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Research Article
Toxicological and Developmental Effects of Selected Insecticides, Plant Volatile Oils and Plant Extracts on House Fly, *Musca domestica* L.

Manal Abdel Raouf Abdel Mageed Abdel Razik

Department of Pesticides, Faculty of Agriculture, Menoufia University, Shebin Elkom, P.O. Box, 32514, Egypt

Abstract

Objective: This study was conducted to evaluate the toxicity, developmental and morphological effects of eight insecticides related to different groups, two plant crude extracts and three plant volatile oils, against *Musca domestica* (*M. domestica*) 2nd instar larvae using 3 treatment techniques. **Methodology:** Experiments were conducted in Toxicology Laboratory, Faculty of Agriculture, Menoufia University. The insecticides as applied as mixed with food media, residual film and dipping techniques. The data were statistically analyzed using one way ANOVA by F test at LSD 5% probability. **Results:** The dipping technique was the most effective one. Methomyl was the most toxic compound recording at LC_{50} 4.93 ppm after 24 h of treatment. Citronella oil was the most toxic one recorded LC_{50} 2663.91 ppm. Khaya crude extract was the most toxic one, recording LC_{50} 3300.25 ppm compared with other plant volatile oils and extracts. Chlorantraniliprole and spinosad showed higher pupicidal activity compared with other tested substances. Nearly all tested substances decreased larval, pupal and adult numbers and duration by three methods. Larval, pupal and adult survival percentages were reduced. Adult survival percentages were sharply decreased in three treated techniques, respectively. There were different forms of larval, pupal and adult abnormalities after treated with sublethal concentartions of tested compounds by three methods. **Conclusion:** The dipping technique is most effective method for control larval stage of house fly, selected insecticides shows more toxicity compared with other tested compounds and tested insecticides, plant volatile oils and plant extracts induced biological and morphological effects.

Key words: Insecticides, bio insecticides, biological effects, morphological effects, *Musca domestica* L.

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Corresponding Author: Manal Abdel Raouf Abdel Mageed Abdel Razik, Department of Pesticides, Faculty of Agriculture, Menoufia University, Shebin Elkom, P.O. Box, 32514, Egypt Tel: +201008179580

Competing Interest: The author has declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

House fly, *Musca domestica* L. is a well-known cosmopolitan insect pest of both farm and house. This species is always found in association with humans or their activities. House fly, not only a nuisance insect, but also can transmit many disease-causing organisms[1]. Until now, control of this important public health pest is mainly relied on the insecticides, which are applied against adults and larvae directly or indirectly to suppress their densities[2]. The organophosphates, carbamates, pyrethroids and insect growth regulators have been used to control house fly while many aerosols containing pyrethroid insecticides are being used[3-9]. Resistance of synthetic pyrethroids in the house fly has also documented. The indiscriminate use of chemical insecticides has given rise to many well-known and serious problems, such as the risk of developing insect resistance and insecticidal residual for humans and the environment. Several studies have also looked at the possibility of using plant extracts in the control of eggs, larvae, pupae and adults of *M. domestica*[10-11]. Plant extracts show a broad spectrum of activity against a wide variety of pests and so they have been touted as attractive alternatives to synthetic chemical pesticides for pest management because they pose little threat to the environment or to human health[12]. Thus, many plants have been reported for their potential insecticidal actions on different stages of *M. domestica* via crude extracts or extracted active compound[13,14].

Many article researches were study the effects of different pesticides on metamorphosis or emergence or fecundity or life span of house flies[14-20]. Consequently, plant products are another alternative that become more interesting. Crude extracts and/or plant essential oils have been reported about their potential insecticidal effects on larvae, pupae and adults of house flies[21-25]. IGRs are a diverse group of insecticides, with a range of effects on insect specific phenomena, disrupting the growth and development of insects and other arthropods. They mainly affect the development of immature stages and disrupt metamorphosis and reproduction[26-27] and are becoming more important in the management of insect pests[28].

This study was conducted to evaluate the toxicity, developmental and morphological effects of eight insecticides related to different groups, two plant crude extracts and three plant volatile oils against *Musca domestica* 2nd instar larvae using mixed with food media, residual film and dipping techniques and their pupicidal activity.

MATERIALS AND METHODS

Rearing of tested insect: Colonies of *Musca domestica* originated from larvae were collected from poultry farm at El Behira province and reared under laboratory conditions of 25-27°C and 55-60% RH. A standard rearing method described by Sawicki[29] was used. The larval rearing medium was consisted of 9 g milk powder, 5 g fresh yeast dissolved in 100 mL water and added to 100 g coarse wheat bran, then the mixture was thoroughly stirred and put into the pots in plastic cages, 40×40×40 cm. The newly emerged flies were fed with full fat fresh milk soaked in cotton wool after that adult flies were given milk sugar solution soaked on cotton wool in petri dishes. After 3 days of fly emergence, glass beakers containing larval food were placed in rearing cages for egg lying. The beakers were removed from cages after 2-3 days when eggs were visible and attached to food along the sides of beakers. The beakers were kept in separate cage to obtain 2nd instar larvae for running bioassay tests.

Tested insecticides: Commercial formulations of eight insecticides were used:

Lambada cyhalothrin (Lambada® 5% EC), deltamethrin (Decis® 5% EC), as pyrethroids. Methomyl (Lannate® 90% WP) as carbamate group. Buprofezin (Applaud® 25% SC) as IGR group. Spinosad (Spintor® 24% EC), Abamectin (Vertemic® 1.8% EC), B.t (Protecto® 9.4% WP) as microbial insecticides, chlorantraniliprole (Coragen®, Rynaxypyr® 20% SC) as diamide group. Indoxacarb (Steward®, Avaunt® 15% SC) as oxidiazine group.

Plants and extraction: Pomegranate (*Punica granatum*) fruits rind and Khaya (*Khaya senegalensis*) leaves were collected from the experimental farm of Faculty of Agriculture, Menoufia University, Shebin Elkom, Egypt.

Fruit rind of pomegranate and Khaya leaves were dried under shade at room temperature (27±2°C) for about 20 days. The completely dried fruit rind of pomegranate and Khaya leaves were powdered with an electrical blender and sieved to get fine powder. One hundred grams of the khaya leaf powder was submerged in 300 mL aqueous 70% ethanol and 100 g of fruit rind pomegranate powders was submerged in 300 mL aqueous 70% methanol at room temperature. After 24 h the supernatants were decants and filtered through Whatman filter paper No. 5 and dried in a rotary evaporator at 40°C for 1 h to obtain crude extract. The crude extracts were kept in brown glass bottles till required for the experiment.

Plant volatile oils: Jojoba oil (*Simmondsia chinensis*), parsley oil (*Petroselinum crispum*) and citronella oil *(Cymbopogon winterianus)* were purchased from El gomhoria Company for medical pharmaceutics.

Larvicidal bioassays: The larval bioassay was evaluated using three methods, mixed with food media (food contamination with toxicants), residual film and dipping methods:

Mixed with food media: Standard methods for the evaluation and testing of new insecticides were conducted according to Wright[30]. Larvicidal tests were based by exposing *M. domestica* 2nd instar larvae to food contaminated with toxicants. The bait was prepared by mixing 2 g coarse wheat bran with 2 mL water containing the five tested concentrations of each compounds. The second instar larvae of *M. domestica* were allowed to feed on batches of freshly prepared baits placed in 250 mL glass beakers, each provided with ten 2nd instar larvae. Five concentrations with 4 replicates were conducted for each compound compared with control where baits were free of any compounds, but supplied with equivalent amount of water.

Residual film method: The 2nd instar larvae of *M. domestica* were treated with residual film method. Solution of different concentrations of tested insecticides, plants extracts and volatile oils were prepared. Ten 2nd instar larvae were used for each treatment and put in petri dishes (9 cm diameter) treated with tested compounds (1 mL/dish). Five concentrations with 4 replicates were prepared for each compounds, compared with control where petri dishes were treated with water only and left at room temperature to dry.

Dipping methods: The larval bioassay was evaluated using dipping method of Sinthusiri and Soonwera[31], Sinthusiri and Soonwera[32]. Ten of the 2nd instar larvae were dipped into 10 mL of each tested concentrations for 30 sec and then transferred to a filter paper (in plastic box, size 7.5×10×7.7 cm). Five concentrations with 4 replicates were prepared, while control larvae were dipped in water for 30 sec.

Pupicidal bioassay: The pupal bioassay was evaluated by dipping method as previously mentioned. Ten pupae were dipped into 10 mL of each tested compound for 20 sec, then transferred to a filter paper (in plastic box, size 7.5×10×7.5 cm). Five concentrations and 4 replicates were used. Mortality was recorded 7 days after treatment.

Biological measurements: Second instar larvae of *M. domestica* was fed on coarse wheat bran bait prepared as previously mentioned treated with amount corresponding to LC_{25} of each tested toxicant. Twenty larvae were transferred to a 250 mL beaker containing the bait (2 g) in the method of food mixed with toxicant, while 20 larvae was transferred to petri dish treated with 1 mL of each tested compounds in residual film method and 20 larvae were dipped in each tested concentration for 30 sec in dipping technique, with 4 replicates for each treatment. All treatments were kept at room temperature and daily examined.

The time required for larvae to develop to pupae and adult stages, larval and pupal survival and adult emergence were recorded. Any abnormalities on the appearance of the different insect stages were also recorded and photographed whenever was possible. All treatments were compared with control as previously mentioned.

Statistical analysis: Larval mortalities after 24, 48 and 72 h and pupal mortalities after 7 days were estimated and corrected according to Abbott[33]. Probit analysis according to Finney[34] was performed to estimate toxicity values and slope of regression line for each tested substance. The data of biological aspects was statistically analyzed using one way analysis of variance (ANOVA) by F-test at 5% probability. The measurements were divided using Duncan's multiple range test.

RESULTS AND DISCUSSION

Larvicidal bioassays

Mixed with food media technique: Data in Table 1 show LC_{50} values, slope and confidence limits of tested compounds against 2nd instar larvae of *Musca domestica* treated by mixed with food media technique. Results indicated that methomyl was the most toxic compound (LC_{50} = 4.93 ppm) followed by deltamethrin, lambada cyhalothrin, indoxacarb, spinosad, abamectin and Chlorantraniliprole 24 h after treatment compared with other tested compounds. On the other side, microbial insecticides, abamectin and spinosad recorded higher toxicity to 2nd instar larvae of *M. domestica* where LC_{50} values were 0.10 and 3.01 ppm, respectively, after 72 h of treatment.

As for LC_{50} of the tested plant extracts and volatile oils against 2nd instar larvae of *M. domestica*, citronella oil recorded the highest toxicity giving 2663.91 ppm, while the least one was Khaya recording 5458.54 ppm.

Table 1: Toxicity of different insecticides, volatile plant oils and plant extracts against house fly 2nd instar larvae treated by mixing with food media technique

Tested compounds	LC_{50} (ppm)	Slope Mean±SE	Confidence limits
Lambada	22.44	0.934±0.184	4.087-52.518
Deltamethrin	16.76	1.313±0.261	2.321-39.428
Methomyl	4.93	0.990±0.151	1.77-11.186
Indoxacarb	39.33	0.737±0.160	6.370-98.856
Chlorantraniliprole	58.88	0.791±0.118	26.062-106.155
Abamectin 24 h	54.62	1.122±0.156	30.724-86.305
Abamectin 48 h	24.66	0.642±0.132	5.586-55.592
Abamectin 72 h	0.10	0.631±0.212	7.114-3.589
Spinosad 24 h	44.10	1.128±0.160	24.010-70.421
Spinosad 48 h	16.04	0.731±0.142	3.709-35.775
Spinosad 72 h	3.01	0.998±0.252	0.199-8.541
Protecto 24 h	6674.46	0.429±0.136	1461.136-1.785 E+6
Protecto 28 h	58.88	0.791±0.118	26.062-106.55
Protecto 72 h	21.60	0.449±0.125	1.318-62.257
Buprofezin 24 h	7096.07	0.940±0.227	4062.014-20330.571
Buprofezin 48 h	970.60	0.929±0.183	522.032-1068.704
Buprofezin 72 h	6.51	0.802±0.313	5.501E-6-48.327
Khaya extract	5484.54	0.692±0.129	2466.163-22597.941
Pomegranate extract	3093.91	0.914±0.125	1907.644-5678.693
Citronella oil	2663.91	1.521±0.247	1864.693-4063.705
Parsley oil	3054.10	0.791±0.118	776.012-6176.518
Jojoba oil	4601.13	0.757±0.213	1345.767-1.322E+5

Table 2: Toxicity of different insecticides, volatile plant oils and plant extracts against house fly 2nd instar larvae treated with residual film technique

Tested compounds	LC_{50} (ppm)	Slope Mean±SE	Confidence limits
Lambada cyhalothrin	39.16	0.859±0.253	14.343-295.148
Deltamethrin	31.71	0.918±0.258	12.300-165.661
Methomyl	14.15	1.002±0.163	5.054-26.935
Indoxacarb	49.57	0.591±0.205	12.470-1828.857
Chlorantraniliprole	53.16	0.657±0.218	12.470-1828.857
Abamectin 24 h	74.26	1.298±0.286	-
Abamectin 48 h	4.59	0.814±0.216	31.744-107.485
Abamectin 72 h	0.20	0.512±0.213	0.481-14.595
Spinosad 24 h	42.67	0.496±0.155	3.618 E-10-2.546
Spinosad 48 h	1.35	0.685±0.178	8.426-1022.760
Spinosad 72 h	0.13	0.594±0.197	0.186-4.664
Protecto 24 h	5126.06	1.071±0.281	3004.832-5898 E+18
Protecto 28 h	101.28	0.902±0.179	2543.565-19868.198
Protecto 72 h	4.94	0.413±0.224	29.617-229.213
Buprofezin 24 h	9193.02	0.485±0.190	96.663-264.442
Buprofezin 48 h	167.71	1.186±0.151	0.004-0.714
Buprofezin72 h	2.26	0.382±0.128	0.024-17.821
Khaya extract	3300.25	0.767±0.111	1875.925-7002.991
Pomegranate extract	3380.04	0.661±0.103	1775.355-8314.565
Citronella oil	5056.53	0.747±0.265	1566.615-3.146 E+5
Parsley oil	5803.09	0.789±0.107	2543.565-19868.198
Jojoba oil	8968.16	1.100±0.161	6158.878-15202.706

Residual film technique: Results in Table 2 show that methomyl was also the most toxic compound to 2nd instar larvae of house fly treated with residual film technique recording LC_{50} value as 14.15 ppm, followed by deltamethrin, lambada cyhalothrin, spinosad, indoxacarb, Chlorantraniliprole and abamectin recording LC_{50} values 71, 39.16, 42.67, 49.57, 53.16 and 74.26 ppm, respectively after 24 h of treatment.

Table 3: Toxicity of different insecticides, volatile plant oils and plant extracts against house fly 2nd instar larvae treated with dipping technique

Tested compounds	LC_{50} (ppm)	Slope Mean±SE	Confidence limits
Lambada cyhalothrin	3.94	0.672±0.121	1.869-7.852
Deltamethrin	1.91	0.661±0.265	-
Methomyl	1.54	0.602±0.127	0.247-3.813
Oxidiazine			
Indoxacarb	0.91	0.460±0.112	0.034-3.174
Chlorantraniliprole	1.14	0.365±0.113	0.247-3.813
Abamectin 24 h	0.23	0.558±0.135	0.026-0623
Abamectin 48 h	0.001	0.378±0.203	-
Abamectin 72 h	0.0001	0.424±0.234	-
Spinosad 24 h	5.01	0.684±0.124	0.005-10.022
Spinosad 48 h	0.25	1.137±0.338	0.008-0.774
Spinosad 72 h	0.21	0.812±0.411	1.000 E-38-0.295
Protecto 24 h	14.85	0.418±0.110	3.765-44.692
Protecto 28 h	0.018	0.601±0.234	1.455E-10-0.255
Protecto 72 h	0.032	0.782±0.330	5.503E-10-0.301
Buprofezin 24 h	2012.83	0.450±0.086	558.776-18872.523
Buprofezin 48 h	65.50	0.249±0.097	0.23-364.255
Buprofezin 72 h	13.74	0.271±0.098	0.001-101.677
Khaya extract	11.42	0.277±0.104	0.0004-81.602
Pomegranate extract	12.62	0.471±0.112	0.582-47.602
Citronella oil	60.97	0.726±0.257	1.000E-38 – 618.006
Parsley oil	684.58	0.299±.099	109.425-4707.766
Jojoba oil	22.33	0.351±0.010	109.425-1040547

Meanwhile, 72 h of treatment, abamectin and spinosad showed high toxicity on 2nd instar larvae of *M. domestica* recording LC_{50} values as 0.20 and 0.13 ppm, respectively, followed by buprofezin and protecto recording LC_{50} values.

As for plant extracts and volatile oils, khaya leave extract was the highest effective against *M. domestica* 2nd instar larvae recording LC_{50} as 3300.25 ppm, while jojoba oil was the least giving LC_{50} 8968.16 ppm.

Larval dipping technique: The data in Table 3 revealed that nearly all tested compounds were very toxic to second instar larvae of *Musca domestica* after 24 h of treating by dipping technique recording LC_{50} ranged between (0.23-60 ppm) except parsley oil and buprofezin. Data revealed that abamectin was the most toxic compound to 2nd instar larvae of *M. domestica* after 24 h of treatment, followed by indoxacarb, chlorantraniliprole, methomyl, deltamethrin, lambada cyhalothrin, spinosad, Khaya, pomegranate, protecto, jojoba oil and citronella oil (Table 3). Abamectin was the most toxic compound after 72 h of treatment recording only 0.0001 ppm as LC_{50}.

Pupicidal bioassay: The toxicity of tested compounds related to different groups against pupal stage of house fly treated by dipping technique are presented in Table 4. The results show that chlorantraniliprole was the most toxic compound followed by spinosad, abamectin, lambada cyhalothrin and

Table 4: Toxicity of different insecticides, volatile plant oils and plant extracts against house fly pupae treated with dipping technique

Tested compounds	LC_{50} (ppm)	Slope Mean±SE	Confidence limits
Lambada cyhalothrin	34.98	0.813±0.239	12.325-270.654
Deltamethrin	206.18	0.252±0.171	-
Methomyl	555.77	0.606±0.275	71.686-1.323E+15
Indoxacarb	397.62	0.293±0.179	-
Chlorantraniliprole	0.85	0.397±0.170	4.181E-5-5.910
Abamectin	28.10	0.581±0.195	7.139-666.494
Spinosad	1.73	0.456±0.172	0.024-2.803E-16
Protecto	113.02	1.070±0.234	40.828-252.176
Buprofezin	343.83	0.669±0.200	64.440-1246.746
Khaya extract	42.26	0.344±0.180	-
Pomegranate extract	563.50	0.736±0.211	151.684-2029.660
Parsley oil	724.75	0.217±0.177	-
Jojoba oil	315.22	0.558±0.191	32.694-1533.846
Citronella oil	357.63	0.666±0.131	182.449-820.901

Khaya, recording LC_{50} values as 0.85, 1.73, 28.10, 34.98 and 42.26 ppm, respectively. On the other side, the other tested compounds recorded LC_{50} values ranged between 206.18-724.25 ppm.

The obtained results are in agreement with El Aziz[35], who found that insecticides showed very high toxicity compared to plant extracts against adult stage of house fly by sugar bait. The LC_{50} concentrations of the plant extracts were 5399.93, 7276 and 8149.33 ppm, respectively for the root of *Calotropis procera*, leaves and root of *Piper longum*. Compared to the plant extracts, the synthetic pyrethroid insecticide cypermethrin had much stronger larvicidal effect with LC_{50} of 239.77 ppm against 2nd instar larvae of *Musca domestica*[36]. Gaaboub *et al.*[37] reported that Esfenvalerate was the most effective insecticide against the 2nd and 4th instar larvae of *S. littoralis* after 24 h on treated leaves, followed by chlorpyrifos, leufenuron, jojoba oil and protecto. Al-Ghamdi *et al.*[38] found that crymazine was the most effective compound against house fly larvae, followed by triflumuron and pyriproxyfen, while plant extracts neem oil was the least effective one after treated with dipping and feeding technique.

Gamil *et al.*[39] found that 2nd instar larvae of *S. littoralis* larvae was more susceptible than the 4th instars, in addition, treated insects exhibited symptoms of toxicity starting by sluggish slow movement, cessation of feeding of followed by regurgitation, tremor of larvae thoracic legs and mouth parts followed by insect paralysis then death. Mansour *et al.*[40] found that the insecticides were highly toxic against larval stage of *Musca domestica* compared with tested plant extracts. Scott[1] detected that spinosad was highly toxic to house fly *Musca domestica* based on 72 h, where LD_{50} values and symptoms of poisoning were consistent with a neurotoxic mechanism of action. The KD_{50} values of cypermethrin, fenpropathrin and fenvalerate when applied to *Musca domestica* alone was more effective than its mixture in ratio 1:1 with sesame oil[41].

Prodhan *et al.*[42] reported that the plant extracts of *P. hydropiper* were highly effective (1205.47 ppm) in comparison with that of *C. procera* (5410.8 ppm) and *P. longum* (10737.43 ppm) on the compactness of salivary gland chromosomes in *M. domestica*. Islam and Aktar[36] found that the aqueous extracts of the plants are capable of killing 2nd instar larvae of *M. domestica* at various concentrations where the root extracts of *C. procera* (LC_{50} = 5399.93 ppm) and *P. longum* (LC_{50} = 8149.93 ppm) and the leaf extracts of *P. hydropiper* (LC_{50} = 7276 ppm) were the most effective ones.

Biological measurements: The effects of tested compounds on the development of 2nd instar larvae of *Musca domestica* treated by food media technique are shown in (Table 5).

Statistical analysis of the obtained results in Table 5 indicated that there were significant differences in the duration period of larval stage of *M. domestica* between most of tested compounds and control where it ranged between 1.25-3 days (LSD = 0.86), while there were no significant differences in larval duration between Citronella oil, Buprofezin, Khaya, Pomegranate and control, where it was ranged between 3.75-4.5 days (LSD = 2.24). Furthermore, survival percentages of treated larvae were ranged between 85-100%.

Regarding to the mean number of observed pupae, there were significant differences between all tested compounds and control treatment ranging between 6.75-15.75 pupa comparing with 19.75 pupa in control (LSD = 2.24). There were significant differences in the duration period of pupal stage of *M. domestica* between most of tested compounds and control where it ranged between 2.25-4.25 days (LSD = 0.82) while there were no significant differences in pupal duration between protecto, jojoba oil, parsley oil, citronella oil, Khaya, pomegranate and control where it was ranged between 4.5-5.75 days (LSD = 0.82). In addition, survival percentages of treated pupae were ranged between 33.75-76.25%, comparing with 98.75% in control.

As for the mean number of observed adults, there were significant differences between all tested compounds and control treatment ranging between 0.25-6.25 adults comparing with 18.5 adult in control (LSD = 1.07). There were significant differences in the duration period of adults of *M. domestica* between most of tested compounds and control where it ranged between 2.25-5.5 days (LSD = 1.07)

Table 5: Biological effects of tested insecticides, plant oils and plant extracts on 2nd instar larvae of house fly, *M. domestica* treated by mixed with food media technique

Tested compounds	Larval duration (days)	Larval survival (%)	Mean No. of observed pupae	Pupal duration (days)	Pupal survival (%)	Mean No. of observed adults	Adult duration (days)	Adult survival (%)
Deltamethrin	1.50^{de}	90.00	8.25^{fg}	2.50^{ef}	41.25	2.50^{de}	3.75^{cd}	12.50
Lambada cyhalothrin	1.75^{cde}	98.75	11.50^{cdef}	2.50^{ef}	57.50	4.50^{c}	3.00^{d}	22.50
Methomyl	2.75^{bcd}	98.75	8.25^{fg}	3.50^{de}	41.25	2.50^{de}	4.75^{bc}	12.50
Indoxacarb	2.00^{cde}	100.00	12.25^{cde}	3.00^{ef}	61.25	2.50^{de}	3.50^{cd}	12.50
Chlorantraniliprole	1.25^{e}	96.25	10.75^{cdef}	2.50^{ef}	53.75	0.25^{f}	3.00^{d}	1.25
Abamectin	1.25^{e}	100.00	11.05^{cdef}	2.25^{ef}	57.50	4.25^{c}	3.25^{d}	21.25
Spinosad	3.00^{bc}	85.00	10.00^{defg}	4.25^{cd}	50.00	4.00^{cd}	5.50^{b}	20.00
Protecto	2.00^{cde}	100.00	9.00^{efg}	5.50^{ab}	45.00	1.25^{ef}	6.00^{ab}	6.25
Jojoba oil	2.25^{cde}	100.00	6.75^{g}	5.00^{abc}	33.75	0.75^{f}	6.00^{ab}	3.75
Parsley oil	2.25^{cde}	100.00	11.50^{cdef}	4.50^{abc}	57.50	5.50^{bc}	5.50^{b}	27.50
Citronella oil	4.25^{a}	97.50	13.75^{bc}	5.00^{abc}	68.75	4.00^{cd}	6.00^{ab}	20.00
Buprofezin	3.75^{ab}	96.25	7.00^{g}	2.25^{ef}	35.00	1.25^{ef}	2.25^{d}	6.25
Khaya extract	4.25^{a}	92.50	13.25^{bcd}	4.75^{abc}	66.25	4.25^{c}	5.50^{b}	21.25
Pomegranate	4.5^{a}	97.54	15.25^{b}	5.50^{ab}	76.25	6.25^{b}	5.25^{b}	31.25
Control	4.25^{a}	100.00	19.75^{a}	5.75^{a}	98.75	18.50^{a}	7.25^{a}	92.50
LSD(0.05)	0.86	-	2.24	0.82	-	1.07	1.07	-

Means in each column followed by the same letter (s) are not significantly different at 5% level

Table 6: Biological effects of tested insecticides, plant oils and plant extracts on 2nd instar larvae of house fly (*Musca domestica*) treated with dipping technique

Tested compounds	Larval duration (days)	Larval survival (%)	Mean No. of live pupae	Pupal duration (days)	Pupal survival (%)	Mean No. of live adults	Adult duration (days)	Adult survival (%)
Deltamethrin	2.00^{bc}	87.50	9.50^{cde}	1.75^{b}	47.50	5.00^{bcd}	3.50^{cde}	25.00
Lambada cyhalothrin	1.75^{bc}	70.00	7.25^{def}	1.50^{b}	36.25	3.25^{cde}	3.00^{cde}	16.25
Methomyl	3.00^{b}	97.50	10.00^{cde}	1.75^{b}	50.00	4.50^{bcde}	4.75^{bcd}	22.50
Indoxacarb	1.5^{bc}	90.00	7.25^{def}	1.50^{b}	36.25	3.00^{cde}	2.5^{de}	15.00
Chlorantraniliprole	1.00^{c}	68.75	3.25^{fg}	1.25^{b}	16.25	0.75^{e}	2.25^{e}	3.75
Abamectin	1.75^{bc}	83.75	6.75^{ef}	1.75^{b}	33.75	1.50^{de}	3.5^{cde}	7.50
Spinosad	2.75^{bc}	91.25	2.5^{g}	1.50^{b}	12.50	0.70^{e}	3.5^{cde}	3.75
Protecto	1.25^{c}	82.50	15.75^{ab}	2.00^{b}	78.75	7.25^{bc}	4.00^{cde}	36.25
Jojoba oil	1.75^{bc}	67.50	10.5^{cde}	1.75^{b}	52.50	6.00^{bc}	2.50^{de}	30.00
Parsley oil	2.5^{bc}	80.00	12.00^{bcd}	2.00^{b}	60.00	5.50^{bcd}	2.25^{e}	27.50
Citronella oil	2.00^{bc}	91.25	8.75^{cde}	1.50^{b}	43.75	4.50^{bcde}	6.25^{b}	22.50
Buprofezin	2.00^{bc}	82.50	13.25^{bc}	2.25^{b}	66.25	5.75^{bc}	5.25^{bc}	28.75
Khaya extract	2.00^{bc}	63.75	6.05^{ef}	1.50^{b}	32.50	4.50^{bcde}	6.00^{b}	22.50
Pomegranate	2.50^{bc}	96.25	14.75^{ab}	2.00^{b}	73.75	8.00^{b}	6.50^{b}	40.00
Control	6.25^{a}	100.00	18.25^{a}	5.00^{a}	91.25	17.25^{a}	8.25^{a}	86.25
LSD (.05)	1.11	-	3.15	1.03	-	2.60	1.51	-

Means in each column followed by the same letter (s) are not significantly different at 5% level

while there were no significant differences in adult duration between Protecto, Jojoba oil, Citronella oil and control, where it was 6 days (LSD = 1.07). In addition, survival percentages of adults were ranged between 1.25-31.25%, comparing with 92.5% in control.

As for the development of 2nd instar larvae of *M. domestica* treated by dipping method, the statistical analysis of the results in Table 6 indicated that there were significant differences in the duration period of larval stage of *M. domestica* between tested compounds and control where it ranged between 1.0-2.75 days compared with 6.25 in control. Furthermore, survival percentages of treated larvae were ranged between 63.75-97.5% compared to 100% in control.

Regarding to the mean number of live pupae, there were significant differences between all tested compounds and control treatment ranging between 2.5-13.25 pupa comparing with 18.25 pupa in control (LSD = 3.15). There were significant differences in the duration period of pupal stage of *M. domestica* between all tested compounds and control where it ranged between 1.5-2.25 days (LSD = 1.03) compared to 5 days in control. Survival percentages of treated pupae were ranged between 12-78.75%, comparing with 91.25% in control.

As for the mean number of live adults Table 6, there were significant differences between all tested compounds and control where it was ranged between 0.7-8 adults comparing with 17.25 adult in control (LSD = 2.6). There were significant

Table 7: Biological effects of tested insecticides, plant oils and plant extracts on 2nd instar larvae of house fly (*M. domestica*) treated with residual film technique

Tested compounds	Larval duration (days)	Larval survival (%)	Mean No. of live pupae	Pupal duration (days)	Pupal survival (%)	Mean No. of live adults	Adult duration (days)	Adult survival (%)
Deltamethrin	2.75^{c}	92.50	11.50^{cd}	4.25^{ab}	57.50	5.00^{c}	7.00^{gh}	25.00
Lambada cyhalothrin	2.25^{c}	90.00	11.25^{cd}	3.75^{b}	56.25	5.50^{c}	6.25^{hi}	27.50
Methomyl	2.75^{c}	100.00	15.25^{bc}	4.50^{ab}	76.25	7.5^{bc}	9.75^{ab}	37.50
Indoxacarb	2.00^{c}	95.00	12.25^{bcd}	5.00^{ab}	61.25	5.75^{c}	6.00^{i}	28.75
Chlorantraniliprole	1.75^{c}	98.75	14.75^{bc}	4.25^{ab}	73.75	7.5^{bc}	7.5^{fg}	37.50
Abamectin	2.75^{c}	92.50	12.75^{bcd}	3.50^{b}	63.75	6.00^{bc}	7.75^{fg}	30.00
Spinosad	5.5^{ab}	96.25	13.25^{bcd}	5.50^{ab}	66.25	8.00^{bc}	9.5^{abc}	40.00
Protecto	2.75^{c}	100.00	11.75^{cd}	3.50^{b}	58.75	6.00^{bc}	8.00^{efg}	30.00
Jojoba oil	5.25^{ab}	92.50	15.00^{bc}	5.25^{ab}	75.00	7.00^{bc}	9.25^{bcd}	35.00
Parsley oil	5.25^{ab}	92.50	14.25^{bc}	5.25^{ab}	71.25	7.75^{bc}	8.5^{cdef}	38.75
Citronella oil	5.5^{ab}	75.00	10.50^{d}	5.25^{ab}	52.50	6.75^{bc}	9.25^{bcd}	33.75
Buprofezin	2.75^{c}	91.25	13.00^{bcd}	4.75^{ab}	65.00	7.25^{bc}	9.00^{bcde}	36.25
Khaya extract	6.00^{ab}	97.50	15.75^{b}	4.00^{ab}	75.00	9.5^{b}	8.25^{def}	47.50
Pomegranate	5.5^{ab}	100.00	15.00^{bc}	6.00^{ab}	75.00	8.75^{bc}	9.50^{abc}	43.75
Control	6.25^{a}	92.50	19.75^{a}	6.75^{a}	98.75	19.25^{a}	10.50^{a}	96.25
LSD 0.05	0.75	-	2.36	1.67	-	2.17	0.78	14.90

Means in each column followed by the same letter (s) are not significantly different at 5% level

differences in the duration period of adults of *M. domestica* between all tested compounds and control where it ranged between 2.25-6.5 days (LSD = 1.51) comparing to 8.25 days in control. In addition, survival percentages of adults were ranged between 3.75-40%, comparing with 86.25% in control.

As for the development of 2nd instar larvae of *M. domestica* treated by residual film method, the statistical analysis of the results in Table 7 indicated that there were significant differences in the duration period of larval stage of *M. domestica* between tested insecticides and control where it ranged between 1.75-2.75 days compared with 6.25 in control (LSD = 0.75), while there were no significant differences in larval duration between spinosad, jojoba, parsley, citronella oils, khaya and pomegranate extracts and control ranging between (5.25-6 days). Furthermore, survival percentages of treated larvae were ranged between 75-100% compared to 92.5% in control.

Regarding to the mean number of live pupae, there were significant differences between all tested compounds and control treatment, where it was ranged between 10.5-15.75 pupa comparing with 19.75 pupa in control (LSD = 2.36). While, there were no significant differences in the duration period of pupal stage of *M. domestica* between all tested compounds and control where it was ranged between 3.5-6 days.

Survival percentages of treated pupae were ranged between 52.5-76.25%, comparing with 98.75% in control.

As for the mean number of live adults, there were significant differences between all tested compounds and control treatment ranging between 5-9.5 adults comparing with 19.25 adult in control (LSD = 2.17). There were significant differences in the duration period of adults of *M. domestica* between all tested compounds and control where it ranged between 6-9.75 days (LSD = 0.78) comparing to 10.5 days in control. In addition, survival percentages of adults were ranged between 25-47.5%, comparing with 96.25% in control.

The obtained results are in agreement with Singh and Kaur[43], who found that castor extracts *Ricinus commumis* induced developmental aberrations such as reduced pupations and non-emergence of adults of *M. domestica* after treating by dipping or thin film technique and reported that plant extract contain active principles that interfere with the development hormone affecting the life cycle of the fly. Khater and Shalaby[44] found that plant oils: Fenugreek, rocket, parsley, mustard and olibanum altered some biological aspects of *Culex pipiens* for instance development periods, pupation rates and adult emergence. Moreover, pupation process was greatly reduced due to plant extracts where *A. monosperma* and *F. aegyptica* reduced pupation of *M. domestica* to 30.765 and 32%, respectively[45]. Assar *et al.*[46] reported that pupation was 88 and 16% when larvae fed 5% concentrations of *C. procera* and *L. termis* while, the pupation of *M. domestica* was 48 and 38% when fed on 50 and 100 ppm of coumarin, respectively while the control was 94%. Coumarin at 50 and 100 ppm concentrations induced 45 and 34% adults' emergence, respectively. Shaalan *et al.*[47] found that fentrothion, lambada cyhalothrin and botanical extract *Callitris glaucophylla* induced sublethal effects on larval mortality, larval duration, Pupicidal activity, pupal duration, adult mortality and malformation of *Aedes aegypti*. Beside immediate toxic larvicidal effects all insecticides significantly reduced the average larval period compared to control. Larvae

were observed to pupate faster as their environment increase in toxicity. Bobi *et al*.[48] observed that the larval of *M. domestica* development tendency for the treated larvae to pupate decreased with an increase in the concentration of the selected plants extracts used in dipping and feeding method. The toxicity of ethanolic extracts of the leaves of *A. indica, C. procera, O. basilicum* and *A. mexicana* were found quite effective against the various developmental stages of housefly especially on the larval development, pupation and the emergence of adult. All the plant materials screened hindered larval-pupal transformation and adult emergence. High significant reduction in pupation percent in larvae of *M. domestica* after treated with LC_{50} and LC_{75} of *M piperita*, respectively compared with control. Adult emergence was reduced to 45 and 27.5% in larvae treated with LC_{50} and LC_{75} of *M. piperita*, respectively comparable to 57.5 and 30% for group treated with LC_{50} and LC_{75} of *L. angustfolia*, respectively compared to 95% in control[49]. High reduction in *M. domestica* emergence were also reported by Abdel Halim and Morsy[50] after using volatile oils of *C. macrocarpa* and *A. officinarum*. Gamil *et al*.[39] Found that percentage pupation and adult emergence were significantly less than their equivalent control after treated 2nd or 4th instar larvae of *S. littoralis* with indoxacarb. El-Kholy *et al*.[51] found that larval duration and mortality was reduced significantly by all tested plant extracts (i.e damssisa, camphor and datura), damssisa extract shows higher effect followed by datura and camphor extracts. Hegab and Abd-El Atty[52] found that the three commercial formulations derived from *Azadirecta indica* (Neem), *Citrullus colocynthis* (Hanzal) and *Thymus vulgaris* (Zaatr) which tested with two concentrations (5 and 10%) adversely affected the mortality of larval and pupal stage (57.33 and 50.67%), (45.33 and 66.66%) in control and significantly decreasing in pupation percentage and influenced on pupal mortality. Also, three tested extracts inhibition the percentage of adult emergence resulted from treated larvae in the two tested concentrations against 1st instar larvae of Spiny bollworm *Earias insulana* (Boisd.)[40]. All tested plant extracts and insecticides caused high shortage in the larval, pupal and adult duration after treated larval instar of *M. domestica* with sublethal concentration of each tested toxicant. A compounded diet of housefly containing these plant materials no doubt contains desirable primary or secondary principles which may have developed from the interactions of the components of the diet. These principles elicit biological activities in respect of larval/pupal transformation and pupal eclosion hindrances and they could be usefull in the formulation of a desirable housefly management strategy[53]. Mansour *et al*.[40] indicated that the average number of pupae resulted from treatment of 3rd instars larvae of *M. domestica* with sublethal concentrations of the tested toxicants was highly decreased compared to the corresponding number in control treatment. Moreover, there was a severe decrease in the percentage of adult emergence of *M. domestica*. In addition, El-Sherbini and Hanykamel[54] found that treated groups of *M. domestica* with LC_{50} and LC75 of *Fortunella crassifolia* significantly reduced pupation percent to 62.50 and 42.50%, respectively and reduced adult emergency to 57.5 and 30%, respectively compared with 95% for control.

Morphological abnormalities: Distinct malformations of larvae, pupae and adults of house fly were induced after treatment the 2nd instar larvae with LC25 of tested insecticides, plant volatile oils and plant extracts using mixed with food media, residual film and dipping methods compared with control. Figure 1 includes abnormalities induced by jojoba, parsley oils, pomegranate, abamectin and buprofezin such as Fig. 1a include black pigmentation of body, weakness in cuticle and irregular body shapes. Morphological abnormalities of pupae Fig. 1b include irregular shape, curved pupae and elongation pupae. Morphological abnormalities in adults Fig. 1c. Developmental effects on the adult were seen as compressed body, disappearance of wings, uncompleted wings and smallest size compared with control Fig. 1d.

The obtained results are in agreement with, Bosly[49], who found that distinct malformations of larvae and pupae of the house fly were induced after treatment of the third larval instar with LC_{50} and LC_{75} of *Mentha piperita* and *Lavandula angustifolia*.

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[55] reported developmental abnormalities in larvae of rice leaf folder, *Cnaphalocrocis medinalis* after treatment with 50% neem oil. Various 10 morphological abnormalities on larvae, pupae and adult stages induced by using essential oils against Culex *pipiens*, Lucilia *sericata* and *M. domestica* were detected by Abd El Monem Atwa[45], Sexena *et al*.[56], Mansour *et al*.[40], respectively. Mansour *et al*.[40] reported that there were different forms of pupal and adult abnormalities, where the treatments of *P. granatum* and *S. oleraceus* caused abnormal pupal size in addition to pupal adult intermediate. In the resulted *Musca domestica* adults, the effects were seen as one winged insects, small size and compressed body and abdomen elongation. Such deformations were attributed to treatments of *C. intybus*, C. *aegyptiaca*, *Piper nigrum* and the IGR flufenoxuron.

Fig. 1(a-d): Different forms of larvae, pupae and adult abnormalities resulted from exposing *Musca domestica* larvae to sublethal concentrations of tested traditional, novel insecticides, microbial insecticides, plant extracts and volatile oils: (a) Larval malformations, (b) Pupal malformations, (c) Adult malformations and (d) Normal stages of house fly

Gaaboub *et al.*[37] reported that application of chlorpyrifos, esfenvalerate, leufenuron and jojoba oil against *S. littoralis* caused high increase in malformations of pupae than protecto, the highest percentage of adults malformation recorded with esfenvalerate, chlorpyrifos, leufenuron and jojoba. In addition, Halawa *et al.*[57] found that the tested insecticides (Beticol, Biosad, Elsan, Lufox, Mani, Match and Radiant) against 1 day old pupae of *Bactrocera zonata* induced different morphological abnormalities. Considerable number of larvae, pupae and adults showed obvious malformations after its treatment as surface contactor sandy soil. Moreover, Abd El Monem Atwa[45] reported that there were malformations in larvae, pupae and adults when *M. domestica* larvae were fed on plant extract, there were larval-pupal intermediates, pigmented pupae and pupae with constriction in their pupal case and curved pupae, adults could not emerge completely and adults with abnormal wings and abdomens.

Bisseleua *et al.*[23] found that seed extract of *Griffonia simplicifolia* reported a very strong regulatory effect against the second larval instars of the housefly, as well as, seed extract induced some morphological abnormalities in larvae, pupae and adult house flies[58]. Recently, El-Sherbini and Hanykamel[54] observed different malformations in larvae and pupae of *M. domestica* after treated with *Fortunella crassifolia*.

CONCLUSION

It is concluded that the dipping technique was most effective method for control larval stage of house fly, selected insecticides shows more toxicity compared with other tested compounds and tested insecticides, plant volatile oils and plant extracts induced biological and morphological effects.

SIGNIFICANCE STATEMENTS

This study discovers that the bio insecticides could be used against the house fly larvae and it achieved good biological and morphological effects. It could be suggested that bio insecticides consider a safe product with a potential in integrated pest management programs especially in urban localities.

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