Atlas Journal of Plant Biology 2024, pp. 115–121
https://doi.org/10.5147/ajpb.vi.254

Mentha pulegium, *Mentha suaveolens* subsp. timija, *Lavandula mairei*, and *Artemisia mesatlantica* Essential Oils: Laboratory Screening for Biological Control Against Red Palm Weevil *Rhynchophorus ferrugineus* Olivier (Coleoptera: Curculionidae)

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Received: September 10, 2024 / Accepted: December 1, 2024

Abstract

The Red Palm Weevil (RPW), *Rhynchophorus ferrugineus* (Olivier) 1790, is currently the most harmful pest on date palm crop. Nowadays, the systematic and abusive use of synthetic pesticides to control insects that ravage crops is questioned following the awareness of the risks they generate for the environment and human health. Therefore, there is a need to search and develop alternative natural environmentally friendly strategies that can replace or reduce the use of chemical fertilizers and synthetic pesticides. In this context, a continuous and rigorous interest has been developed for the use of plant essential oils (EOs) and extracts that could constitute healthy alternatives to chemicals. This study aims to evaluate the insecticidal activity of essential oils obtained from four aromatic and/or medicinal plants, namely, *Mentha suaveolens* subsp. timija, *Lavandula mairei*, *Artemisia mesatlantica*, and *Mentha pulegium*, against larvae and adults of RPW, *R. ferrugineus* (Oliver) by contact. The four essential oils were tested at two different concentrations (0.5 µl/cm^2 and 1.5 µl/cm^2). A toxic effect was demonstrated for each essential oil tested. This effect depends on the concentration of the essential oil tested and the duration of exposure. For the toxicity of the EOs to larvae, EOs of *Mentha pulegium* developed the most important insecticide activity with very low LT50 (time required to kill 50% of the exposed insects) and LT90 (time required to kill 90% of the exposed insects) values of 9.45h and 22.16h respectively for 1.5 µl/cm^2 concentration. *Mentha suaveolens* subsp. timija is in the second position with values of LT50 and LT90 of 19.35 h and 34.80 h respectively for a concentration of essential oil of 1.5 µl/cm^2. The longest lethal times 50 and 90 are recorded with Lavandula mairei oils which required 52.44h and 107.25h to cause adult mortality of 50% and 90%, respectively. The essential oils of *Artemisia mesatlantica* recorded average values of LT50 and LT90 of 35.60h and 91.53h respectively. On the other hand, the shortest lethal times LT50 and LT90 for adult were recorded after 8.27h and 19.39h respectively of treatment with EOs of *Mentha pulegium* and *Mentha suaveolens* subsp. timija using the dose of 1.5 µl/cm^2. The essential oil of *Artemisia mesatlantica*, classified as the second most toxic oil, showed average values of LT50 and LT90 of 22.68h and 30.09h, respectively for the concentration 1.5µl/cm^2. Treatment with *Lavandula mairei* EO gives average values of LT50 and LT90 of 30.95h and 57.77h, respectively. Our study could be a basic guide for the possible field examination of the preventive (oviposition repellency) and curative action of the essential oils tested.

Keywords: Date palm, Red palm weevil, *Rhynchophorus ferrugineus*, Essential oils, Plant extracts, Aromatic and medicinal plants, *Mentha pulegium*, *Mentha suaveolens* subsp. timija, *Lavandula mairei*, *Artemisia mesatlantica*.

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Atlas Journal of Plant Biology - ISSN 1949-1379. Published By Atlas Publishing, LP (www.atlas-publishing.org)

Atlas Journal of Plant Biology - ISSN 1949-1379. Published By Atlas Publishing, LP (www.atlas-publishing.org)

Introduction

Date palm (*Phoenix dactylifera* L.) is often infested with multiple insect pests. The red palm weevil, *Rhynchophorus ferrugineus* (Olivier) 1790, is the most destructive pest of many palm species grown in Asia, the Middle East and the Mediterranean basin. The hidden life of the larvae, as well as the eggs and the nymphs of this insect in the trunk of the palm trees is the serious obstacle of the control of this pest. Despite the considerable amount of work and publications done on alternative methods of control of *R. ferrugineus*, the field control of this pest is still done mainly by the use of synthetic insecticides. The application of synthetic insecticides by injection and chemical control by fumigation are used to treat the infested palms (Rao et al., 1973; Abraham et al., 1975; Muthuraman, 1984; Avand-Faghih et Gharib, 1995). However, this massive use of synthetic insecticides has negative consequences on the environment (biodiversity loss, water and soil pollution, etc) and human health (Hoy and Dahlsten 1984; Kamel et al., 2007; EL-Saeid and AL Dosary 2010; Al-Ayedh et al., 2016). For these reasons, it is necessary to develop alternative methods to replace chemical insecticides for the management of red palm weevil. Among these methods, biological control through the use of essential oils (EOs) which have received significant attention from the scientific community because of their acute toxicity, repellency, antifeedant effect and fumigant toxicity against many stored-grain pests and weevils (Regnault-Roger and Hamraoui, 1995; Shaaya et al., 1997, Tozlu et al. 2011). Indeed, various research has been conducted in recent years to control RPW, using essential oils (Abdel-Raheem et al., 2020; Reyad et al., 2020; Al Dawsari, 2020; Ali et al., 2019; Abdel Kareim et al., 2017; Ahmed et al., 2015; Abdullah, 2009). Furthermore, Hussain and AlJabr (2014) found that African myrrh's essential oil, *Commiphora myrrha* (Burseraceae) can be used as a bio-pesticide in an integrated management plan to control this pest. Shukla et al (2012) demonstrated the antifeedant activity of crofton weed flower essential oil against the adult of RPW. An investigation done by Yan et al (2021) showed that eugenol and thymol derivatives exhibited antifeedant activity against RPW larvae. Essential oils of many plant species were tested and showed strong toxicity against many other insect species (Bouda et al., 2001; Isikber et al., 2006).

In this context, we initiated this study with the objective of investigating the insecticidal activity of essential oils obtained from four aromatic and/ or medicinal plants against larvae and adults of RPW, *R. ferrugineus* (Oliver) one of the most widespread and destructive date palm pests. Therefore, our study could provide valuable information on the potential of essential oils tested as biological control agents against this insect, and it will be a starting point for further research on the feasibility of their field application as biological preventive (oviposition repellency) and curative agents. To our knowledge, there has been no previous report evaluating the insecticidal properties of the essential oils of these four plants on the larvae and adults of this insect.

Materials and Methods

Plant Material

The plant material used for our experiments consists of the aerial parts of four following aromatic and medicinal plants: *Mentha suaveolens* subsp. timija, *Lavandula mairei*, *Artemisia mesatlantica* and *Mentha pulegium*. The first three plants were harvested from their natural populations located in the western center of Morocco; *Mentha pulegium* was cultivated then harvested in Nafeesa organic farm of Yousef Abdul Latif and Sons Agriculture Ltd. (YALA) Company. Qassim. Saudi Arabia. . Plant samples were air-dried in the shade and then stored in the dark at 4°C until their use.

Extraction of Essential Oils

The extraction of EOs from dried aerial plant materials was carried by hydrodistillation using a Clevenger type apparatus (1928) for 4 hours. By density difference, the extracted essential oil separates from water and rises to the surface. The obtained EOs were dried over anhydrous sodium sulfate and stored away from light in brown glass bottles at 4°C until used.

Insect Collection and Mass Rearing Conditions

In the present study larvae and adults of the RPW were obtained from infested palm trees in Nafeesa organic farm of Yousef Abdul Latif and Sons Agriculture Ltd. (YALA) Company, Qassim province, Saudi Arabia. About 300 adults and 225 larvae were collected carefully through mechanically scrapping the infested and surrounding palm tissue. The collected study larvae and adults were kept in the laboratory. Larvae and adults of both sexes are reared in oblong and transparent plastic boxes (20*10*15 cm). These boxes have been covered with a plastic lid riddled with 1.5*1.5 mm diameter holes to allow aeration of the insects and prevent their exit. Date palm leaf parts were cleaned and cut into small pieces of about 10cm and were used as a food source. The boxes were then placed on shelves in the laboratory. Mass rearing is done at room temperature. Only adults and larvae are used for the contact biological testing. They were checked as repeatedly as possible for good health before their use. The tests aim to determine the values of LT50 and LT90 (time required to kill 50% and 90% of the exposed insects respectively).

Insecticidal Activity

This test consists of studying the effect of direct and permanent contact between adults and larvae of R. ferrugineus from one side and essential oils from the other side. For each essential oil, two doses were prepared 0.5 and 1.5 µl/cm² by diluting each time in 1/5 ml of acetone the respective volumes of 31.8 and 95.37µl of the essential oil. Each of the solutions prepared was spread evenly using a micropipette on a disc of filter paper (Whatman n°1) 9 cm in diameter (63.62 cm² of surface) placed in a glass Petri dish of the same diameter. The disc was then left at room temperature for 15 minutes to allow uniform diffusion of the essential oil and complete evaporation of the dilution solvent. For the negative control, the discs were treated only with acetone as when applied to insects, it did not have any significant effects on insect mortality (Singh and Jain, 1987; Tunc et al., 1997). Adults and larvae were collected from their rearing environment and divided into batches of 5 individuals each. The 5 adult insects were transferred to each petri dish, which is then immediately closed. The larvae underwent the same treatment with 3 individuals for each petri dish. The test was performed in triplicate repeating each assay twice. The boxes are then maintained at 25°C. The insects and the larvae are fed with small pieces of the palm leaf. For each treatment, mortality was checked after 2, 4, 6, 8 and 24 h, then periodically every 24 h. The larvae were considered dead if no movement was recorded, while adult insects were considered dead when no leg or antennal movements were recorded.

Mean mortality data were used to calculate lethal time causing 50% and 90% of mortality (LT50and LT90) for each essential oil.

In order to have a mortality gradient as a function of time, the mortality percentages obtained by each dose of essential oil tested were calculated after 24h, 48h and 72h of contact. From these data, the lethal times 50 and 90 were determined. They are calculated from the probit regression line corresponding to the percentages of mortalities corrected according to the logarithms of treatment times. The Schneider formula and the problem table are used (Ramade, 2007).

$$MC=[M2 -M1/100 -M1] \text{ x}100$$

With: MC: % of mortality corrected, M2: % of mortality in treated population, M1: % of mortality in control population.

Data Analysis

Data were subjected to statistical analysis (anova, p= 0.05.) where treatment f, treatment/error df and true p values were computed using the randomised block design through STATISTICA software, (V6.0) (Stat-Soft, 2001, Creteil, France). To determine the LT50 and LT90 lethal time values with their confidence limits as well as the Chi-two (χ2), the results obtained were analyzed by the SPSS 12.0 Statistical Software from the probit log model. Two values are significantly different if their confidence limits (LC) do not overlap.

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Results

Toxicity of Essential Oils to Larvae

The analysis of the data allowed the determination of the plants and their essential oils concentration of high efficiency. This analysis revealed the presence of a very highly significant difference between the EOs (Table 1). The results showed that the EOs of *Mentha pulegium* and *Mentha suaveolens* subsp. timija have developed the most important insecticide activities. For *Mentha pulegium* EO, the mean mortality rate for both concentrations (0.5 and 1.5 µl/cm²) was 55.56% and 88.89%, respectively, during the first 24 hours of treatment. The EOs of Mentha suaveolens subsp. timija resulted in a mortality of 55.56% for 0.5 µl/cm² and 66.67% for 1.5 µl/cm², after 24 hours of treatment. The lowest mean mortality rate (00.00%) after 24 hours of treatment for 0.5 µl/cm² was obtained by the EOs of *Lavandula mairei* and *Artemisia mesatlantica*. At a concentration of 1.5 µl/cm², the EOs of *Lavandula mairei* showed the lowest average mortality rate (11.11%) after 24 hours of treatment.

After 48h of treatment and at the lowest concentration of 0.5µl/cm², the results of the highest mortality rates are recorded by *Mentha pulegium* and *Mentha suaveolens* subsp. timija with 77.78% and 66.67%, respectively. At the same concentration, the essential oils of *Lavandula mairei* and *Artemisia mesatlantica* recorded the lowest values of mortality, that is 22.22% and 33.33% respectively. However, for the high dose (1.5 µl/cm²), the EOs of the four plants showed an increase in mortality rates after 48 hours of treatment.

After 72 hours of treatment, the effectiveness of the EOs tested is most notable for *Mentha pulegium* and *Mentha suaveolens* subsp. Timija with mortality rates between 88.89% and 100% for both doses. The acetone used in the negative control generated no mortality to the larvae.

The increase in the applied concentration is positively correlated with the increase in mortality rates related to each essential oil.

Toxicity of Essential Oils to Adult

For each EO tested, there is a toxic effect depending on the concentration and duration of exposure. The results obtained (Table 2), showed that the survival of adults decreases as the concentration of essential oils increases. After 24 hours of exposure, the most effective EOs, by contact against adults of R. ferrugineus are those of Mentha suaveolens subsp. timija, with an average mortality rate, for both concentrations (0.5 and 1.5 µl/cm²), of 86.67% and 100% respectively. Mentha pulegium EOs resulted in 33.33% mortality for 0.5 µl/cm² and 100% for 1.5 µl/cm² after 24 hours of treatment. While the EOs of Lavandula mairei showed the same mortality rate (33.33%) after 24 hours for both concentrations. While Artemisia mesatlantica gave a mortality of 13.33% for 0.5 µl/ cm² and 60.00% for 1.5 µl/ cm² after 24 hours of treatment.

After 48 hours of exposure, *Mentha suaveolens* subsp. timija showed the same average adult mortality after 24 hours of exposure for both concentrations. While *Mentha pulegium* EOs developed a 100% mortality rate for both concentrations. *Lavandula mairei* EOs caused lower mortality, about 60.00% in adults for 0.5 µl/cm² and 73.33% for 1.5 µl/cm², while *Artemisia mesatlantica* caused 26.67% mortality for 0.5 µl/cm² and 100% for 1.5 µl/cm².

After 72 hours of exposure at a concentration of 0.5 µl/cm², only *Mentha pulegium* and *Mentha suaveolens* subsp. timija showed high toxicity, causing 100% of mortality. At the same concentration, EOs of *Lavandula mairei* and *Artemisia mesatlantica* generated moderate mortality rates of 73.33% and 46.67%, respectively. However, at the concentration of 1.5 µl/cm², the EO of all plants generated high mortality rates of 100%. Acetone used in the negative control resulted in negligible mortality in adults of 13.33%.

As for the effect of concentration, the increase in concentration is positively correlated with the increase in mortality rates related to each essential oil. The concentration of 1.5 µl/cm² was found to be the most toxic.

The decreasing order of insecticide activity of the essential oils tested can be summarized as follows:

M. pulegium L. ≥ *M. suaveolens* > *A. mesatlantica* ≥ *L. mairei*

Lethal time LT50 and LT90 in larvae

All essential oils of the studied plants showed insecticidal activity against the larvae of the insect studied. The highest mortality is always observed in the insects treated at a concentration of 1.5 µl/cm² (Table 3). The results showed that EO of *Mentha pulegium* developed the most important insecticide activity with very low LT50 and LT90 values of 9.45 h and 22.16 h respectively for the concentration of 1.5 µl/cm². This means that a concentration of 1.5 µl/cm² of *Mentha pulegium* EO, used for 9.45 h, leads to the death of 50% of the larvae population of the red palm weevil, and that the same concentration of *Mentha pulegium* EO, used for 22.16 h leads to the death of 90% of the RPW larvae population. *Mentha suaveolens* subsp. timija essencial oil is in the second position with values of LT50 and LT90 equal to 19.35 h and 34.80 h for a concentration of essential oil of 1.5 µl/cm². At this same concentration, the longest lethal times 50 and 90 are recorded with *Lavandula mairei* essential oils that required 52.44 h and 107.25 h to cause adult mortality of 50% and 90%, respectively. The essential oils of *Artemisia mesatlantica* recorded average values of 35.60 h and 91.53 h, respectively for LT50 and LT90. The acetone used for the negative control had no effect on larvae. LT50 and LT90 results are grouped in Table 3.

The analysis of the variance with two classification criteria reveals a very highly significant difference for the essential oils and dose factors and their interaction.

Table 1. Average mortality caused by each essential oil for larvae. Means that do not share a letter within the same row indicate significant differences ($p < 0.05$).

	Concentration (µl/cm²)	Time	Average mortality rate (%)				
			M. suaveolens	*M. pulegium*	*L. mairei*	*A. mesatlantica*	Control
Larvae	0,5	after 24 h	55.56[c]	55.56[c]	0.00[f]	0.00[f]	0.00[f]
		after 48 h	66.67[c]	77.78[b]	22.22[d]	33.33[d]	0.00[f]
		after 72 h	88.89[a]	100.00[a]	44.44[c]	55.56[c]	0.00[f]
	1,5	after 24 h	66.67[c]	88.89[a]	11.11[e]	33.33[d]	0.00[f]
		after 48 h	100.00[a]	100.00[a]	33.33[d]	55.56[c]	0.00[f]
		after 72 h	100.00[a]	100.00[a]	77.78[b]	88.89[a]	0.00[f]

Table 2. Average mortality caused by each essential oil for adult. Means that do not share a letter within the same row indicate significant differences ($p < 0.05$).

	Concentration (µl/cm²)	Time	Average mortality rate (%)				
			M. suaveolens	*M. pulegium*	*L. mairei*	*A. mesatlantica*	Control
Adult	0,5	after 24 h	86.67[b]	33.33[d]	33.33[d]	13.33[e]	0.00[f]
		after 48 h	86.67[b]	100.00[a]	60.00[c]	26.67[d]	6.67[e]
		after 72 h	100.00[a]	100.00[a]	73.33[b]	46.67[c]	13.33[e]
	1,5	after 24 h	100.00[a]	100.00[a]	33.33[d]	60.00[c]	0.00[f]
		after 48 h	100.00[a]	100.00[a]	73.33[b]	100.00[a]	13.33[e]
		after 72 h	100.00[a]	100.00[a]	100.00[a]	100.00[a]	13.33[e]

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Table 3. LT50 and LT90 values of essential oils against red palm weevil larvae.

Concentration (µl/cm^2)	LT50 (h) (95 % CI)	LT90 (h) (95 % CI)
M. suaveolens **(timija)**	-	-
0.5	22.31	99.13
1.5	19.35	34.80
M. pulegium	-	-
0.5	23.23	54.53
1.5	9.45	22.16
L. mairei	-	-
0.5	74.45	138.45
1.5	52.44	107.25
A. mesatlantica	-	-
0.5	64.05	115.63
1.5	35.60	91.53

CI: Confidence Interval.

Table 4. LT50 and LT90 values of essential oils against red palm weevil adult.

Concentration (µl/cm^2)	LT50 (h) (95 % CI)	LT90 (h) (95 % CI)
M. suaveolens **(timija)**	-	-
0.5	27.04	35.97
1.5	8.27	19.39
M. pulegium	-	-
0.5	25.82	32.28
1.5	8.27	19.39
L. mairei	-	-
0.5	37.36	141.44
1.5	30.95	57.77
A. mesatlantica	-	-
0.5	83.80	331.57
1.5	22.68	30.09
Control	-	-
0.5	156,00	388.16
1.5	169.66	537.11

CI: Confidence Interval.

Lethal time LT50 and LT90 in Adult

The highest and most effective concentration (1.5µl/cm^2) is related to the shortest 50 and 90 lethal times for all essential oils (Table 4). The shortest lethal times 50 and 90 were recorded after 8.27 h and 19.39 h of treatment with EOs of *Mentha pulegium* and *Mentha suaveolens* subsp. timija at the dose of 1.5 µl/cm^2. The essential oil of *Artemisia mesatlantica,* classified as the second most toxic essential oil, showed average values of LT50 and LT90 equal to 22.68 h and 30.09 h, respectively for the concentration of 1.5µl/cm^2. Lavandula mairei EO gives average values of LT50 and LT90 equal to 30.95 h and 57.77 h, respectively. The highest mortality is always observed with insects treated at 1.5 µl/cm^2.

The analysis of the variance with two classification criteria reveals a very highly significant difference for the essential oils and dose factors and their interaction.

On the other hand, the results show that the speed of action and the degree of toxicity of essential oils are higher for adults than for the larvae of the red palm weevil.

For a healthy and environmental-friendly agricultural production, aromatic and medicinal plants, through their insecticide activities, are promising as biological control agents, through the use of their essential oils with insecticidal effect, against crop pests (Ahmed et al., 2015; Barud et al., 2014; Benelli et al., 2013).

As shown in Table 5, in *M. pulegium* EO aerial parts, 36 compounds were recognized, bringing the total of identified compounds to 91.6%, in which pulegone (83.24%) accounted for nearly the totality of the EO composition (Hallouti, 2019. The EO *Mentha suaveolens* subsp. timija accounted for a total of 99.00% of identified compounds. Menthone (39.4%), pulegone (34.3%) and isomenthone (7.8%) were found as the main components for this EO (Kasrati et al., 2014). Up to Twenty-three compounds were identified from *Lavandula mairei* EO, representing more than 98% of EO which characterized by high amount of carvacrol (78.29%) (El Hamdaoui et al., 2018). The chemical composition of *Artemisia mesatlantica* EO is dominated by β-thujone with a percentage of 77.77%, followed by 1.8-cineol (6.31%), and camphor (3.52%) (Hinane et al., 2020).

Discussion

In this study, essential oils extracted from four plants (*Mentha suaveolens* subsp. timija, *Mentha pulegium, Lavandula mairei* and *Artemisia mesatlantica*) were tested for their toxicity to larvae and adults of R. ferrugineus by contact. To our knowledge, this is the first work that illustrates the effect of the essential oils of these four plants on the larvae and adults of this insect. The lower the LT values are, the greater the toxicity of the essential oil is. Many authors have also highlighted the insecticidal effect of EOs in other aromatic and medicinal plants against the red palm weevil (Abdel-Raheem et al., 2020; Al Dawsari, 2020; Reyad et al., 2020; Ali et al., 2019; Abdel Kareim et al., 2017; Ahmed et al., 2015; Abdullah, 2009). The analysis of our data revealed a sensitivity of this insect to the essential oils of the plants studied. However, we noted a difference in this sensitivity depending on the stage of development of the insect (larva or adult), the essential oil and the concentration / dose tested. The results obtained in this investigation also indicated that the essential oils of *Mentha pulegium* and *Mentha suaveolens* subsp. timija were the most toxic against this pest. While the lowest insecticidal effects were observed with the essential oils of *Artemisia mesatlantica* and *Lavandula mairei*. This difference in the insecticidal activity from one essential oil to another is related to the differences in their chemical composition. In fact, the insecticidal activity of these EOs can be attributed to the synergistic effect of their chemical compounds or in particular to the activity of their major compounds (Franzios et al., 1997). Lee et al. (2001) reported that the insecticidal activity of essential oils is influenced by the chemical composition of these essential oils. In another study done by Park et al. (2002) evaluating the effectiveness of *Ocimum gratissimum* essential oil on *Callosobruchus chinensis* (Chinese bruche) and *Sitophilus oryzae* (rice weevil), it has been shown that the toxicity of these essential oils is mainly due to their high content of oxygenated monoterpene molecules. The important insecticidal activity of the essential oil of *M. suaveolens* subsp. timija can be attributed to the presence of large amounts of menthone and pulegone. These two oxygenated monoterpenes are reported to be majority compounds with significant insecticidal activity (Kasrati et al., 2014; Benayad et al., 2012; Liu et al., 2011; Koul et al., 2008). Nevertheless, other studies have reported that pulegone's insect toxicity is more interesting than that of menthone (Lee et al., 2003; Pavlidou et al., 2004). *Mentha pulegium* EO has already been mentioned for its remarkable insecticidal activity by several authors (Franzios et al., 1997; Boulogne et al., 2012). This toxicity is strongly linked to its chemical composition rich in monoterpenes (pulegone, menthone, and carvone). The chemical composition of the essential oils of this plant is dominated, in Morocco, by pulegone with a percentage of 83.24% (Hallouti, 2019) and 80.3% (Bouchra et al., 2003). Franzios et al. (1997) showed that pulegone is more toxic than carvone and menthone, causing mortality of *Drosophila melanogaster* even at low doses. The high toxicity of pulegone has also been proven against other insects in numerous studies: against larvae of *Spodoptera eridania* (Gunderson et al., 1985) and *Spodoptera littoralis* (Pavela, 2005). *Mentha pulegium* EO showed important contact toxicity (100%) at 3 hours of exposure with the 0.25 µL/cm^2 concentration against *Plodia interpunctella* (Lepidoptera: Pyralidae) (Moullamri et al., 2024). The important toxic effect of essential oils of *M. suaveolens* subsp. timija and *Mentha pulegium* demonstrated in this study, is consistent with that already reported for other species of the genus *Mentha* against various insects (Upadhyay et al., 2018; Kasrati et al., 2014; Aggarwal et al., 2001; Pavlidou et al., 2004; Pavela, 2008; Kumar et al., 2011).

With regard to EO of *Artemisia mesatlantica*, very little is reported in the literature. The toxic effects of its essential oils against *R. ferrugineus* are generally moderate. The chemical composition of *A. mesatlantica* essential oil, an endemic plant to Morocco has been investigated (Sekkat et al., 2017; Bencheqroun et al., 2012; Ouyahya et al., 1990). In this context, Bencheqroun et al. (2012) reported that ß-thujone is the main component (56.33 %) of *A. mesatlantica* essential oil followed by camphene (7.48 %) and camphor (4.17 %). The presence of a significant content of these oxygenated monoterpenes (thujones, camphene and camphor) may be responsible for its pronounced activity against larvae and adults of *R. ferrugineus*. Compounds isolated from *Artemisia* species are known primarily for their insecticidal properties (Kordali et al., 2005, 2006). The essential oils of *Artemisia herba-alba* and *Artemisia absinthium* have toxic properties against adults of *Orysaephilus surinemensis* and *Tribolium castaneum* (Bachrouch et al., 2015). The composition of the essential oil of *Artemisia absinthium* L exhibits a large intraspecific variability. In addition to the widely recognized -thujone, at least 17 other major compounds have been identified in the essential oil, in-

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Table 5. Chemical composition of essential oils obtained from the four plants tested (%). Tr = Traces.

Compound	*M. pulegium*	*M. suaveolens* (timija)	*L. mairei*	*A. mesatlantica*
α-Pinene	-	0.6	0.35	-
α-Humulene	-	0.3	-	-
α-Eudesmol	-	-	-	2.75
α -Phellandrene	-	-	0.19	-
α -Terpinene	-	-	0.16	-
α -Terpineol	-	-	0.38	-
α- thujone	-	-	-	0.23
β-Bourbonene	-	0.2	-	-
β -Caryophyllene	-	-	1.45	-
β-Copaene	-	0.3	-	-
β-Dihydoagarofuran	-	0.1	-	-
β-Elemene	-	0.2	-	-
β -Myrcene	-	-	1.00	-
β-Pinene	-	0.5	-	-
β– thuyone	-	-	-	77.77
δ-3-Carene	-	-	0.54	-
γ-Amorphene	-	0.1	-	-
γ-Cadinene	-	0.1	-	-
***p*-Cymene**	-	-	0.77	-
***p*-Cymen-8-ol**	-	-	1.81	-
Bicyclogermacrene	-	0.7	-	-
Borneol	-	1.4	-	-
Cadinene (delta)	1.5	-	-	-
Carvacrol	-	-	78.29	-
Carvacrol methyl ether	-	-	1.36	-
Caryophyllene oxide	-	-	2.31	-
Camphene	-	0.3	-	1.44
Camphor	-	-	-	3.52
***Cis-β*-dihydroterpineol**	-	-	-	2.94
***Cis-β*-Guaiène**	-	0.2	-	-
***Cis*-Jasmone**	-	0.1	-	-
***Cis- β* -Ocimene**	-	-	1.72	-
Davanone	-	-	-	0.13
(E)-Caryophyllene	-	3.0	-	-
Germacrene D	-	4.1	-	-
Humulene epoxide II	-	-	-	2.68
Isomenthone	-	7.8	-	-
Limonene	0.39	0.6	0.31	-
Linalool	-	-	0.33	-
Manoyloxide	-	-	0.44	-
Menthone	-	39.4	-	-
Mentha-1.8-dien-4-ol	-	-	0.36	-
Myrcene	-	0.1	-	-
Neomenthol	1.96	-	-	-
Octanol 3	0.69	-	-	-
Octen-3-ol	-	-	1.84	-
Octan-3-one	-	-	0.45	-
Para mentha-3,8-diene	0.16	-	-	-
Pinene (alpha)	0.21	-	-	-
Pinene (beta)	0.18	-	-	-
Piperitone	-	0.6	-	-
Piperitenone	-	1.8	-	-
Pulegone	83.24	34.3	-	-
Sabinene	-	0.4	-	-
Spathulenol	-	0.1	1.51	-
Terpinen-4-ol	2.06	-	0.36	2.10
Terpineol (alpha)	1.21	-	-	-
Terpinolene	-	-	2.96	-
***Trans- β* -Ocimene**	-	-	Tr	-
Valerianol	-	0.1	-	-
Z-β-Ocimene	-	0.4	-	-
10-Epi-γ-eudesmol	-	-	-	0.09
1,8-Cineol	-	0.5	-	6.31
Total (%)	91.6	99.0	98.64	99.96

cluding myrcene, sabinene, sabinyl acetate, epoxyocimene, chrysanthenol, and chrysanthenyl acetate, among others. (Nguyen and Németh, 2016). As for *L. mairei*, which is an endemic species of Morocco (El Hamdaoui et al., 2021), its toxicity, even low, is probably due to the high percentage of carvacrol which is well known for its biological activities including insecticide activity (Szczepanik et al., 2012; Ahn et al., 1998). El Hamdaoui's results (2019) reported that the two plants *L. mairei* and *T. leptobotrys* have carvacrol as their main compound and show the most important antioxidant and antibacterial activities and lower insecticidal activity compared to other plants tested.

In relation to their biological mode of action, essential oils would be the safest alternative to synthetic insecticides. They are promising candidates for the development of novel bioinsecticides due to their strong insecticide activity. Large-scale trials on these compounds are needed to assess their application and formulation as bioinsecticides and as oviposition deterrent agents in combination with efficient trapping system, for developing a sustainable agricultural pest management system against red palm weevil (Kedia et al., 2015). However, there are so many gaps and challenges, particularly with regard to their high volatility, which necessitates the development of appropriate formulations to minimize their high volatility and optimize effectiveness while permitting field application. The second challenge is to determine their modes of action as they are a complex mixture of biologically active molecules capable of affecting multiple targets at the same time. More work is needed to elucidate their availability and consistency of the source plant and their cost-effectiveness.

Conclusion

In the light of this study, there is a potential for the use of essential oils extracted from four aromatic and medicinal plants namely: *Mentha suaveolens* subsp. timija, *Lavandula mairei, Mentha pulegium* and *Artemisia mesatlantica*, as effective bioinsecticides against larvae and adults of *R. ferrugineus*. Among the four essential oils tested, the essential oils of *Mentha pulegium* and *Mentha suaveolens* subsp.

timija showed very high insecticide activity against adults and larvae of this pest in vitro, while *Artemisia mesatlantica* and *Lavandula mairei* were less effective.

The insecticide performances highlighted deserve to be studied by very thorough research and in complementary in vivo tests in order to confirm these performances and to consider the prospects of application of these essential oils as natural insecticides, alternatives to synthetic insecticides to control R. ferrugineus. Our study could be a basic guide for the possible field examination of the preventive (oviposition repellency) and curative action of the essential oils tested. They can be used also after any leaf, offshoot and aerial offshoots removal. This is because the periodically removal of offshoots go a long way in enhancing early detection of infested palms by RPW besides facilitating treatment of the palm trees. The application of these EOs in an integrated agricultural pest management plan against the red palm weevil seems to be interesting.

Acknowledgements

The authors would like to deeply thank Mr Youssef Jameel the Chairman of Yousef Bin Abdul Latif and Sons Agriculture Co. Ltd for his financial support and for allowing us to use the farm and laboratory facilities of his Company during this research.

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