.* 2022; Gharbi and Tay 2022; Kirişik 2022). Although some EOs are not toxic to non-target organisms and can be used in combination with biological control agents, assessment of the potential side effects that pesticides have on the natural enemies is an important part of IPM programs (Consoli *et al.* 1998; Takada *et al.* 2001; Miresmailli *et al.* 2006). The functional response of predators in predator-prey interactions describes individual predation rates, and it is often used to estimate the efficacy of predators (Holling 1959). In this regard, the purpose of this study was to determine the functional response of predatory mite, *A. swirskii* under EOs obtained from *Mentha piperita* (Lamiaceae) and *Laurus nobilis* (Lauraceae), fed on the first instar larvae of *F. occidentalis*.

MATERIALS AND METHODS

Stock culture of Frankliniella occidentalis and Amblyseius swirskii

The initial colony of *F. occidentalis* was established with adult individuals collected from strawberry flowers (*Fragaria ananassa* L., Rosaceae) from a greenhouse in the Hashtgerd region, Alborz, Iran. They were reared on common bean pods (*Phaseolus vulgaris* L., Fabaceae) and maintained in ventilated plastic bottles (15 cm in height, 5 cm in diameter). To obtain a sufficient cohort of individuals of the same age for the experiments, approximately 100 adult females were transferred from the initial colony to each bottle with five bean pods and allowed to lay eggs for 24 hours. The *F. occidentalis* adults were removed and bean pods were transferred to a new bottle containing fresh bean pods and monitored daily until the first instars emerged.

The initial population of *A. swirskii* was obtained from Koppert biological systems (The Netherlands) and subsequently reared on units including a piece of hard black-green plastic placed on a water-saturated sponge in a plastic tray filled with water. The borders of the plastic tray were covered with water-saturated tissue paper to ensure a constant water supply for the mites and to prevent them from escaping (Abad-Moyano *et al.* 2009). Cotton threads (4 cm long) were provided on the rearing units to serve as oviposition sites. Twice a week, mites were fed with maize pollen, *Zea mays* L. (Poaceae), and mixed stages of *Tetranychus urticae* Koch (Acari: Tetranychidae) (Ragusa and Swirski 1975). To obtain a cohort of individuals of the same age, approximately 100

gravid females were randomly selected from the initial colony and transferred to the new arenas for egg-laying; females were removed after 24 hours. The laid eggs were monitored at daily intervals. The rearing arenas were kept in growth chambers under controlled conditions at 25 ± 1 °C, $60 \pm 10\%$ RH, and a photoperiod of 16:8 h (Light:Dark).

The essential oils extraction

The fresh leaves of *L. nobilis* were collected in April (2021) in the region of Iran-Karaj (College of Agriculture and Natural Resources) and leaves of *M. piperita* were purchased from Pakguiah company (Iran-Tehran (www.pakguiah.com)). The dried leaves of peppermint (*M. piperita*), and bay leaf (*L. nobilis*), were powdered using an electric grinder, and 50 gr of powder with 500 ml of distilled water were extracted for three hours using a Clevenger-type apparatus (Heating Mantle-MODEL: MS-E105) by hydrodistillation method. The oil was dried over anhydrous Na_2SO_4 to remove water and stored in clean dark glass bottles at 4 °C in the refrigerator until utilized for experiments (Milojević *et al.* 2008).

Toxicity bioassays of essential oils against Frankliniella occidentalis

To determine the fumigant toxicity of *M. piperita* and *L. nobilis* EOs on young females of *F. occidentalis* (under 24 hours), five concentrations of EOs that caused 10 to 90% mortality in Preliminary tests were dissolved in ethanol (98 %, Merck, Germany) with Triton™ X-100 (100%, Merck, Germany) (0.02) and applied to Whatman filter papers (No.4, 1×1 cm diameter). Each filter paper was attached to the lid of the Petri dishes (6 cm in diameter) and allowed to dry for two minutes at room temperature (Choi *et al.* 2004). To reduce the effect of EO contact toxicity, the entry of the lid was covered with a net and the lid was immediately closed. In each Petri dish, 20 adult females were released; Triton X-100 (0.02%) was utilized in the control treatment. All treatments were replicated three times. Adults' mortality was determined using a stereomicroscope after 24 hours. If thrips' bodies or appendages did not move when touched with a fine brush, they were considered dead. The final concentrations utilized in the bioassays were 7.94, 10.96, 15.13, 20.89, 28.84 µL/L air for *M. piperita*, and 12.59, 15.85, 19.95, 25.12, 31.62 µL/L air for *L. nobilis*. The 50% Lethal Concentration (LC₅₀) and 30% Lethal Concentration (LC₃₀) and their 95% confidence limits were calculated by using probit regressions in Polo-Plus V.2 software (Robertson *et al.* 1980).

Functional response experiments

In order to determine the functional response, five-day-old *A. swirskii* females were exposed to LC₅₀ and LC₃₀ concentrations of *M. piperita* and *L. nobilis* EOs and were starved for 12 hours (Döker *et al.* 2015). EOs were put on filter papers (2 × 2 cm) by using a micropipette in Petri dishes (6 cm in diameter). All procedures were performed for the control treatment with Triton X-100 (0.02). After 12 hours, the treated mites were individually transferred by a thin paintbrush to the experimental arenas.

Each experimental arena consisted of a dark green plastic sheet (5 × 5 cm) placed on a thick sponge pad of similar size (5 × 5 cm) and placed in a plastic container (5 cm in height, 10 cm in diameter) half-filled with water. The edges of the plastic sheet were covered with moisture tissue paper that provided the mites moisture and prevented them from escaping (Vantornhout *et al.* 2005). The predators were provided with one of the six densities of the first instar larvae of *F. occidentalis* (2, 4, 8, 16, 32, and 64). The functional response experiments were carried out in 15 replicates for each prey density and each EO treatment. The number of consumed prey by predatory mites was recorded after 24 hours. Adult thrips were used in the bioassay to determine lethal concentrations of EOs, however, larvae were used in functional response experiments. *Amblyseius swirskii* preferentially fed on this pest's larval stage, especially first instar larvae. Moreover, adult thrips are expected to be more resistant to essential oils. In other words, higher concentrations are needed to kill adult thrips than that of their larvae. Therefore, the concentrations determined for adults allow us to

determine the toxic effects of EOs in a worst-case scenario.

Data analyses

Juliano's two-step procedure was utilized to analyze the functional response data. In the first step, to determine the type of functional response (II or III), the positive or negative sign of the linear coefficient was determined by logistic regression of the proportion of prey consumed (N_a/N_0) as a function of prey density (N_0):

$$\frac{N_a}{N_0} = \frac{\exp(P_0 + P_1N_0 + P_2N_0^2 + P_3N_0^3)}{1 + \exp(P_0 + P_1N_0 + P_2N_0^2 + P_3N_0^3)}$$

The intercept, the linear, the quadratic, and the cubic coefficients are P_0 , P_1 , P_2 , and P_3 , respectively. The signs of P_1 and P_2 indicated the type of functional response. If the linear coefficient is significantly negative ($P_1 < 0$), indicating a type II functional response the proportion of prey consumed decreases as a result of the increasing initial prey density. These parameters are estimated by using the CATMOD procedure in SAS 9.4 software (Juliano 2001). In the next step, the handling time (T_h) and the attack rate (α) coefficients of type II functional response were estimated using Rogers' random predator equation (Rogers 1972):

$$N_a = N_0[1 - \exp(\alpha(T_h N_a - T))]$$

where N_a is the number of prey consumed, N_0 is the initial prey density, α is the attack rate (searching efficiency), T_h is the amount of time that a predator handles each prey individual (handling time) and T is the total time available for predators during the experiment (in this study 24 hours).

Non-linear regression was utilized to estimate the attack rate and the handling time parameters (Proc NLIN, SAS Institute 2013). In addition, the general linear model was utilized to investigate prey consumption and differences between means were compared by Tukey's test ($\alpha = 0.01$) in SPSS V.25 software. Curves of the observed number and percentage of prey consumed by *A. swirskii* females to different densities of the first instar of *F. occidentalis* were depicted in Microsoft Excel V. 2019. The indicator variable equation (Juliano 2001) was used to perform pairwise comparisons for all possible pairs of treatments:

$$Na = N_0\{1 - \exp[(a + D\alpha(j))(T_h + DT_h(j))Na - T]\}$$

Where j is an indicator variable with a value of 0 for the first treatment and 1 for the second. The parameters D_α and DT_h estimate the differences in the values of the parameters α and T_h between the two treatments. For example, the handling time for *A. swirskii* untreated with EOs is T_h , and the handling time for *A. swirskii* treated with EOs is $T_h + DT_h$. The null hypothesis that $DT_h = 0$ is tested to see if there is a significant difference in handling times between the two treatments.

All pairwise comparisons between the treatments were performed. Similarly, the attack rate between treatments is also compared using the D_α parameter (Juliano 2001; Allahyari *et al.* 2004).

RESULTS

Toxicity bioassays of Mentha piperita and Laurus nobilis essential oils on adult females of Frankliniella occidentalis

The LC_{30} and LC_{50} concentrations of *M. piperita* and *L. nobilis* for *F. occidentalis* were 12.489, 16.494 and, 17.071, 20.564 $\mu\text{L/L}$ air, respectively (Table 1).

Table 1. Toxicity of two concentrations of *Mentha piperita* and *Laurus nobilis* essential oils on adult females of *Frankliniella occidentalis*.

Treatments	n	LC ₃₀ (µl/L air) CI (95%)	LC ₅₀ (µl/L air) CI (95%)	Slope ± SE	χ ² (df)	Heterogeneity
<i>Mentha piperita</i>	360	12.489 (11.192–13.662)	16.494 (15.138–18.065)	4.341 ± 0.460	205(13)	0.477
<i>Laurus nobilis</i>	360	17.071 (15.835–18.155)	20.564 (19.395–21.842)	6.488 ± 0.661	300(13)	0.562

CI = Confidence Intervals; df = degrees of freedom; χ² = chi-square.

Functional response of *A. swirskii* to six densities of the first instar larvae of *F. occidentalis*

Maximum likelihood estimates from logistic regression analysis of consumed prey by *A. swirskii* at different densities showed that P_1 values were significantly negative for the control and the two EOs in all treatments (Table 2). In addition, the rectangular hyperboles obtained for all treatments indicated that the proportion of food consumption of *A. swirskii* was decelerating with increasing *F. occidentalis* densities, and the predator was limited by its capacity to process food (Fig. 1). Therefore, *A. swirskii* females exhibited a type II functional response to the first instar *F. occidentalis* larvae, regardless of EO applications. The highest attack rate coefficient (α) (0.105 ± 0.011) was found in the control treatment. The attack rates obtained at LC₃₀ and LC₅₀ concentrations were 0.101 ± 0.010 and 0.089 ± 0.082 , for *M. piperita*, and 0.098 ± 0.011 and 0.087 ± 0.084 for *L. nobilis* EOs, respectively (Table 3). The shortest handling time (1.62 ± 0.046) was found in the control treatment. The handling times obtained at LC₃₀ and LC₅₀ concentrations were 1.85 ± 0.047 and 1.91 ± 0.049 for *M. piperita*, and 1.82 ± 0.054 and 1.84 ± 0.052 for *L. nobilis* EOs, respectively. The theoretical maximum predation rate (T/T_h) of *A. swirskii* was higher (14.81 preys/day⁻¹) in the control treatment and lower (12.56 preys/day⁻¹) in the LC₅₀ treatment of *M. piperita*.

Table 2. Estimated coefficients by logistic regression of proportion of prey consumption of adult female *Amblyseius swirskii* to first instar larvae of *Frankliniella occidentalis* in two concentrations of *Mentha piperita* and *Laurus nobilis* essential oils in 24 hours.

Treatments	Parameters	Estimate (± SE)	χ ²	P-value	
Control	Triton X-100	P ₀	3.7680 ± 0.5311	50.33	< 0.0001
		P ₁	-0.3414 ± 0.0694	24.22	< 0.0001
		P ₂	0.00886 ± 0.0024	13.23	0.0003
		P ₃	-0.00007 ± 0.0000	10.27	0.0013
<i>Mentha piperita</i>	LC ₃₀	P ₀	2.1972 ± 0.3824	33.02	< 0.0001
		P ₁	-0.1967 ± 0.0543	18.13	0.0003
		P ₂	0.00441 ± 0.00199	9.05	0.0266
		P ₃	-0.00004 ± 0.000019	3.28	0.0699
	LC ₅₀	P ₀	2.8498 ± 0.8112	43.46	< 0.0001
		P ₁	-0.2630 ± 0.0593	19.65	< 0.0001
		P ₂	0.00657 ± 0.00214	9.43	0.0021
		P ₃	-0.00005 ± 0.000021	7.08	0.0078
<i>Laurus nobilis</i>	LC ₃₀	P ₀	3.4551 ± 0.3964	38.37	< 0.0001
		P ₁	-0.2374 ± 0.0577	18.13	< 0.0001
		P ₂	0.00612 ± 0.00204	9.05	0.0026
		P ₃	-0.00005 ± 0.00002	7.13	0.0076
	LC ₅₀	P ₀	2.7074 ± 0.4229	40.98	< 0.0001
		P ₁	-0.2425 ± 0.0584	17.27	< 0.0001
		P ₂	0.00587 ± 0.00211	7.74	0.0054
		P ₃	-0.00005 ± 0.00002	5.64	0.0175

P₀, P₁, P₂, and P₃ are the intercept, linear, quadratic, and cubic coefficients, respectively. SE: Standard Error; χ²: Chi-square.

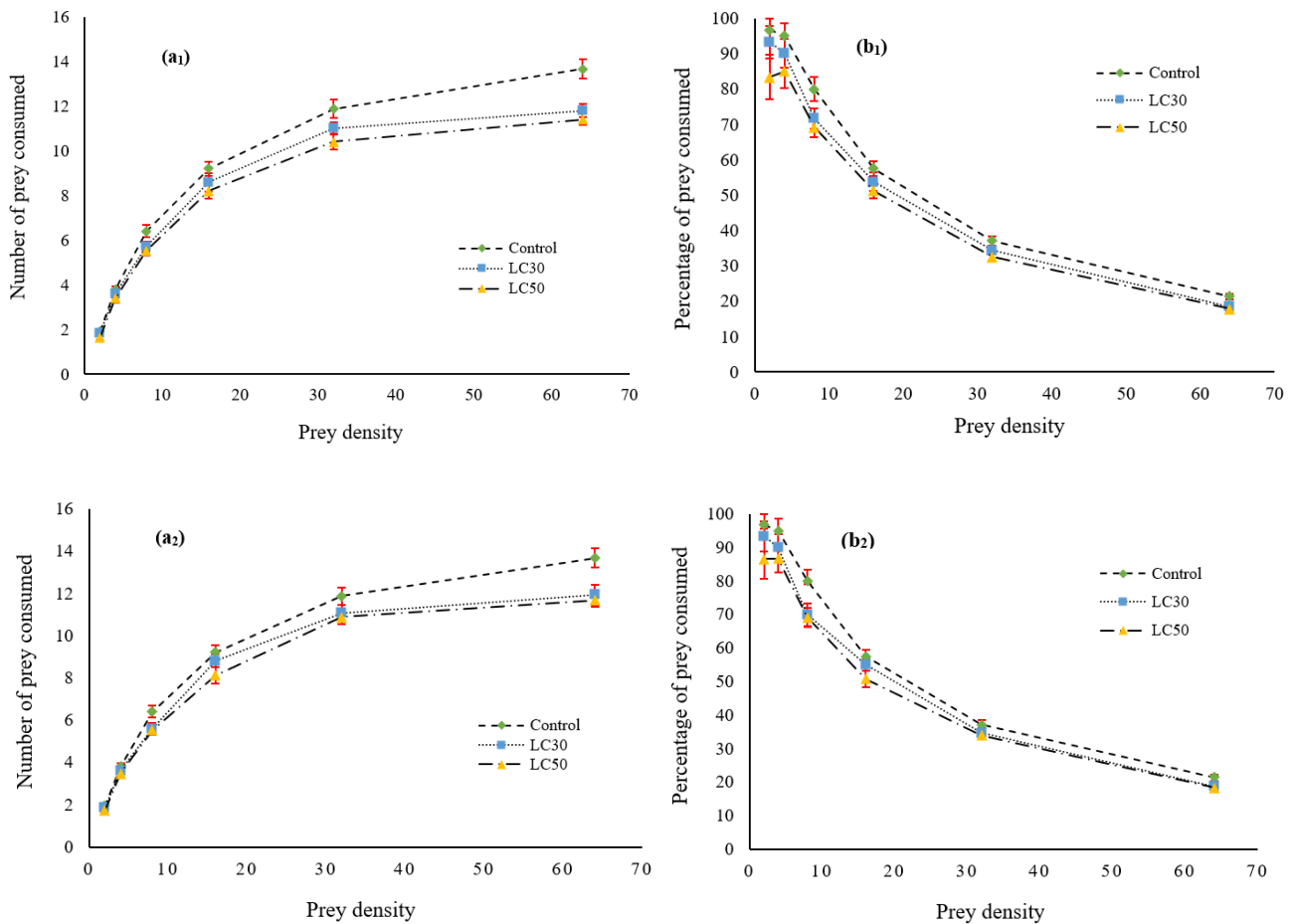


Figure 1. The functional response curves and feeding percentages of *Amblyseius swirskii* on the first instar *Frankliniella occidentalis* after exposure to *Mentha piperita* (a₁, b₁), and *Laurus nobilis* (a₂, b₂) essential oils.

Table 3. Estimate (\pm SE) of attack rate (α) and handling time (T_h) of *Amblyseius swirskii* on the first instar *Frankliniella occidentalis* larvae using Rogers' random predator equation.

Treatments	Type of functional response	Attack rate (h) $\alpha \pm$ SE (Lower–Upper)	Handling time (h ⁻¹) $T_h \pm$ SE (Lower–Upper)	Theoretical maximum attack rate (T/ T_h)	R ²
Control Triton X-100	II	0.1046 \pm 0.0108 (0.0830–0.1262)	1.6177 \pm 0.0461 (1.5261–1.7093)	14.81	0.75
<i>Mentha piperita</i>	LC ₃₀	0.1005 \pm 0.0096 (0.0815–0.1195)	1.8469 \pm 0.0469 (1.7528–1.9391)	12.97	0.73
	LC ₅₀	0.0898 \pm 0.0082.0 (0.0726–0.1106)	1.9088 \pm 0.0494 (1.8107–2.0069)	12.56	0.72
<i>Laurus nobilis</i>	LC ₃₀	0.0977 \pm 0.0109 (0.0781–0.1213)	1.8196 \pm 0.0536 (1.7131–1.9261)	13.19	0.81
	LC ₅₀	0.0873 \pm 0.0843 (0.0706–0.1041)	1.8385 \pm 0.0523 (1.7346–1.9424)	13.04	0.71

Values in parentheses are 95% confidence intervals. α , attack rate; T_h , handling time; T/T_h , maximum attack rate; $R^2 = 1 - (\text{residual sum of squares}/\text{corrected total sum of squares})$.

In all prey densities, *A. swirskii* consumed more prey in the control treatment than in the other treatments. The prey consumption between control and LC₅₀ of *M. piperita* EO treatments was significant at 8 ($F = 3.58$; $P = 0.037$) and 32 ($F = 4.43$; $P = 0.018$) prey densities. A significant

difference was also found between the control treatment and two concentrations of *M. piperita* EO at 64 prey density ($F = 9.87$; $P < 0.001$). In addition, significantly lower prey consumptions compared to the control were also detected in both concentrations of *L. nobilis* EO at 8 ($F = 3.41$; $P = 0.042$) and 64 prey densities ($F = 6.78$; $P = 0.003$) (Table 4).

In LC_{30} and LC_{50} concentrations of both EOs treatments, the asymptotic 95% confidence interval for D_a included 0 but that of D_{Th} was greater than 0, indicating that there was a significant difference between T_h and $T_h + D_{Th}$ in comparison with control (Table 5). In contrast, when LC_{30} and LC_{50} treatments are compared the asymptotic 95% confidence interval for D_a and D_{Th} included 0, indicating that there was no significant difference between the two parameters (Table 5).

Table 4. Daily prey consumption (mean \pm SE) of *Amblyseius swirskii* at different densities of the first instar *Frankliniella occidentalis* larvae in control and two concentrations of *Mentha piperita* and *Laurus nobilis* essential oils.

EOs	Prey density	Control	Treatments		F	P-value
			LC_{30}	LC_{50}		
<i>Mentha piperita</i>	2	1.9 \pm 0.24	1.86 \pm 0.35	1.66 \pm 0.48	2.02	0.145
	4	3.8 \pm 0.54	3.60 \pm 0.63	3.41 \pm 0.73	1.43	0.250
	8	6.4 \pm 1.01 ^a	5.73 \pm 0.88 ^{ab}	5.53 \pm 0.83 ^b	3.58*	0.037
	16	9.2 \pm 1.22	8.60 \pm 1.59	8.20 \pm 1.26	1.99	0.150
	32	11.9 \pm 1.54 ^a	11.01 \pm 1.07 ^{ab}	10.40 \pm 1.35 ^b	4.43*	0.018
<i>Laurus nobilis</i>	64	13.7 \pm 1.65 ^a	11.80 \pm 1.08 ^b	11.80 \pm 1.08 ^b	9.87**	0.000
	2	1.9 \pm 0.24	1.86 \pm 0.35	1.73 \pm 0.46	1.17	0.321
	4	3.8 \pm 0.54	3.60 \pm 0.63	3.46 \pm 0.64	1.13	0.334
	8	6.4 \pm 1.01 ^a	5.60 \pm 1.05 ^b	5.53 \pm 0.92 ^b	3.41*	0.042
	16	9.2 \pm 1.22	8.80 \pm 1.21	8.13 \pm 1.51	2.45	0.098
	32	11.9 \pm 1.54	11.06 \pm 1.39	10.87 \pm 1.30	2.04	0.143
	64	13.7 \pm 1.65 ^a	11.93 \pm 1.83 ^b	11.67 \pm 1.23 ^b	6.78**	0.003

*, ** The means followed by different letters in the same row are significantly different ($P < 0.01$; $P < 0.05$, Tukey's test).

Table 5. Comparison between functional response parameters of *Amblyseius swirskii* treated with sub-lethal concentration of *Mentha piperita* and *Laurus nobilis* essential oils.

Essential oils	treatments	Parameter	Estimate	SE	95% confidence interval	
					Lower	Upper
<i>Mentha piperita</i>	Control- LC_{30}	D_a	-0.00407	0.0145	-0.0327	0.0246
		D_{Th} *	0.2282	0.0668	0.0963	0.3602
	Control- LC_{50}	D_a	-0.0148	0.0135	-0.0414	0.0118
		D_{Th} *	0.2911	0.0695	0.1539	0.4282
<i>Laurus nobilis</i>	LC_{30} - LC_{50}	D_a	-0.0111	0.0126	-0.0359	0.0137
		D_{Th}	0.0582	0.0691	-0.0781	0.1945
	Control- LC_{30}	D_a	-0.0049	0.0154	-0.0352	0.0254
		D_{Th} *	0.2019	0.0708	0.0621	0.3417
	Control- LC_{50}	D_a	-0.0173	0.0136	-0.0442	0.00963
		D_{Th} *	0.2207	0.0706	0.0814	0.3601
LC_{30} - LC_{50}	D_a	-0.0124	0.0137	-0.0393	0.0146	
D_{Th}	0.0189	0.0751	-0.1293	0.1670		

* sign shows a significant difference between functional response parameters.

DISCUSSION

Over the last few decades, studies have shown that plant EOs, as lipophilic chemical compounds

produced by plant secondary metabolites, can effectively control plant pests as an alternative to chemical pesticides. Depending on their composition and dose, they can cause mortality or have sub-lethal effects such as repellence, reduced fertility, and fecundity, or affect the behavior of phytophagous arthropods (Plata-Rueda *et al.* 2018). Due to their low toxicity, and selectivity to non-target organisms, as well as their short persistence in the environment, EOs are promising candidates for agricultural pest management (Chae *et al.* 2014). Although EOs are considered to be non-toxic to beneficial arthropods including predatory mites, in general (Amer and Momen 2002), they may cause a series of sub-lethal effects on development, longevity, fecundity, and the functional response parameters of the phytoseiid predatory mites (Amer and Momen 2002; Amer *et al.* 2016; Shirvani *et al.* 2023b). This situation should be taken into consideration when evaluating the potential of EOs to be used in pest management programs. Although functional response studies are frequently used to estimate the potential of the predators, studies on the effects of various plant compounds, such as EOs, on the functional response of *A. swirskii* are rarely addressed. In the present study, we showed the toxic effects of *M. piperita* and *L. nobilis* EOs on *F. occidentalis* females. These findings are similar to those found by Kirişik (2022) and Stepanycheva *et al.* (2018, 2019) who reported the toxic effects of these EOs on *F. occidentalis*. Our results also demonstrated that even after being exposed to sub-lethal concentrations of *M. piperita* and *L. nobilis* EOs determined for adult thrips, *A. swirskii* females exhibited a type II functional response to the first instar *F. occidentalis* larvae. The predator causes the most mortality at low prey densities in this type of response (Khanamani *et al.* 2014); species that exhibit type II functional response are considered to be successful predators (Fathipour and Maleknia 2016).

Similar to our results, the functional responses of *Typhlodromus ornatus* and *Neoseiulus baraki*, both preying on the coconut mite, *Aceria guerreronis* Keifer (Acari: Eriophyidae), did not change when they were exposed to coconut oil (Freitas *et al.* 2018) and various acaricides (Lima *et al.* 2015). In addition, Shirvani *et al.* (2023a) reported the type II functional response for *A. swirskii* when it preyed on western flower thrips larvae, and the type of functional response was stable when the mites were exposed to *Zataria multiflora* EO. In contrast, Li *et al.* (2006) reported that the functional response of the predatory thrips, *Scolothrips takahashii* Priesner (Thysanoptera: Thripidae) feeding on *Amphitetranychus viennensis* (Acari: Tetranychidae) eggs changed from Type II to Type III when it was exposed to abamectin and fenpropathrin. Likewise, the functional response of a parasitoid wasp, *Habrobracon hebetor* (Hymenoptera: Braconidae) to *Ephestia kuehniella* Zeller (Lepidoptera: Pyralidae) changed from type II to type III when exposed to garlic (*Allium sativum*) and rosemary (*Rosmarinus officinalis*) extracts (Asadi *et al.* 2018).

The attack rate (α) and the handling time (T_h) are considered to be two fundamental components in determining the potential of natural enemies (Juliano 2001). The estimated prey handling time of the *A. swirskii* in this study under the sub-lethal concentrations of both EOs is significantly longer compared to the control. In other words, the predatory mite spent more time handling the prey. The difference in handling time may be due to the decrease in speed of the predator but there were no significant differences between LC₃₀ and LC₅₀ treatments. There was no significant difference among the attack rates of *A. swirskii* under sub-lethal concentrations of both EOs in comparison with the control.

The prey consumption under EOs treatments was also decreased in our study. The number of prey consumed becomes constant at high prey densities probably due to predators spending their time handling the prey, thus causing a decrease in consumption (Jeschke *et al.* 2002). Generally, attack rate and handling time are related to the prey size and predator-prey encounter rate (Hammill *et al.* 2015). These findings could be attributed to the physical or chemical properties of EOs, which might interfere with prey capture. *Mentha piperita* and *L. nobilis* EOs contain large quantities of monoterpenes with proven repellent activity against pests especially *F. occidentalis* (Gharbi and Tay 2022; Kirişik 2022). The use of EOs in pest management requires predator conservation to enhance the success of the management (Regnault-Roger *et al.* 2012; Raveau *et al.* 2020). Previous studies

have shown that pests are more sensitive to EOs than their predators (Gospodarek *et al.* 2023; Shirvani *et al.* 2023a, b). Similarly, the study by Momen *et al.* (2001) showed that *Mentha viridis* was toxic to *T. urticae* since it caused high mortality and reduction in fertility, but less toxic to phytoseiid predatory mites. The concentrations of 0.2 and 0.05% of Neemazal-F (a commercial product of neem seed extract) were, in contrast, very toxic for *A. swirskii* (Momen *et al.* 1997). Amer and Momen (2002) investigated the direct toxicity of four EOs, marjoram, rosemary, peppermint, and lavender to adult females of the predatory mite *A. swirskii*. The authors found that peppermint oil was the most toxic to the females of this predator, while lavender oil was the least toxic. Nevertheless, more investigation on the side effects of these two EOs on *A. swirskii* is needed to use this predator in *T. urticae* management.

CONCLUSION

In summary, based on the present results we can conclude that LC₃₀ and LC₅₀ concentrations of *M. piperita* and *L. nobilis* EOs didn't change the functional response type in *A. swirskii*. In comparison to the control, there was no significant difference in the attack rate of *A. swirskii* while the handling time was significantly longer at sub-lethal concentrations of both EOs. Further research is therefore needed before *M. piperita* and *L. nobilis* EOs can be recommended for application in combination with *A. swirskii* for *F. occidentalis* management under realistic field conditions especially in greenhouses.

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اثرات دو اسانس بر واکنش تابعی *Amblyseius swirskii* (Acari: Phytoseiidae) تغذیه شده با *Frankliniella occidentalis* (Thysanoptera: Thripidae)

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چکیده

گونه *Amblyseius swirskii* Athias-Henriot (Acari: Phytoseiidae) شکارگر عمومی مهمی است که در مهار چندین آفت مهم گیاهان زراعی مانند کنه تارتن، سفیدبالک و تریپس استفاده می‌شود. در این پژوهش، غلظت‌های کشنده اسانس‌های نعنای فلفلی *Mentha piperita* L. (Lamiaceae) و برگ بو *Laurus nobilis* L. (Lauraceae) روی حشره کامل *Frankliniella occidentalis* (Pergande) (Thysanoptera: Thripidae) محاسبه شد. اثرهای جانبی اسانس‌های گیاهی روی واکنش تابعی *A. swirskii* روی لاروهای سن یکم *F. occidentalis* بررسی شد. ماده‌های *A. swirskii* به مدت ۱۲ ساعت در معرض غلظت‌های زیرکشنده (LC₅₀ و LC₃₀) اسانس‌ها با روش تدخینی قرار گرفتند. در شاهد، تیمار با استفاده از Triton X-100 (۰/۰۲) انجام شد. سپس کنه‌های شکارگر تیمار شده به طور تصادفی انتخاب و به واحدهای آزمایشی حاوی ۲، ۴، ۸، ۱۶، ۳۲ و ۶۴ تریپس منتقل شدند. شکارگرها در واحدهای آزمایشی به مدت ۲۴ ساعت در دمای ۲۵ ± ۱ درجه سلسیوس، رطوبت ۱۰ ± ۶۰ درصد و دوره نوری ۸:۱۶ ساعت (تاریکی: روشنایی) نگهداری شدند. نتایج نشان داد که *A. swirskii* تحت تاثیر اسانس‌ها، واکنش تابعی نوع II از خود نشان داد. کوتاه‌ترین (۰/۰۴۶ ± ۱/۶۲ ساعت) و طولانی‌ترین (۰/۰۴۹ ± ۱/۹۱ ساعت) زمان دستیابی (Th) به ترتیب در تیمار شاهد و تیمار LC₅₀ اسانس *M. piperita* ثبت شد. تفاوت معنی‌داری بین ضریب نرخ حمله (α) در غلظت‌های زیرکشنده وجود نداشت. بر اساس نتایج، دو اسانس، قابلیت استفاده در مدیریت *F. occidentalis* را دارند، اما عوارض جانبی آنها بر *A. swirskii* باید در برنامه‌های IPM در نظر گرفته شود.

واژگان کلیدی: میزان حمله، آفتکش‌های گیاهی، زمان دستیابی، مدیریت تلفیقی آفات، کنه شکارگر.

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