

Full Length Research Paper

Evaluation of insecticidal activities of *Mentha piperita* and *Lavandula angustifolia* essential oils against house fly, *Musca domestica* L. (Diptera: Muscidae)

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The essential oils of peppermint, *Mentha piperita* and lavender, *Lavandula angustifolia* were tested for their larvicidal and pupicidal activities against the house fly, *Musca domestica* L. (Diptera: Muscidae). The effects of the two lethal concentrations LC50 and LC75 on the larval duration, pupation percent, pupal duration and adult emergence were also determined. In addition, the induced malformation at larval and pupal stages were recorded and photographed. The results about *M. piperita* showed higher toxicity against *M. domestica* larvae than *L. angustifolia*. The LC50 and LC75 values were 2.5% (225 ppm) and 3% (270 ppm), respectively, for *M. piperita* and 3% (264 ppm) and 4% (352 ppm) respectively, for *L. angustifolia*. Moreover, a significant prolongation in both larval and pupal duration, reduction in pupation and adult emergency percent in addition to various morphological abnormalities of larvae and pupae were detected post treatment of the third larval instar with LC50 and LC75 of *M. piperita* and *L. angustifolia*. The present results revealed that the essential oils of peppermint and lavender have a control potential against *M. domestica* and should be further explored as a component of integrated vector management programs.

Key words: *Musca domestica*, *Mentha piperita*, *Lavandula angustifolia*, larvicidal pupicidal.

INTRODUCTION

The house fly, *Musca domestica* L. is considered as one of the most important pests which cause health problems in the environment as it accompanies humans during their daily activity everywhere, both indoors and outdoors. It is recognized as a public health pest causing a serious threat to human and livestock by vectoring several pathogenic organisms such as protozoa cysts, helminth parasites, enteropathogenic bacteria, and enterovirus (Emerson et al., 1999; Douglass and Jesse, 2002; Mian et al., 2002; Barin et al., 2010). Chemical control method commonly used against this pest produce the risk of developing pest resistance and bioaccumulation. Bio-insecticides, especially those derived from plant origin are recently considered eco-friendly alternatives to conventional synthetic pesticides (Scott et al., 2000;

Shono and Scott, 2003; Srinivasan et al., 2008). Essential oils are odorous components and secondary metabolites that can be extracted from plant tissues through steam distillation. Essential oils and their volatile constituents are widely used in the prevention and treatment of human illnesses. They are also documented for exhibition of acute toxicity, anti-feeding and oviposition deterrents against a wide variety of insect-pests. The risk lower level of the volatile essential oils to the environment and their minimal residual activity against predator, parasitoid and pollinator insect populations, making essential-oil-based pesticides compatible with integrated pest management programs (Regnault-Roger, 1997; Isman, 2006; Koul et al., 2008). Bio-efficacy of the essential oils of *Mentha piperita*, *Zingiber officinalis*, *Emblica officinalis*,

Cinnamomum verum, *Eucalyptus globulus*, *Cymbopogon citratus*, *Pogostemon cablin*, *Vetiver zizanoides* and *Curcuma longa* were evaluated for their larvicidal activity against *Spodoptera littoralis*, *M. domestica*, *Leptinotarsa decemlineata*, and *Bovicola ocellatus* by Rice and Coats (1994), Isman (2000), Pavela (2005 and 2006), Sajfrtova et al. (2009), Kumar et al. (2011) and Talbert and Wall (2012).

The present work aimed to evaluate the toxicity effect of two essential oils of *M. piperita* and *L. angustifolia* against the house fly larvae for the possibility of using these oils as larvicides for controlling the insects by treating insect breeding places. The study designed to determine the effects of the two lethal concentrations LC50 and LC75 on the larval duration, pupation percent and adult emergence.

MATERIALS AND METHODS

Collection and maintenance of flies

Adults of *M. domestica* were collected from the garbage site of the Abu Arish area (Eastern Jazan), southern Saudi Arabi, ($16^{\circ}58'N$, $42^{\circ}47'E$), by using a sweeping net and transported into a small cage to the Department of Biology, Faculty of Science, Jazan University, for identification and reared in laboratory for four generations before experiment. Adult flies were maintained in cages (30x 30 x 30 cm) and provided with granulated sugar, petri dishes containing cotton pads soaked in milk powder dissolved in water (10% w/v) and jars (500 mL) containing larval media for egg laying. Larval media consisted of yeast, dry milk powder, wheat bran and water according to the method described by Pavela (2006). The jars were removed from cages after 2-3 days when eggs were visible and were provided with wood dust for pupation and kept in separate cages for fly emergence.

Essential oils

The essential oils of peppermint, *M. piperita*, and lavender, *L. angustifolia* were purchased from Sigma Company. *M. piperita* L. was of 98% purity, 0.90 g/ml density and $n_{20}/D_1.46$ refractive index, while, *L. angustifolia* L. was of 98% purity, 0.88 g/ml density and $n_{20}/D_1.46$ refractive index. Four concentrations (4, 3, 2.5 and 1%) representing (360, 270, 225 and 90 ppm) for *M. piperita* L. essential oil and (352, 264, 220 and 88 ppm) for *L. angustifolia* essential oil, respectively. The two essential oils were prepared using acetone and stored in glass bottles at 4°C until they were used.

Biological studies

The second instar larvae used in this experiment were 3-days-old after hatching from the same egg batch. Larval treatment carried out in petri dishes according to the method explained by Brady (1966), in which interior of each petri dish treated with 1 ml from each of the four aforementioned concentrations of the tested essential oils. Each experiment was conducted in four replicates (20 larvae/ replicate) along with the control group. After treatment, the larvae were transferred to the rearing jar and the mortality assessed by touching each larva with a paint brush (no. 0), and those not responding were considered dead. The LD50 and LD75 toxicity determined based on mortality data at 24 h assessments. Living

larvae were further examined daily to estimate the effect on the larval duration after treatment, percentage of pupation and the successfully emerged adults according to Sripong (2008) and Peydro et al. (1995). In addition, any morphological abnormalities recorded were photographed at all developmental stages.

Statistical analysis

The observed mortality was corrected by Abbott's formula (Abbott, 1987). Data analyses were performed using a one-way ANOVA (Least Significant Difference (LSD)) in SPSS version 20 and significant differences were determined at $P<0.05$.

RESULTS AND DISCUSSION

Assessment of *M. piperita* and *L. angustifolia* toxicity against *M. domestica* larvae revealed that the LC50 values were 2.5 and 3% for *M. piperita* and *L. angustifolia*, respectively. The LC75 values recorded for the two oils were 3 and 4%, respectively. The results exhibiting promising larvicidal activity of peppermint and lavender which are in line with Sajfrtova et al. (2009), Kumar et al. (2011) and Morey and Khandagle (2012) against *S. littoralis*, *M. domestica* and *L. decemlineata*. Kumar et al. (2011) used formulated *M. piperita* essential oil to perform on a par in housefly control with chemical larvicides, such as novaluron and benzoylureas. Seo and Park (2012) studied the larvicidal activity of medicinal plant extract from 27 plant species against *M. domestica*, including *Phryma leptostachya*, *Atractylodes japonica*, *Saussurea lappa*, *Asiasarum sieboldi*, and *Gleditsia japonica* and also Morey and Khandagle, (2012) recorded that the highest larvicidal activity LC50 (104 ppm) was shown by *M. piperita*. The insecticidal action of essential oils related to their active recorded natural pesticide ingredients which reported for *M. piperita* essential oil as menthol (40.7%) and menthone (23.4%) and further components were (+/-)-menthol acetate, 1,8-cineole, limonene, beta-pinene pulegone, and beta-caryophyllene (Palacios et al., 2009; Schmidt et al., 2009) and for *L. angustifolia* essential oil was Linalool, (27.3–42.2 %), linalyl acetate (27.2–46.6 %), (Z)- β -ocimene (0.2–11.6 %), terpinen-4-ol (0.70–4.6 %), lavandulyl acetate (0.50–4.8 %), β -caryophyllene (1.8– 5.1 %), (E)- β -ocimene (0.30–3.8 %), α -terpineol (0.30–2.0 %) and 1,8-cineole (0.10–1.2 %) (Behnam et al., 2006; Zheljazkov et al., 2013) supporting the insecticidal potential of the used essential oils as previously mentioned by Koul et al. (2008).

Larval duration

Results in Table 1 revealed that the duration of the control larvae of *M. domestica* was 4.46 ± 0.51 days; a significant prolongation was observed in the duration of larvae treated with LC50 and LC75 of the two examined oils, which were 5.61 ± 0.69 and 5.56 ± 0.73 days, respectively for *M. piperita* and 5.21 ± 0.63 and $5.55 \pm$

Table 1. Effect of the tested plant oils on the larval duration of *M. domestica* treated as 3rd larval instar at 29°C.

Treatment	Group	Larval duration (days ±SD)	% Change	F value
<i>Mentha piperita</i>	Control	4.46 ± 0.51 ^a		
	LC50	5.61 ± 0.69 ^b	25.784	
	LC75	5.56 ± 0.73 ^b	24.663	***
<i>Lavandula angustifolia</i>	Control	4.46 ± 0.51 ^a		
	LC50	5.21 ± 0.63 ^b	16.816	
	LC75	5.55 ± 0.52 ^b	24.439	

Data expressed as mean± SD, significance at p<0.05 is between different superscripts.

Table 2. Effect of the tested plant oils on the pupation percentage, and pupal duration (days) of *M. domestica* treated as 3rd larval instar at 29°C.

Treatment	Group	Pupation (%±SD)	F value	Pupal duration (days ±SD)	Percentage (%) change	F value
<i>Mentha piperita</i>	Control	95.00±5.77a		4.43 ± 0.50a		
	LC50	75.50±5.00b		4.94 ± 0.68b	11.512	
	LC75	32.50±9.57b	***	5.44 ± 0.53b	22.799	***
<i>Lavandula angustifolia</i>	Control	95.00±5.77a		4.43 ± 0.50a		
	LC50	62.50±9.57b		4.82± 0.53b	8.803	
	LC75	42.50±9.57b		5.00 ± 0.67b	12.866	

Data expressed as mean± SD, significance at p<0.05 is between different superscripts.

Table 3. Effect of the tested plant oils on the adult emergence percent *M. domestica* treated as 3rd larval instar at 29°C.

Treatment	Group	Adult emergence (%±SD)	F value
<i>Mentha piperita</i>	Control	95.00±5.77a	
	LC50	45.00±5.77b	
	LC75	27.50±5.00b	***
<i>Lavandula angustifolia</i>	Control	95.00±5.77a	
	LC50	57.50±9.57b	
	LC75	30.00±8.16b	

Data expressed as mean± SD, significance at p<0.05 is between different superscripts

0.52 days, for the same doses of *L. angustifolia*, respectively. The benefit of elongation is that housefly larvae numbers are reduced due to longer life cycle of houseflies. These results are in line with that of Mansour et al. (2011) who evaluated the toxicity against the larval stage of the *M. domestica* L. for the ethanolic plant extracts of *Piper nigrum*, *Azadirachta indica*, *Conyza aegyptiaca* and *Cichorium intybus* and The same findings were reported by Khater and Shalaby (2008) on *Culex pipiens* after treatment of the 4th larval instars with extracts of *Boswellia serrata* and *Trigonella foenum-grecum*.

Pupation percent and pupal duration:

Data in Table 2 showed a high significant reduction in pupation percent that decreased to 75.50 and 32.50% at LC50 and LC75 of *M. piperita*, comparable to 95.00% for control group. Also highly significant reductions in pupation percent (62.50 and 42.50%) were reported for LC50 and LC75 of *L. angustifolia*, respectively. Data in the same table revealed that the pupal duration of the control group was 4.43 ± 0.50 days which significantly prolonged in groups treated with LC50 and LC75 of the two tested oils and reached 5.44 ± 0.53 and 5.00 ± 0.67 days at LC75 *M. piperita* and *L. angustifolia*, respectively. The current results are in agreement with findings of Kumar et al. (2012) during their evaluation of insecticidal activity of *E. globulus* (Myrtaceae) against the house fly.

The adult emergence percentage

Adult emergence reduced from 95% for control group to 45 and 27.5% for groups treated with LC50 and LC75 of *M. piperita*, respectively, comparable to 57.5 and 30% for groups treated with LC50 and LC75 of *L. angustifolia*, respectively (Table 3). High reduction in *M. domestica* emergence were also reported by Abdel Halim and Morsy (2005) after using volatile oils of *C. macrocarpa* and *A. officinarum* against *Synthesiomyia nudiseta*. Also, Kumar

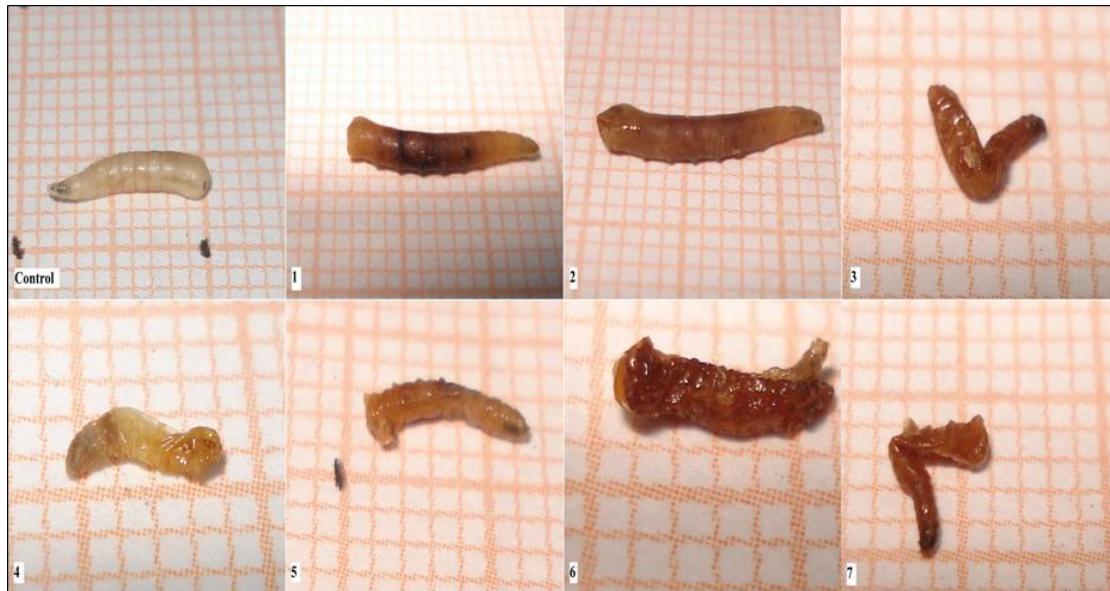


Plate 1. The control figure represented the normal larva and Figures 1-7 represented the malformations at larval stage.



Plate 2. The control figure represented the normal pupa and figures (1-11) represented the malformations at pupal stage.

et al. (2011) found that crude oils of *M. piperita* and *E. globulus* suppressed the emergence of adult flies by 100%.

Morphologic abnormalities

Plates 1 and 2 showed that distinct malformations of larvae and pupae of the house fly were induced after

treatment of the third larval instar with LC50 and LC75 of peppermint and lavender. Morphological larvae abnormalities (Plate 1: 1-7) include swelling of body, brown pigmentation, weakness in cuticle and irregular body shapes. Morphologic abnormalities of pupa (Plate 2: 1-11) include larviform puparium, larval-pupal interme-diates, irregular-shaped and shrinkage pupae. Com-parable with these results. Sexena et al. (1981) reported developmental

abnormalities in larvae of *Cnaphalocrocis medinalis* after treatment with 50% neem oil. Various morphological abnormalities on larvae, pupae, and adult stages induced by using essential oils against *Culex pipiens*, *Lucilia sericata* and *M. domestica* were detected by Khater and Shalaby (2008), Khater and Khater (2009) and Mansour et al. (2011), respectively. The abnormalities could be attributed to the metamorphosis inhibiting effect of the essential oils, as a result of the disturbance of hormonal control. Khater and Khater (2009) suggested that the larviform puparia could be caused by the failure of larvae to contract to the pupal form, as a result of muscle paralysis, but their ability to acquire melanization of the pupal cuticle is attributed to the continuation of the enzymatic process of tanning.

In conclusion, *M. piperita*, showed a higher toxicity effect against *M. domestica* larvae than *L. angustifolia*. Moreover, treatment of larvae with LC50 of *M. piperita* caused higher prolongation in the larval and pupal duration and higher reduction of adult emergence than in the case of *L. angustifolia*. On the other hand, reductions of pupation percent were higher in groups treated with LC50 of *L. angustifolia* than in groups treated with the same dose of *M. piperita*. It is imperative to explore new active natural products to introduce an effective method against houseflies and further studies concerning the application of these oils in control of house fly in field are recommended.

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