



The effectiveness of essential oils derived from *Salvia officinalis* and *S. rosmarinus* (Lamiaceae) against *Panonychus ulmi* (Acari: Tetranychidae)

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ABSTRACT

European red mite, *Panonychus ulmi* (Acari: Tetranychidae) is one of the main pests of apple trees worldwide. The main purpose of this research was to study the lethal and sub-lethal effects of *Salvia officinalis* and *S. rosmarinus* essential oils (EOs) on the European red mite. It was found that the LC₅₀ values of EOs of *S. officinalis* and *S. rosmarinus* were 581.95 and 460.67 $\mu\text{L l}^{-1}$ for contact toxicity, and 3.40 and 4.03 $\mu\text{L l}^{-1}$ air, for fumigant toxicity respectively. With increasing the concentration of EOs, the death rate of mites was increased. To study the lethality of *S. officinalis* and *S. rosmarinus* EOs on the European red mite, concentrations of the LC₂₅ values of the mentioned EOs were 443.32 and 273.48 $\mu\text{L l}^{-1}$, respectively. The net reproductive rate (R_0) in the control and in the treatment with *S. officinalis* and *S. rosmarinus* EOs were 5.18 ± 0.94 , 3.12 ± 0.49 and 3.28 ± 0.51 offspring/female/ individual, respectively. The intrinsic rate of increase (r) in the control and in the treatment with *S. officinalis* and *S. rosmarinus* were 0.10 ± 0.01 , 0.07 ± 0.01 , and $0.07 \pm 0.01 \text{ day}^{-1}$, respectively. The finite rate of population increases (λ) in the control and the *S. officinalis* and *S. rosmarinus* treatments were 1.11 ± 0.13 , 1.07 ± 0.07 and $1.08 \pm 0.07 \text{ day}^{-1}$, respectively, which were significantly smaller than the control. The results showed that both tested EOs have lethal and sub-lethal effects on the European red mite. Also, the effect of *S. officinalis* EO in reducing parameters such as the immature longevity, female longevity, and fertility was estimated to be greater than that of *S. rosmarinus* EO, and there was no statistical difference between them. Finally, the mentioned plant compounds have noticeable effects on the age-stage specific survival and fertility rates in the European red mite and possess the capability of significantly reducing its population under controlled condition.

KEYWORDS

Biological characteristics, European red mite, lethal property, medicinal plants, pest control, toxicity

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INTRODUCTION

Many species of insects and mites have been identified as apple pests. Mites are an extremely diverse group of organisms and among them, Tetranychidae family has a wide range of hosts (Zhang 2003; Alberti 2005) and are an important damaging group of pest mites in agricultural ecosystems (Migeon and Dorkeld 2025). Damaging species from the mentioned family of the mites on the apple trees known as the European red mite, *Panonychus ulmi* (Koch) have attracted interest (Rodrigues 2005).



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This species is distributed worldwide (Schruft 1985) and its main hosts are fruit trees and bushes of the Rosaceae family including apple trees (Baker and Tuttle 1994; Bolland *et al.* 1998). High densities of *P. ulmi* population may result in browning, early leaf drop, and the production of small, discolored fruits in apple trees, ultimately leading to significant economic losses (Arbabi *et al.* 2001). Acaricides play the main role in *P. ulmi* control. However, indiscriminate application of synthetic acaricides for pest control may lead to adverse consequences, including health risks, environmental pollution, threats to mammals, and the development of resistance in highly fecund and rapidly proliferating pests like spider mites (Cavalcanti *et al.* 2010). Such concerns have prompted an increased focus on naturally derived acaricides, with numerous essential plant oils (EOs) identified as potential alternatives to their synthetic counterparts. EOs are aromatic oily liquids sourced from multiple plant parts, including leaves, seeds, fruits, buds, flowers, wood, herbs, barks, and roots (Burt 2004). The pesticidal efficacy of these oils is linked to the presence of monoterpenes, diterpenes, and sesquiterpenes, which have been the subject of extensive evaluation for many years, revealing their antifeedant, repellent, and ovicidal activities against a range of pests (Govindarajan *et al.* 2016; Reddy *et al.* 2016). Although, acaricidal properties of many EOs have been studied against spider mites especially, *Tetranychus urticae* Koch (Acari: Tetranychidae), but not much work has been done for European red mite.

The Lamiaceae family encompasses a vast number of species found across the globe and is well-known for its antimicrobial properties that are effective against various arthropods (Biljana *et al.* 2007; Tong and Coats 2010; Spinozzi *et al.* 2024). Among these aromatic plants, Rosemary (*Salvia rosmarinus* Spenn.) is a native woody perennial herb to the Mediterranean region (Szumny *et al.* 2010) whose extracts are widely used in cosmetics and aromatherapy and which holds an important place due to its antibacterial activity and its insecticidal/acaricidal effects (Martinez-Velazquez *et al.* 2011; Laborda Cenjor *et al.* 2013; Ben Slimane *et al.* 2015). *Salvia*, a genus characterized by its broad distribution, can be found in a variety of regions, including those with temperate and warmer temperatures. Sage (*Salvia officinalis* L.) has been used since ancient times for medicinal purposes, as a food seasoning, and in the cosmetics industry. Its strong antibacterial and antifungal activities have been demonstrated, too (Wu *et al.* 2012). The primary goal of this research is to evaluate both the lethal and sub-lethal effects of these plant essential oils on the European red mite, aiming to assess their potential as eco-friendly control agents. The findings of this study are anticipated to be applicable in the management of the European red mite within apple orchards.

MATERIAL AND METHODS

This research was carried out during 2023–2024 in the greenhouse and entomology laboratory of the Department of Plant Protection, Faculty of Agriculture, University of Maragheh (East Azerbaijan Province, Iran). Apple seedlings were obtained from the orchards of Maragheh.

Mite colonies

The culture of the European red mite began with individuals collected from apple orchards in Maragheh, which were then transferred to a greenhouse. The leaves of the apple seedlings were subsequently infested with the pest mites. To prepare a laboratory colony, apple seedlings were placed in a climate chamber at 26 ± 2 °C, 16:8 h L:D, and $50 \pm 5\%$ RH. Seedling leaves were infested with mites and were maintained for 60 days before starting the experiments.

Plant material and preparation of essential oils

Plant samples were collected in September and March 2023 from various regions of Fars Province, where the species either occur naturally or are cultivated in agricultural fields. Sages were collected from Marvdasht Shiraz (29.52° N, 52.48° E, 1595 m a.s.l.) and rosemary were collected from Kazerun Shiraz (29.51° N, 23.51° E, 509 m a.s.l.). Following their identification, the specimens were subjected to a drying process within a controlled laboratory setting maintained at a temperature of 25 ± 1 °C. To extract EOs from the dried plants, 300 grams of each plant were powdered using an electric

mill. Then, 50 grams of the powdered plants were mixed with 600 mL of distilled water and extracted using a Clevenger-type apparatus with a distillation method, employing water for three hours. The EO was dried over anhydrous sodium sulfate. Plant EOs were kept in 1.5 mL containers, which were covered with aluminum foil, stored in a refrigerator at 4 °C temperature until use (Gulluce *et al.* 2007).

Analysis of essential oil samples

Essential oils components were identified using an Agilent GC-MS instrument (Agilent 5975C) at the University of Maragheh according to the procedure introduced by Morshedloo *et al.* (2018). An HP5-MS column type with a length of 30 meters and an inner diameter of 0.25 micrometers was used for the separation of the components. The oven temperature was programmed for 5 min at 60 °C, and rinsing from 60 to 240 °C at 3 °C/min, and then held isothermal for 10 min at 240 °C; carrier gas was He (flow rate of 1 mL/min); split ratio 1:24; acquisition mass range 40–400 m/z; ionization voltage, 70 eV.

Contact and fumigant toxicity bioassay

To check the contact toxicity of each EO separately in the preliminary tests, concentrations of 50, 100, 200, 400, 600, 800 and 1000 $\mu\text{L l}^{-1}$ were prepared for each essential oil. For prepared concentrations of 1000 $\mu\text{L l}^{-1}$, 200 mL distilled water and 20 mL ethanol were added. For the control treatment, only ethanol and distilled water were used, and the concentrations that caused between 10-90% losses were determined and used in the final test. The immersion method was used to conduct the test (Roh *et al.* 2011). After preparing the concentrations, apple leaves were placed in Petri dishes with dimensions of 2×9 cm and volume of 40 mL, containing different concentrations of EOs and after 5 seconds the leaves were removed from the solution and kept for a period of 20 minutes to dry, then placed in Petri dishes on wet pads. 20 adult mites were placed on the leaves with a fine brush in Petri dishes. All Petri dishes were kept at 26 ± 2 °C, 16:8 h L:D, and $50 \pm 5\%$ RH. Four repetitions were considered for each concentration. After 24 hours, the mite mortality rate was calculated.

To investigate fumigant toxicity separately in preliminary tests of concentrations 0.1, 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9 and 1 $\mu\text{L l}^{-1}$ or 2.5, 5, 7.5, 10, 12.5, 15, 17.5, 20, 22.5 and 25 $\mu\text{L l}^{-1}$ air were used and concentrations which caused between 20 and 90% losses were used in the final test. To achieve this, 15 adult female mites were placed on apple leaf discs under controlled conditions, without exposure to any treatments, as well as in the presence of *S. officinalis* essential oil at concentrations of 2.5, 3.13, 3.91, 4.88, 6.08, and 7.5 $\mu\text{L l}^{-1}$ air. Essential oil (EO) concentrations of 2.5, 3.31, 4.37, 5.75, 7.59, and 10 $\mu\text{L l}^{-1}$ air for *S. rosmarinus* were introduced using a Hamilton syringe onto filter paper measuring two square centimeters in diameter, which was then positioned within a 40 mL glass container to minimize the potential loss of EO vapors. From inside the containers, the doors were completely blocked by parafilm and placed in an incubator at 26 ± 2 °C, 16:8 h L:D, and $50 \pm 5\%$ RH. After 24 hours, the mortality rate of mites was calculated. Mites that exhibited total immobility as a result of contact with the brush were deemed deceased.

Sub-lethal effect (LC₂₅) of essential oils on the biological parameters of the European red mite

The basis of this test is the two sex age-stage life table (Chi and Liu 1985; Chi 1988), which is based on the method of (Yin *et al.* 2013; Li *et al.* 2017) and used in order to study the lethal effects of *S. officinalis* and *S. rosmarinus* EOs on the European red mite. Concentrations of the LC₂₅ values of the mentioned EO_s were prepared with values of 443.32 and 273.48 $\mu\text{L l}^{-1}$ respectively, while in the control, distilled water was used. Thereafter, leaf discs were prepared after immersion. The samples were subjected to open air exposure for one hour to facilitate drying at these concentrations. Then, 50 pairs of one-day-old adults were placed individually on the dorsal surface of leaf discs, placed on wet filter paper, and then Petri dishes were transferred to the controlled conditions (26 ± 1 °C, 70 ± 10 RH and 16:8 L:D). After 24 hours, 50 eggs for each treated mite were randomly selected and the rest of the eggs and adult mites were removed. Each of the eggs was placed separately in a Petri dish on the leaf discs. Experimental arenas were checked daily to record the survival and development of the different life stages. Each newly

emerged female was coupled with an untreated male under the same conditions. Survival and fecundity rate were recorded until the death of the last individual.

Statistical analysis

All the bioassay data were analyzed to calculate LC_{25} , LC_{50} , and LC_{90} values with 95% confidence limits using the Probit procedure of the SAS program (SAS Institute 2002) and mean comparisons carried out using LSD test ($p \leq 0.05$). Abbott's formula (Abbott 1925) was used for corrections in relation to blank control mortality. Life table and reproduction parameters were estimated using Twosex-Mschart software (Chi 2022) and the figures were prepared using Sigma Plot software.

RESULTS

Lethal effect of essential oil on the European red mite

Although the effect of the abovementioned EOs on the European red mite has not been studied, studies of their EOs on other mites, including *T. urticae*, have shown that these compounds have a variety of effects, such as increasing mortality of the various mite stages, reducing the life span of the females and decreasing their fertility (Abd-Elhady and El-Zahi 2011; Amer *et al.* 2011; Laborda Cenjor *et al.* 2013; Bakkali Aissaoui *et al.* 2021). In this research, it was found that the LC_{50} values of EOs of *S. officinalis* and *S. rosmarinus* for contact toxicity were 581.95 and 460.67 $\mu\text{L l}^{-1}$, and for fumigant toxicity were 3.40 and 4.03 $\mu\text{L l}^{-1}$ air, respectively (Table 1). With the increase in the concentration of EOs, the death rate of mites increased. Due to the lack of overlap, the 95% confidence interval values for the toxicity of EOs of *S. officinalis* and *S. rosmarinus* are significantly different at 5% significance level. While both EOs had low LC_{50} values, they showed high toxicity against the European red mite (Figs 1–4).

Table 1. Toxicity of tested *Salvia officinalis* and *S. rosmarinus* essential oils (EOs) on adult of *Panonychus ulmi*.

Essential oils	Methods	N	df	Chi-square	Slope $\pm$ SE	LC_{25}	LC_{50}
<i>S. officinalis</i>	contact toxicity	560	4	5.07	0.27 ± 1.26	443.32 (407.33–471.41)	581.95 (555.56–610.33)
<i>S. rosmarinus</i>	contact toxicity	560	4	10.75	2.68 ± 0.29	273.48 (168.53–343.92)	460.67 (597/8–371.31)
<i>S. officinalis</i>	fumigant toxicity	420	3	1.0273	0.342 ± 0.794	1.86 (1.01799–2.40234)	3.40 (2.78511–3.89626)
<i>S. rosmarinus</i>	fumigant toxicity	420	3	5.2034	1.734 ± 0.157	2.34 (1.65243–2.83779)	4.03 (3.48688–4.58480)

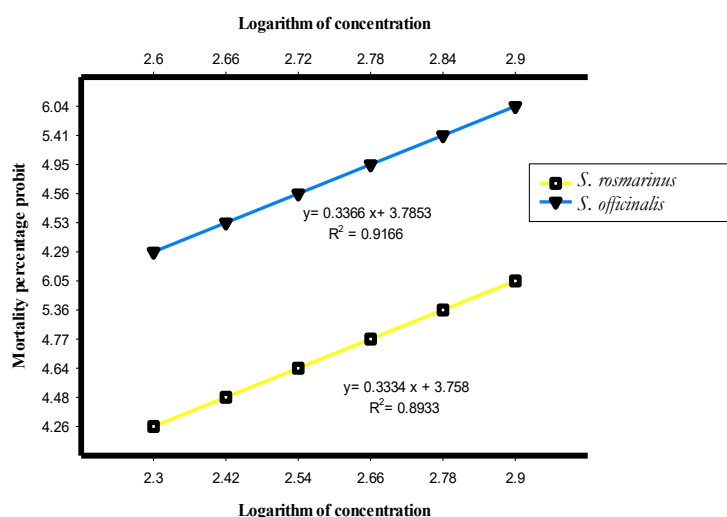


Figure 1. Probit diagram of *Salvia officinalis* and *S. rosmarinus* EOs for contact toxicity.

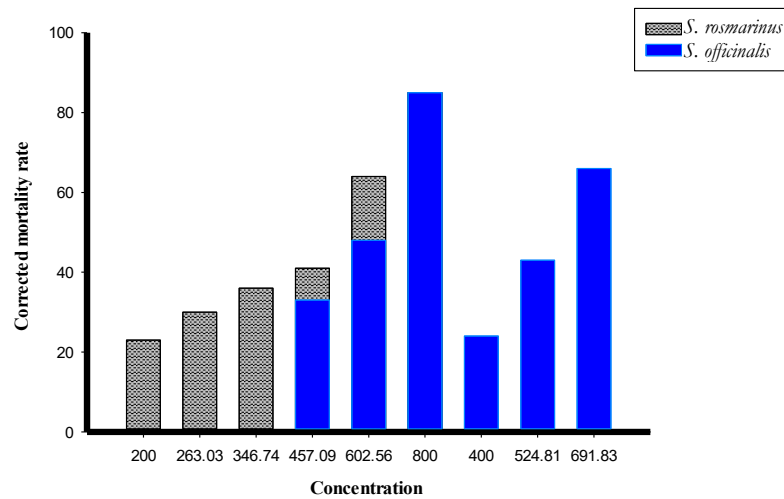


Figure 2. The corrected percentage mortality of adult *Panonychus ulmi* caused by the contact toxicity of *Salvia officinalis* and *S. rosmarinus* essential oils.

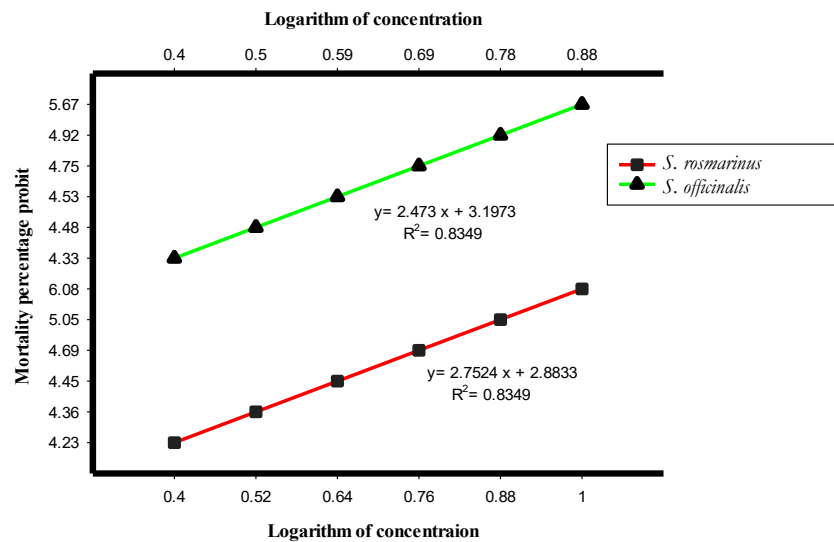


Figure 3. Probit diagram of *S. officinalis* and *S. rosmarinus* EOs for fumigant toxicity.

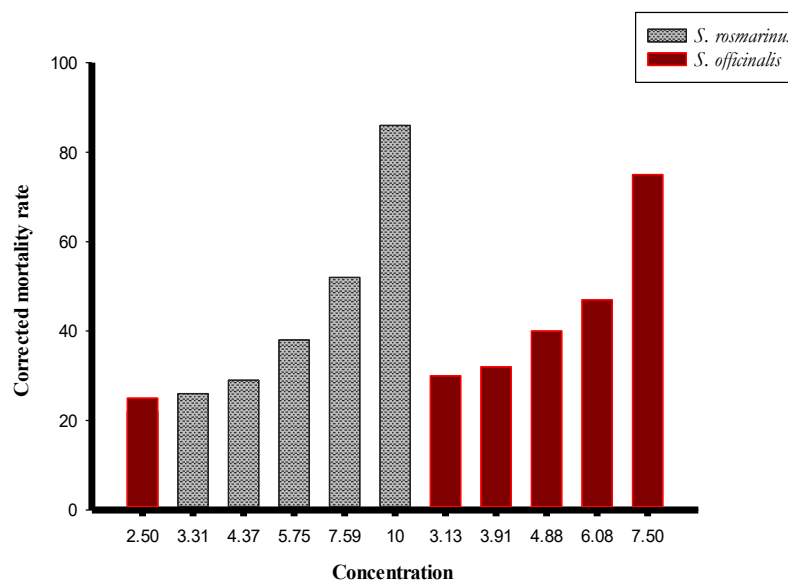


Figure 4. The percentage mortality of adult *Panonychus ulmi* resulting from the fumigant toxicity of *Salvia officinalis* and *S. rosmarinus* essential oils.

Chemical components of essential oils

Identification of EO compounds was carried out by GC-MS chromatography. Thus, 40 compounds were identified in *S. rosmarinus* EO while the majority of the compounds include 1.8-cinEOle, verbenone, vampfhor and α -pinene. In Comparison, 86 compounds were identified for *S. officinalis* EO, while its main compounds identified trans-thujone, cis-thujone and 1.8-cinEOle (Table 2).

Table 2. The main components of *Salvia officinalis* and *S. rosmarinus* essential oils were identified using GC-MS analysis.

No	RI	RI Reference	Components	<i>S. rosmarinus</i> (%)	<i>S. officinalis</i> (%)
1	899	900	n-Nonane	0.37	-
2	919	921	Tricyclene	0.31	-
3	931	932	α -Pinene	10.87	3.57
4	944	946	Camphene	2.83	2.99
5	950	961	Verbenene	0.16	-
6	972	974	β -Pinene	0.17	3.2
7	976	977	1-octen-3-one	0.28	-
8	984	983	3-Octanone	2.7	-
9	987	988	Myrcene	1.87	1.18
10	999	1000	n-Decane	0.2	-
11	1014	1014	α -Terpinene	0.31	0.906
12	1021	1024	p-Cymene	1.95	-
13	1026	1026	1,8-Cineole	11.61	10.45
14	1055	1059	γ -Terpinene	0.99	-
15	1063	1065	cis-Sabinene hydrate	0.3	-
16	1085	1086	Terpinolene	0.19	-
17	1098	1096	Linalool	4.08	0.88
18	1121	1124	Chrysanthenone	0.9	-
19	1140	1141	Camphor	10.64	8.73
20	1156	1155	Isoborneol	0.37	-
21	1162	1165	Borneol	9.81	3.13
22	1169	1172	cis-Pinocamphone	1.4	0.98
23	1173	1174	Terpinen-4-ol	1.44	0.54
24	1187	1186	α -Terpineol	3.19	0.32
25	1202	1204	Verbenone	16.22	-
26	1232	1232	Thymol, methyl ether	1.673	-
27	1283	1284	Bornyl acetate	5.81	0.99
28	1292	1289	Thymol	1.9	6.4
29	1300	1298	Carvacrol	0.3	-
30	1413	1417	(E)-Caryophyllene	1.61	3.99
31	1578	1582	Caryophyllene oxide	0.8	-
32	923	924	α -Thujene	-	0.991
33	1001	1002	α -Phellandrene	-	0.141
34	1020	1022	o-Cymene	-	0.94
35	1096	1098	trans-Sabinene	-	0.65
36	1105	1101	cis-Thujone	-	19.3
37	1113	1112	trans-Thujone dihydro	-	6.99
38	1194	1191	cis-Carvone	-	0.35
39	1149	1152	Humulene	-	6.65
40	1590	1592	Viridiflorol	-	6.92
Total				95.25	91.18

Sub-lethal effects (LC_{25}) of essential oils on the biological parameters of the European red mite

In order to study the sub-lethal mortality of *S. officinalis* and *S. rosmarinus* EOs on the European red mite, concentrations of the LC_{25} of the mentioned EOs were prepared using values of 443.32 and 273.48 $\mu\text{L l}^{-1}$, respectively. Life table parameters and standard error were estimated based on bootstrap technique. The results showed that the net reproduction rate (R_0) in the control and treatments with *S.*

officinalis and *S. rosmarinus* EOs were recorded as 5.18 ± 0.94 , 3.12 ± 0.49 and 3.28 ± 0.51 offspring/female/individual, respectively. Furthermore, intrinsic rate of natural increase (r) in control and treatment of *S. officinalis* and *S. rosmarinus* were estimated as 0.10 ± 0.01 , 0.07 ± 0.01 and 0.07 ± 0.01 d⁻¹, respectively. The mean generation time (T) in the control and in the treatments using sage and rosemary EOs were 15.74 ± 1.93 , 14.79 ± 1.02 and 14.97 ± 1.07 d, respectively, which were shorter than the control (Table 3). The length of different developmental stages of female European red mite in control and treated with EOs of *S. officinalis* and *S. rosmarinus* were 18.79 ± 0.16 , 15.29 ± 0.25 and 15.75 ± 0.23 d, and in male it was 17.00 ± 0.71 , 13.40 ± 1.44 and 13.40 ± 1.12 d, respectively, which was less than the control. The longest period of the egg stage was seen in the *S. rosmarinus* treatment, which was more than control. The longevity from egg to death in all three tested populations was higher in females than in males. The longest longevity of females and males was in control. The average length of the different developmental stages of the European red mite in control and treated with LC₂₅ concentrations of *S. officinalis* and *S. rosmarinus* EOs (Table 4, Fig. 5).

Table 3. Comparison of the mean ($\pm$ SE) parameters of *Panonychus ulmi* life table in treatments including the control and exposing to LC₂₅ value of *Salvia officinalis* and *S. rosmarinus* EOs.

Treatment	r	λ	R_0	T	GRR
Control	0.10 ± 0.01^a	1.11 ± 0.13^a	5.18 ± 0.94^a	15.74 ± 1.93^a	7.93 ± 1.19^c
<i>S. officinalis</i>	0.07 ± 0.01^b	1.07 ± 0.07^b	3.12 ± 0.49^b	14.79 ± 1.02^b	8.37 ± 1.45^b
<i>S. rosmarinus</i>	0.07 ± 0.01^b	1.08 ± 0.07^b	3.28 ± 0.51^b	14.97 ± 1.07^{ab}	8.84 ± 1.54^a

SE was estimated using 100000 bootstrap replications. The means followed by different letters in each column are significantly different ($P \leq 5\%$).

Table 4. The mean ($\pm$ SE) duration (days) period of different developmental stages of *Panonychus ulmi* in control and exposed to LC₂₅ value of *Salvia officinalis* and *S. rosmarinus* EOs.

Treatment	Sex	N	Egg	Larva	Nymph	Adult	Longevity
Control	M	4	5.75 ± 0.25^b	2.00 ± 0.00^a	5.75 ± 0.25^a	3.50 ± 0.50^a	17.00 ± 0.71^a
<i>S. officinalis</i>	M	5	6.00 ± 0.32^a	1.40 ± 0.24^b	3.80 ± 0.58^b	2.20 ± 0.58^b	13.40 ± 1.44^b
<i>S. rosmarinus</i>	M	5	6.00 ± 0.32^a	1.40 ± 0.24^b	3.80 ± 0.71^b	2.20 ± 0.58^b	13.40 ± 1.12^b
Control	F	29	5.97 ± 0.03^b	1.83 ± 0.07^a	5.48 ± 0.09^a	5.52 ± 0.20^a	18.79 ± 0.16^a
<i>S. officinalis</i>	F	28	5.96 ± 0.06^b	1.61 ± 0.09^b	4.86 ± 0.14^b	2.86 ± 0.13^b	15.29 ± 0.25^b
<i>S. rosmarinus</i>	F	28	6.04 ± 0.06^a	1.61 ± 0.09^b	4.93 ± 0.13^b	3.18 ± 0.14^b	15.75 ± 0.23^b

SE was estimated using 100000 bootstrap replications. The means followed by different letters in each column are significantly different ($P \leq 5\%$).

In the study of population growth, determining the time and age of the start of oviposition is of great importance and can have a significant effect on population growth (Chi and Li 1985). Most entomologists only calculate the pre-oviposition period (APOP) of complete female insects. In determining the effect of the pre-oviposition period on the population growth, it is stated that the calculation of the total pre-oviposition period (TPOP) provides more significant figures than the adult pre-oviposition period of female insects (APOP), because the total pre-spawning period (TPOP) more precisely defines the effect of the length of time from birth to the first reproduction on population parameters (Ebrahimi *et al.* 2013). The mean total pre-oviposition period (TPOP) in different treatments including the control and treatments of *S. officinalis* and *S. rosmarinus* EOs, were 14.28 ± 0.1 d and 13.43 ± 0.1 d and 13.57 ± 0.0 d. The fecundity rate in control and treatments of *S. officinalis* and *S. rosmarinus* EOs were calculated as 8.93 ± 1.24 , 5.57 ± 0.54 and 5.86 ± 0.55 offspring/female/individual, respectively. The highest age-specific fecundity was recorded for control females. The average length of the reproductive period (days) and the total fecundity of the European red mite in control and treated with LC₂₅ concentrations of *S. officinalis* and *S. rosmarinus* EOs are represented in Table 5 and Figure 6.

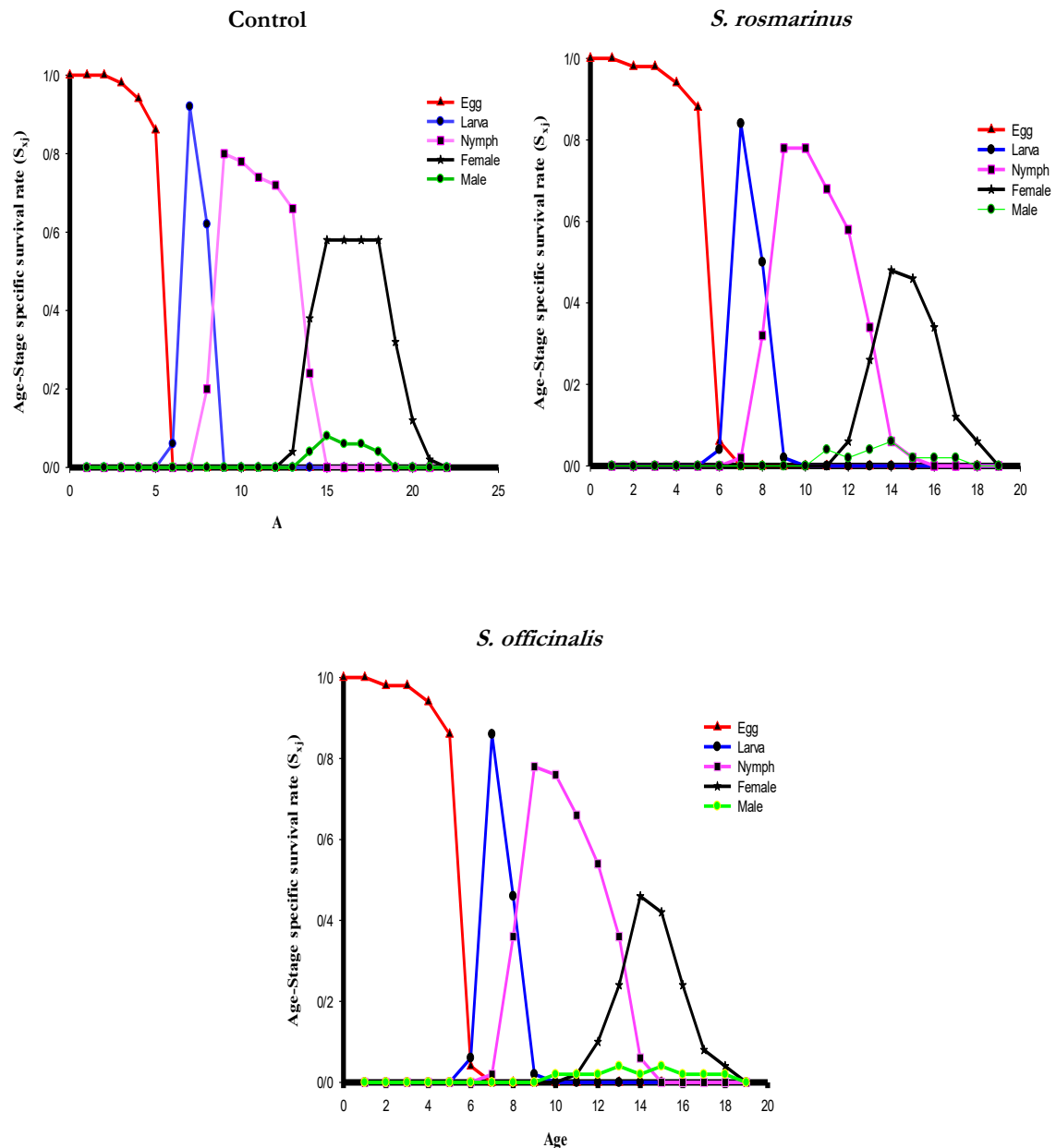


Figure 5. Age-Stage specific survival rate (s_{xj}), for different developmental stages of *Panonychus ulmi* in control and exposed to LC₂₅ value of *S. officinalis* and *S. rosmarinus* EOs.

Table 5. The mean ($\pm$ SE) duration (days) period of reproduction and total fertility of *Panonychus ulmi* in control and exposed to LC₂₅ value of *Salvia officinalis* and *S. rosmarinus* EOs.

Treatment	Oviposition days	APOP	TPOP	Total Fecundity
Control	2.03 $\pm$ 0.13 ^a	1.00 $\pm$ 00 ^a	14.28 $\pm$ 0.1 ^a	8.93 $\pm$ 1.24 ^a
<i>S. officinalis</i>	1.67 $\pm$ 0.14 ^b	1.00 $\pm$ 00 ^a	13.43 $\pm$ 0.1 ^b	5.57 $\pm$ 0.54 ^b
<i>S. rosmarinus</i>	1.67 $\pm$ 0.14 ^b	1.00 $\pm$ 00 ^a	13.57 $\pm$ 0.0 ^b	5.86 $\pm$ 0.55 ^b

SE was estimated using 100000 bootstrap replications. The means followed by different letters in each column are significantly different ($P \leq 5\%$).

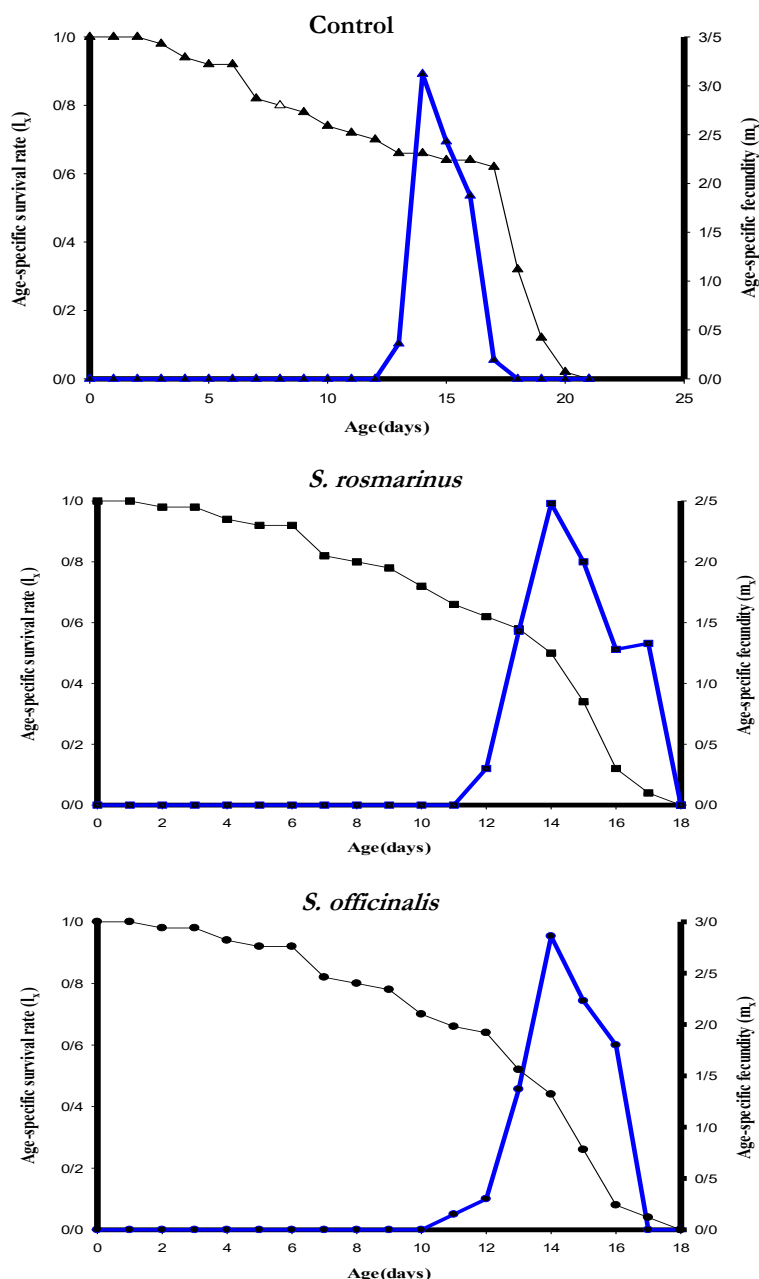


Figure 6. Age-specific survival rate (l_x) and Age-specific fecundity (m_x) of *Panonychus ulmi* in control and exposed to LC₂₅ value of *Salvia officinalis* and *S. rosmarinus* EOs.

DISCUSSION

EOs and their constituents demonstrate fumigant and contact toxicities towards insects which besides having direct influence on mortality, also have several secondary impacts, such as oviposition, repellency and antifeedancy (Motazedian *et al.* 2011; Gholamzadeh Chitgar *et al.* 2013). In this research, it was found that the LC₅₀ values of EOs of *S. officinalis* and *S. rosmarinus* for contact toxicity were 581.95 and 460.67 $\mu\text{L l}^{-1}$, and for fumigant toxicity were 3.40 and 4.03 $\mu\text{L l}^{-1}$ air, respectively. Laborda Cenjor *et al.* (2013) studied effects of *S. officinalis* and *S. rosmarinus* EOs on *T. urticae*. Slide-dip and leaf-disk bioassays were employed to study the mortality caused by these plant oils on two-spotted spiders. Different dilutions of both EOs (0.10–0.25%, v/v) caused acute contact toxicity, although the sage extract showed greater acaricidal activity than rosemary oil. Mortality rates of 95–100% were observed at all the sage oil dosages and when rosemary emulsions contained at least 0.20% of EO. In the residual contact experiments (leaf-disk assays), 0.15–0.25% of sage oil or 0.25% of rosemary extracts significantly reduced

mite survival. However, in this study, it was determined that the toxicity of the EO of *S. rosmarinus* was higher than that of the EO of *S. officinalis*, which could be due to the different compositions of the plant EOs and the different type of test. Talebi Jahromi *et al.* (2015) studied the combined effect of orange peel EO with two chemical acaricides, spiromeclofen and propargite, against adult female spider mites *T. urticae*. In this study, the fumigant and contact toxicity of Citrus sinensis orange peel EO and the contact effect of a mixture of EOs with two acaricides, spiromeclofen and propargite, against two-spotted spider mites, *T. urticae*, were investigated. The EO was extracted from fresh Dezful orange peel by steam distillation. The median lethal concentration (LC₅₀) values of orange peel EO for fumigant and contact were estimated to be 48.75 $\mu\text{L l}^{-1}$ air and 48.48 $\mu\text{g/cm}^2$, respectively.

Nouri Ghonbolani *et al.* (2015) studied chemical components and toxicity of two plant EOs against adult insects *Tribolium castaneum* Herbst (Col.: Tenebrionidae) and *Callosobruchus maculatus* F. (Col.: Bruchidae). In this study, the contact and fumigation toxicity of wormwood, *Salvia pratensis* L. and *Artemisia absinthium* L. EOs on adult insects of *Tr. castaneum* and *C. maculatus* was investigated. The results showed that the EOs had appropriate contact and fumigant toxicity. Wormwood EO with a lethal concentration of 50% (7.093–10.033) was 8.518 $\mu\text{L l}^{-1}$ air compared to sage EO with a lethal concentration of 50% (13.904–11.121). 12.539 $\mu\text{L l}^{-1}$ air significantly increased fumigant toxicity on *Tr. castaneum* after 72 hours. This difference was not significant for the *C. maculatus* due to overlapping confidence limits. In contact toxicity of wormwood EO, the *Tr. castaneum* was more sensitive to 50% lethal concentration (0.1170/196) 0.150 $\mu\text{L/cm}^2$ than the *C. maculatus* to 50% lethal concentration (0.2300/356) 0.281 $\mu\text{L/cm}^2$. Kaveh (2014) conducted a study on evaluation of the acaricidal activity of several EOs from plants of the Lamiaceae family on the two-spotted spider mite *T. urticae*. Their results showed that LC₅₀ values of EOs of *S. officinalis* and *S. rosmarinus* for contact toxicity were 602.13 and 530.71 $\mu\text{L l}^{-1}$, and for fumigant toxicity were 6.73 and 7.49 $\mu\text{L l}^{-1}$ air, respectively. The results of this research are close to the results of Kaveh. Fumigant toxicity of plant EOs is much higher than their contact toxicity. Since plant EOs have a nervous mechanism of action in the mortality of the terminal section (Enan 2001), the high fumigant toxicity compared to contact toxicity may be due to the fact that in fumigant toxicity, the EO compounds easily enter the body of insects and mites through the respiratory pores and directly affect nerve cells, but in the contact state, it must travel through the cuticle and haemolymph and then reach the nerve cells, during which a small amount of the EO enters the body of the insect and some of the EO is metabolized and loses its lethal effect. However, given that there is no closed space in orchard and filed ecosystems and if plant EOs are consumed as pesticides never reach a vapor pressure high enough to cause fumigant toxicity, therefore despite the use of high concentrations of EOs in contact toxicity, research on the contact effects of EOs is more useful than fumigant toxicity (Negahban *et al.* 2010). In order to study the sub-lethal mortality of *S. officinalis* and *S. rosmarinus* EOs on the European red mite, concentrations of the LC₂₅ of the mentioned EOs were prepared using values of 443.32 and 273.48 $\mu\text{L l}^{-1}$, respectively. Life table parameters and standard error were estimated based on bootstrap technique. The mean generation time (T) in the control and in the treatments using sage and rosemary EOs were 15.74 ± 1.93 , 14.79 ± 1.02 and 14.97 ± 1.07 d, respectively, which were shorter than the control.

Herbert (1981) observed an oviposition period for *P. ulmi* on apple leaves of 16 days, 22 eggs per female, and 20 days of longevity at 18 and 21 °C. Gotoh *et al.* (2003) observed an average of 17.5 days, 52.8 eggs per female, and 20 days of longevity. Johann *et al.* (2018) studied the life table parameters of *P. ulmi* on two grape varieties and apple leaves, and reported that the intrinsic rate of population increase (r) were 0.11 and 0.09 d⁻¹, the finite rate of population increase (λ) were 1.11 and 1.10 d⁻¹, the average duration of one generation (T) were 17.05 and 16.94 d, respectively. In the present study, the third summer generation beginning in early July in East Azerbaijan Province, Iran was examined under controlled conditions at 26 ± 2 °C; the results of this research are close to the results of Johan *et al.* (2018). The life cycle of the European red mite on apples in Iran, in West Azerbaijan Province, Urmia city, were as follows: the first generation 35 to 40 days in the second generation 25 to 30 days, the third generations, fourth and fifth generations from 10 to 20 days, the sixth generation, 25 days in the seventh generation 30 days and the eighth generation takes 35 to 40 days (Mostaan 1991). Tomczyk and Suszko (2011)

reached similar conclusions in trials with *Salvia* extracts, which were conducted to evaluate their toxicity to adult females, the eggs and larvae of two-spotted spider mite. They observed that the negative effect of sage extracts on *T. urticae* was not due only to the contact toxicity for the mobile mite stages, but also to the decreased fecundity of surviving females. Thus, the number of eggs laid by females on ivy leaves treated with sage extracts was more than three-fold as low if compared to the controls. Goldansaz *et al.* (2012) tested inhibition of carob moth damage using *Ferula assafoetida* EO in pomegranate orchards of Iran. The EO of *Ferula assafoetida* was tested in four pomegranate orchards. In each garden 10 trees were treated with three concentrations of oil. The oil was diluted by ethanol as solvent. Three concentrations, 1:1 (oil: solvent), 1:3, and 1:5 were prepared and sprayed on the canopy of the plants (5 ml/plant) every two weeks. All three concentrations of the EO of *F. assafoetida* significantly ($P < 0.001$) reduced fruit infestation by carob moth, *Ectomyelois ceratoniae* (Zeller) (Lep.: Pyralidae). There were no significant differences between the experimental sites ($P > 0.05$). The rotten fruits in treated plots that fell to the ground during the growing season were significantly lower ($P < 0.001$) than the control. The percentage of infected fruits by the pest larvae in treated gardens were also significantly lower ($P < 0.001$) than the control in the end of season. The reduction in pomegranate fruits infestation may be due to the direct repellent effect of the oil on the adult carob moth, or a disruption of reproductive behavior of the adult carob moth by volatile compounds emitted by the EO, or combination of the two effects. Their study revealed some EOs can be used in integrated pest management programs as the safe compounds for human health.

Bibi *et al.* (2021) studied the effect of profenofos and citrus oil on *Cryptolaemus montrouzieri* Mulsant (Col.: Coccinellidae) and *Chrysoperla carnea* Stephens (Neurop.: Chrysopidae), key predators of citrus mealybug, *Planococcus citri* (Risso) (Hem.: Pseudococcidae), under laboratory conditions. They found higher prey consumption rates of citrus mealybugs by *Cr. montrouzieri* adults when scales were treated with citrus oil, compared to mealybugs treated with the organophosphate insecticide profenofos. Additionally, mortality of *Cr. montrouzieri* adults was significantly higher following ingestion of citrus mealybugs treated with profenofos compared to citrus oil treatment, which indicate that citrus oil may be applied against *P. citri* populations in the presence of *Cr. montrouzieri* adult predators in citrus orchards. Mansour *et al.* (2022) tested toxicity of *Mentha pulegium* EO and chemical pesticides toward citrus pest scale insects and the coccinellid predator *Cr. montrouzieri*. They evaluated the contact toxicity of *Mentha pulegium* EO (applied at either 2.73 mg/L, 9.56 mg/L, 13.65 mg/L, 27.31 mg/L, or 40.96 mg/L) toward three pest scales, *Pl. citri*, *Aonidiella aurantii* Maskell, and *Chrysomphalus aonidum* (L.) (Hem.: Diaspididae), and two chemical insecticides, chlorpyrifos (100 mL/ hL) and spirotetramat (120 mL/hL), against *P. citri* and *A. aurantii* under laboratory conditions. Toxicity of *M. pulegium* EO and both insecticides was also assessed on the coccinellid predator *Cr. montrouzieri*. The highest mortality rates for all scale insect nymphs ($> 97\%$ for *A. aurantii* and 100% for *P. citri* or *Ch. aonidum*) were obtained following EO application at a dose of 40.96 mg/L. Sharifiyan *et al.* (2024) studied lethal and sublethal effects of *Mentha piperita* L. and its nanoformulation form on the biological and population growth parameters of *Trialeurodes vaporariorum* (Westwood) (Hem.: Aleyrodidae) under laboratory conditions. In this study, the lethal and sublethal effects of *M. piperita* L. EO and its nanoformulation were investigated on the *Tr. vaporariorum* by considering the biological and population growth parameters. The leaf dipping was used for the bioassays. The bioassay results showed that, the nanoformulation of EO (LC_{50} : 3375.411 ppm) was more toxic than the peppermint (LC_{50} : 4536.118 ppm) on the *Tr. vaporariorum* adults. The life table data were analyzed based on the age-stage, two-sex life table theory. Also, the sublethal concentration (LC_{25}) *M. piperita* L. EO and nanoformulation were 2145.91 and 1762.79 $\mu\text{L l}^{-1}$, respectively. Giordano *et al.* (2025) conducted a study on toxicity EOs of *Origanum vulgare*, *Salvia rosmarinus*, and *Salvia officinalis* against *Aculops lycopersici* (Acari: Eriophyidae). In this study, they evaluated the acaricidal effects of EOs extracted from three officinal plants, *O. vulgare*, *S. rosmarinus*, and *S. officinalis*, cultivated using precision aromatic crop (PAC) techniques. They tested multiple concentrations (320-5000 $\mu\text{L l}^{-1}$) and exposure times (1–4 days) to assess mite mortality. In this research two EOs showed significant reduction on life table parameters like r , R_0 , T and λ . The EOs caused a reduction in longevity, survival and fecundity of *P. ulmi*. We used *S. officinalis*

and *S. rosmarinus* EOs, which have high efficiency in EO extraction and have the ability to be widely cultivated. Therefore, it is not far-fetched that in the near future, commercial products of these EOs will be launched on the market against various pests. However, more research is needed in this field. The results of this study can be used to increase the toxicity and durability of these EOs, which can be nanoencapsulated, or the synergistic effect of the mixture of EOs and acaricides can be used to increase the effect of these compounds on the *P. ulmi* and reduce the dose of acaricides used, and also reduce the environmental impact of chemical pesticides.

CONCLUSIONS

The results of this research showed that both tested EOs have lethal and sublethal effects on the European red mite. *Salvia rosmarinus* EO caused more mortality Compared to *S. officinalis* EO. Also, the effect of *S. officinalis* EO in reducing parameters such as the longevity of the immature stages, female life span, and fertility was estimated to be greater than that of *S. rosmarinus* EO. Therefore, the application of these EOs led to a decline in mite populations, suggesting their potential for use in pest management, pending confirmation through field experiments.

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اثربخشی اسانس‌های استخراج شده از *Salvia officinalis* و *S. rosmarinus* (Lamiaceae) روی *Panonychus ulmi* (Acari: Tetranychidae)

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چکیده

کنه قرمز اروپایی (*Panonychus ulmi* (Acari: Tetranychidae)) یکی از آفات اصلی درختان سیب در سراسر جهان است. هدف اصلی این پژوهش مطالعه اثر کشندگی و زیرکشندگی اسانس گیاهان مریم‌گلی (*Salvia officinalis*) و رزماری (*S. rosmarinus*) روی کنه قرمز اروپایی است. در این پژوهش مشخص شد که LC_{50} اسانس گیاهان مریم‌گلی و رزماری به ترتیب در سمیت تماسی برابر است با $581/95$ و $460/67$ میکرولیتر بر لیتر و در سمیت تدخینی برابر است با $3/40$ و $4/03$ میکرولیتر بر لیترها و با افزایش غلظت اسانس‌ها میزان مرگ‌ومیر کنه‌ها افزایش یافت. برای مطالعه اثر زیرکشندگی اسانس‌های مریم‌گلی و رزماری روی کنه قرمز اروپایی غلظت‌هایی از مقادیر LC_{25} اسانس‌های ذکر شده به ترتیب $443/32$ و $273/48$ میکرولیتر بر لیتر تهیه شد. میزان خالص تولید مثل (R_0) در شاهد و در تیمار با اسانس مریم‌گلی و رزماری به ترتیب برابر $5/18 \pm 0/94$ ، $3/12 \pm 0/49$ و $3/28 \pm 0/51$ تخم به ازای هر فرد بود. میزان ذاتی افزایش جمعیت (r) در شاهد، تیمار مریم‌گلی و رزماری به ترتیب $0/10 \pm 0/01$ ، $0/07 \pm 0/01$ و $0/07 \pm 0/01$ بر روز بود. میزان متناهی افزایش جمعیت (λ) در شاهد و تیمار مریم‌گلی و رزماری به ترتیب $1/11 \pm 0/13$ ، $1/07 \pm 0/07$ و $1/08 \pm 0/07$ بر روز بود، که به‌طور معنی‌داری کمتر از شاهد بود. نتایج نشان داد که هر دو اسانس مورد آزمایش دارای اثر کشندگی و زیرکشندگی روی کنه قرمز اروپایی می‌باشند. همچنین اثر اسانس مریم‌گلی در کاهش پراستجه‌هایی همچون طول دوره مراحل نابالغ، طول عمر جنس ماده و باروری بیشتر از اسانس رزماری است و از نظر آماری هیچ تفاوتی با هم ندارند. در نهایت ترکیب‌های گیاهی مذکور بر میزان منحنی زنده‌مانی و باروری ویژه سنی کنه اثر گذاشته و به‌طور معنی‌داری توانایی کاهش جمعیت را در شرایط کنترل شده دارند.

واژگان کلیدی: ویژگی‌های زیستی، کنه قرمز اروپایی، خاصیت کشندگی، گیاهان دارویی، کنترل آفت، سمیت

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