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EFFECT THE EXTRACTS OF PEPPERMINT (*MENTHA SPICATA* L.) IN SOME BIOLOGICAL ASPECTS OF MOSQUITOES *CULEX MOLESTUS* FORSKAL (1775) (DIPTERA: CULICIDAE)

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Abstract

The effect of aqueous cold, boiling and chloroform of roots, stems and leaves extracts of peppermint plant *Mentha spicata* was studies by using concentrations (1,5 and 10) mg/ml of aqueous extracts and (0.25, 0.5 and 1) mg/ml of chloroform extract on some biological aspects of *Culex molestus* by treated fourth instar larvae of the insect, and the results were as follows :

The results of the study showed that the above mentioned extracts have an effect in inhibition of emergence and that effect is positively co-related with the concentrations used. The most of these effects were demonstrated by the usingthe chloroform extract of the leaves as the value of $LC_{50}$was 0.29 mg/ml, as for the effect of the mortality of the fourth instar larvae and pupae, we note that the highest percentage of mortality occurred when using aqueous cold, boiled and chloroform for leaves as the value 90% to mortality of fourth instar in concentration 10 mg/ml, in the aqueous extract of boiled in the stem as the mortality percentage 75.6% in concentration 5 mg/ml.

The results indicated that the extracts under study had an effect on the age of male and female, the results indicated to reduce in age due to the treatment of these extracts as it reached 0,3.5 day in control treatment and 2,4 day in chloroform extract of stem compared with 7.9, 9.2 day in control treatment .

Keywords: mosquito, active compounds in plants, *Mentha spicata*

Introduction

Mosquitoes one of the insect that feeding blood by sucking, return to Diptera, [1] medically and veterinary importance because transmits for manydangerous pathogens. [2] One of the most important parasites transmitted by mosquitoes is malaria parasites and ring viruses such as Dengue fever virus, yellow fever virus, and West Nile virus [4, 3]. As a result of the rapid proliferation of this insect and the production of large numbers of eggs, a wide spread of these diseases has been observed, causing an epidemic, so attention has been paid to developing programs to combat and reduce their spread. Mosquitoes come back to the Culicidae family which contain 3601species and subspecies[5], which The Culex genus is found in tropical and temperate regions with favorable environmental conditions for the transmission of many viral diseases. [6] *Culex molestus* Forskel mosquitoes are the most prevalent in the human environment [7, 8]. However, the use of chemical pesticides is the fastest method among many control methods. [9] However, many studies have found that the use of parts of plants or their extracts [10], plant extracts have toxic effects to larvae, pupae and adult mosquitoes in general. [11] *Mentha spicata* to the oral family Lamiaceae has important functions from the medical point of view and insect repellent as mosquitoes [12], and given the importance of combating some of the water stages to control the adult stages. And the follow-up of the individuals resulting from them until reaching to adults and calculate the proportion of the cumulative mortality of the effect of these extracts in the longivity of the fourth larvae and pupae and adults resulting from different treatments.

Materials and Methods

1. Collect and diagnose plant samples

Samples of peppermint plant were collected from the orchards of the province of Karbala in December 2016 and separated into leaves, stems and roots, and after drying and grinding separately, put in glass bottles and the name of each vegetable section was recorded and stored in the refrigerator until use. The diagnose of plant in Department of biology, college of Sciences, University of Babylon.

2. Preparation of plant extracts

The modified method [13] was adopted for [14] in the preparation of water extracts. In the preparation of organic solvent extracts, the organic solvent chloroform was used in the preparation of organic plant extracts. The method [15] and the modified [14] were adopted in the extraction process for roots, stems and leaves of peppermint.

3. Collection and diagnosis of mosquito samples:

samples of mosquitoes *Cx. Molestus* Forskal were collected from one of the exposed sewage areas of open heavy water measuring (2 x 2) m and containing its limbs on reed plants *Typha sp*. In the province of Karbala - district of Ramadan - the role of stone on 20/3/2017. And to diagnose mosquitoes within the study area where samples were taken from the fourth instar larvae and identified using the taxonomic key. [16] To confirm the diagnosis of this type of mosquito was taken advantage of the phenomenon Autogenous and the existence of the ability to put the first boat eggs without the need to feed a blood meal [17].

4. Mosquito breading:

The egg boats collected from the site were isolated and each boat was placed in a 400 mL plastic container containing tap water. After hatching the eggs to larvae, they were fed with bread crumbs and when they were turned into pupae, the pupae were transferred to the breeding cage (30X30 x 30 cm) was enclosed by a metal wire on four sides and a fifth-line tulle. The adults were fed on a 10% glucose solution and don't fed to the blood meal. The colony was purified for three generations before the experiments began. On fourth-generation larvae and adults used for diagnosis and Conduct experiments .

5. Effect of water extracts and chloroform of roots, stalks and leaves of peppermint plant in the biological aspects of the insect:

Took (10) larvae / replicate of fourth instar larvae and (3) replicates per concentration. transferred it to 100 mL plastic containers containing the above mentioned extracts (1, 5 and 10) mg / ml for the water extract and 0.25, 0.5 and 1 mg / ml for the chlorofuramic extract With bread crumbs for the purpose of feeding them. Used distilled water and solvent in control experiments, recorded The percentage of mortality in the fourth instar larval and the pupal stage (produced after treatment of the fourth instar larval) and until it reached the adult stage and calculated the longevity of the fourth instar larvae, pupae and adult. The percentages of mortality are calculated according to equation [18].

%mortality corrected = (mortality% in treatment - mortality% in control) / (100-mortality% in control) x 100

The percentage of inhibition of adultsemergence (IE%). was calculated According to equation [19].

IE% = 100- (T X100 / C)

T = the percentage of the emergence of the treatment

C = Percentage of the emergence in the control.

The relationship between these values and concentrations was graphically represented on logarithmic leaves to draw the toxicity line and calculate the LC_{50} value.

The results of the study experiments were analyzed according to the model of global experiments and with full randomization design, the loss ratios were adjusted according to equation [20.18] and the corrected values were converted to angle values for inclusion in the statistical analysis.

Results and Discussion

1. Effect of interaction concentrations of cold and boiling water extracts of *M. spicata* in the mortality percentage of the fourth larval instar of mosquitoes. *Cx. molestus*

The results of Table (1) showed that the effect of the cold water extract on the plant was more effective than the boiling water extract. The effect varied with the different concentrations of the extract with the highest rate of fourth larvae loss in the cold and boiled water extract of the roots and stalks of peppermint leaves (90, 89.6, 90) respectively and in higher concentration(10) mg / ml compared to 2.8, 5.2 and 5.6% respectively in control treatment. The percent mortality of fourth instarlarvae in the boiling water extract of roots, stalks and leaves was (40.5 and 80.1 and 90)% respectively, with a concentration of 10 mg / ml compared with 2.8, 4.6 and 5.6%, in control treatment.

Table 1 : Effect the interaction of the cold and boilingaqueousextract of *M. spicata*in the mortality percentage of the fourth larval instar of *Cx. molestus*

Concentrations mg/ml	% mortality of fourth –instar larvae					
	cold aqueous extracts			boiled aqueous extracts		
	Roots	Stems	Leaves	roots	Stems	Leaves
Control	2.8	5.2	5.6	2.8	4.6	5.6
1	15.6	53.6	68.3	25.2	46.8	27.6
5	5.6	83.3	80.2	35	47.2	80.4
10	27.6	89.6	90	40.5	80.1	90
Value of LC_{50}	4.216	1.318	0.6	6.309	0.891	1.445
L.S.D. interaction between effect of difference of the type of extract and type of plant part in the percentage mortality of fourth-instar larvae 10.72						

The results of the statistical analysis showed significant differences in the results obtained.

A study (21) of *Peganum harmala* confirmed that the cold water extract of the plant is superior to the hot water extract in the various biological performance parameters of mosquitoes *Cx. pipiens* L.

The lethal effect of essential oils extracted from leaves, flowers and roots of aromatic plants was studied against the fourth instar of mosquito larvae *Cx. molestus* Forskal The most commonly used extracts are *Foeniculum vulgare* Mill, the most toxic, followed by *Ferula hermonis boiss, Citrus sinensis osbeck, Pinus pinea* L., *Laurus nobilis* L. and *Eucalyptus spp*. The LC50 values were 24.5, 44.0, 60.0, 75.0, 117.0 and 120.0 mg/L, respectively [6].

A study [22] showed the effect of the water extract of leaves of *Calotropis procera* Ait. For mosquito control, with LC_{50} (273.53, 366.44 and 454.99) ppm in the treatment of the third and fourth instar larvae respectively for mosquitoes *Anarabiensis* in the concentrations (200, 400, 600, 800 and 1000) ppm and for *Cx. quinquefasciatus* (187.93, 218.27 and 264.85) ppm respectively in the treatment of the third and fourth instar larvae in the concentrations (100, 200, 300, 400, 500 and 600) ppm. The value of IE50 was the percentage of inhibition of adult emergence 277.90 and 183.65 ppm Respectively

A study [23] of the cold and boiling water extract of *R. communis* confirmed that the mortality percentage of the fourth instar larval of mosquitoes *Cx. pipiens* were 0, 33.76, 36.84, 40.39, 46.30%, 0, 44.43, 47.30, 53.77 and 58.07% respectively in the concentrations (0, 2, 5, 10 and 20 mg / ml).

[24] indicates that the effect of boiling water causes the inhibition of the esterase, phenolase and hydrolase enzymes of active compounds that are not affected by the cold water extract or as a result of feeding the larvae to the toxic compounds found in the plant extract that affect the target tissues. [25, 11], The cause of themortality of the larval stage may be due to its sensitivity to the presence of toxic substances in the leaves of the plant, these materials may not lead to the rapid and direct destruction of larvae, but to the impact on the inefficiency of food conversion and therefore the destruction of the insect as a result of its impact on the movement of the digestive tract and the rate of digestion effectiveness and absorption Leading to reduced growth [26], or the cause of the destruction of the fourth larvae stage may

be affected by the group of enzymes Microsomal oxidase enzyme in the epithelial cells of the gastrointestinal tract, which works to remove the toxicity of natural compounds found in the plant extract and thus poisoning the insect and then its destruction [27], Or may be due to contain the water extract of the plant under study to the presence of compounds that prevent the formation of chitin in immature stages, and thus unable instar larvae to build the new cuticle leading to the death of the insect [28], or may be the reason to discourage the growth and development of new It is hardened due to effective inhibition Mechanism Tyrosinase enzyme found in mosquito larvae as it is directly responsible for cuticle seclerotization [29].

2. Effect of interaction concentrations of the chloroform extracts of *M. spicata* in the mortality percentage of the fourth larval instar of mosquitoes *Cx. molestus*

The results of Table 2 show the effect of chloroform extract on the roots, stalks and leaves of the peppermint plant in the mortality rate of fourth instar larvae, with the highest mortality rate (90, 90 and 89.7%) respectively with the highest concentration of extract (1 mg / ml)) compare with (4.2)% In control treatment.

Table 2 : Effect the interaction of the chloroform extract of *M. spicata* in the mortality percentage of the fourth larval instar of *Cx. molestus*

Concentrations mg/ml	%mortality of fourth –instar larvae		
	Roots	Stems	Leaves
Control	4.2	4.2	4.2
0.25	40.3	37.8	78.3
0.5	90	60.1	80.6
1	90	90	89.7
Value of LC_{50}	0.28	0.631	0.209
L.S.D. interaction between effect of difference of the type of extract and type of plant part in the percentage mortality of fourth-instar larvae 7.829			

The results of the above table showed that the peppermint extract had an effect on the fourth larvae. The effect was increase with increasing the extract concentrations. The results of the statistical analysis showed the significance of the differences in the obtained results.

[30] obtain the alcoholic extract of the seeds of the black bean *Nigella sativa* and leaves of eucalyptus *Eucalyptus spp*. toxic of the larval stage of mosquito *Cx. pipiens*

[31] showed the value of LC_{50} in the petrolium ether extract of *Mentha microphylla* Koch 39 mg / L when treated forth instar larvae of *Cx. molestus* with plant essential oils

The reason for its resistance to pesticides and some plant extracts may be due to the increased thickness of the cuticle layer when advancing larvae to the fourth larval stage [32] or as a result of feeding the larvae to the toxic compounds found in the plant extract that affect the target tissue [25.11].

3. Effect of interaction concentrations of cold and boiling water extracts of *M. spicata* in the mortality percentage of pupal mortality resulting from treatment the fourth instar larvae of mosquitoes *Cx. molestus*

The highest mortality rate of pupae resulting from cold water extract for roots, stalks and leaves of peppermint (15.6%) respectively in concentration (5 and 1) mg / ml compared with 0, 1.2 and 0% respectively in control treatment. While the pupal mortality from the treated of boiling water extract of roots, stalks and leaves (15.6, 27.6 and 25.6%) in the concentrations (5 and 1) mg / ml respectively compared to (1.2, 0 and 0)% in control treatment.

Table 3: Effect the interaction of the water extract of *M. spicata* in the mortality percentage of pupae resulting from treatment of the fourth larval instar of mosquitoes *Cx. molestus*

Concentrations mg/ml	%mortality of pupae resulting from treatment of fourth –instar larvae					
	cold aqueous extracts			boiled aqueous extracts		
	Roots	Stems	Leaves	roots	Stems	Leaves
Control	0	1.2	0	1.2	0	0
1	15.6	15.6	25.6	15.6	27.6	35
5	15.6	15.6	15.6	15.6	75.6	25.6
10	0	0	0	0	25.7	0
L.S.D. interaction between effect of difference of the type of extract and type of plant part in the percentage mortality of pupae 2.013						

The results of Table (3) showed that the effect of the boiling water extract of the plant was more effective than the cold water extract with different effect of different concentrations of the extract. The results of the statistical analysis showed the significance of the differences in the obtained results**.**

A study [23] confirmed the superiority of the hot water extract on the cold water extract of the leaves of the rhododerma *R. communis* on pupa of *Cx. pipiens*; the pupal mortality percentage when use hot water extract were (0, 45, 48.54, 53.15, 65.70%). In the cold water extract, the percentage of pupal mortality (0, 42.12, 43.27, 45.57 and 56.82%) was in the concentrations (0, 2, 5, 10 and 20) mg / ml

4. Effect of interaction concentrations of chloroform extracts of *M. spicata* in the mortality percentage of pupal mortality resulting from treatment the fourth instar larvae of mosquitoes *Cx. molestus.*

The results indicated in Table (3) the highest percentage of pupal mortality resulting from the treatment of the fourth instar larvae in the chloroform extract of roots, stalks and leaves of peppermint was (29.2, 0 and 15.6%) in concentration 0.25 mg / ml compared with 1.2% in control treatment.

Table 4 : Effect the interaction of the chloroform extract of *M. spicata* in the mortality percentage of pupae resulting from treatment of the fourth larval instar of mosquitoes *Cx. molestus*

Concentrations mg/ml	% mortality of pupae resulting from treatment of fourth –instar larvae		
	Roots	Stems	Leaves
Control	1.2	1.2	1.2
0.25	29.2	0	15.6
0.5	0	0	15.6
1	0	0	0
L.S.D. interaction between effect of difference of the type of extract and type of plant part in the percentage mortality pupae 3.412			

The results of Table (4) showed that the chloroform extract of the peppermint plant had an effect on the percentage of pupae mortality resulting from the treatment of the fourth instar larvae of *Cx. molestus* mosquitoes, and the results of the statistical analysis showed the significance of the differences in the results obtained

The results of a study [33] confirmed that the concentration of 500 ppm for the chloroform extract of *Terminalia chebula* Retz. Was the most effective, giving pupae mortality in mosquito *Culex quinquefasciatus* 56%, and the percentage of pupal mortality in the concentration of 250 ppm was 39.20%

5. Effect the interaction of the concentrations the cold and boiling water extracts of the peppermint plant *M. spicata* in the percentage of cumulative mortality of the immature stages of the *Cx. molestus*

Table (5) shows the effect of the concentrations of the extract on the mortality of immature stages of the insect treated. The increase in the concentration of the extract resulted in an increase in the mortality rates of the immature stages of the treated insect. A positive relationship was observed between the concentration of the extract and the percentage of mortality. The cumulative mortality of immature stages when use 10 mg/ml concentration in cold water extracts for root; steam and leaves peppermint plant were (90, 89.6 and 90)% respectively compare with (2.8, 6.4 and 5.6)% in control treatment, whereas Cumulative mortality rate in hot water extract were (40.9; 90.90)% respectively in the concentration (10) mg / ml for root extract, stems and leaves compared with 4.0, 4.6 and 5.6% respectively in the control treatment

Table 5: Effect the interaction of the water extract of *M. spicata* in the percentage of cumulative mortality of immature stages of mosquitoes *Cx. molestus*

Concentrations mg/ml	mortality cumulative of the immature stages%					
	cold aqueous extracts			boiled aqueous extracts		
	Roots	Stems	Leaves	roots	Stems	Leaves
Control	2.8	6.4	5.6	4.0	4.6	5.6
1	31.2	69.2	93.9	40.8	74.4	62.6
5	44.5	98.9	80.2	50.6	90	80.4
10	90	89.6	90	40.9	90	90
L.S.D. interaction between effect of difference of the type of extract and type of plant part in the percentage mortality cumulative of the immature stages 12.733						

Table (5) shows that the cold and boiling water extract of the plant had a high effect in the percentage of cumulative mortality, and the results of the statistical analysis showed the significance differences in the results obtained

A study [34] confirms the superiority of the water extract of leaves of the *Nerium oleander* plant in the biological efficiency of the cumulative mortality of mosquito larvae *Cx. quinquefasciatus* given a concentration of 3000 ppm mortality of 100% killing

This may be due to the effect of these extracts on insect stages because they contain phenol compounds, toxic substances or other effective compounds that act as feeders, causing the insect to die. The killer cause of insect stages may be due to the nervous and digestive system of the insect by touching these extracts to the surface [35, 34], the reason for the existence of high cumulative loss rates in the plant under study for larvae is an indication of the presence of poisoning and accumulation of active substances in the studied plant in the digestive channels of larvae and the appearance of many deformities , The reason may be the effect of these substances on food enzymes analyst for the substance found in the gut. [27] The presence of a group of enzymatic microsomal oxidase enzymes in the gut cells of insects are important in removing the toxic effect of natural compounds of the plant, and that any effect on these enzymes by these compounds causes poisoning of the digestive tract tissues of the insect

6. Effect the interaction of the concentrations the of chloroform extract of peppermint plant *M. spicata* in the percentage of cumulative mortality the immature stages of the *Cx. molestus*

The percentage of cumulative mortality for the immature stages in the chloroform extract for the roots, stalks and leaves of the peppermint plant were highest concentration of extract (1) mg / ml was (90, 89.7 and 90) %respectively, compared with 5.4% in the control treatment

from the results of Table (6) showed that the chloroform extract of the plant had a high effect in the percentage of cumulative mortality, and the results of the statistical analysis showed the significance of the differences in the results

It is noted in the present study that the high concentration and the length of time, caused higher percentage of cumulative mortality of immature stages, this may be due to the effect of these extracts in the stages of the insect because of the containment of toxic substances or other effective compounds act as anti-feeding cause the insect to die, or The reason of die may be due to the impact of the nervous system and the digestive of the insect by touching these extracts to the surface of the body of the insect or enter these substances through the respiratory openings [35], or may explain the cause of the mortality of the fourth larvae to starvation because the active compounds of the plant extract works as anti-feeding [36] This disease affects the epithelial cells of the gastrointestinal tract and causes cases of insect poisoning, combined with the fatty substances present in the digestive system and causing the death of the larvae [37, 38]

The results of [39] confirmed a cumulative mortality of the immature stages of mosquitoes *Cx. pipiens* 100% when treated with *Turmine ra mousissima* Turbine extract for concentrations (2.5, 5, 10 and 20) mg / ml, respectively.

Table 6 : Effect the interaction of the chloroform extract of *M. spicata* in the percentage of cumulative mortality of immature stages of mosquitoes *Cx. molestus*

Concentrations mg/ml	**% mortality cumulative of the immature stages**		
	Roots	**Stems**	**Leaves**
Control	5.4	5.4	5.4
0.25	69.5	37.8	93.9
0.5	90	60.1	95.6
1	90	89.7	90
L.S.D. interaction between effect of difference of the type of extract and type of plant part in the percentage mortality of cumulative of immature stages 11.241			

7. Effect the interaction of concentrations of cold and boiling water extract of *M. spicata* in longevity of fourth instar larvae, pupae and adult in *Cx. molestus*

Table (7) shows the effect of cold water extract on roots, stalks and leaves of peppermint plant in the growth rate the fourth instar larvae, pupae and adultit reached at the highest concentration (10) mg/ml (2.6, 3 ,-and 4) day, (3, 2,-and-) day, (1, 2,- and -)day respectively compared to(5, 6, 5.5 and 9.2) day, (6, 6, 8.8 and 13.2) day, (3, 6, 7.25 and 9.75) day respectively in controls treatments. when use boiling water extract on roots, stalks and leaves of peppermint plant the longevity reached to (3.8,-,-and-)day, (1, 2, - and -) day, and (1,-,- and -)day respectively compared to (5, 6, 5.5 and 9.2) day, (2.7, 4.3, 6 and 9) days, (3, 5.38, 7.25 and 9.75) day respectively in the control treatments.

Table 7 : Effect the interaction of the water extract of *M. spicata* in longevity of fourth instar larvae, pupae and adult in *Cx. molestus*

Concentrations mg/ml	**cold aqueous extract/duration of growth of insect stages(day)**									**boiled aqueous extract/duration of growth of insect stages(day)**								
	Fourth-instar larvae			**pupae**			**Male and female**			**Fourth-instar larvae**			**pupae**			**Male and female**		
	roots	**stems**	**leaves**	**roots**	**stems**	**leaves**	**roots**	**stems**	**leaves**	**roots**	**stems**	**leaves**	**roots**	**stems**	**leaves**	**roots**	**Stems**	**leaves**
Control	5	6	3	6	6	6	5.5/9.2	8.8/13.2	7.25/9.75	5	2.7	3	6	4.3	5.38	5.5/9.2	6/9	7.25/9.75
1	4.3	2.6	2.3	2.6	5.5	5.5	3.4/11	5.3/18.25	8/6	1.3	1.5	1.9	2.57	3	3.25	5.6/10.5	5.5/9.5	10/-
5	3	2	2.29	2	1	1	3.5/0	11/0	0/8	2	2	1.43	5	2.14	2	5.4/-	-/-	-/5
10	2.6	3	1	3	2	2/	-/4	-/-	-/-	3.8	1	1	-	2	-	-/-	-/-	-/-
L.S.D. interaction between effect of difference of the type of extract and type of plant part in period of the fourth-instar larvae=0.912,pupae=1.21 , male and female adult non-significant difference.																		

Table (7) shows a decrease in the age of the fourth instar larvae and pupae and adult stage of insect in most of the extract concentrations. The results of the statistical analysis showed the significance of the differences in the obtained results

A study [40] confirmed the effect of water extracts of *Peganum harmala* and *R. communis* in the growth of the different stages of the *Cx. pipiensmolestus* the longevity of larvae, pupae and adult stage reached to (10.1, 3.5 and 15.7) day respectively in the concentration of 2 g / kg where as In the concentration of 5 g/ml, the longevity of insect stages was (10.5, 3.5 and 16.3) day and in concentration 10 gm / ml the stage longevity was (11.3, 3.9 and 17.2) day, respectively, compared to the control treatment of (9.5, 3 and 14.1) day. The longivity of the larval, pupal and adult stage in the castor plant when use 2 gm / ml was (10.1, 3.5 and 14.7) day, and in the 5 gm / mL concentration the longivity of insect stages was(10.4, 3.6 and 16.5) day and in concentration 10gm / ml The longevity reached to (10.7, 3.8 and 17.2) day, respectively with control treatment (9.7, 3.2 and 14.7) day.

The reason of increase of longevity in different stages of insect when treated with water extract from peppermint plant may be due to inhibition of the oxidative phosphorylation of the gut tissue in some *lepidoptera* insects [41].

8. Effect the interaction of concentrations of chloroform extract of *M. spicata* in the longevity of fourth instar larvae, pupae and adult in *Cx. molestus*

Table (8) shows the effect of chloroform extract on roots, stalks and leaves of peppermint plants in longevity of fourth instar larvae, pupae and adultit reached at the highest concentrations (1) mg / ml (1, -,-and-) day, and (1.16, 2, 2, 4) day, and (1,-,-,-) day respectively compared to (2, 6, 7.9 and 9.2) days in the control treatment.

Table 8 : Effect the interaction of the chloroform extract of *M. spicata* in longevity of fourth instar larvae, pupae and adult in *Cx. molestus*

Concentrations mg/ml	Fourth-instar larvae			pupae			Male and female		
	roots	stems	leaves	roots	stems	leaves	roots	stems	leaves
Control	2	2	2	6	6	2	9.2/7.9	9.2/7.9	9.2/7.9
0.25	2.25	2.18	1.6	5	2.5	8	7/5	8/3.2	8/2.6
0.5	2	1.6	1.5	-	2.5	3	-/-	5.5/2	5.3/3
1	1	1.16	1	-	2	-	-/-	4/2	-/-
L.S.D. interaction between effect of difference of the type of extract and type of plant part in period of the fourth-instar larvae=1.3,pupae=2.001 , male=0.912 and female=2.41.									

Table (8) shows a decrease in the age of the fourth larval instar and pupae and adult stage of the insect in most concentrations of peppermint extract. The results of the statistical analysis showed the significance of the differences in the obtained results

A study [40] confirmed the effect of the organic extracts of *P. harmala* and *C. communis* in the growth period of the different roles of mosquitoes *Cx. pipiensmolestus*. The longivity of the larval and pupal and adult stage when use the concentrate 2 g/ml (12.2, 3.7 and 18.3). In the concentration, 5 g/ml was the longevity of different stages of the insect was (13.0, 4.1 and 20.1) day and in concentration 10gm/ml reached to (13.7,4.6 and 21.8)day respectively, compared with control treatment (9.5, 3.0 and 14.1) day. The longevity of larval, pupal and adult stage when use castor plant with concentration 2 gm (10.0, 3.3 and 12.3) day and (10.8, 3.7 and 16.6). In the 10 g/mL concentration, and reached to (11.5, 4.0, 17.8) day respectively compare with (9.7, 3.2, 14.7) in control treatment

9. Effect the interaction of concentrations of cold and boiling water extract of *M. spicata* in the percentage of adults emergence inhibition

Cold and boiling water extracts of *M. spicata* leaves, stalks and roots significantly affected the percentage of adults emergence inhibition of mosquito *Cx. molestus* and in different concentrations used. Table (4-12) showed, The percentage of inhibition in adults emergence in the cold water extract for roots, stalks and leaves of peppermint in highest concentration (10) mg/ml reached to (95.4, 100 and 100)% respectively compared with (13, 33 and 30)% in control treatment, and in boiling water extract reached to (77.02, 100 and 100)% respectively in the same concentration compared with (13, 10 and 33)% respectively in control treatment. The results of Statistical analysis showed the significant differences in the results.

Table 9: Effect of the interaction of cold and boiling water extract of *M. spicata* in the percentage of adults emergence inhibition in mosquitoes *Cx. molestus*

Concentrations mg/ml	Cold aqueous extract						Boiled aqueous extract					
	%Inhibition of emergence			Emergence%			%Inhibition of emergence			%emergence		
	root	stem	leaves	root	stem	leaves	root	stem	leaves	root	stem	leaves
Control	13	33	30	87	67	70	13	10	33	87	90	67
1	8.05	40.30	75.72	80	40	17	14.95	55.56	40.30	74	40	40
5	22.9	89.56	94.29	67	7	4	31.04	100	74.11	60	0	14
10	95.4	100	100	4	0	0	77.02	100	100	20	0	0
L.S.D. interaction between effect of difference of the type of extract and type of plant part in percentage of inhibition of emergence =5.624 , emergence =3.625												

It was observed in the table (10) that peppermint extract had an effect on the percentage of inhibition of adult emergence, and the cold water extract was superior to the extract of the boiling water.

A study [22] showed the effect of water extract of leaves of *Calotropis procera* Ait. that used in mosquitoes control , with LC50 (273.53, 366.44 and 454.99) ppm when treated second, third and fourth larval instar respectively in *An arabiensis* ,in concentrations (200, 400, 600, 800 and 1000) ppm and in *Cx. quinquefasciatus* (187.93, 218.27 and 264.85) ppm respectively in treated third and fourth larval instar in concentrations (100, 200, 300, 400, 500 and 600) ppm, IE50 value was (277.90 and 183.65) ppm, respectively.

The difference may be due to differences in the types of plants used in the study or the insect species, or the difference may be due to the polarity of the solvent that used. The polarity index of water (9) and chloroform (4.1) [42] Lowering the polarity of the solvent relative to another solvent may increase its effectiveness in dissolving the active compounds [43].

10. Effect the interaction of concentrations of chloroform extract of *M. spicata* in the percentage of adults emergence inhibition

The results of Table 11 showed that the mint extract had an effect on the percentage of adult emergence inhibition. The highest percentage of inhibition of adult emergence in chloroform extract of the roots, stalks and leaves of peppermint plant at the highest concentration of extract(1) mg / ml was (100, 81.82, 100)% compared with (23)% in control treatment. The results of the statistical analysis showed the significance differences in results.

Table 10: Effect the interaction of the chloroform extract of *M. spicata* in the percentage of adults emergence inhibition in mosquitoes *Cx. molestus*

Concentrations mg/m	%Inhibition of emergence			Emergence%		
	Root	stem	leaves	root	stem	leaves
Control	23	23	23	77	77	77
0.25	48.06	4	74.03	40	74	20
0.5	100	39	78	0	47	17
1	100	81.82	100	0	14	0
L.S.D. interaction between effect of difference of the type of extract and type of plant part in percentage of inhibition of emergence =1.062 , emergence =4.200						

References

[1] Ilango, S.; Athirstalaxmi, D.; Manonmani, P. and Malarvizhi, M. (2016). Repellency of aromatic plants based ingredients against mosquitoes. *Int. J. Appl. Biol. Pharm. Tech.* 7(2): 55-61.

[2] Kassim, N.F.A.; Webb, C.E. and Russell, R.C. (2012). The importance of males: larval diet and adult suger feeding influences reproduction in *Culex molestus*. *J. Amir. Mosq.Contr. Associ*., 28(4):312-316.

[3] Abul-hab, J.K. (1979). Medical and Veterinary insects in Iraq (Theoretical section). Baghdad University press.450 p.

[4] Service, M. (2012). Medical Entomology for students. 5th edition. Cambridge University Press. 303 pp.

[5] Wilkerson, R.C.; Linton, Y.M.; Fonseca, D.M.; Schuftz, T.R.; Price, D.C. and Strickman, D.A. (2015). Making mosquito taxonomy useful :a stable classification of tribe aedini that balances utility with current knowledge of evolutionary relationships .*PLOS ONE*.10 (7).638p.

[6] Traboulsi, A.F.; El-Haj, S.; Tueni, M.; Taoubi, K.; AbiNader, N. and Mrad, N. (2005). Repellency and toxicity of aromatic plant extracts against the mosquito *Culexpipiens molestus* (Diptera : Culicidae). *Pest Manag Sci*. 61: 597-604.

[7] Alahmed, A.M.; AlKuriji, M.A.; Kheir, S.M.; AlHaqawi, H.M. and Sultan, E.A.A. (2010). Mosquitoes (Diptera: Culicidae) and their seasonal activity in Jazan region, Saudi Arabia. J. Saudi Soc. Agr. 9(2a).

[8] Fodeal, N.K. (2014). Bioeffect of two strains of *Bacillus thuringensis* and growth regulator Applaud and pesticide Abte to control of mosquito insect *Culex molestus* Forskal (Diptera : Culicidae)in the province of Karbala. Thesis. Science in Agriculture - plant protrction. 75 pp.

[9] Al-Jinabi, A.A. and Al-Essa, R.A. (2013). Effect of water and alcohol extract from *Nerium oleander* L. s *Meloidogyne incognita* and some in control nematode larvae non-target organisms. J. Univer. Kerb., 11(3).

[10] Al-Kahfagi, H.S.; Al-Essa, R.A. and Al-Rubiei, T.H.T. (2017). Study of repellent and attractant effect of aqueous and ethanolic of same plant extracts against adults of house flies *Musa domistica* (Diptera : Muscidae). J. Univer. Kerb. 15(1).

[12] Habiba, B. (2010). The medicinal plants traded in the northern region of the state of Setif are anatomical studies of genera Mentha, and bactericidal bacteria against their essential oils. Thesis. college of science. University of Farhat Abbas. 116p.

[13] Al-Mansour, N.A.A. (1995). Effect of different extracts of *Ibiceilalutea* in biological aspects in *Bemisatabaci*. PhD. College of science, University of Basra.

[14] Harborne, J.B. (1973). Phytochemical methods. Halsted Press. John and Wiley Sons New York. 278 PP.

[15] Samurai, Kh.W. (1983). The distribution of alkaloids and their taxonomic importance in some wild species of the family Solanaceae in Iraq .Thesis. colloge of science /Baghdad University.157p.

[16] Abul-hab, J. and Kassal, S. (1989). Lecture notes; Regional training center, Advanced course in mosquito control.

[17] Hayes, J.A. (1973). Study of *Culexpipiens* Linnaeus. In the Ohio Mississippi river basin in relation to St. Louis encephalitis (SLE). *Mosq. News*. 33(2): 447-454.

[18] Abbott, W.S. (1925). Amethod of computing the effectiveness of an insecticide. J. Econ. Entomol. 18: 265-267.

[19] Mulla, M.S.; Darwazeh, H.A. and Norland, R.L. (1974). Insect growth regulation : evaluation procedures and activity against mosquitoes. J. Ecom. Entomol., 67(3): 329-332.

[20] AlRawi, K.M. and Khalf Allah, A.M. (2000). Design and Analysis of Experiments Agricultural. Ministry of Higher Education and Scientific Reserarch. Dar Al-Kutub for Printing and Publishing. University of Mosul. Ed. 2nd .488 pp.

[21] Khafaji, A.A. (2004). Effect of extracts in some biological aspects of *Culexpipiens* L. (Diptera: Culicidae). Thesis college of science, University of Kufa.

[22] Elimam, A.M.; Elmalik, K.H. and Ali, F.S. (2009). Efficacy of leaves extract of *Calotroppis procera* Ait. (Asclepiadaceae) in controlling Anopheles arabiensis and *Culexquinquefasciatus mosquitoes*. Saudi J. Biol.Sci.16(2):95-100.

[23] Al-Badairi, F.SH.I. (2017). Effect of water and alcohol extracts of leaves *Ricinus communis* L. in the life of *Culexpipiens* L.(Diptera: Culucidae). search BA. college of science. University of Qadisiyah. 36 p.

[24] Harborne, J.B. (1984). Phytochemical methods .Chapman and Hall. New York. 2nd ed. 288pp.

[25] Kuusik, A.; Tartes, U.; Vanatoa, A.; Metspalu, L. and Hiiesaar, K. (2001). Body stereotypic movements and their possible role as triggers of heart activity in pupae of Colorado potato beetle *Leptinotarsa decemlineata* (Say). Physiological Entomology, 26(2):152-158.

[26] Al-Rubaie, H.M.K. (1999). Effect of extract of *Datura innoxia* Mill. in some biological aspects of *Musca domestica* L. (Diptera: Muscidae). Ph.D. college of science. University of Babylon. 126p.

[27] Wigglesworth, V.B. (1972). The principle of insects physiology chapman and Hall, London, 872 pp.

[28] Chalabi, B.M. (1998). Effect of extracts of *Euphorbia granulate* in biological life of *Culex pipiens* (Diptera : Culicidae). PhD. college of science. Baghdad University.216p.

[29] Evans, D.A. and Kaleysa, R.R. (1992). Effect of quassin on the metabolism of catechol amines different life cycle stages of *Culex quinquefasciatus* . Indian. J. Biochem. Biophys., 29(4): 360-363.

[30] Hamza, A.K. (2001). The study of the repellents effect of extracts of three species of plants against C*ulex pipiens* (Diptera :Culicidae). Thesis. college of Education. University of Qadisiyah.107p.

[31] Traboulsi, A.F.; Taoubi, K.; El-Haj, S.; Bessiere, J.M. and Rammal, S. (2002). Insecticidal properties of essential plant oils against the mosquito *Culex pipiensmolestus* (Diptera : Culicidae). Pest Manag. Sci., 58: 491-495.

[32] Abdul Hamid, Z.H. and Abdul Majid, M.I. (1988). Modren Trends in Pesticides and Pest Control. Part 2: Environmental Presence and Integrated Control. Dar Al-Arabia for Publishing and Distribution/605 p.

[33] Narayanan, P.C.S.; Devi, R.K.; Pushpalatha, M.; Raveen, R. and Lingathurai, S. (2014). Bio-Efficacy of *Terminalia chebula* Retz. (Combretaceae) against *Culex quinquefasciatus* Say (Diptera : Culicidae). Biosci. Plant Biol. 1(3): 41-49.

[34] Saleh, Th.A.; Abid T.M. and Thaker, Abid A. (2010). The accumulative and non accumulative effects of water and some organic extracts of *Eucalyptus microtheca* and *Datura innoxia* on the larvae of *Culex quinquefasciatus*. J. Agri. Sci., 8(4): 321-333.

[35] Halawa, Z.A.; Mohamed, R.A. and El-Kashlan, I.I. (1998). Labaratory evaluation plant and insecticides against beetle *Callobruchus maculates* infesting stored product. Eygpt. J. Agr. Res. 79(1): 85-93.

[36] Frankel, G. (1969). Evalution of our thought on secondary plant substance. Entomol. Exp. Appli. 12: 473-486.

[37] Pederson, M.W.; Barner, D.K.; Sorensen, E.I.; Griffin, G.P.; Nickon, M. and Howath, E. (1976). Effect of low and high saponin selection in alfa on organomic and pest Resistance traits and the interrelationship of these traits. Crop. Sci., 15: 254-256.

[38] Metspalu, L.; Hiiesaar, K.; Joudu, J. and Kuusik, A. (2001). The effects of certain toxic plant extracts on the larvae of Colorado potato beetle *Leptin trasadecemlineala* (Say). Institute of plant protection, Estonian Agriculture University, 93-100.

[39] AL-fetlawy, G.A.A. (2014). Effect of crude secondary compound extracts for *Tamarix ramosissma* on some biological aspects of *Culex pipiens* L. (Diptera :Culicidae) and study their role in transmission of hepatitis C virus. thesis. college of Science for Women, University of Babylon.102.

[40] Mahdi, N.Kh. (2010). The effects of aquous extracts and organic solvents of *Peganum harmala* and *Ricinus communison* the different instars of mosquito *Culex pipiensmolestus*. J. Tik. Univer. Agri. Sci. 10(2): 119-128.

[41] Tangiuch, M.; Yamaguchi, M.; Kubo, I. and Kubota, T. (1979). Inhibitory effect of isodonditerpenoids on growth and mitochondrial oxidative phosphorylation in lepidopterous insects. Agric. Biol. 43(1): 71-74.(Abstract).

[42] Gailliot, F.G. (1998). Initial Extraction and Product capture. In : Cannell, R.J.P.(Eds.) Natural Products Isolation. Methods in Biotechnology . Humana Press. Totowa, New Jersey. 4: 53-109.

[43] Alwan, A.A.; Mansour, N.A. and Salim, A.H. (2011). Effect of some plant extracts on the mortality of larvae *Culex pipiensmolestus* Forskal. J. Basra Sci., 29(1): 47-61.