

Screening for Bioactive Plant Materials against House Fly, *Musca domestica* L. (Diptera: Muscidae)

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ABSTRACT

Ground parts of various plants viz *Cupressus sempervirens*, *Cupressus macrocarpa*, *Euphorbia pereskiiifolia*, *Pelargonium zonale*, *Plumeria rubra* (leaves), *Lantana camara* (leaves and flowers); *Cyperus rotundus* (whole plant); *Acacia nilotica* (seeds); *Simmondsia chinensis*, *Eucalyptus globulus*, *Amygdalus communis*, *Citrus maxima*, *C. sempervirens* (essential oils, EOs) were evaluated for their repellent potential, antifeedant activity and larvicidal activity towards *Musca domestica* larvae. In filter paper repellency tests, *A. communis* EO produced a repellent activity of class II, *P. rubra* and *C. rotundus* powders showed no repellent activity (class 0), and the remaining plant materials produced repellent activity of class I. In food preference tests, antifeedant activity, *P. zonale* and *E. globulus* were the most effective repellents while *C. macrocarpa* powder came in second followed by *C. rotundus* and *A. nilotica*. During preliminary screening, the tested plants showed high larvicidal effects. The most outstanding possible plant candidates for controlling *M. domestica* were *L. camara*, *P. zonale*, *A. nilotica*, *C. rotundus*, *C. macrocarpa* and *C. sempervirens* with LC₅₀s of 1.37, 1.24, 3.78, 3.24, 12.16 and 20.59 g/100 g, respectively. The LC₅₀s of *A. communis* and *S. chinensis* were 1.274 and 1.67 mL/100 g, respectively.

Keywords: bioassay, botanical pesticides, insect control, repellency, susceptibility test

Abbreviations: EO, essential oil; PR, percentage repellency; RH, relative humidity

INTRODUCTION

Botanical pesticides are known to have toxic effects against insect pests. Their use has increased with an increase in the awareness of health and environmental hazards of conventional pesticides. They are safe to use as they are natural products, and are easily disintegrated into harmless compounds in a relatively short period by physical and/or biological systems. Using botanical materials as alternatives or adjuvants to chemical insecticides is an ideal method for control. They have repellent, insecticidal, antifeedant and/or insect growth regulator effects (Hashem and Youssef 1991; Pavela 2008).

Musca domestica L. is a cosmopolitan pest in farm and home environments. The insect has followed man over the entire globe and constitutes a major problem in a variety of industries (Axtell and Arends 1990). Its potential for transmitting pathogens was also demonstrated. More than 100 pathogens are associated with the house fly as they transmit diseases to human and animal, including typhoid, cholera, bacillary dysentery, tuberculosis, anthrax and ophthalmia, as well as some parasitic worms (Akinboade *et al.* 1984; Iwasa *et al.* 1999). Pathogenic organisms are picked up by the fly from garbage, sewage and other sources of filth. They are carried by its mouthparts, through their vomiting, feces and/or on external body parts to food and beverage (Fotedar 2001; Zurek *et al.* 2001).

M. domestica showed different percentage repellency values when exposed to citral ((Z,E)-3,7-dimethyl-2,6-octadienal) and eugenol (4-allyl-2-methoxyphenol) (Vartak *et al.* 1994), pine essential oil (EO), known for containing myrcene, *p*-cymene, γ -terpinene, and (\pm)-linalool (Maganga *et al.* 1996). A similar effect was observed when treated with citrus EOs, of which δ -limonene, α -pinene, and myrcene are the principal components (Liao 1999), catnip, *Nepeta cataria* EO (Schultz *et al.* 2004, 2006), *Artemisia vulgaris* EO (Wang *et al.* 2005). Antifeedant activity against *M. domes-*

tica was detected in the extract from *Plectranthus coesta* (Sharma *et al.* 1992). Antonious and Gomaa (1988) reported that *M. domestica* flies were deterred by the bad taste of solutions containing neem EO, *Azadirachta indica*. Studies about the potential insecticidal actions of many plants on larvae and/or adults house fly, are many, among them are: *Libocedrus bidwillii* (Russell *et al.* 1976); *Ageratum conyzoides* (Gonzalez *et al.* 1991); *Derris urucu* and *Derris nicou* (Costa *et al.* 1997); *Matricaria chamomilla* (Shoukry 1997); *Lupinus termis* and *Calatropis procera* (Assar 2002); *Artemisia monosperma*, *Conyza dioscoridis*, *Eichhornia crassipes*, *Clerodendron inerme*, *Clocasia antiquorum*, *Zygophyllum coccineum* and *Farestia aegyptia* (Bakr *et al.* 2003); eucalyptol (Sukontason *et al.* 2004); *Trigonella foenum-graecum* (Abdel Halim and Morsy 2006) and *Illicium verum* (Sripongpun 2008).

A conventional method for fly control in the short term is the use of insecticides. Nevertheless, the widespread and massive applications of chemical insecticides frequently produce risks in the development of insect resistance and leaving residues harmful to human and its environment (Scott *et al.* 2000). Thus, the aim of this research was to find novel, effective, safe and cheap bio-insecticides which would feasibly be used to control house fly.

MATERIALS AND METHODS

Tested plants

The tested plants selected for this study are listed in Table 1. Fresh green leaves and flowers of *Lantana camara* and leaves of *Cupressus macrocarpa*, *Cupressus sempervirens*, *Euphorbia pereskiiifolia*, *Pelargonium zonale* and *Plumeria rubra* were collected in 2008 from the gardens of the Faculty of Education, Cairo, Egypt. Plants were identified using voucher specimens kept in the department Herbarium. Tested plants were washed with distilled water, air-dried at room temperature then ground using an electric mill.

Table 1 List of plants and plant materials tested in this study.

Scientific name	Family name	English name	Plant material
<i>Cupressus sempervirens</i> L.	Cupressaceae	Italian cypress	Leaf powder and EO
<i>Cupressus macrocarpa</i> Hartw. ex Gordon	Cupressaceae	Monterey cypress	Leaf powder
<i>Euphorbia pereskiifolia</i> Houllet ex Baill.	Euphorbiaceae	African milk bush	Leaf powder
<i>Pelargonium zonale</i> L'Her.	Geraniaceae	Geranium	Leaf powder
<i>Plumeria rubra</i> L.	Apocynaceae	Temple tree	Leaf powder
<i>Lantana camara</i> L.	Verbenaceae	Spanish flag	Leaf and flower powders
<i>Cyperus rotundus</i> L.	Cyperaceae	Nut grass	Whole plant powder
<i>Acacia nilotica</i> (L.) Wild. ex Delile	Fabaceae	Scented thorn	Seed powder
<i>Simmondsia chinensis</i> (Link) C. K. Schneid	Simmondsiaceae	Jojoba	EO
<i>Eucalyptus globulus</i>	Myrtaceae	Camphor	EO
<i>Citrus maxima</i> (Burm. ex Rumph.) Merr.	Rutaceae	Sweet orange	EO
<i>Amygdalus communis</i> L.	Rosaceae	Bitter almond	EO

Cyperus rotundus (whole plant), *Acacia nilotica* (seeds) and *Simmondsia chinensis*, *Eucalyptus globulus*, *Amygdalus communis*, *Citrus maxima*, *Cupressus sempervirens* EO's were all purchased from Harraz Co. (Agriculture Seeds, Spices and Medicinal Plants Co., Cairo, Egypt). All plant materials were air-dried at room temperature then ground using an electric mill. Powdered dried material was kept separately in a tightly closed jar kept at room temperature until used.

Insect rearing

A *Musca domestica* L. colony was obtained from the Medical Insect Research Center, Dokki, Giza. The adults were allowed free access to sugar and cotton pads soaked in milk powder dissolved in water (10% w/v). Larvae were reared according to the method described by Pavela (2008) and Huang *et al.* (2008) on a mixture of sterilized bran (38 g), milk powder (2 g) and water (60 mL), and maintained at 27 ± 1°C and 70 ± 5% relative humidity (RH).

Repellency tests on filter paper

Double choice area-preference test

The repellent effect of the tested plants against the larvae of *M. domestica* was evaluated using the area preference method (McDonald *et al.* 1970). Test areas consisted of 9 cm Whatman No. 1 filter paper cut in half. Test solutions were prepared by diluting 0.01 g or 10 µL of the desired plant powder or EO in 5 mL of acetone. Each solution was uniformly applied to a half-filter paper disc. The treated half discs were then left to air dry for 15 min. Full discs were then remade by attaching treated halves to untreated (filter paper discs impregnated with acetone only) halves of the same dimensions with cellulose tape. Each filter paper was then placed in a 9 cm Petri dish. Ten larvae (5-days old) were released at the center of each filter paper disc; the Petri dish was covered and kept in the dark at 27 ± 1°C and 50-70% RH, with six replicates for each tested plant. Observations were made after 1, 3, 5 and 24 h from the beginning of the test, the number of insects present on the control (Nc) and treated (Nt) areas of the discs were recorded.

Percentage repellency (PR) values were computed by the formula used by Tapondjou *et al.* (2005) as follows:

$$PR = [(Nc - Nt) / (Nc + Nt)] \times 100$$

The mean repellency value of each tested plant was calculated and assigned to repellency classes (Juliana and Su 1983) from 0 to V: class 0 (PR < 0.1%), class I (PR = 0.1–20%), class II (PR = 20.1–40%), class III (PR = 40.1–60%), class IV (PR = 60.1–80%), class V (PR = 80.1–100%).

Multiple-area preference test for pupation

Sixteen pieces of filter paper, each consisting of 3 layers, 4 × 8 cm in dimensions and folded several times to furnish pupation site for the larvae. Each piece was treated with 5 mL of acetone containing 10 mg or 10 µL of the desired plant powder or EO, then left to dry in the air for 1 h. The papers were arranged randomly in a 34 cm

in diameter, circular plastic plate. Last larval instars (wandering larvae), 100 larvae, were introduced into the center of the circular plate contained larvae feeding medium. The plate was covered and left in dark at 27 ± 1°C and 70 ± 5% RH until pupation. The number of pupae on each piece of filter paper was recorded. The experiment was repeated five times.

Food preference tests

1. Multiple-choice bioassay

The food preference test developed by Golob *et al.* (1999) was adopted in the present study. A 34 cm diameter plastic plate was radially divided into 16 equal sections. In each section, 0.01 g of the tested plant powder was mixed thoroughly with larvae feeding medium (38 g sterilized wheat bran, 2 g milk powder, 60 mL of water). For essential oils, 10 µL of the tested oil was dissolved in 5 mL acetone, then mixed with feeding medium and left for 1 h at room temperature for the complete evaporation of the solvent. 200 larvae (3-days old) were introduced into the center of the circular plate. The plate was covered and left in the dark at 27 ± 1°C and 70 ± 5% RH for 24 h after which the content of each section was gently retrieved and the number of larvae was recorded. The experiment was repeated four times.

2. Double-choice bioassay

The two halves of several Petri dishes (Anumbra®, 12 cm diameter) were marked with marking pen. Each half of a Petri dish received a different type of treated feeding medium. Always, one half received control medium, while the other received medium treated with a specific plant material. Each Petri dish received 20 larvae (3-days old), then covered and sealed with parafilm. Petri dishes were left in the dark at 27 ± 1°C and 70 ± 5% RH for 24 h, after which, the number of larvae in each half were counted and recorded. The experiment was repeated three times.

Screening for insecticidal activity

Insecticidal activities of the different plant materials were done following a method adopted after Ruiu *et al.* (2008). 10 mg of each dried plant powder were mixed thoroughly with 100 g of the larval feeding medium, divided into 3 equal aliquots in labeled 250 mL beakers. For EOs, 10 µL of the tested oil was dissolved in 5 mL acetone, mixed with the larval medium and left for 1 h at room temperature to evaporate acetone. The control container was prepared in the same manner using normal or 5 mL acetone-treated medium. Each beaker received 20 larvae (1-day old), covered with a sterile thick layer of sawdust and double layer of sterile muslin. The number of pupae that formed was recorded.

Susceptibility of the larvae to the tested plant materials and products

Bioassay was conducted using the food contamination method adopted after Ruiu *et al.* (2008). The susceptibility levels of larvae toward 8 plant materials, proved to be the most effective during screening for insecticidal activity, were done and the regression

lines were constructed. These botanicals were powdered leaves of *L. camara*, *C. sempervirens*, *C. macrocarpa* and *P. zonale*, *C. rotundus* powdered whole plant, *A. nilotica* powdered seeds and EOs of *A. communis* and *S. chinensis*.

Serial concentrations were prepared for each material and product, mixed thoroughly with the rearing media, and then dispensed in 250 mL beakers. Normal media and media treated with acetone only were simultaneously prepared as control for plant powders and plant EOs, respectively. Each beaker received 20 larvae (3-days old), a sterile layer of sawdust and covered with double layer of muslin. Mortality percentages were calculated from the differences between the number of tested larvae and those of emerged adults. Each concentration was replicated 4 or 5 times.

Statistical analysis of data

Results were expressed as mean \pm SD. The statistical significance of differences between means was determined by the student's *t*-test for paired observations. The results of the bioassays were corrected for control mortality using Abbott's formula (Abbott 1925). They were represented graphically as Probit Log Regression lines. Statistical analysis of the data was made using the χ^2 test, the different lethal concentrations and the confidence limits values were all done using SPSS 12.0. In all cases, the percentage of change was calculated using the following equation: Percentage of change = {(test-control)/control} $\times 100$.

RESULTS AND DISCUSSION

Repellency tests on filter paper

The present results (**Table 2**) revealed that treatment with leaf powders of *C. macrocarpa* and *L. camara*, and the whole plant powder of *C. rotundus* produced a highly significant ($P<0.01$) reduction in the mean percentage of formed pupae, amounting to 87.14, 77.14 and 87.45%, respectively, compared to controls. On the other hand, leaf powders of *C. sempervirens*, *E. pereskiifolia*, *P. zonale*, seed of *A. nilotica*, and the EOs of *S. chinensis*, *A. communis* and *C. sempervirens* reduction this percentage by 54.816, 38.461, 35.96, 48.92, 53.86, 38.10 and 44.43%, respectively, from that observed in controls ($P<0.05$). The remaining tested plants and plant products caused insignificant effects ($P>0.05$) on the mean percentage of pupae formed. *P. rubra* had no repellent effect compared with the control or even with other botanical materials and products. These results were in accordance with some other findings. Leaf powder of *L. camara* acted as a physical barrier against *Phthorimaea operculella* (Raman *et al.* 1987; Lal 1988). The EO of

L. camara flowers showed repellent activity against *Aedes aegypti* (Dua *et al.* 1996). Burning dry leaves of *E. globulus* and *L. camara* showed deterrence rates of 88.1 and 79.4%, respectively on *Anopheles arabiensis*, and 86.1 and 71.2%, respectively against *Culex quinquefasciatus* (Kweka *et al.* 2008). The hexane extract of *C. rotundus* was also effective in repelling three dipteran disease vectors, *A. culicifacies*, *A. stephensi* and *C. quinquefasciatus*. The percent repellency at different observation periods (0, 1, 2, 4 and 6 hrs) ranged from 80 to 100%, for different concentrations against different species (Singh *et al.* 2009).

The double-choice test on filter paper (**Table 3**) revealed that *A. communis* EO was the most effective repellent, generating repellency of class II (30%) while the leaf powder of *P. rubra* and *C. rotundus* whole plant powder were least effective. The remaining tested plant materials produced class I repellency. Similar results were reported for *Pelargonium citrosum* against populations of *Aedes* mosquitoes (Matsuda *et al.* 1996). The EOs from *Eucalyptus saligna* and *C. sempervirens* had a strong repellent activity against *Sitophilus zeamais* and *Tribolium confusum* (Tapondjou *et al.* 2005). Also, the EOs of eucalyptus induced the same repellency effects against *Culex pipiens* adult females (Erler *et al.* 2006). The same moderate level of repellency of *A. communis* EO and *S. chinensis* was reported by Al-Jabr (2006) against *Oryzaephilus surinamensis* and *Tribolium castaneum*. Bitter almond EO has a strong fumigation activity against *M. domestica vicina*, *Aedes albopictus*, *S. zeamais* and *Mythimna separata* adults (Ma *et al.* 2007).

Antifeedant activity

The influence of tested plant materials on the level of food preference of *M. domestica* larvae (**Table 4**) revealed that larvae suffered varying degrees of antifeedant activity. Powders of dried leaves of *C. sempervirens*, *C. macrocarpa*, *P. zonale*, whole plant of *C. rotundus* and EOs of *S. chinensis*, *E. globulus* and *C. maxima*, all induced a highly significant decrease ($P<0.01$) in feeding preference as observed a reduction of the mean number of larvae found in treated media. *A. nilotica* seeds and *A. communis* and *C. sempervirens* EOs caused significant ($P<0.05$) antifeedant activity. On the other hand, insignificant antifeeding activity was encountered using *E. pereskiifolia*, *P. rubra* and *L. camara* leaves and flowers compared with controls. Another species of *Euphorbia*, *E. caracasana*, is reported to be not-toxic towards *M. domestica*, *Pieris brassica* and *Artemia salina* (Moreno *et al.* 1995). *L. camara* reduced feeding of *C. quinquefasciatus* the most (Kweka *et al.* 2008). *C. rotundus*,

Table 2 Repellency effect of the tested plant materials on *M. domestica* larvae as experimented by using the multiple-area preference test for pupation.

Treatment	Number of pupae (%)			Change (%) ²	Statistical analysis ³	
	Min.	Max.	Mean ¹ \pm SD		P-value	Significance level
Control (normal diet)	9.375	17.500	12.880 \pm 3.215			
<i>C. sempervirens</i> leaves	1.087	10.112	5.820 \pm 3.765	-54.816	0.013	*
<i>C. macrocarpa</i> leaves	0.000	3.614	1.656 \pm 1.410	-87.143 a	0.000	**
<i>E. pereskiifolia</i> leaves	4.494	11.957	7.926 \pm 2.923	-38.461	0.034	*
<i>P. zonale</i> leaves	3.371	10.843	8.249 \pm 2.907	-35.957	0.044	*
<i>P. rubra</i> leaves	3.371	30.435	14.254 \pm 10.262	10.669 a, b, c	0.782	ns
<i>L. camara</i> leaves	1.205	5.435	2.944 \pm 1.666	-77.142 b	0.000	**
<i>L. camara</i> flowers	2.174	16.854	8.260 \pm 6.209	-35.866	0.178	ns
<i>C. rotundus</i> whole plant	1.042	2.500	1.616 \pm 0.700	-87.452 c	0.000	**
<i>A. nilotica</i> seeds	2.500	12.048	6.579 \pm 3.510	-48.924	0.018	*
Control (acetone-treated diet)	4.494	10.417	7.417 \pm 2.268			
<i>S. chinensis</i> EO	0.000	6.742	3.423 \pm 2.770	-53.855	0.037	*
<i>E. globulus</i> EO	6.250	7.229	6.598 \pm 0.408	-11.038	0.450	ns
<i>C. maxima</i> EO	2.410	7.500	4.591 \pm 2.014	-38.102	0.071	ns
<i>A. communis</i> EO	2.174	5.618	3.665 \pm 1.556	-50.583	0.016	*
<i>C. sempervirens</i> EO	2.174	6.742	4.122 \pm 1.752	-44.427	0.033	*

¹: Mean of five replicates.

²: Figures with similar letters indicate significant differences between means as determined by the Bonferroni test ($P<0.05$).

³: Student *t*-test, levels of significance: ns, insignificant ($P>0.05$); *, significant ($P<0.05$); **, highly significant ($P<0.01$), as compared with the controls.

EO = essential oil

Table 3 Repellency effect of the tested plant materials on *M. domestica* larvae by using the double-choice area preference test.

Treatment ¹	Percentage repellency ² of the observed larvae after 4 time intervals				Overall average (%)	Repellency class ³
	1 h	3 h	5 h	24 h		
<i>C. sempervirens</i> leaves	20.000	10.000	20.000	30.000	20.000	I
<i>C. macrocarpa</i> leaves	0.000	3.333	13.333	6.667	5.833	I
<i>E. pereskiifolia</i> leaves	23.333	13.333	10.000	20.000	16.667	I
<i>P. zonale</i> leaves	16.667	26.667	23.333	10.000	19.167	I
<i>P. rubra</i> leaves	-26.667	3.333	23.333	-36.667	-9.167	0
<i>L. camara</i> leaves	6.667	13.333	3.333	33.333	14.167	I
<i>L. camara</i> flowers	26.667	3.333	-3.333	3.333	7.500	I
<i>C. rotundus</i> whole plant	10.000	-3.333	0.000	-10.000	-0.833	0
<i>A. nilotica</i> seeds	-20.000	-3.333	3.333	33.333	3.333	I
<i>S. chinensis</i> EO	-3.333	-3.333	-6.667	23.333	2.500	I
<i>E. globulus</i> EO	-6.667	16.667	16.667	18.333	11.250	I
<i>C. maxima</i> EO	0.000	-10.000	16.667	23.333	7.500	I
<i>A. communis</i> EO	26.667	30.000	43.333	20.000	30.000	II
<i>C. sempervirens</i> EO	-6.667	23.333	10.000	6.667	8.333	I

¹: Plant material versus the corresponding control: Normal diet and acetone treated diet for plant powder and EO, respectively.²: Average of 6 replicates, 10 larvae per replicate.³: Repellency classes, from 0 to V: class 0 (Percentage repellency <0.1%), class I (PR = 0.1–20%), class II (PR = 20.1–40%), class III (PR = 40.1–60%), class IV (PR = 60.1–80%), class V (PR = 80.1–100%).

EO = essential oil

Table 4 The influence of the tested plant materials on food preference of *M. domestica* larvae as experimented by using multiple-choice bioassay.

Treatment	Number of larvae (%)			Change (%) ²	P-value	Statistical analysis ³
	Min.	Max.	Mean ¹ ± SD			
Control (normal diet)	9.160	14.194	11.064 ± 2.310			
<i>C. sempervirens</i> leaves	3.053	7.595	4.959 ± 1.957	-55.179	0.007	**
<i>C. macrocarpa</i> leaves	3.817	6.452	5.501 ± 1.165	-50.279	0.005	**
<i>E. pereskiifolia</i> leaves	8.054	17.557	12.793 ± 3.881	15.626 !	0.473	ns
<i>P. zonale</i> leaves	3.226	7.383	5.221 ± 1.760	-52.810	0.007	**
<i>P. rubra</i> leaves	6.107	14.194	10.162 ± 3.916	-8.157 a, b, c	0.705	ns
<i>L. camara</i> leaves	5.063	9.396	7.209 ± 2.323	-34.846	0.057	ns
<i>L. camara</i> flowers	4.430	14.504	8.018 ± 4.461	-27.534	0.271	ns
<i>C. rotundus</i> whole plant	1.935	4.430	3.385 ± 1.062	-69.410 a	0.001	**
<i>A. nilotica</i> seeds	1.527	8.861	5.068 ± 3.018	-54.193	0.020	*
Control (acetone-treated diet)	5.806	9.160	8.138 ± 1.565			
<i>S. chinensis</i> EO	1.935	3.817	2.910 ± 0.839	-64.243 b	0.001	**
<i>E. globulus</i> EO	1.527	3.797	2.647 ± 0.928	-67.470 c	0.001	**
<i>C. maxima</i> EO	3.053	5.696	3.833 ± 1.248	-52.903	0.005	**
<i>A. communis</i> EO	1.935	7.634	4.032 ± 2.557	-50.457	0.034	*
<i>C. sempervirens</i> EO	9.160	14.194	5.060 ± 0.754	-37.823	0.012	*

¹: Mean of four replicates.²: Figures with similar letters indicate significant differences between means as determined by the Bonferroni test ($P<0.05$). (!) indicates significant difference between this material and the others used in this list.³: Student t-test, levels of significance: ns, insignificant ($P>0.05$); *, significant ($P<0.05$); **, highly significant ($P<0.01$), as compared with the controls.

EO = essential oil

Table 5 The influence of tested plant materials on food preference of *M. domestica* larvae as experimented by using double -choice bioassay.

Treatment ¹	Percentage of larvae (Mean ² ± SD)		<i>t</i> -Test	
	Control	Treated	P-value	Significance level ³
<i>C. sempervirens</i> leaves	66.667 ± 12.583	33.333 ± 12.583	0.149	ns
<i>C. macrocarpa</i> leaves	93.333 ± 2.887	6.667 ± 2.887	0.001	**
<i>E. pereskiifolia</i> leaves	65.000 ± 13.229	35.000 ± 13.229	0.188	ns
<i>P. zonale</i> leaves	96.667 ± 5.774	3.333 ± 5.774	0.005	**
<i>P. rubra</i> leaves	68.333 ± 20.207	31.667 ± 20.207	0.257	ns
<i>L. camara</i> leaves	55.000 ± 27.839	45.000 ± 27.839	0.785	ns
<i>L. camara</i> flowers	83.333 ± 7.638	16.667 ± 7.638	0.017	*
<i>C. rotundus</i> whole plant	88.333 ± 10.408	11.667 ± 10.408	0.024	*
<i>A. nilotica</i> seeds	90.000 ± 13.229	10.000 ± 13.229	0.035	*
<i>S. chinensis</i> EO	83.333 ± 12.583	16.667 ± 12.583	0.044	*
<i>E. globulus</i> EO	96.667 ± 5.774	3.333 ± 5.774	0.005	**
<i>C. maxima</i> EO	76.667 ± 7.638	23.333 ± 7.638	0.026	*
<i>A. communis</i> EO	75.000 ± 8.660	25.000 ± 8.660	0.038	*
<i>C. sempervirens</i> EO	70.000 ± 15.000	30.000 ± 15.000	0.147	ns

¹: Plant material versus the corresponding control: Normal diet and acetone treated diet for plant powder and EO, respectively.²: Mean of four replicates.³: Significance levels: ns, insignificant ($P>0.05$); *, significant ($P<0.05$); **, highly significant ($P<0.01$), as compared with the controls.

EO = essential oil

E. globulus and *S. chinensis* plants and their products were the most effective antifeedant substances, while the leaf powder of *E. pereskiifolia* was the least effective.

In the double-choice test (**Table 5**), the leaves of *C. macrocarpa* and *P. zonale* and *E. globulus* EO produced a

highly significant ($P<0.01$) antifeedant activity in larvae feeding on the treated media compared with the controls. *L. camara* flowers, *C. rotundus*, *A. nilotica*, *S. chinensis*, *C. maxima* and *A. communis* produced significant ($P<0.05$) repellency against the tested larvae compared with the

Table 6 The larvicidal effect of tested plant materials on 2nd instar larvae of *M. domestica*.

Treatment	Mortality (%)			Change (%)	P-value	t-Test Significance level ²
	Min.	Max.	Mean ¹ ± SD			
Control (normal diet)	10.000	20.000	15.000 ± 5.000			
<i>C. sempervirens</i> leaves	70.000	85.000	76.667 ± 7.638	411.111	0.000	**
<i>C. macrocarpa</i> leaves	55.000	90.000	75.000 ± 18.028	400.000	0.005	**
<i>E. pereskiifolia</i> leaves	70.000	95.000	81.667 ± 12.583	444.444	0.001	**
<i>P. zonale</i> leaves	70.000	80.000	75.000 ± 5.000	400.000	0.000	**
<i>P. rubra</i> leaves	75.000	90.000	80.000 ± 8.660	433.333	0.000	**
<i>L. camara</i> leaves	80.000	90.000	85.000 ± 5.000	466.667	0.000	**
<i>L. camara</i> flowers	85.000	95.000	88.333 ± 5.774	488.889	0.000	**
<i>C. rotundus</i> whole plant	60.000	95.000	76.667 ± 17.559	411.111	0.004	**
<i>A. nilotica</i> seeds	75.000	95.000	85.000 ± 10.000	466.667	0.000	**
Control (acetone treated diet)	20.000	30.000	25.000 ± 5.000			
<i>S. chinensis</i> EO	95.000	95.000	95.000 ± 0.000	280.000	0.000	**
<i>E. globulus</i> EO	90.000	100.000	96.667 ± 5.774	286.666	0.000	**
<i>C. maxima</i> EO	90.000	100.000	96.667 ± 5.774	286.666	0.000	**
<i>A. communis</i> EO	85.000	100.000	95.000 ± 8.660	280.000	0.000	**
<i>C. sempervirens</i> EO	90.000	100.000	95.000 ± 5.000	280.000	0.000	**

¹: Mean of three replicates.²: Student t-test, Significance level: **, highly significant ($P<0.01$), as compared with the controls.

EO = essential oil

controls. Hou *et al.* (2002) showed that *Citrus sinensis* EO had high antifeedant activity against the 3rd instar larvae of both *Helicoverpa armigera* and *Plutella xylostella*. In conclusion, *P. zonale* and *E. globulus* were the most effective repellent against *M. domestica* larvae with a feeding inhibition rate of 96.667%. The leaf powder of *C. macrocarpa* was second rank (93.33%) followed by *C. rotundus* and *A. nilotica* (88.33 and 90.00%, respectively).

Insecticidal activity

The present investigation confirms that house fly larvae were highly sensitive to plant materials and EOs, even at substantially low concentrations. All the tested plant materials and EOs produced a highly significant ($P<0.01$) increase in the mortality percentages of the treated larvae at a concentration of 10 mg (powder) or 10 µL EO per 100 g of rearing medium (**Table 6**). The larvicidal activity of some of the tested plants against *M. domestica* was previously reported. Orange volatile oil produce remarkable toxic effects on *M. domestica* females (Mesbah *et al.* 1990). A reduction on emergence of *M. domestica* adults from larvae treated with eucalyptol was also recorded (Sukontason *et al.* 2004). Abdel Halim and Morsy (2005) evaluated the insecticidal activity of *E. globulus* EO against the 3rd larval instar of *M. domestica*. They reported that concentrations of 100, 70, 50, 25, 5, 2, 1, 0.9 and 0.7% all produced 100% larval mortality. Huang *et al.* (2010) found that the extract of *Cupressus funebris* (leaves and stems), another species of *Cupressus*, produced 90% mortality rate of the *M. domestica* adults within 24 h after treatment when the applied dose was 10,000 mg/L.

Also, the potential insecticidal actions against other insects of the tested plants have also been reported. Mwaiko (1992) suggested that extracts of the peel of bitter oranges of three different species of *Citrus* spp. contain potentially useful larvicidal agent against 3rd and 4th instar larvae of *C. quinquefasciatus*. Dwivedi and Karwasara (2003) found the acetone extract of *L. camara* to be effective against *C. quinquefasciatus* larvae at a dose of 1 mL/100 mL.

Susceptibility of *M. domestica* larvae to plant materials

The data obtained from the susceptibility test of larvae to a list of selected plant materials and products is shown in **Table 7**. The LC₅₀s at 95% probability were 1.37, 3.78, 3.24, 1.24, 12.16 and 20.59 g/100 g for *L. camara* leaves, *A. nilotica*, *C. rotundus*, *P. zonale*, *C. macrocarpa* and *C. sempervirens*, respectively. The LC₅₀s for *A. communis* and *S. chinensis* EOs were 1.274 and 1.67 mL/100 g, respectively.

Table 7 Susceptibility levels of *M. domestica* larvae (3-day-old) towards selected plant materials.

Plant material	LC ₅₀ (%) ¹	Goodness of Fit χ^2
<i>L. camara</i> (leaves)	1.366 (0.019, 42.274) ¹	39.098
<i>A. nilotica</i> (seeds)	3.780	13.282
<i>C. rotundus</i> (whole plant)	3.236 (0.029, 8.115) ¹	18.410
<i>P. zonale</i> (leaves)	1.235	7.325
<i>C. macrocarpa</i> (leaves)	12.157 (4.782, 334.645) ¹	10.272
<i>C. sempervirens</i> (leaves)	20.592	11.267
<i>A. communis</i> (EO)	1.274 (0.953, 1.708) ¹	5.188
<i>S. chinensis</i> (EO)	1.67 (0.78, 4.55) ¹	6.887

¹: Lower and upper Limits for LC₅₀ at 95% confidence.

EO = essential oil

For comparison, **Table 8** summarizes a list of plant materials proved to have larvicidal activities against different larval instars of *M. domestica*. The larvicidal effects of some plants have been reported against various insect species at different considerable mortality rates. The volatile oil extracted from the leaves and flowers of *L. camara* induced a mortality rate that ranged from 80-100% in *M. domestica* treated with 0.0125, 0.025, 0.05, 0.1 and 0.2% of the EO (Abdel Hady *et al.* 2005). The LC₅₀ of fixed oil extracts of *Acacia* sp. was 320 ppm against *C. pipiens* larvae (Hussein 1999). The total number of adults of *Chrysomya chloropyga* that separately emerged from the 1st and 2nd instar larvae in different diets treated with 5% leaf powder of *L. camara* was 0.7 ± 0.30 and 6.7 ± 0.58, respectively which are significantly less than those of the control, 7.7 ± 0.33 and 9.7 ± 0.33, respectively (Muse *et al.* 2003). EO extracted from the leaves of *C. sempervirens* had LD₅₀s of 0.84 and 0.74 mL/cm² against *Sitophilus zeamais* and *Tribolium confusum*, respectively (Tapondjou *et al.* 2005).

CONCLUSIONS

Some of the tested plants have a promising effect as a house fly control agent. *P. zonale*, *L. camara*, *C. macrocarpa*, *C. rotundus*, and *A. nilotica* were a source of repellents and toxicants against the house fly, *M. domestica*. The present investigation showed their potential use as natural insecticides. The most striking fact was the simplicity of the method of application which seemed suitable and economical for controlling house fly larvae in most breeding sites. More studies and extensive research are needed to isolate and identify the larvicidal components in the candidate plant parts and materials.

Table 8 Susceptibility levels of different larval instars of *M. domestica* towards different plant materials.

Plant name	Plant material	Target instar	Method of application	Mortality rate	Reference
<i>Artemisia monosperma</i>	Water extracts of leaves	2 nd instar larvae	Diet contamination	LC ₅₀ =13.0%	Bakr et al. 2003
<i>Conyza dioscoridis</i>	Water extracts of leaves			LC ₅₀ =19.9%	
<i>Eichhornia crassipes</i>	Water extracts of leaves			LC ₅₀ =19.0%	
<i>Clerodendron inerme</i>	Water extracts of leaves			LC ₅₀ =15.0%	
<i>Clocasia antiquorum</i>	Water extracts of leaves			LC ₅₀ =18.0%	
<i>Zygophyllum coccineum</i>	Water extracts of aerial parts			LC ₅₀ =9.9%	
<i>Faresta aegyptia</i>	Water extracts of leaves			LC ₅₀ =12.0%	
<i>Eucalyptus</i> spp.	Eucalyptol (Commercial, Sigma-Aldrich®)	3 rd instar larvae	Dipping	LD ₅₀ =101 µg/L	Sukontason et al. 2004
<i>Lantana camara</i> L	Volatile oil of leaves and flowers	3 rd instar larvae	Diet contamination	Conc. of 0.0125, 0.025, 0.05, 0.1 and 0.2% caused mortality rate from 80-100%	Abdel-Hady et al. 2005
<i>Trigonella foenum-graecum</i>	Crude extract of seeds	3 rd instar larvae	Diet contamination	Conc. of 5, 2 and 1% caused 44.4, 33.3 and 22.2% mortality, respectively.	Abdel Halim and Morsy 2006
<i>Cinnamomum zeylanicum</i>	Crude extract of bark	3 rd instar larvae	Diet contamination	Conc. of 25, 5, 2 and 1% caused of 88.9, 55.6, 33.3 and 22.2% mortality, respectively	Abdel Halim 2008
<i>Illicium verum</i>	Ethanol crude extract of dry fruits	2 nd instar larvae	Dipping	LC ₅₀ =7.4 × 10 ⁴ , 4.1 × 10 ⁴ and 3.2 × 10 ⁴ mg/L, after 24, 48 and 72 h, respectively.	Sripongpun 2008

ACKNOWLEDGEMENTS

Thanks to Prof. Dr. M. Abou-Al-Ennain for identifying some of the collected plants to species level.

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