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The Insecticidal Activity of *Artemisia absinthium* leaves Extracts Against Blowfly,*Chrysomya albiceps* (Calliphoridae)

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Chrysomya albiceps, *Artemisia absinthium*, Larvicidal, Development, Fecundity.

ABSTRACT

This study was carried out to evaluate the biological effects of methanolic, acetone and petroleum ether leaves extract of *Artemisia absinthium* against the 1st instar larvae of *Chrysomya albiceps*. All extracts have toxicity against the larvae of *C. albiceps*. The larvicidal, pupicidal and adulticidal activity of these extracts was varied depending on the solvent used in the extraction and the dose of the extract. Complete larval mortality percent (100%) was caused only by the petroleum ether extract of at 0.4g/ml with (Lc_{50} 0.11 g/ml). Methanol and acetone extract had delayed toxic effects on pupae resulted from treated larvae. The toxicity of all plant extracts was extended to the adults that resulted from the treated larvae. Larval and pupal duration showed a significant increase. The growth index for treated larvae and resulted pupae was greatly affected by methanol and acetone extract. The highest retardation in growth was recorded at the highest concentration of 0.4g/ml by acetone leaves extract (2.2 vs. 8.4 for control).

INTRODUCTION

A number of Diptera species have veterinary and medical importance, as they cause myiasis and act as vectors for some bacterial, protozoans, helminth eggs pathogens. Families Muscidae, Sarcophagidae, and Calliphoridae contain the major flies which are vectors to many diseases such as bacillary dysentery, trachoma virus, tuberculosis, cholera, enteric infection spoliomyelitis, typhoid fever, etc. (Greenberg 1973, Zumpt 1965, Patton and Cushing 1934). The main reason for the economic loss of livestock farmers in the world is due to the worldwide dominant fly species *Chrysomya bezziana* and *Chrysomya albiceps* which cause coetaneous myiasis in the tropical regions all over the world. These fliesare known for producing myiasis in humans and animals and transmitting pathogens mechanically (Reigada and Godoy 2005; Marinho et al., 2006; Al-Ghamdi*et al*., 2015). The fly pest *C. albiceps*spread throughout Southeast Asia, India, Africa and Arabian Peninsula (Norris and Murray 1964; Spradbery and Kirk 1992). It had been reported in Jeddah, (Al-Shareef 2016), North, center, east, south of KSA (Setyaningrum and Al Dhafer 2014). *Wohlfahrtia nuba* (Wied), *C. albiceps*and *C. bezziana*are the major species responsible for inducing coetaneous myiasis in KSA (Badawi 1994 and Alahmed, 2001).

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Myiasis by maggot causes a nuisance, less productivity and reproductivity, milk production decrease, blindness, muscle damage, hobble and mortality in progeny (Sotiraki and Hall 2012). To minimize the economic losses for the cattle industry, alternative techniques for population eridication of the flies causing myiasis are needed.

Until the present time, in general, controlling myiasis has used synthetic pesticides such as Spiramycin, amitraz, coumaphos, ivermectin, fenthion, enrofloxacin, diazinon by dipping methods and topical treatment (Nielson, 2003, Osorio et al. 2006, Wardhana, A.H., 2006). Moreover, the applied of synthetic pesticides are recorded to have unfavorable effects, as insect resistance, the mortality of natural enemies of pests, toxicity for humans and animals, tumour, the remnant in milk and meat, as well as causing ecological contamination. (Gealh et al., 2009). So, it is necessary to have a medicine alternative that has some characteristics, among others, simple, attainable, safe for humans and animals and no residue is left in animal products. Plant-derived materials are strongly considered the alternative insecticides to synthetic chemicals in controlling myiasis-producing flies and their larvae.

Plant fauna grown in the Saudi Arabia deserts involve large numbers of plants that have medicinal importance. There are few studies concerned with these plants, most of them were conducted on phytophagous pests and few investigations were established on medical and veterinary pests, for example, mosquitoes. Although there are a number of references that are interested in flies causing myiasis control among animals in some regions of the world (Alahmed, 2004, Mohamed*et al.*2016,Singh and Kaur 2017) no studies on the control of flies causing myiasis in the Kingdom of Saudi Arabia is available. No reports on the use of wild plant *Artemisia absinthium* which grown locally in Saudi Arabia deserts for the control of *C. albiceps* were found in literatures. Therefore, this study was performed to evaluate the toxicity and oviposition deterrent activity of this plant in control of the first instar larvae of *C. albiceps* and their development.

MATERIALS AND METHODS

Flies Rearing:

The blowfly, *Chrysomyaalbiceps* adults were collected from Abu Arish slaughterhouse and transferred to insect laboratory, Biology Department, Faculty of Science, Jazan University, Kingdom Saudi Arabia (KSA) and maintained for several generations under controlled conditions, at a temperature of (27±2ºC), relative humidity (60±10%) and photoperiods (12h light: 12h dark). Collected insects were identified to species by using a key (Shaumar*etal*., 1989). Flies were reared according to method Singh and Kaur (2017) with some modifications. Adults were reared in mesh cages (45×45×45cm) with three sides of the wire and feed on milk powder and sucrose solution. A plastic dish containing fresh cow liver was provided inside the cages for oviposition and as a diet for the larvae. The late 3rd instar larvae were picked up from the plastic dish and transferred into plastic pans (25 x 30 x 15 cm) containing 300cm of softwood sawdust and left to pupate and adult emergence.

Extraction of Plant Materials:

Medicinal plant *Artemisia absinthium* (Asteraceae) was selected to be used as insecticides for the first time against *C. albiceps.*The plant was collected from Hashr mountains (17° 27' 02 " N 43° 02' 26" E),Jazan Governorate (KSA). The leaves were dried at room temperature (27-31ºC) in the shadow until constant weight. Then, pulverized to powder in a hammer mill. One hundred grams of powder were extracted four times with 300 ml of 70% methanol or acetone or petroleum ether solvents at room temperature. After 24 h., the supernatants were decanted, filtrated through whatman filter paper No. 5. and

dried in a rotary evaporator at 40 ºC for (1-2) hours to methanol and (40 – 60) minutes to acetone and petroleum ether. The dry extracts were weighed and kept in deep freezer (4ºC) till used for experiments.

Experimental Bioassay:

1. Larval Bioassay:

The larval bioassay was carried out according to Singh and Kaur (2015) with some modifications. The first instar larvae of *C. albiceps*, used in this experiment were 1-day-old after hatching from the same egg batch. The larvae were grouped into three groups (20 larvae/group) and reared in separate rearing boxes. Plant extracts solutions were immediately prepared in a ceramic bowl by the serially 2-fold dilution method using distilled water and one drop of Tween80 as an emulsifier to facilitate the dissolving of tested material in water. A bowl containing each concentration of the extract was tightly covered with the lid until they were used for the dipping method. For the experiment, the larvae of each group were wrapped in a voile cloth and gently dipped into extract solution, whereas those of the controls were dipped in distilled water and one drop of Tween80. After being dipped for exactly 30 sec, the larvae were transferred to the rearing box containing food. The mortality of each larva was assessed each 24 h by touching each one with a paint brush (no. 0), and those not responding were considered dead. Mortality was recorded daily and dead larvae and pupae removed until adult emergence.Abnormal larvae, pupae and adults were removed daily and placed in labeled glass vials containing 70% ethanol then photographed under abinocular microscope (GX Microscope, GXMXTL3T10) at the Biology Department, Jazan University.

Criteria Studied:

The larvae were observed daily until pupation and adult emergence to estimate the following parameters:

Mortality of the larvae or pupae was indicated by a failure to respond to mechanical stimulation (Williams *et al*. 1986).Larval or pupal mortality percent was estimated by using the following equation (Briggs, 1960):

Larval mortality%= Number of dead larvae / Number of tested larvae x 100.

Pupal mortality% = Number of dead pupae / Number of resulted pupae from treated larvae x 100

The larval and pupal duration was calculated for each one and then the mean value was taken. The adult emergence percent was calculated by using the following equation:

Adult emergence % = A / B × 100

Where **:**A = number of emerged adults, B = number of introduced pupae.

Larval, Pupal and adult malformation was estimated by any change in color, size, shape, or failure to develop to the next stage.

The Growth index was estimated by using the following equation:

Growth index = a / b,

Where**:** A = % of adult emergence, B = mean development (days)

2. Reproductive Potential of Resulted Females:

The adult females that succeeded to emerge from the 1st instar larvae treated with each concentration were collected and transferred with normal adult males obtained from the colony to the wooden cages (30×30×30 cm) by using an electric aspirator recommended by (WHO), and feed on milk powder and sucrose solution for three days, then, the adult females transferred separately to a jar (15×10×10 cm) with a piece of liver to lay egg raft. The number of egg/raft (fecundity) was counted after hatching by using binocular and then the mean value was taken.

The Egg-hatchability (fertility) was calculated by using the following equation:

Egg-hatchability % = A / B × 100,

Where: A = total No. of hatched eggs, **B** = total No. of eggs laid. Sterility index (SI), the Sterility percentage was estimated according to the formula of Toppozada *et al*. (1966):
Sterility percentage = 100 – [a×b /A×B×100]
Where: a = number of eggs laid / female in treatment, b = percentage of hatched eggs in treatment, A = number of eggs laid / female in control, B = percentage of hatched eggs in control**.**

3. Statistical Analysis:

The data on larval, pupal duration and females fecundity will be subjected to statistical analysis by one-way ANOVA followed by Tukey's HSD test ($P < 0.05$) to test for the differences between various concentrations and control using SPSS (19.0) software. The LC_{50} of larvae was determined based on mortality data and Probit analysis (Excel Program) was used in analyzing the dosage mortality response.

RESULTS

Tested Plant Extracts:

Data given in table (1) indicated the amounts of dry 70% methanol, acetone and petroleum ether soluble material from 100g of leaves part. The extracts were varied from one solvent to another. Generally, methanolic plant extracts produced higher weights than acetone and petroleum ether extracts.

Table 1: The weight of dry 70% methanol, acetone and petroleum ether soluble material from 100 gm of *Artemisia absinthium* leaves.

Plant	Part	Weight of extract (gm)		
		Methano	Acetone	Petroleum ether
Artemisia absinthium	Leaves	11.2	7.7	3.6

Toxic Effects of Methanolic, Acetone and Petroleum Ether Extract:

Data given in table (2) indicated the biological activity of methanolic, acetone and petroleum ether extracts of *A. absinthium* (leaves) against the 1st instar larvae of *C. albiceps*. Complete larval mortality percent (100%) was caused by petroleum extract followed by acetone (70%) and methanol extract (50%) at the highest concentration (0.4g/ml) compared to 0.0% for the untreated larvae. A delayed toxic effect of methanol and acetone extract against pupae was observed. The pupal mortality percent was 30.0 and 28.6% for methanolic extract and 66.7, 38.1% for acetone extract at concentrations 0.4 and 0.2g/ml, respectively compared to 1.7% at the control group. Data given in table (2) revealed that there is no effect of petroleum ether extract on the mortality percent of pupae and adult emergence percentages at all concentrations. A remarkable reduction in the percentage of adult emergence was observed by acetone extract especially at the highest concentrations of 0.4, 0.2 and 0.1g/ml, the adult emergence percent was 33.3, 61.9 and 82.5%, while; the adult emergence percent for the untreated group was 98.3%. Results showed that methanol, acetone and petroleum ether extract had a delayed toxic effect against the survival adults resulted from treated larvae, the adult mortality percent was 42.8, 30.0, 33.3 and 50.0, 30.7, 9.1% for methanol and acetone extracts at 0.4, 0.2 and 0.1g/ml, respectively while it was 60.0, 24.1, 15.0% for petroleum ether extract at 0.4, 0.2 and 0.1g/ml, compared to 0.0% for the control group. At the two highest concentrations 0.4 and 0.2g/ml, methanolic extract showed malformative effect among the larvae (16.6 ,11.1%) and pupae (44.4, 25.0%) compared to 0.0 for control group. Also, some malformative effect (58.3, 37.5 and 28.5%) among pupae was observed by acetone extract.

From the aforementioned results it is obvious that the toxicity values of the tested leaves extracts of *A. absinthium* based on Lc_{50} values (Table 3 and Fig. 1) may be arranged in descending order as follows: Pet. ether > Acetone > Methanol. In general, larvicidal evaluation of the tested extracts against *C. albiceps*larvae revealed them to possess low, moderate and high larvicidal activity, with Lc_{50} values ranging from 0.11–0.39g/ml.

Table 2: Effectof methanolic, acetone and petroleum ether extracts of *A. absinthium* (Leaves) on mortality percent of different stages of *C. albiceps.*

Plant extract	Conc. (g//ml)	Larval mortality		Malformed Larvae		Pupation		Pupal mortality		Malformed Pupae		Total Larval and Pupal Mortality		Adult emergence %	Adult mortality %
		No.	%	No.	%	No.	%	No.	%	No.	%	No.	%		
Methanol	0.4	30	50	5	16.6	30	50	9	30.0	4	44.4	39	65.0	70.0	42.8
	0.2	18	30	2	11.1	42	70	12	28.6	3	25.0	30	50.0	71.4	30.0
	0.1	15	25	0	0	45	75	00	00	0	0	15	25	100	33.3
	0.05	12	20	0	0	48	80	00	00	0	0	12	20	100	16.7
	0.025	6	10	0	0	50	90	00	00	0	0	6	10	100	00
Control		00	00	0	0	60	100	00	00	0	0	00	00	100	00
Acetone	0.4	42	70	0	0	18	30	12	66.7	7	58.3	54	90.0	33.3	50.0
	0.2	39	65	0	0	21	35	8	38.1	3	37.5	47	78.3	61.9	30.7
	0.1	20	33	0	0	40	66.	7	17.5	2	28.5	27	45.0	82.5	9.1
	0.05	15	25	0	0	45	75.	4	8.9	0	0	19	31.6	91.1	00
	0.025	10	16	0	0	50	83.	00	00	0	0	10	16.6	100	00
Control		00	00	0	0	60	100	1	1.7	0	0	00	00	98.3	00
Petroleum ether	0.4	60	10	0	0	00	00	-	-	0	0	60	100	-	-
	0.2	45	75	0	0	15	25	00	00	0	0	45	75.0	100	60.0
	0.1	31	51	0	0	29	48.	00	00	0	0	31	51.7	100	24.1
	0.05	20	33	0	0	40	66.	00	00	0	0	20	33.3	100	15.0
	0.025	15	25	0	0	45	75	00	00	0	0	15	25.0	100	4.4
Control		00	00	0	0	60	100	1	1.7	0	0	00	00	98.3	00

Table (3): Relative efficiency of *A.absinthium*leaves extracts against *C. albiceps* larvae

Solvent	LC_{50} (g/m)	Slope (b)	Correlation coefficient (r)
Methanol	0.39	94.8	0.951
Acetone	0.21	144.7	0.838
Petroleum	0.11	196.6	0.949

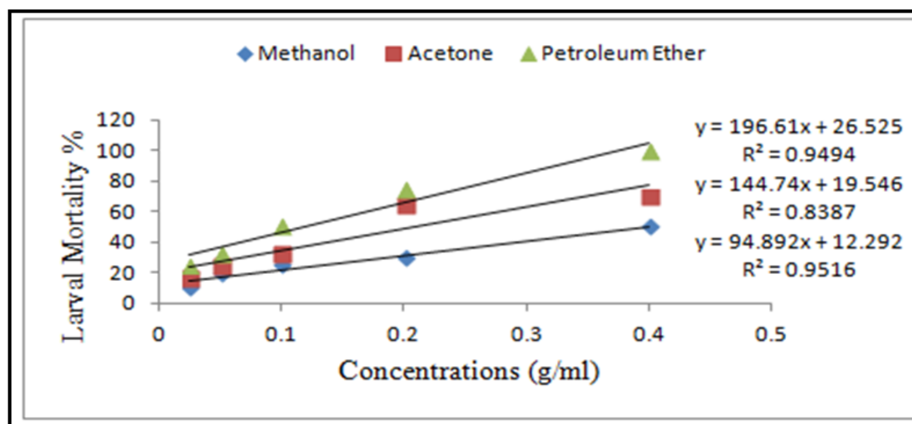

Fig. 1. Regression line of larval mortality of *C. albiceps* treated with leaves extracts of *A. absinthium.*

Morphogenetic Effects:

The different forms of morphogenetic effects as induced by the different plant extracts tested against the 1st instar larvae of *C. albiceps* are illustrated in **Fig.** (2) and can be summarized as follows:

A – Dead deformed larvae partially exuviated attached to the moulting skin. This feature was induced by methanol extract (0.4,0.2g/ml).

B –Larviform (Larval pupal intermediate) resulted from larvae treated with methanol extract (0.4,0.2g/ml).

C – Dead puparium with opening posterior portion and not adult emergence. This feature was induced by acetone extract (0.4,0.2,0.1 g/ml), methanolic extract (0.4,0.2 g/ml).

D – Dead puparium with shrunk appearance induced by acetone extract (0.4,0.2,0.1 g/ml), methanolic extract (0.4,0.2 g/ml).

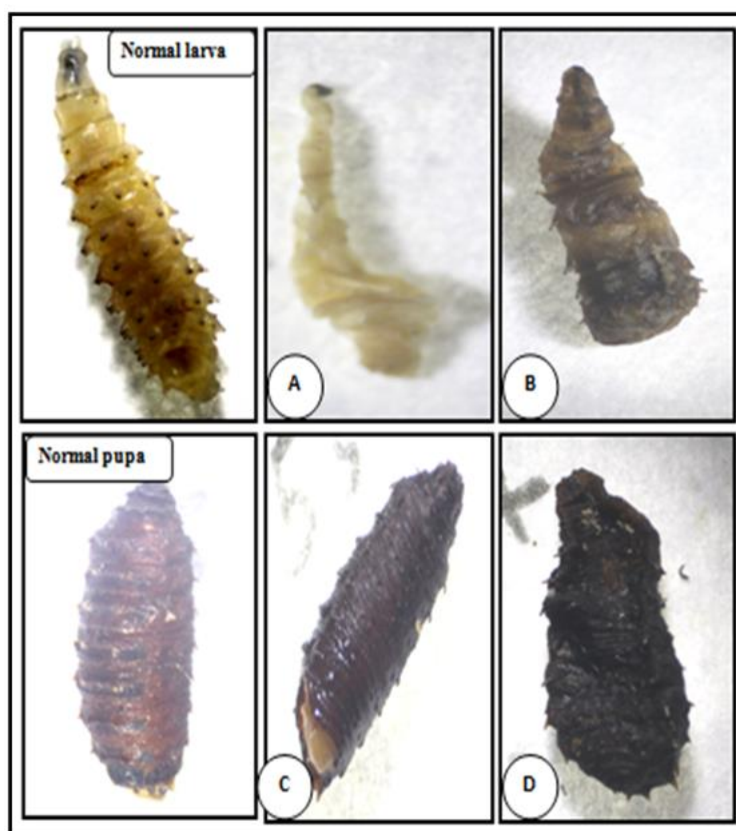

Fig. 2. Malformative effects in *C. albiceps* larvae and pupae as induced by the plant extract tested (160 X). Explanations of figures (see results).

Effect Plant Extracts on Larval and Pupal Development:

The profoundly retarded larval and pupal development through the action of methanolic extract at all concentrations used were observed. The larval period significantly (P<0.05) prolonged to 9.53± 4.25, 7.59± 4.39, 6.06± 2.04, 6.26± 0.66, and 6.03± 0.83 days at the concentrations: 0.4, 0.2, 0.1, 0.05 and 0.025g/ml, respectively, vs. 5.09± 0.29 days for the control group. The pupal period significantly (P<0.05) prolonged to 6. 87± 0.33, 6. 56± 0.49, 6. 93± 0.69, 7. 39± 0.80 and 6. 85± 1.11 vs. 5. 36± 0.84 days for the control group. The growth index decreased as the concentration increased, where it recorded 4.2, 5.0, 7.5, 7.7 and 7.8 at the concentrations: 0.4, 0.2, 0.1, 0.05 and 0.025g/ml, respectively compared to 9.6 for the control group (Table 4). A prolonged larval duration was observed at all concentrations by acetone extract. The highest concentration level

caused amore remarkable prolongation (9.44± 0.83 vs. 5.60± 0.47 days for control). The mean duration of pupae was insignificantly (P>0.05) affected at all concentrations except at the highest concentration 0.4g/ml, it was significantly decreased to 5.66± 0.47 days as compared with the untreated group 5.96± 0.55days (Table 3). The growth index decreased as the concentration of acetone extract increased, where it recorded 2.2, 5.1, 5.9, 7.1 and 8.4 at the concentrations: 0.4, 0.2, 0.1, 0.05 and 0.025g/ml, respectively compared to 8.4 for the untreated group (Table 4). Larval treatment with petroleum extract at only the high concentration (0.2g/ml) increased the larval duration promoting these larvae to develop in a slow rate than that of their control congeners (6.65± 2.49 vs. 4.88± 0.53 days). Meanwhile, this duration was insignificantly affected at the lowest concentrations as compared with the control. On the other hand, Pupal duration was significantly (P<0.05) prolonged to 5.93± 0.68, 7.17± 0.59, 6.27± 0.63 and 6.77± 0.91 days by the concentrations: 0.2, 0.1, 0.05 and 0.025g/ml, respectively compared to 5.55± 0.49 days for the control group. The growth index recorded 7.8, 8.4, 9.1, and 8.8 compared to 9.6 for the untreated group (Table 4).

Table 4: Larval and pupal developmental periods (days) of 1st instar larvae of *C. albiceps* treated with methanolic, acetone and petroleum ether leaves extract of *A. absinthium*

Plant extract	Conc. (%)	Larval duration Mean ± SD	Range	Pupal duration Mean± SD	Range	Total Development ± SD (Range)	Growth Index
Methanol	0.4	9.53± 4.25^{e}	4-13	6. 87± 0.33^{c}	6-7	16.4±4.58^{d}	4.2
	0.2	7.59± 4.39^{d}	4-14	6. 56± 0.49^{b}	6-7	14.2±4.88^{c}	5.0
	0.1	6.35± 2.04^{c}	5-10	6. 93± 0.69^{c}	6-8	12.99±2.73^{b}	7.7
	0.05	6.12± 0.66^{b}	5-7	6. 81± 0.80^{c}	5-8	13.65±1.46^{b}	7.3
	0.025	6.03± 0.83^{b}	5-7	6. 85± 1.11^{c}	5-8	12.88±1.94^{b}	7.8
Control		5.09± 0.29^{a}	5-6	5. 36± 0.84^{a}	5-6	10.45± 1.13^{a}	9.6
Acetone	0.4	9.44± 0.83^{f}	9-11	5.66± 0.47^{a}	5-6	15.1± 1.3^{d}	2.2
	0.2	6.24± 0.42^{c}	6-8	5.93± 0.85^{b}	5-7	12.17± 1.27^{b}	5.1
	0.1	7.85± 2.31^{e}	5-11	5.99± 0.44^{b}	6-7	13.84± 2.75^{c}	5.9
	0.05	6.66± 1.82^{d}	5-7	6.09± 0.61^{b}	5-7	12.75± 2.43^{b}	7.1
	0.025	5.90± 0.53^{b}	5-7	6.0± 0.44^{b}	5-7	11.90± 0.98^{ab}	8.4
Control		5.60± 0.47^{a}	5-6	5.96± 0.55^{b}	5-7	11.65± 1.02^{a}	8.4
Petroleum ether	0.4	-	--	-	-		
	0.2	6.65± 2.49^{b}	4-9	5.93± 0.68^{b}	5-7	12.85± 3.13^{c}	7.8
	0.1	4.76± 1.27^{a}	4-9	7.17± 0.59^{e}	6-8	11.93± 1.86^{bc}	8.4
	0.05	4.77± 2.46^{a}	5-11	6.27± 0.63^{c}	5-7	11.04± 3.09^{ab}	9.1
	0.025	4.81± 0.71^{a}	4-6	6.56± 0.91^{d}	6-8	11.37± 1.62^{ab}	8.8
Control		4.88± 0.53^{a}	4-5	5.55± 0.49^{a}	5-6	10.43± 1.02^{a}	9.6

Means followed by different letter(s) are significantly different from each other (p < 0.05) by Tukey HSD test

Effect of Plant Extracts on Fecundity, Fertility and Sterility Index (S.I.):

Data presented in table (5) showed that at the lowest concentrations (0.05 and 0.025g/ml) the fecundity percent was insignificantly (P>0.05) compared to control, while at the highest concentrations (0.4,0.2 and 0.1g/ml), there was significant (P<0.05) decrease in the mean number of eggs laid (75.00±39.08,123.5±43.43 and 139.11±42.44 eggs/♀; respectively vs. 177.42±29.12eggs/♀ for the control). No effect of the methanolic extract on fertility. The sterility index recorded 58.3, 32.3, 28.4, 9.7 and 12.5% at 0.4, 0.2, 0.1,0.05 and 0.025g/ml, respectively compared to 0.0% for control.On the other hand, there was an

insignificant (P>0.05) difference in fecundity by acetone extract at all concentrations used as compared with the control group. Reduction in fertility which was concentration-dependent criterion, where it was 0.0, 50.0 and 86.6% at the concentrations 0.4,0.2 and 0.1g/ml, respectively. The (S.I.) for females resulted from treated larvae with acetone extract was 100.0% at the highest concentration (0.4g/ml) decreased to 14.4% at the lowest concentration (0.025g/ml compared to 0.0% for the control.Moreover, petroleum ether extract showed a significant (P<0.05) reduction in the fecundity of the females resulted from treated larvae at the highest concentrations 0.2 and 0.1g/ml was observed, where the average number was 78.0±0.0 and 151.2±39.9 eggs/♀ respectively vs. 175.0±28.7 eggs/♀ for the control. A very slight reduction in the hatchability percent was observed, Also the (S.I.) recorded 61.3 and 18.1% at the concentrations 0.2 and 0.1g/ml.

Table 5: Effect of *A. absinthium* (Leaves) methanolic, acetone and petroleum ether leaves extract on fecundity, fertility and sterility index of female *C. albiceps*.

Plant Extract	Conc. (g/ml)	No. of Tested Females	Eggs laid		Hatched eggs		Sterility index (S.I) %
			Total	Mean ± SD	Total	%	
Methanol	0.4	3	225	75.00±18.08^{a}	220	97.7	58.3
	0.2	7	865	123.5±19.43^{b}	833	96.3	32.3
	0.1	12	1670	139.11±16.4^{c}	1605	96.1	28.4
	0.05	17	2793	164.19±14.4$^{d.}$	2703	96.7	9.7
	0.025	13	2207	169.76±12.7^{d}	2012	96.8	12.5
Control		21	3726	177.42±11.12^{d}	3691	99.06	00
Acetone	0.4	1	85	85.0±00	00	00.0	100
	0.2	4	507	126.7±21.6^{a}	253	50.0	64.6
	0.1	9	1300	144.4±19.4^{b}	1127	86.6	30.1
	0.05	12	1817	151.4±17.6$^{b.c}$	1708	94.0	20.4
	0.025	13	2023	155.6±18.3^{c}	1989	98.3	14.4
Control		17	3040	178.8±12.8^{d}	3040	100	00
Petroleum ether	0.2	1	78	78.00±00	67	85.9	61.3
	0.1	4	605	151.2±19.9^{a}	567	93.7	18.1
	0.5	13	2130	163.84±14.9^{a,b}	2077	97.5	7.7
	0.025	20	3385	169.25±12.3^{b}	3303	97.5	4.6
Control		22	3850	175.0±11.7^{b}	3811	98.9	00

Means followed by different letter(s) are significantly different from each other (p < 0.05) by Tukey HSD test

DISCUSSION

An insecticide does not have to cause high mortality on target organisms in order to be acceptable. Antifeedant and development inhibiting activity reduce pest damage to products even without killing the pest. Further, in the long run, populations are reduced through disrupted metamorphosis or reduced fecundity (Ajayi and Muse 2015). Fraternale, *et al*. (2015) suggested the existence of variations in toxicities of phytochemical

compounds on target species depending on the plant part from which they are extracted. In addition, noted that other variations were due to responses by species and developmental stages of species to the specified extract, solvent of extraction, geographical origin of the plant, photosensitivity of compounds in the extract, effect on growth and reproduction, and other factors.

The plant tested in the present study are known to be eco-friendly and are not toxic to vertebrates.No previous data studied the effect of *A. absinthium* against *C. albiceps*. Therefore this plant was chosen to evaluate against the 1st instar larvae of *C. albiceps*. Moreover, it is clearly proved that crude or partially purified plant extracts are less expensive and highly efficacious for the control of flies rather than the purified compounds or extracts (Jang *et al*., 2002 and Cavalcanti *et al*., 2004). The choice of dipping techniques is based on the method that used in controlling of external parasites in large livestock.The active constituents may penetrate into the body of larvae through ingestion or through the cuticle. Previous studies have shown that the plant extracts can penetrate the larval gut thereby damaging its epithelial lining which can either kill them or alter their feeding behavior.

The toxicity of the tested plant extracts against the larval instar was varied according to the solvent used in extraction and concentration of the extract. The larval mortality percent was increased by increasing extract concentration. Generally, based on Lc_{50} values, the present results indicated that the toxicity of petroleum ether > acetone extracts > methanolic extract. The tested plant extracts on larval mortality of *C. albiceps* were in agreement with the results obtained by Mustika (2016), Khater (2017), Suwannayod*et. al*., (2018) and Chil-Núñez *et. al.,* (2018). The genus *Artemisia* which was used in extractions tested against the 1st instar larvae of *C. albiceps* in the present investigation was tested against the larvae of different Calliphoridae species by some other authors. Abdel-Shafy*et. al*, (2009) tested hexane, diethyl ether, ethyl acetate and ethanol extracts of *A. herba-alba*, *A. monosperma*,*Euphorbia aegyptiaca* and *Francoeuria crispa* for toxicity against the 3rd instar larvae of *C. albiceps* using dipping technique.They recorded that all extracts had toxic effects on larvae. Hexane and diethyl ether extracts of *A. herba-alba* and ethyl acetate extract of *A. monosperma* recorded the highest effect. Hexane and diethyl ether extract of *A. herba-alba* caused 100% larval mortality at the concentration 0.06 g/ml, while Hexane and diethyl ether extract of *A. monosperma*100 and 6.7 % larval mortality at the concentrations 0.08 and 0.05 g/ml. Such results may be comparable with the present results on the larvicidal activity for *A. absinthium* plant species used in the present study against *C. albiceps*, whereas the petroleum ether, acetone and methanol extract caused 100, 70.0 and 50% larval mortality at concentrations of 0.4 g/ml. The existence of variations in toxicities of phytochemical compounds on target species confirms the previously mentioned suggestions of Fraternale, *et al*. (2015). Using crude petroleum ether, chloroform, ethyl acetate and methanol extracts of the plant neem,*Azadirachta indica, against* 3rd instar larvae *C. bezziana,* Singh and Kaur (2015) recorded that in the dipping method all the extracts had a toxic effect on the larvae, the highest mortalities were recorded in methanol extract followed by chloroform, petroleum ether and ethyl acetate extracts with LC_{50} values 1.07g/100ml, 1.7g/100ml, 3.39g/100ml and 4.9g/100ml, respectively. However, the present study showed that the highest mortalities were recorded in petroleum ether extract of *A. absinthium* leaves with LC_{50} 0.11 g/ml. It is possible that the assay methods, species differences, and exposure time to test substances may be responsible for the variations between the assay outcomes. Singh and Kaur (2017) reported thataccording to larval mortalities the effects of the crude extracts of *R. communis* on third instar larvae of *C. bezziana* were arranged as chloroform > ethyl acetate> methanol > petroleum ether.

Exposure of *C.albiceps*1st instar larvae to a sub-lethal dose of methanolic, petroleum ether and acetone extracts significantly prolonged the duration of larvae and pupae and the prolongation of development was dependent on the solvent used in extraction and concentration of the extract. These results are in agreement with Ajayi and Muse (2015) using 10% concentration ofneemseed and neem seed kernel powdersagainstfirst, second and third instar larvae of the blowfly,*Chysomya chloropyga*, Mohamed *et al*. (2016) using two medicinal plant extracts (*Commiphora molmol* and *Balanites aegyptiaca*) against 3rd instar larvae of *L.sericata*, *Carriçoa et. al*., (2014) using crude leaf aqueous extract of *Pouteria sapota* on the post-embryonic development of *Chrysomya putoria*, Singh and Kaur (2017) using four solvents of *R. communius* extracts against 3rd instar larvae of *C. bezziana*. Chil-Núñez *et. al.,* (2018) found that, *Ocimum sanctum* essential oil shortening the duration of all post-embryonic stages having a direct impacton the viability of *C. putoria* fly estimating the LC_{50} in 7.47 mg/ml of concentration. The pupal period is when the most important hormonal changes are taking place in holometabolous insects, such as blowflies. Cabral *et. al*., (2007) suggested that compounds extracted from plants and tested to control insects could modify specific physiological processes such as the endocrine control of insect growth, the neuroendocrine system, or the production of some hormones.

In the present study, *A. absinthium* methanol and petroleum ether extract had delayed toxic effects extended to the pupae. In addition, these plant extracts induced a reduction in the % of adult emerged from the pupae produced from treated larvae. The reduction was concentration and solvent used in the extraction – dependent.These results are comparable to earlier results of Mohamed*et. al.* (2016) using medicinal plant extracts of *C. molmol*and *B. aegyptiaca* against *L. sericata* larvae. Ajayi and Muse (2015) reported that neem seed powder at 10 % concentration prevented the emergence of the adult. The percentage of pupal mortality and emergence of adult flies were significant with crude extracts of *Azadirachta indica* (Singh and Kaur 2015) and with crude extracts of *R. communis* against *C. bezziana* larvae (Singh and Kaur 2017) at all the concentrations as compared with the control. The adult emergence percentage of *L. sericata* was markedly decreased after treatments with acetone extracts from the tree of Heaven *(Ailanthus altissima*), Dill *(Anethum graveolens*) and Coriander *(Coriandrum sativum*) (Khater 2017). Plant extracts that accelerate larval development or prolonged it probably caused ahormonal imbalance in the organism (Muse *et al*., 2002) which probably affected the normal development of structures that facilitate adult ecdysis.

Till now very few studies on the fecundity and sterility of Calliphoridae species were examined. All plant extracts tested in the present study especially at the highest concentrations of 0.4 and 0.2 g/ml significantly reduced the fecundity and increased the sterility % of females developed from treated larvae as compared with the untreated control. The fecundity and sterility percents were solvent used in extract– and conc. – dependent. Moreover, a remarkable decrease in the hatchability % of eggs laid by females resulted from treated larvae with petroleum ether extract was observed. The hatchability % of eggs decreased as the conc. of the extract increased.These results are inconsistent with those obtained by Ajayi and Muse (2015)using different concentrations (1, 2, 3, 4, 6 and 10%) of neem seed and neem seed kernel powders as diet against *C. chloropyga*.

In the present study methanol and acetone extract of plant tested against the 1st instar larvae of *C. albiceps* induced some morphological abnormalities in larvae and pupae. It is generally accepted that the developmental anomalies induced by plant compounds are due to an interference with the neuroendocrine control of molting and ecdysis (Schmutterer, 1990). In larva insects three endocrine glands are known to be responsible for releasing neurohormones essential for growth, development and

differentiation; the prothoracic gland (PTG), the corpus allatum (CA), and the corpus cardiacum (CC). It has been shown that plant compounds cause progressive degeneration of all these endocrine glands in larvae (Meurant*et. al*., 1994). This morphological degeneration implies a generalized dysfunction of the neuroendocrine systemleading to prolonged larval and pupal periods. Similar observations were obtained by different plant extracts against different Calliphoridae species in earlier studies. Mohamed *et al*.(2016) observed that *C. molmol*oil and *B. aegyptiaca* alcoholic extracts induced morphological abnormalities in *L. sericata* such as malformation of larvae included small-sized, contractile, and damaged larvae with weak cuticle. Abnormalities of pupa included small-sized, distorted and larviform pupae. Singh and Kaur (2017) recorded varying degrees of morphogenetic abnormalities in immature stages of *C.bezziana* (shrunk larva and dark coloured, dead puparium with anterior portion resembling normal larva when larvae were treated with thecrude extracts of *A. indica* in different solvents (methanol, petroleum ether, chloroform, ethyl acetate).

Conclusion

The activity of these extracts used extends beyond the larval stage. They kill larvae when used at high concentrations, and lead to a series of morphologic abnormalities that inhibit metamorphosis. Consequently, they may prevent adult emergence and protect against re-infestation. The results of the present study may have great importance as today the environmental safety of an insecticide is considered to be of paramount importance.

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