

Insecticidal Activity of Essential Oils Derived from Lavender, Laurel and Peppermint Against Lesser Grain Borer, *Rhyzopertha dominica* (Fabricius, 1792) (Coleoptera: Bostrichidae)

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ABSTRACT

Fumigant effects of the essential oils from *Laurus nobilis* L., 1753 (Lurales: Lauraceae), *Mentha piperita* L., 1753 and *Lavandula x intermedia* Emeric ex Loisel, 1828 (Lamiales: Lamiaceae) were determined against the lesser grain borer [*Rhyzopertha dominica* (Fabricius, 1792) (Coleoptera: Bostrichidae)], an important pest species of stored grains.

Essential oils were extracted by steam distillation by Clevenger apparatus and analyzed by capillary gas chromatography-mass spectrometry. The major compounds of the essential oils were detected as linalool (43.33%), 1,8-Cineole (57.67%) and menthone (53.51%) in *Lavandula x intermedia*, *L. nobilis* and *M. piperita*, respectively. The experiments were conducted in a controlled-climate chamber at 27 ± 1 °C, 50 ± 10% RH and in the dark. The fumigant effects of essential oils were determined at five concentrations (1, 5, 20, 50, and 200 µl/L air) with larval, pupal, and adult stages of *R. dominica*. The highest concentrations of *L. nobilis* and *M. piperita* essential oils completely killed the adults of *R. dominica*, while *L. nobilis* killed 100% of the larval stage of the pest. However, the pupal stage was less susceptible, in which the highest mortality was 49% with *L. nobilis* essential oil. As a result, it is considered that both *L. nobilis* and *M. piperita* essential oils have the potential to be used as effective biofumigants for stored product pests.

Keywords: Essential oil, fumigant, *Laurus nobilis*, *Lavandula intermedia*, *Mentha piperita*

Çilgin, E. & Keçeci, M. (2024). Insecticidal activity of essential oils derived from lavender, laurel and peppermint against lesser grain borer, *Rhyzopertha dominica* (Fabricius, 1792) (Coleoptera: Bostrichidae). *Journal of the Entomological Research Society*, 26(1), 1-16.

Received: March 13, 2023

Accepted: November 18, 2023

INTRODUCTION

Cultural, mechanical and chemical methods are employed to prevent contamination and eliminate potential damage for controlling stored product pests. In Türkiye, fumigation is common control method for *Rhyzopertha dominica*, (Fabricius, 1792) (Coleoptera: Bostrichidae), which is similar to the control of many other pests active in stored products.

Chemical control methods in stored product pests can lead to negative effects for humans, animals, plants and other organisms, and troubles affiliated with the occurrence of pest resistance, residual toxicity, and the demolition of natural balance. There is worldwide worry about the prejudicial effects of fumigants and synthetic insecticides, including as depletion of the ozone layer, ecological pollution, toxicity on non-target organisms and pest resistance (Ogendo et al., 2008; E. Shaaya, Kostjukovski, Eilberg, & Sukprakarn, 1997; Kalpna, Hajam, & Kumar, 2022; Ngegba, Cui, Khalid, & Zhong, 2022). Due to prevalent use of synthetic chemical insecticides, insects develop resistance, complicating pest control (Whalon, Mota-Sanchez, & Hollingworth, 2008). It has been revealed that several pests of stored product can develop resistance to synthetic pyrethroid and organophosphorus insecticides (Attia et al., 2020; Collins, Lambkin, Bridgeman, & Pulvirenti, 1993; Lee & Lees, 2001; Subramanyam, Harein, & Cutkomp, 1989; Zettler & Cuperus, 1990). Especially after the methyl bromide ban, phosphine gas has been widely utilized as a fumigant against stored product pests. In addition to mistakes made during application of this gas, the employment of phosphine gas as a single control method for several years led to the development of resistance by stored product pests. The coleopter pests *Tribolium castaneum* (Herbst, 1797) (Coleoptera: Tenebrionidae) (Opit, Phillips, Aikins, & Hasan, 2012; Pimentel, Faroni, Tótola, & Guedes, 2007; Pimentel, Faroni, Batista, & Silva, 2008; Wakil, Kavallieratos, Usman, Gulzar, & El-Shafie, 2021), *Sitophilus granarius* (Linnaeus, 1758) (Wakil et al., 2021), *Sitophilus oryzae* (Linnaeus, 1763) (Holloway, Falk, Emery, Collins, & Nayak, 2016), *Sitophilus zeamais* Motschulsky, 1855 (Coleoptera: Curculionidae) (Pimentel et al., 2008), *Oryzaephilus surinamensis* (Linnaeus, 1758) (Coleoptera: Silvanidae) (Pimentel et al., 2007), *Trogoderma granarium* Everts, 1898 (Coleoptera: Dermestidae) (Wakil et al., 2021), *Lasioderma serricorne* (Fabricius, 1792) (Coleoptera: Anobiidae) (Baliota, Athanassiou, & Cohnstaedt, 2021; Sağlam, Edde, & Phillips, 2015) have been stated to have evolved resistance to phosphine gas. Similarly, other studies reported that *R. dominica* has also developed resistant to phosphine gas (Afful, Elliott, Nayak, & Phillips, 2018; Opit et al., 2012; Pimentel et al., 2007; Pimentel et al., 2008; Wakil et al., 2021).

Due to the increasing difficulties in pesticide resistance management, alternatives are required to control the pests in storage of new and environment friendly products (Duke et al., 2003). The share of herbal extracts and essential oils (EOs) is significant among alternative control methods and these products have a potential in the protection of agricultural crops. There is estimated to be 17 500 aromatic plant species in nature (Bruneton, 1995), and 3 000 of these species are known to include EOs, and over 300 plant EOs are commercially available in medicine, cosmetics and perfumery industries

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(Franzios et al., 1997), in addition to the pesticide industry (Bakkali, Averbeck, Averbeck, & Idaomar, 2008). EOs exhibit fumigant effects and contact toxicity (Abdelgaleil, Mohamed, Badawy, & El-arami, 2009; Alkan, 2020; Baş & Ersoy, 2020; Göktürk, Kordali, Ak, Kesdek, & Bozhüyük, 2020; Sahaf, Moharramipour, & Meshkatalsadat, 2008), repellency (Alkan, 2020; Alkan et al., 2021; Nerio, Olivero-Verbel, & Stashenko, 2009; Wang, Zhu, Zhou, Niu, & Lei, 2006), and can inhibit feeding (Huang, Tan, Kini, & Ho, 1997) growth (Tomova, Waterhouse, & Doberski, 2005; Waliwitiya, Kennedy, & Lowenberger, 2009) and sublethal effects (such as decreasing digestive enzyme activities; reduction in protein, lipid, and carbohydrate level; low consumption rate or etc. (Ebadollahi, Naseri, Abedi, & Setzer, 2022; Ebadollahi, Naseri, Abedi, Setzer, & Changbunjong, 2022; Ebadollahi, Ziaee, & Palla, 2020) when used in the control of insect pests.

The plants from which these essential oils come are mostly in the families Lamiaceae, Lauraceae, Asteraceae, Cupressaceae, Myrtaceae, Rutaceae, Piperaceae and Poaceae (Campolo, Giunti, Russo, Palmeri, & Zappalà, 2018). The relative constituent of essential oils varies considerably and depends on geographical location, climatic conditions and various other factors. There are many studies related to the essential oil's chemical composition. Previous studies reported 1,8-cineole 46.5 % (Baratta, Dorman, Deans, Biondi, & Ruberto, 1998) and 41.9 % (Simić et al., 2004) as the main constituents of *L. nobilis*. However, the same compound was reported as 24.55 % in the plant cultivated in Tunisia (Mediouni Ben Jemâa, Tersim, Toudert, & Khouja, 2012). Although menthol was the main compound (37.3%) in *M. piperita* collected from Amazonia, Brasil (Miura et al., 2021), it was not high (24.71%) as much as from Ardabil province, moreover main constituents was limonene (27.28%) (Ebadollahi, Davari, Razmjou, & Naseri, 2017). It is therefore assumed that the insecticidal effect of the essential oils of plants that grow in different regions could be significant.

Several studies have reported effects of herbal EOs on stored product pests. However, studies on fumigants for *R. dominica* did not target larval or pupal stages of the pest. Thus, the present study aimed to assess the fumigant toxicity of EOs extracted from three plants for controlling of *R. dominica* larvae, pupa and adults, given this species is a significant stored product pest.

MATERIALS AND METHODS

The experiments were conducted in the 2021 in the Plant Protection Department, Agricultural Faculty, Malatya Turgut Özal University, Türkiye.

Plant essential oils extraction

Lavandula x intermedia Emerica ex Loisel, 1828 (Lamiales: Lamiaceae) collected from Muratpaşa district of Antalya province and *Laurus nobilis* L., 1753 (Lurales: Lauraceae) collected from Anamur district of Mersin province were extracted from the Bati Akdeniz Agricultural Research Institute. *Mentha piperita* L., 1753 (Lamiales: Lamiaceae) collected from Ovacik district of Tunceli province was extracted from

Malatya Turgut Özal University, Faculty of Agriculture. Both EOs were extracted by steam distillation for 3 h by Clevenger apparatus. The EOs were kept in tightly closed glass tubes at 4 °C until used.

Essential oils content

Essential oil analyzes were conducted at the Batı Akdeniz Agricultural Research Institute. *Lavandula x intermedia*, *L. nobilis* and *M. piperita* EOs (diluted by 1:100 hexane) were characterized by GC/GC-MS (gas chromatography (Agilent 7890A) and mass detector (Agilent 5975C)) with a capillary column (HP Innowax Capillary; 60.0 m x 0.25 mm x 0.25 µm). Helium gas was used as a carrier at a constant flow rate of 0.8 ml/min, and 1 µl samples were applied by injection with a split ratio of 40:1. The injector temperature was maintained at 250°C, and the column temperature was set to 60°C (10 min), increased from 60 to 220°C at a rate of 4°C/min and set again to 220°C (10 min). The total analysis duration was 60 min. The scanning range (m/z) was 35-450 atomic mass units and 70 electron volt (eV) electron bombardment ionization was used. The essential oil components were identified based on the data provided by Wiley and Adams oil libraries by comparison of their RT. The component rates were determined with the FID detector and the components were identified with the MS detector (Topuz et al., 2016; Gölükcü, Toker, Tokgöz, & Çınar, 2016; Syraiyl, Ydyrys, Ahmet, Aitbekov, & Imanaliyeva, 2022).

Insect rearing

Rhyzopertha dominica adults were procured from Kahramanmaraş Sütçü İmam University, Faculty of Agriculture, Plant Protection Department Entomology Laboratory. The adults were placed in 200 g wheat flour for 2-3 days to oviposit eggs, and then separated from the flour using a 500 µm sieve. To separate the eggs, the flour was sieved through a 212 µm sieve. The eggs were incubated in 500 g bread wheat grain in 1 L plastic jars with a gauze lid. Rearing was conducted in a dark controlled-climate chamber at 27 ± 1°C and 50 ± 10% RH (Cinco-Moroyoqui, Rosas-Burgos, Borboa-Flores, & Cortez-Rocha, 2006; Chanbang, Arthur, Wilde, & Throne, 2007).

Fumigant tests

EOs were tested at 1, 5, 20, 50, and 200 µl/L. An insecticide, dimethoate (50 µl/L) (Poligor EC) (Hektaş Ticaret Türk A.Ş.) was used as the comparator, since there is no available effective fumigant that could be practically applied in small volumes. Tests with no treatment were used as a negative control. The tests were conducted in a 135 ml autoclave bottle with a screw cap containing 10 g wheat grain. Filter paper was stuck with silicon adhesive to the inside of the caps, and the filter paper was impregnated with pure EOs with a micropipette.

Fumigant tests with *Rhyzopertha dominica* larvae

Fumigant tests were conducted with third instar larvae of *R. dominica*. To obtain third instar larvae, the eggs (without counting) obtained from 8,000-10,000 pests for 3 days were placed in plastic jars containing 500 g wheat grain. Edde (2012) reported that at

28°C, the egg stage lasted 7 days, first immature stage 9 days, second immature stage 6 days and third immature stage 5 days. Twenty-five to 27 days after the eggs were cultured, 25 g wheat grain, assumed to include third stage larvae, was transferred into autoclave bottles and 1, 5, 20, 50 or 200 µl/L essential oil was applied to filter paper in the autoclave bottle caps. *Rhyzopertha dominica* larvae were exposed to the EOs for 48 h at 27°C. The bottle lids were then removed, and the wheat was ventilated, and the contents were transferred into other plastic containers (with a tulle lid) and incubated at 27°C until adult emergence. The pest adults that emerged 5 weeks after the application were separated from the wheat grain with a sieve and counted. The adults that remained in the grain were collected after a further week. The trial was set up in a randomized plot design with five replicates.

Fumigant tests with *Rhyzopertha dominica* pupae

To determine the fumigant effects on *R. dominica* pupae, eggs were placed in plastic jars containing 500 g wheat grain. Forty to 42 days after the addition of eggs, 25 g wheat grain, assumed to contain pest pupae, was transferred to autoclave bottles. One, 5, 20, 50 or 200 µl/L of essential oil was applied the filter paper in the bottle caps and *R. dominica* pupae were exposed for 48 h at 27°C. The wheat was then aerated, transferred to plastic containers with gauze lids and incubated at 27°C until adult emergence. Adults that emerged 3 weeks after treatment were separated from the wheat grain with a sieve and counted. The adults that remained in the grain were collected after a further week. The assessment was set up in a randomized plot design with five replicates.

Fumigant tests with *Rhyzopertha dominica* adults

Twenty mixed-sex *R. dominica* adults (1 to 10 days old) were placed in autoclave bottles containing 10 g wheat grain. Essential oil was applied the filter paper in the bottle caps and the bottles were tightly closed. The adults were exposed to EOs for 24 and 48 h at 27°C. The viability of the adults was then determined. The trial was set up in a randomized plot design with five replicates.

Data analysis

The adult mortality rate was corrected with the Abbott (1925) formula based on the counts conducted 5-6 weeks after the essential oil application in the larval test and 3-4 weeks after the application in the pupal test. The data on the effect of fumigation with essential oil on adult pests were also corrected using Abbott (1925) formula. Percentage mortality data then converted to an arcsine transformation. They were subjected to a one-way analysis (ANOVA). The differences between the mean values were determined using the Tukey test at the 5% significance level. The analysis of variance and the multiple comparison tests were performed using the SPSS program.

Dose response modeling was performed with *drc* function in the R package “drc” (Ritz, Baty, Streibig, & Gerhard, 2015; Ritz & Streibig, 2005) in R version 4.2.0. The fumigant essential oil concentration in the treatments were log transformed using

Equation (1). Then, essential oil concentration causing 50% *R. dominica* mortality (LC_{50}) in the treatments were obtained using the single three-parameter log-logistic regression model LL.3 (Equation 2) for each essential oil (Arena, Merlo, Defagó, & Zygadlo, 2020; Taquet et al., 2020).

$$x = \text{Log}10(\text{Fumigant concentration} + 0.1) \quad (1)$$

$$U = \frac{d}{1 + \left(\frac{x}{EC_{50}}\right)^b} \quad (2)$$

where, b is the slope, d the upper-limit determined by the fitted concentration-response model with lower limit fixed at 0 (3 parameters), and U the response.

RESULTS

Compositions of the essential oils employed in the trials

The chemical composition of the EOs examined is presented in Tables 1. Twenty-six components were determined in the *L. intermedia* essential oil with the main components were linalool (43.3%), linalyl acetate (19.8%), camphor (6.7%), and 1,8-cineole (4.4%) (Table 1). Nineteen components were characterized for the *L. nobilis* essential oil with the main constituents were 1,8-cineole (57.7%), alpha-terpinyl acetate (9.92%), and alpha-pinene (6.4%) (Table 1). Twenty-one components were detected in the *M. piperita* essential oil with the main constituents were menthone (53.5%), menthol (17.2%) and menthofuran (5.4%) (Table 1).

Table 1. Chemical composition of *Lavandula x intermedia*, *Laurus nobilis* and *Mentha piperita* essential oils

Compound	percentage (%)	Compound	percentage (%)
<i>Lavandula x intermedia</i>			
α -Bisabolol	0.84	Lavandulyl acetate	1.91
Borneol	2.38	Limonene	1.08
β -Caryophyllene	0.63	Linalool	43.33
Camphene	0.32	Linalyl acetate	19.82
Camphor	6.66	β -Myrcene	1.44
1,8-Cineole	4.38	Nerol	0.89
β -Citronellol	0.59	Neryl acetate	0.98
Cryptone	0.42	<i>cis</i> -Ocimene	1.76
Geraniol	2.18	<i>trans</i> -Ocimene	2.03
Geranyl acetate	1.79	3-Octanone	0.92
Germacrene-D	0.25	Octen-1-ol, acetate	0.34
1-Hexyl acetate	0.54	α -Terpineol	3.73
Hexyl butyrate	0.39	α -Terpinolene	0.40
<i>Laurus nobilis</i>			
Camphene	0.33	beta-Pinene	4.56
1,8-Cineole	57.67	Sabinene	4.47
Eugenol	0.33	<i>gamma</i> -Terpinene	2.10
<i>para</i> -Cymene	2.19	Terpinen-4-ol	4.67
Linalool	0.88	Terpinolene	0.58

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Table continued

Compound	percentage (%)	Compound	percentage (%)
Limonene	2.24	<i>alpha</i> -Terpineol	0.38
Methyl eugenol	0.54	<i>alpha</i> -Terpinyl acetate	9.92
Myrcene	0.48	<i>delta</i> -Terpinyl acetate	0.37
<i>alpha</i> -Phellandrene	1.19	<i>alpha</i> -Thujene	0.70
<i>alpha</i> -Pinene	6.40		
<i>Mentha piperita</i>			
β -Caryophyllene	0.89	β -Pinene	1.07
1,8-Cineole	4.61	Sabinene	0.59
Germacrene	0.80	neoisomenthyl acetate	0.28
Isomenthol	5.22	β -Ocimene	0.33
Limonene	1.30	3-Octanol	0.41
Linalool	0.30	α -Pinene	0.75
Menthol	17.21	Piperitone	0.89
Menthofuran	5.43	Pulegone	3.99
Menthone	53.51	<i>cis</i> -Sabinene hydrate	0.35
Menthyl acetate	1.10	Terpinen-4-ol	0.21
β -Myrcene	0.76		

The fumigant effects of the essential oils on the *Rhyzopertha dominica*

The mortality effect of the essential oils on different stages of *R. dominica* is shown in Fig. 1. The toxicity of the fumigant increased progressively with increasing concentration of the essential oils in the larval, pupal and adult stages. *Laurus nobilis* essential oils applied to the larvae of *R. dominica* had a very strong fumigant activity (Fig. 1a). At a dosage of 200 μ l/L, larval mortality reached 100% and was statistically different from the other plant oils ($F_{15,79}=65.717$, $P=0.000$). At the same dose, the larval mortality of *M. piperita* was 59.04%.

The larvae and adults of *R. dominica* were more susceptible than the pupal stages of the pest. None of the applied concentrations or the standard toxic insecticide dimethoate caused 100% mortality. However, the highest mortality (%49) was achieved by the highest dose of *L. nobilis* and was significantly different from all *L. intermedia* doses ($F_{15,79}=4.298$, $P=0.000$) (Fig. 1b).

Compared to *L. intermedia*, *M. piperita* and *L. nobilis* caused higher mortality in adult *R. dominica* at each dose (Fig. 1c-d). At a dose of 200 μ l/L, both *M. piperita* and *L. nobilis* caused the highest adult mortality of *R. dominica*, while *L. intermedia* caused significantly low (54%) adult mortality at 24-h exposure ($F_{15,79}=44.369$ $P=0.000$). It was similar in the 48-hour exposure experiment. At a dose of 50 μ l/L, the highest adult mortality was achieved by *L. nobilis* (72%), followed by *M. piperita* (38%) and *L. intermedia* (15%) ($F_{15,79}=48.404$, $P=0.000$).

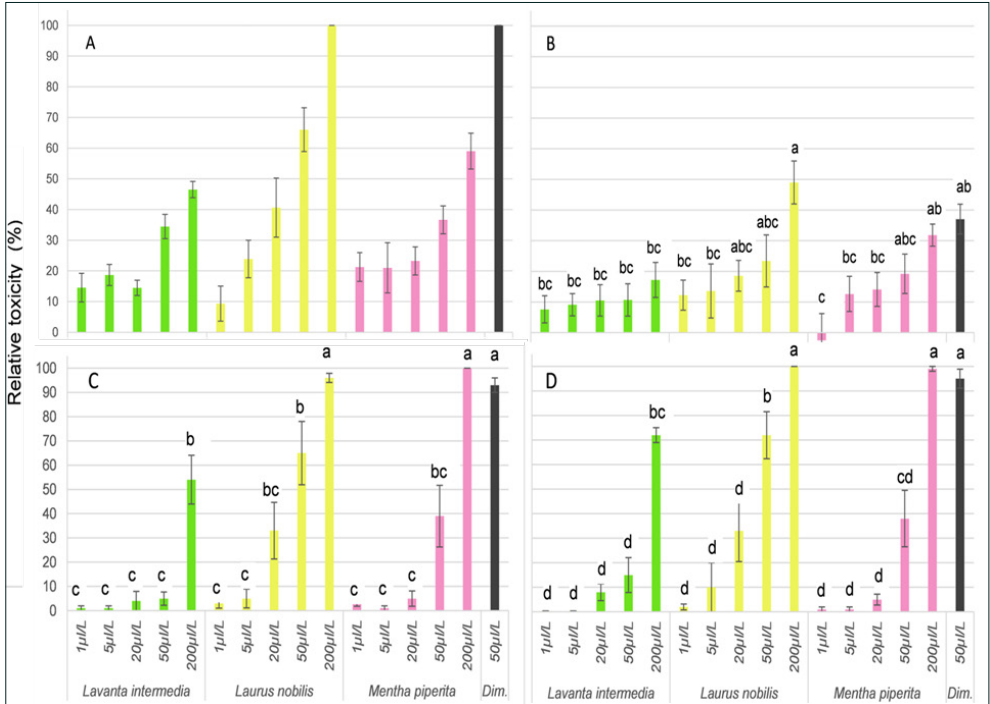


Figure 1. Fumigant effect of essential oils on the mortality of different *Rhyzopertha dominica* stages; A) larva, B) pupa, C) adult (24h exposure), D) adult (48h exposure) (different lower-case letters show statistical differences according to Tukey test at $P \leq 0.05$; bars represent standard errors; Dim.: Dimethoate).

The three-parameter log-logistic models, as presented in Equation 2, fitted to the data are as shown in Fig. 2. This demonstrated that the essential oil concentrations had different fumigant effects on *R. dominica* larvae. The mortality (determined from the number of adults obtained) strongly increased with the increased *L. nobilis* oil concentration but somewhat less with the other two EOs. Differences between the efficacy of *L. nobilis* and other EOs were evident from the 50 µl/L. LC_{50} estimates were 2.1, 7.0 and 7.4 (\log_{10} µl/L + 0.1) for *L. nobilis*, *L. intermedia* and *M. piperita*, respectively (Table 2). The back-transformed *L. nobilis* LC_{50} was approximately 128 µl/L and 5 orders of magnitude more effective than the other essential oil. The fumigant tests on larvae gave the highest mortality (100%) at 200 µl/L *L. nobilis* essential oil (Fig. 2a).

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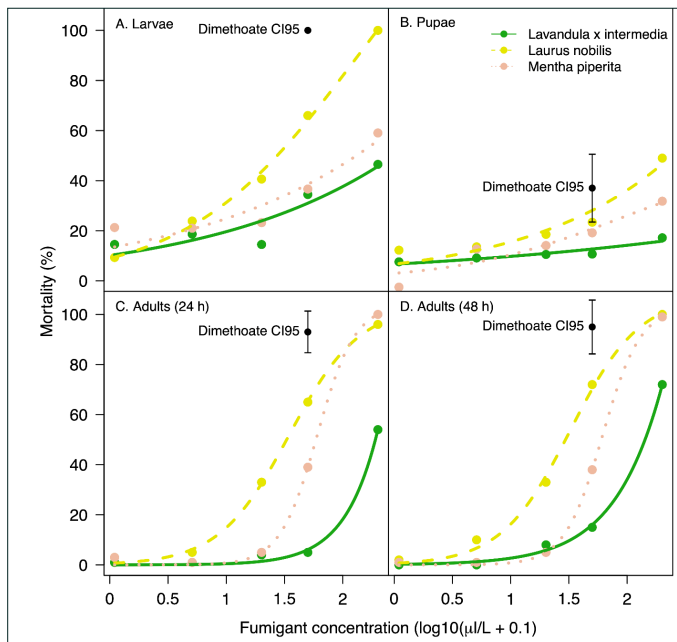


Figure 2. Essential oil dose response on mortality of *Rhizopertha dominica* (data were fitted to a three-parameter log-logistic model). The mean mortality and 95% confidence interval for dimethoate is plotted at the concentration applied as a comparator.

Table 2. Dose–response parameters of the log-logistic model for essential oils on *Rhizopertha dominica*.

Treatments	Upper limit (d)	Curve slope (b)	LC ₅₀
Larval stage			
<i>Lavandula x intermedia</i>	1072.103 (5419.319)*	-0.669 (0.185)	6.956 (8.572)
<i>Laurus nobilis</i>	176.954 (88.372)	-1.389 (0.423)	2.108 (0.738)
<i>Mentha piperita</i>	1490.234 (5333.455)	-0.640 (0.142)	7.365 (6.275)
Pupal stage			
<i>Lavandula x intermedia</i>	151.611 (1154.299)	-0.405 (0.456)	7.599 (24.709)
<i>Laurus nobilis</i>	2944.843 (24036.059)	-0.852 (0.228)	7.150 (10.119)
<i>Mentha piperita</i>	49.836 (62.979)	-1.422 (1.199)	1.935 (1.900)
Adult stage (24 h exposure)			
<i>Lavandula x intermedia</i>	1761.8 (18497.0)	-3.672 (1.763)	3.242 (3.112)
<i>Laurus nobilis</i>	103.60 (11.999)	-3.315 (0.965)	1.539 (0.108)
<i>Mentha piperita</i>	104.00 (11.600)	-6.203 (3.466)	1.782 (0.0899)
Adult stage (48 h exposure)			
<i>Lavandula x intermedia</i>	1423.9 (15582.0)	-2.563 (0.863)	3.446 (4.804)
<i>Laurus nobilis</i>	107.63 (10.810)	-3.367 (0.965)	1.513 (0.909)
<i>Mentha piperita</i>	103.32 (11.625)	-6.108 (3.282)	1.788 (0.092)

* Standard errors are in parentheses.

Dose-response parameters for *R. dominica* pupae after 48-h exposure to the EOs in Table 2. In contrast to the results for the larvae, the mortality caused by all EOs was low and adult emergence was not strongly suppressed, even at the highest concentrations. Pupal mortality was the lowest with *L. intermedia* essential oil. The

pupal mortality (based on the number of emerged adults) increases gradually with increasing concentration of *M. piperita* and *L. nobilis* oils. However, these EOs on *R. dominica* pupae gave well under 50% mortality. Only *L. nobilis* oil gave mortality close to 50% at the highest concentration, a result similar to dimethoate (Fig. 2b).

Dose-response parameters for *R. dominica* adults after 24-h exposure to different essential oil doses are shown in Table 2. LC_{50} estimates were 1.5, 1.8 and 3.2 ($\log_{10} \mu\text{L} + 0.1$) for *L. nobilis*, *M. piperita* and *L. intermedia*, respectively. Back-transformed estimates for *L. nobilis* and *M. piperita* estimates were similar at 35 and 60 $\mu\text{L/L}$, respectively. Both EOs there were more efficacious than that of *L. intermedia*. The mortality at two lowest concentration (1 and 5 $\mu\text{L/L}$) was negligible. However, the differences between the three oils changed with the concentration. The oil from *L. nobilis* was clearly better at 50 $\mu\text{L/L}$, but by 200 $\mu\text{L/L}$ *M. piperita* oil was equally effective.

The comparison of the mortality rates after 24 and 48 h of exposure to plant EOs revealed similar curves based on the exposure time. The three model parameters were similar between the two assessment times (Table 2) and consequently the fitted curves were quite similar (Fig. 2c, d). The extra 24 h of exposure provided minimal additional benefit.

DISCUSSION

The insecticidal (fumigant) effects measured for the EOs from studies plants (mint, lavender, laurel) indicated they have potential for control of *R. dominica*. They had significant effects on the mortality of *R. dominica* at various developmental stages (larva, pupa and adult). The fumigant activity tests on larvae gave complete mortality with 200 $\mu\text{L/L}$ *L. nobilis* oil. Similar results were obtained with adults. Complete mortality was obtained at high concentrations of *L. nobilis* and *M. piperita* oils. However, the effects of the EOs were generally quite low on the pupal stage. The fumigant effect tests on pupae had the highest effect with *L. nobilis* oil (49% mortality), followed by *M. piperita* and *L. intermedia* EOs. It is reported that the efficacy of fumigants can be low for pupae, as pupal respiration rate is especially low in insects found in grain kernels (Hagstrum, Phillips, & Cuperus, 2012). Similarly, the efficacies of various EOs and chemicals have been investigated in *Callosobruchus maculatus* (Fabricius, 1775) (Coleoptera: Chrysomelidae) (Güdek & Çetin, 2016), *Acanthoscelides obtectus* Say, 1831 (Coleoptera: Bruchidae) (Papachristos & Stamopoulos, 2002), *Ephestia cautella* (Walker, 1863) (Lepidoptera: Pyralidae), *O. surinamensis* and *T. castaneum* (Isikber et al., 2004) control, and pupae were generally more resistant. In the present study, similarly, the essential oil activities in the larval and adult stages were higher than the pupal stages at all concentrations.

Across all the developmental stages, *L. nobilis* oil gave the highest activity and *L. intermedia* the lowest.

The mortality from 50 $\mu\text{L/L}$ *L. intermedia* oil was 15% in adults stage 48 h whereas it was 72% at the highest dose (200 $\mu\text{L/L}$). Similar to these findings, Eli Shaaya et al. (1991) reported that 15 $\mu\text{L/L}$ lavender essential oil had fumigant effects on *R. dominica*

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adults with complete mortality. Rozman, Kalinovic, & Korunic (2007) stated that the fumigant effect of lavender essential oil was high on *R. dominica*, and the linalool and camphor in the lavender essential oil led to complete mortality within 24 h at the lowest concentration (0.1 µl/720 ml). In another study, Ebadollahi, Safaralizadeh, & Pourmirza (2010) reported that the fumigant effect of *Lavandula stoechas* L., 1753 (Lamiales: Lamiaceae) essential oil on *R. dominica* after 24 and 48 h with LC₉₅ being 80.6 and 28.9 µl/L, respectively. Main constituents reported were 1,8-cineole (7.02%), γ-cadinene (5.33%), t-cadinol (5.07%), p-mentha-1-en-8-ol (5.02%) and caryophyllene (5.01). In the present study, however, *L. intermedia* essential oil containing menthone (53.5%), menthol (17.2%) and menthofuran (5.4%) did not produce complete mortality of *R. dominica* adults. It is concluded that this lower effect might be due to the variations in the chemical compositions of the essential oil even though coming from plants in the same genera.

Mentha piperita essential oil LC₅₀ was almost 60 µl/L in adults after 24 h of, and the mortality at 200 µl/L was complete. Similar to these findings, Tripathi, Veena, Aggarwal, & Sushil (2000) reported that *M. piperita* essential was lethal to *T. castaneum* and *C. maculatus*. Mackled, El-Hefny, Bin-Jumah, Wahba, & Allam (2019) demonstrated that 70 µl/L *M. piperita* essential oil, the main components of which included menthol (33%) and menthone (20%), had a fumigant effect on *R. dominica* after 72 h giving complete mortality. In the present study, it was found that the fumigant effect of 200 µl/L *M. piperita* oil was 100% in the shorter time (24 h). In another study, Çam, Karakoç, Gökçe, Telci, & Demirtaş, (2012) reported that 10 µl/L *M. piperita* essential oil had fumigant effects on *S. granarius* with mortality of 58% after 24 h. Khani et al. (2017) reported 95% mortality in *S. oryzae* adults after exposure to 299 µl/L *M. piperita* essential oil. In the present study, 200 µl/L *M. piperita* essential oil gave complete mortality in *R. dominica* adults. Although the pest species tested were different, the effects were similar. Akkuş, Gözüaçık, & Gültekin (2021) investigated the fumigant toxicity of *Mentha longifolia* L., 1759 (Lamiales: Lamiaceae) essential oil on *R. dominica* adults with 10 µl/petri dish giving complete mortality after 24 h. It is suggested that the lower response seen in the current study was due to the variations between the chemical compositions of the plant essential oil.

In the present study, the mortality caused by 35 µl/L *L. nobilis* essential oil was about 50% in adults but at 200 µl/L was mortality was complete in both larval and adult stages of *R. dominica*. Similarly, Mediouni Ben Jemâa et al. (2012) reported that the essential oil of *L. nobilis* from Morocco, Algeria, and Tunisia was lethal to *R. dominica* with LC₅₀ of 67.9, 99.0 and 113 µl/L, respectively. Major components also reported as 1,8 cineole 38.86, 34.62 and 24.55% and Linalool 9.45, 12.57 and 17.67% for the essential oils obtained in these countries. Kırpık, Kılıçlı, & Yıldız Asker (2019) reported that 100 µl/L *L. nobilis* essential oil gave complete mortality of *R. dominica* after 24 h. In the present study, the mortality with *L. nobilis* essential oil with high 1,8-cineole content (58%) was 96% at the highest concentration (200 µl/L) after 24 h. It is suggested that the different chemical compositions of plant EOs are likely to lead to different findings.

Data from current experiments indicate that all the EOs of all three plants have fumigant effects on *R. dominica* at promising levels, especially the oils of *L. nobilis* and *M. piperita*. The EOs of both plants could be employed as a substitute instrument to chemical insecticides in pest control.

Despite the high effects obtained with the administration of EOs in larval and adult stages, the same effect was not observed on the pupal stage. Thus, the developmental stage of the pest should not be ignored in the recommendations about treating infested products.

Finally, further studies should be conducted on the developmental stages inside the kernel, i.e. the larval and pupal stages. It is recommended that future studies also be conducted to test the penetration of the essential oils into large quantities of the product and to prepare new formulations to improve insecticidal duration and efficacy.

ACKNOWLEDGEMENTS

This study is a part of the master degree thesis of the first author and was sponsored by the Scientific Research Fund of Malatya Turgut Özal University (Grant Project No: BAP-YL3). We thank Batı Akdeniz Agricultural Research Institute (Turkey) for supplying the *L. intermedia* and *L. nobilis* essential oils and analyzing all oils. We are also grateful to Prof. Dr. Ian T. Riley for his assistance during the statistical analysis and manuscript preparation.

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