Composition and Acaricidal Activities of *Rosmarinus Officinalis* Essential Oil against *Tetranychus urticae* and Its Predatory Mite *Phytoseuilus persimilis*

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ABSTRACT

Searching for pesticidal activity of plant extracts with some known medicinal attributes could lead to the discovery of new agents for pest control. The essential oil (EO) was extracted from aerial parts of *Rosmarinus officinalis* by hydrodistillation method and tested for its toxicity against adult females of two-spotted spider mite *Tetranychus urticae* Koch (Acari: Tetranychidae) and its predatory mite *Phytoseuilus persimilis* Athias-Henriot (Acari: Phytoseiidae) under laboratory conditions. Exposure periods were 24, 48, and 72 h. The results indicated that adults of *T. urticae* were more susceptible to EO(LC_{50} ranged from 10.70 to 17.30 ml/L) than *P. persimilis* (LC_{50} ranged from 28.05 to 42.09 ml/L). Laboratory bioassay results indicated that the EO caused high mortality of spider mites with high fumigant and repellent activities. EO with the highest used concentration (10%) caused no phytotoxicity to some host plants of *T. urticae* (greenhouse-grown cotton, bean, tomato and cucumber). The chemical composition of hydrodistilled essential oil of *R. officinalis* was identified through GC/MS. The main constituents of the oil were 1,8-cineole (39.74%), camphor (22.76%), cis-ocimene (8.89%) and camphene (7.01%) which may provide the acaricidal properties against *T. urticae*. Results of the present study suggested that *Rosmarinus officinalis* entiaessl oil could be useful in promoting new agents for mite control from the medicinal plants.

Key words: *Rosmarinus officinalis*, Essential oil, Acaricidal activity, *Phytoseuilus persimilis*, *Tetranychus urticae*, Chemical composition

INTRODUCTION

The two-spotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae) is one of the most serious pests in some of agricultural systems (Deligeorgidis *et al.,* 2006, 2007). It ingests leaf cell contents, thus reduces plant photosynthesis (Park and Lee, 2002) and potentially decreases yield quality and quantity (Flaherty and Wilson, 1999). This pest is commonly controlled by applications of synthetic acaricides (Pontes *et al.,* 2007a). The number of confirmed resistant mite species to synthetic pesticides has continued to rise, apart from risks associated with the use of these chemicals (Aslan *et al*., 2004). Unfortunately, spider mites have developed resistance to most available pesticides and the weakness of acaricidal efficacy is considered the major problem encountered (Ay, 2005). The spider mite developed up to 100-fold resistance to dicofol and over 460-fold to parathion (Dagli and Tunc, 2001). As a result, the efficacy of many miticides has been reduced and the cost of chemical control has increased. Therefore, there is an urgent need to develop safer and efficient alternatives that have potential to replace synthetic pesticides and are convenient to use for control of *T. urticae.* Alternatives to conventional acaricides are needed, because many are being banned from the market. Essential oils of aromatic plants are considered as good control alternative tools. The search for efficient natural pesticide substances with low environmental toxicity has increased (Kabir *et al.,* 2003; Silva *et al.,* 2004). The essential oils of aromatic plants are among the most efficient pesticides of botanical origin and often constitute the bioactive fraction of plant extracts (Cosimi *et al.,* 2009). Plant essential oils have a broad spectrum of activity against insects and mites due to their several modes of action, including repellent and antifeedant activities, inhibition of molting and respiration and reduction in growth and fecundity (Enan, 2001; Akhtar and Isman, 2004).

The predatory mite *Phytoseiulus persimilis* Athias-Henriot (Acari: Phytoseiidae) is well known as a predatory mite that specializes on the *Tetranychid* species, and it may considered a tool in the integrated pest management program for controlling spider mite *T. urticae* in many countries throughout the world (Cote *et al.,* 2002; Kim and Yoo, 2002). Use of insecticides and natural enemies has become essential components of IPM in field and greenhouses (Yi *et al*., 2007). Essential oils derived from plants also may have minimal direct and/or indirect effects on natural enemies (Isman, 2006; Bostanian *et al.,* 2005). They are more compatible with the environmental components than synthetic pesticides (Isman and Machial, 2006). Choi *et al*. 2004, tested a total of 53 essential oils against *T. urticae* and *P. persimilis* as a fumigant. Rosemary oil is relatively effective against mite also found to be toxic to the predaceous mites *Amblyseius barkeri* Hughes, *A. zaheri* and *Typhlodromus athiasae* Porath (Momen and Amer, 1999). The oils are generally composed of complex mixtures of monoterpenes, biogenetically related

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phenols, and sesquiterpenes. Examples include 1,8-cineole, the major constituent of rosemary oil (Opender *et al.,* 2008).

Rosemary *(Rosmarius officinalis* L.) is a woody, perennial herb native to the Mediterranean region. It is cultivated mainly in Mediterranean countries (Szumny *et al.,* 2010). Rosemary, from family Labiatae, is an aromatic shrub with an intense pleasant smell reminiscent of pine wood. Rosemary oil has been traditionally used as a medicine for colic, nervous disorders and painful menstruation. Among bioactive natural compounds, several plant essential oils (Calmasur *et al.,* 2006) were evaluated as acaricides. The insecticidal and larvicidal properties of rosemary essential oil are documented (Waliwitiya *et al.,* 2009; Pavela, 2008). Natural products have been used as templates for semi-synthetic acaricidal agents (Tsukamoto *et al.,* 1997a, b). Moving toward green chemistry processes and developing new crop protection tools with novel modes of action to control the two-spotted spider mite are needed.

The aim of this work was (i) to evaluate the acaricidal activities of the EO of *R. officinal*is on the two-spotted spider mite *T. urticae* and its predatory mite *P. persimilis* . (ii) to determine the chemical composition of the EO of *R. officinalis.*

MATERIALS AND METHODS

2.1. Plant material and extraction of the essential oil

2.1. Plant material

Fresh aerial parts of *R. officinalis* were collected during its flowering period from the research farm of National Research Center (NRC), Dokki, Giza, Egypt in 2009 and shadow dried for 7-9 days at room temperature (25±2°C). The EO of rosemary was extracted by hydro-distillation using a Clevenger-type apparatus for 3 h as described by Negahban *et al.,* 2006. The oily layer obtained on top of the aqueous distillate was separated and dried with anhydrous sodium sulfate (0.5 g). The extracted EO was kept in sealed airtight glass vials and covered with aluminum foil at 5°C until further analysis.

2.2. GC/MS analysis of the essential oil

Major constituents of the essential oil were identified by gas chromatography/mass spectrometry (GC/MS) using a Shimadzu GC-17A gas chromatograph (Shimadzu Corp., Kyoto, Japan), coupled with a Shimadzu mass spectrometer detector (GC-MS QP-5050A). The GC-MS system was equipped with a TRACSIL Meta X5 column (Teknokroma S. Coop. C. Ltd., Barcelona, Spain; 30 m × 0.25 mm i.d., 0.25 µm film thickness). Analyses were carried out using helium as carrier gas at a flow rate of 1.0 mL/min at a split ratio of 1:10 and the following temperature program: 40°C for 5 min; rising at 3.0°C/min to 200°C and held for 1min; rising at 15°C/min to 280°C and held for 10 min. The injector and detector were held at 250 and 300°C, respectively. Diluted samples (1:10 pentane, v/v) of 0.2 µL of the extracts were always injected. Mass spectra were obtained by electron ionization at 70 eV, using a spectral range of *m/z* 45-450. The identification of individual compounds of essential oils was accomplished using two different analytical methods: (a) KI, Kovats indices in reference to *n*-alkanes (C_8-C_{32}) by National Institute of Standards and Technology (NIST) 2009; and (b) mass spectra (authentic chemicals and Wiley spectral library collection). Identification was considered to be tentative when it was based on mass spectral data only. The relative concentration of each component of the essential oil was quantified according to the peak area integrated by the analysis program.

2.3. Acaricidal activity

2.3.1. Origin and rearing of mites

Field strains of mites were established from field collections of spider mite *T. urticae* and *P. persimilis* from cotton plantations at the farm of Sakha Agricultural Research Station, Egypt. Mite cultures were maintained in climatic rooms at 27± 1° C and 60 ± 5 % R.H., with 16h L: 8 hD photoperiod for two generations to increase the cultures populations. Spider mites were reared on bean plants (*Phaseolus vulgaris* L.), a more suitable host using Dittrich (1962) technique. The predacious mite *P. persimilis* was reared in plastic boxes (22 x 15 x 10 cm); a cotton pad was put in the middle of each box, provided with water as a barrier to prevent predatory mite individuals from escaping. Predatory mites were provided with highly infested bean leaves (mixed prey stages) three times per week.

2.3.2. Toxicity tests

LC_{50} for adult females of *T. urticae:* Leaves of bean plants, *P. vulgaris* L., were punched for preparing leaf discs (3.0 cm diameter). Leaf discs were placed on wet cotton pads in Petri dishes (12 cm diameter). Seven concentrations (i.e. 2.5, 5.0 10.0, 20.0, 40.0, 80.0 and 100.0 ml /L) of EO each with nine replicates were prepared using tap water plus the spreader (Tween 20, 0.5%). Tested concentrations of the EO were sprayed on leaf discs with aid of hand sprayer even complete coverage. Control treatments were made using tap water and the spreader only. After drying of leaf discs at room temperature, ten adult females of *T. urticae* were transferred to the lower surface of each treated leaf disc. Mortality percentages were calculated after 24, 48 and 72 h from exposing *T. urticae* individuals to treated leaf discs. *T. urticae* adults were considered dead if they looked black and no movement was observed when probing with the tip of fine haired brush.

LC_{50} for adult females of *P. persimilis:* Four adult females of predacious mite were transferred to each bean leaf disc (3 cm diameter) previously infested with 30 individuals of *T. urticae* and places on moistened cotton pad in a Petri dish (12 cm diameter). The leaf discs were sprayed as previously mentioned with the same concentrations and replicates. Mortality of predatory mites was recorded 24, 48 and 72 h after treatments.

Repellent activity: To study the preference response of *T. urticae* when given a choice between EO-treated and untreated plants, the deterrent activity of rosemary oil at concentrations of 0.25, 0.50, 1.00 and 1.50% against *T. urticae* was assessed. Bean leaves were cut into discs (3.0 cm diameter) of symmetrical portion along the midrib obtained per each disc. Half of each disc was sprayed with the EO tested concentration, with aid of hand sprayer even complete coverage, while the other half was sprayed with water plus the spreader (Tween 20, 0.5%) as check. The treated discs were left to dry and put on moistened cotton wool in Petri dishes (10.0 cm diameter). Twenty adult females were transferred on the midrib of each disc. Three discs were used as replicates for each concentration. The mites were left to move freely across the two portions of the disc. *T. urticae* individuals in each portion were counted after 12, 24 and 48 h. The number of eggs laid on both sides was recorded after 48 h. The deterrence index (DI) was calculated according to Pascual and Robledo, 1998 using the equation:

$$DI = [(C - T) / (C + T)] \times 100$$

Where: T and C represent the mean number of adult female on the treated and untreated portions of leaf disc, respectively.

Fumigant activity: The method used to evaluate the fumigant activity of the oil was adapted from Aslan *et al.* (2004). Glass jars receptacles with a capacity of 3.0 L were used as test chambers. One Petri dish (7.5 cm diameter) was introduced to each chamber. Thirty adult females of *T. urticae*, 10 mites in each leaf disc (2.5 cm diameter) of bean, were put in each Petri dish. Six discs were used as replicates for each concentration. Filter paper discs saturated with water were used under the leaf discs. Essential oil at 0.25, 0.50, 1.00, 2.00, 3.00, 4.00 and 5.00 ml were applied on Whatman filter paper pieces (2 cm × 3 cm),and fixed on the inner surface of the glass chambers, which corresponding to 0.08, 0.17, 0.33, 0.67, 1.00, 1.33 and 1.67 ml oil /L of air, respectively. No material was applied to the control glass chamber. The treatments were kept in a holding chamber of 27 ± 1°C and 60 ± 5 % R.H., with 16hL :8 hD photoperiod. Exposure periods were 24, 48 and 72 h and the numbers of dead adults were counted.

2.4. Phytotoxic effects

The foliages of greenhouse-grown cotton, bean, tomato and cucumber, which represent some host plants of *T. urticae*, with four weeks old were visually examined to discover any phytotoxic effects after treatment with the essential oil. The highest concentration of the essential oil used in the experiments (10%) was dissolved in water plus the spreader (Tween 20, 0.5%) and sprayed uniformly with a hand sprayer on the surface of whole host plant leaves (10 ml for each plant). Each plant in control groups was sprayed uniformly with 10 ml of water plus the spreader (Tween 20, 0.5%) solution. The differences in the appearance of treated plants compared with controls were considered as indication of phytotoxicity.

2.5. Data analysis

The mortality percentage was corrected using Abbott's formula 1925; the observed data were then analyzed by probit analysis PC (Finney, 1971). Means of *T. urticae*t or *P. persimilis* predatory mite numbers among treatments were calculated and compared with a single factor analysis of variance (ANOVA). Duncan's multiple range test was used to determine significant differences ($p < 0.05$) between treatments by CoStat system for Windows, version 6.311, CoStat Program (2006), Berkeley,CA, USA).

RESULTS AND DISCUSSION

3.1. Essential oil constituents

The hydrodistillaton of the dried aerial parts of *R. officinalis* gave EO with yield of 1.2% (w/w). GC/MS analysis indicated that there are thirty-one compounds, representing 99.00% of the essential oil. Their retention indices and percentage composition, listed in order of elution in the column, are given in Table (1). It is obvious from data in this Table that the major components of *R. officinalis* essential oil were identified as 1,8-cineole (39.74%), camphor (22.76%), cis-ocimene (8.89%) and camphene (7.01%). The essential oil of *R. officinalis* was rich in some monoterpenoids which may provide the acaricidal properties of this oil against *T. urticae*. Like other essential oils, natural rosemary oil is a complex mixture of monoterpenoids. Considering that target site resistance is an important problem for mite control, it is more probable that mites will evolve resistance faster to an acaricide based on a single active ingredient than to one based on a mixture of different active compounds as *R. officinalis* essential oil (EO) used in this study. The major compounds found in the essential oil have previously been reported

Table 1. Chemical composition of essential oil isolated by hydrodistillation from aerial parts of *Rosmarinus officinalis* analyzed by GC/MS

Number	RI [a]	Compound [b]	Peak Area (%) [c]
1	9.068	alpha pinene	0.15
2	9.319	Cis-ocimene	8.89
3	9.790	alpha-fenchene	0.47
4	9.873	camphene	7.01
5	10.430	sabinene	0.11
6	10.714	beta-pinene	3.32
7	11.801	isocineol	0.15
8	12.176	para cymene	2.68
9	12.353	1-limonene	1.64
10	12.638	1,8-cineole	39.74
11	13.908	linalool oxide	0.25
12	14.890	alpha-terpinolene	0.58
13	16.366	alpha-campholene aldehyde	0.39
14	17.557	camphor	22.76
15	18.115	pinocarvone	0.46
16	18.322	alpha-terpineol	0.40
17	18.813	terpineol	0.32
18	19.012	p-cymen-8-ol	0.19
19	19.428	alpha-terpineol	0.91
20	19.633	myrtenal	0.28
21	20.190	verbenone	0.40
22	21.736	carvone	0.16
23	22.335	cis sabinene hydrate	0.16
24	23.549	bornyl acetate	1.34
25	23.720	isobornyl acetate	0.63
26	24.310	trans-caryophyllene	0.79
27	24.726	trans -verbeol	1.90
28	25.243	myrtenol	1.19
29	32.426	germacrene D	0.10
30	37.450	caryophyllene oxide	1.42
31	38.618	humuladienone	0.21
		Total	99.00

[a] RI, retention index on a TRACSIL Meta X5 column
[b] Compounds are listed into order of their elution from a TRACSIL Meta X5 column
[c] Compound percentag

to have activity against a variety of insects, mites, weeds and plant pathogens (Bakkali *et al*., 2008; Koschier, 2008). Individual activities of eight monoterpenoids (anethole, carvacrol, 1,8-cineole, p-cymene, menthol, gamma-terpinen, terpinen-4-ol and thymol) were investigated against three major greenhouse pests, i.e. females and eggs of the carmine spider mite, females of the cotton aphid *Aphis gossypii* and second instar larvae of the western flower thrips *Frankliniella occidentalis* (Erler and Tunc, 2005). The composition of *R. officinalis* essential oil obtained in this study was similar to that one characterized by Miresmailli *et al.* (2006), with 1,8-cineole and camphor as major compounds.

3.2. Acaricidal activity

Toxic effects:

The LC_{50} values of rosemary EO along with their confidence limits (CL) and slope values against adult females of *T. urticae* and adult females of *P. persimilis* after 24, 48 and 72 hours exposure periods were listed in Table (2). It is clear that adult females of *T. urticae* were more susceptible to EO (LC_{50} ranged from 10.70 to 17.30 ml/L) than adult females of *P. persimilis* (LC_{50} ranged from 28.05 to 42.09 ml/L). With exposure period increase ,the acaricidal activity of EO against adult females of *T. urticae* increased with significant differences (LC_{50} values were 10.70 and 17.30 ml/L after exposure of 72 and 24 hours, respectively.) and the same results obtained in case of adult females of *P. persimilis*.

Our results clearly confirmed that essential oil from *R. officinalis* posses acaricidal activity against the *T. urticae* adults under laboratory conditions. Pontes *et al.*(2007b) reported that plant essential oils have been found to be active against *T. urticae* . Plant extracts contain compounds that show ovicidal, repellent, antifeedant, sterilant and toxic effects in insects (Isman, 2006; Bakkali *et al.*, 2008). The toxicity may be by contact, ingestion or through fumigant action. Plant essential oils and their constituents invariably have higher boiling points and such plant products that show insect toxicity in the vapour state have been recently reviewed by Rajendran and Sriranjini (2008). Although some studies have demonstrated the contact or volatile efficacy of essential oils against different mite species of the family Tetranychidae, chronic exposure to rosemary oil in high concentration has caused contact dermatitis in some rare cases (Cockayne and Gawkrodger, 1997; Hjorther *et al.*, 1997).

Repellent activity:

The deterrence index (DI) of the EO at 0.25, 0.50, 1.00 and 1.50 % on the adult females of *T. urticae* after 12, 24 and 48 hours of exposure, and average number of eggs laid per adult female on treated and untreated (control) leaves after 48 h. exposure were listed in Table(3). It is intelligible that, adult females of *T. urticae* preferred the untreated section of the leaves to feed and deposit eggs. After 12 h of exposure, few numbers of *T. urticae* adult females were recorded on leaves treated sections with essential oil concentrations. The higher concentrations of the oil used revealed a higher repellent effect on adult females of *T. utricae.* Females of the mite showed an ovipostion preference for residue-free substrate where the mean number of eggs laid on the water treated control halves of the leaf discs were higher (8.4) than that on the EO treated halves (1.1),at the higher used concentration (1.50%).

Table 2. Toxicity of extracted essential oil from *Rosmarinus officinalis* to adult females of *Tetranychus urticae* and its predatory mite *Phytoseuilus persimilis*

Mite sp.	Exposure period(h)	LC_{50}* (ml/L)	95% CL	Slope ± SE
T.urticae	24	17.30 [b]	(13.24 - 20.58)	0.9006± 0.0955
	48	12.23 [ab]	(9.83 – 15.83)	1.1092± 0.0987
	72	10.70 [a]	(8.62 – 13.14)	1.1293± 0.0988
P. persimilis	24	42.09 [c]	(33.15 – 57.03)	1.4321± 0.2001
	48	31.43 [c]	(24.15 – 40.71)	1.5373± 0.2061
	72	28.05 [c]	(21.38 – 36.22)	1.5313± 0.2039

* LC_{50} values followed by the same letter within each vertical column are not significantly different (95% CL do not overlap).

Table 3. Repellent effect of extracted essential oil from *Rosmarinus officinalis* on adult females of *Tetranychus urticae*

Exposure Period (h)	Oil Concentration (%)	Deterrence Index (DI)	Average number of eggs	
			Treatment	Control
12	0.25	50.00	n.d.	n.d.
	0.50	55.56	n.d.	n.d.
	1.00	61.11	n.d.	n.d.
	1.50	66.67	n.d.	n.d.
24	0.25	72.22	n.d.	n.d.
	0.50	77.78	n.d.	n.d.
	1.00	88.98	n.d.	n.d.
	1.50	94.44	n.d.	n.d.
48	0.25	72.22	3.0 [c]	5.2[a]
	0.50	77.78	2.1 [b]	6.4[b]
	1.00	83.83	1.8 [b]	6.3[b]
	1.50	83.83	1.1 [a]	8.4[c]

Values followed by the same letter within each column are not significantly different, n.d., not determined.

Table 4. Fumigant effect of extracted essential oil from *Rosmarinus officinalis* against adult females of *Tetranychus urticae*

Exposure period (h)	LD_{50}* (ml/L)	95% CL	Regression equation
24	1.0615 [a]	(0.6794 – 2.2908)	Y = 4.965 + 1.338X
48	0.8563 [a]	(0.5605 – 1.5592)	Y = 5.101 + 1.497X
72	0.6646 [a]	(0.4036 – 1.1823)	Y = 5.290 + 1.632X

* LD_{50} values followed by the same letter within each column are not significantly different (95% CL do not overlap).

The obtained results agreed with that of Park *et al.*(2005) and Trongtokit *et al.*(2005).they stated that a number of EO showed repellent effect against mosquitoes and usually attributed to their main compounds. A concentration of 12.5% of *R. officinalis* showed the longest repellency time for mosquitoes (Gillij *et al.,* 2008). Comparisons of the principal components of *R. officinalis* suggest that camphor was the main component responsible for the repellent effects (Gillij *et al.,* 2008).

Fumigant effect:

The LD_{50} values, 95% confidence limits and the regression line equations are shown in Table (4). It is clear that , the LD_{50} values were calculated as 1.0615, 0.8563 and 0.6646 ml/L of air with 95% confidence limits from 0.6794 to 2.2908, from 0.5605 to 1.5592 and from 0.4036 to 1.1823 ml/L of air after 24, 48 and 72 h of treatment, respectively. With the increase of exposure periods, the fumigant effect of the essential oil from *R. officinalis* against adult females of *T. urticae* increased. Our results coincided with Isman, 2000 when reported that, the aromatic vapor of rosemary affected the two-spotted spider mite as a fumigant. Koschier and Sedy, (2001) studied the antifeedant effect of rosemary essential oil against onion thrips. Several reports indicated that monoterpenoids caused insect mortality by inhibiting acetylcholinesterase enzyme activity (Houghton *et al.,* 2006). In this aspect, acaricidal activities of the essential oil used in our study on adult mortalities could be due to the fumigant toxicities of the major components of the essential oil. The effect of the essential oils on aphid mortality was attributed primarily to starvation or oral and fumigant toxicity (Hori, 1999).

3.3. Phytotoxic effects:

The foliages of greenhouse – grown cotton, bean, tomato and cucumber with four weeks old, which represent some host plant species of *T. urticae*, were visually examined to discover any changes in the growth of these host plants after treatment with the highest used concentration (10%) of the essential oil in the present study. No phytotoxic effects of the essential oil were observed on treated plants comparing with untreated plants. In a few cases, essential oil-treated plants have become attractive to plant-damaging insects and phytotoxic effects on some cultivated plants have been observed (Miresmailli and Isman, 2006). Ibrahim *et al.* (2001) reported the phytotoxic effects in limonene-treated plants, whereas Chiasson *et al.* (2004) did not observe any phytotoxicity among lettuce, roses, and tomatoes that were treated with a *Chenopodium*-based pesticide.

CONCLUSION

The obtained results suggested that the essential oil have the potential for use in the control of *T. urticae.* In accordance with the present conclusions, *R. officinalis* essential oil may be used as ecologically safe alternative miticides against *T. urticae,* and its content could lead to the development of new classes of miticidal compounds. However, further studies are needed to evaluate the mode of action and cost-efficacy of this EO on wide range of pests. Natural miticides are a desirable alternative to synthetic miticides because they have little environmental effect. In addition, no phytotoxic effects of rosemary oil to foliage of cotton, bean, tomato and cucumber, as host plants, have been observed with the highest tested concentration.

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الملخص العربي

التركيب الكيميائي والنشاط الابادي للزيت الأساسي لإكليل الجبل المصري ضد أكاروس العنكبوت الأحمر ذو البقعتين *Tetranychus urtica* ومفترسه الأكاروسي *Phytoseuilus persimilis*

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إن البحث عن مستخلصات نباتات طبية لها خواص ابادية للآفات قد يؤدى إلى اكتشاف عناصر جديدة في مكافحة الآفات. الدراسة الحالية هي محاولة لإيجاد بديلا للمبيدات المصنعة المستخدمة حاليا في مكافحة العنكبوت الأحمر ذو البقعتين. تم استخلاص الزيت الأساسي من الأجزاء الخضرية لنبات إكليل الجبل المصري. واختبر مدى سميته على العنكبوت الأحمر ذو البقعتين *Tetranychus urtica* وكذلك العنكبوت المفترس *Phytoseuilus persimilis* تحت الظروف المعملية. وكانت فترة تعريض الحيوانات للزيت 24، 48، 72 ساعة. وأوضحت النتائج أن العنكبوت الأحمر ذو البقعتين أكثر حساسية للزيت عن العنكبوت المفترس. وفى تجارب التقييم الحيوي سبب الزيت نسبة موت عالية للعنكبوت الأحمر ذو البقعتين مع وجود خواص للزيت كمادة مدخنة وكمادة طاردة. ووجد أن الزيت غير سام لبعض النباتات العائله للعنكبوت الأحمر ذو البقعتين(القطن-الفاصوليا-الطماطم-الخيار) بأعلى تركيز مستخدم من الزيت(10٪). تم التعرف على مكونات الزيت الكيماوية بواسطة استعمال جهاز الكروماتوجرافى الغازي المرتبط بمطياف الكتلة GC/MS وكانت39.74 ٪ من 8,1- سينيولبالاضافة الى كامفور (22.76 ٪)وسيس- بينين (8.89 ٪) وكامفين(7.01 ٪) وهى التي ربما تعطى الخواص الابادية للزيت ضد العنكبوت الأحمر ذو البقعتين. وخلاصة هذه الدراسة أن الزيت الأساسي لإكليل الجبل المصري ربما يكون مفيدا في تطوير عناصر جديدة لمكافحة العنكبوت الأحمر ذو البقعتين.