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LARVICIDAL POTENIAL OF DIFFRENT PLANTS EXTRACTS AGAINST THE LARVAE OF MOSQUITO (*AEDES AEGYPTI* (L.) (CULICIDAE: DIPTERA).

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ABSTRACT

Larvicidal potential of five indigenous plants extracts viz., *Eucalyptus globulus*, *Calotropis gigantea*, *Coriandrum sativum*, *Mentha* sp. and *Azadirachta indica* against *Aedes aegypti* larvae were studied in the present work. The methanol extract of incubator dry leaves were used for the study. Different concentrations were made of all these extracts and 20-25 larvae were placed in different concentrations for different time of intervals. After 24 hrs of exposure, 15-45% mortality was observed at different concentrations of *A. indica* followed by *C. sativum* (10-35%) and *C. gigentea* (5-30%). After 48 hrs of exposure, 35-70 % mortality was observed at different concentrations of *A. indica* followed by *C. sativum* (20-60%) and *E. globulus* (20-30%). The results revealed that the extract of *A. indica* proved to be the most toxic against the tested larvae. The results can be helpful in devising an effective environment friendly approach for the management of *Ae. Aegypti* larvae. However, further studies are required to check the efficacy of these plant extracts under field conditions.

Keywords: Biopesticde, *Aedes aegypti,* Mosquito, Plant extracts, Larvicidal

INTRODUCTION

During the study by different scientists, it has been proved that mosquitoes are sources of many diseases as compared to other arthropod groups. After the survey and seeing the effect of mosquito, WHO declared it as number one across the globe (Lopez and Pascual, 2010). Due to dangerous effects of the mosquito from last few years, a lot of synthetic chemicals for mosquito managing program have been restricted due to the deficiency of introducing of new insecticides, high price of insecticides and negative effects on environment and human beings (Russell *et al.,* 2009).

The mosquito which is famous as a yellow fever mosquito was irritating species in the United State. In most of the areas, *Ae. aegypti* is known as primary and major vector of yellow fever. It is common in tropical south America and Africa in summer months. During war between Spain and USA, USA army faced yellow fever which was transmitted by *Ae. aegypti* (Tabachnick, 1991). The larva of this mosquito is known as wigglers, for the reason that they come into view to wiggle at irregular intervals in the water after disturbing. Larval stages of *Ae. aegypti* spent their time on the water surface if anybody disturb them, they move towards the bottom of container. The larvae can be separated from other genera by naked eye by their short siphon (Nelson, 1986).

On the basis of previous information, it can be concluded that use of pesticides has steadily increased by about 10 % (Bailey and Mupondwa, 2006). No doubt the control through chemical pesticides showed the good effects against the pest and disease, but it also produces the resistance in the pest and diseases.The use of chemical disturbed both the environment human health and non target animals (Shen and Zhang, 2002.

Many natural materials derived from plants are called bio-pesticides. Bio-pesticides are taking place in all over the

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world as new technique for the control of insect pest. The biopesticde have less risk to people and environment than synthetic pesticides (Sexena and Panday, 2001).

During the last few years, this mosquito caused sever epidemic of dengue fever in Pakistan. So, instead of using synthetic larvicides, the use of plant- derived products for controlling mosquito larvae are inexpensive and environment- friendly (Das NG *et al.,* 2007). In this study different plant extracts were evaluated control *Ae. aegypti.*

MATERIALS AND METHODS

Selection of plants

The experiment was carried out at the Institute of Agricultural Sciences, University of the Punjab and Government College University Faisalabad in Randomized Complete Block (RCBD) design. Five plants Eucalyptus globules, *Calotropis gigantean*, *Coriandrum sativum*, *Mentha* sp. and *Azadirachta indica* were used in this experiment.

Procedure of extraction

Fresh leaves of selected plants were taken and rinsed with water for further extraction and preparation of concentrations. All the dust and debris were removed from the leaves. After washing, the leaves were placed in incubator at 30°C for two days for drying. The dried leaves were grinded in to powder form with the help of grinder machine. For better extraction, a 100 ml of methanol was used for the preparation of extracts. Conical flask was used for this purpose. Leaves powder and methanol were mixed together, placed in the conical flask and covered with aluminum foil. The flask was agitated at 100 oscillations for 1 min a day. For better shaking, refrigerator shaking incubator was used for the whole night at the temperature of 30°C. The oscillation produced suspension which was filtered with the help of filter paper. The extract was separated by keeping the solution on magnetic stirrer. After obtaining the extraction, distilled water was used to form different concentrations.

Preparation of stock solution

The 10% stock solution was prepared from the plant material. For this purpose 2.5 gram extract of leaves was taken and mixed with 25 ml water, to prepare the solution. Different dilutions (Table 1-5) of solution were prepared from stock solution.

Table 1

Different concentrations prepared for *Calotropis gigantea.*

Dilutions (%)	Stock solution (ml)	Distilled water (ml)
5	1	19
15	3	17
25	5	15
35	7	18
40	10	19

Table 2

Different concentrations prepared for *Eucalyptus globulus.*

Dilutions (%)	Stock solution (ml)	Distilled water (ml)
10	3	15
20	5	18
30	8	20
40	10	25
50	15	30

Table 3

Different concentrations prepared for *Coriandrum sativum*

Dilutions (%)	Stock solution (ml)	Distilled water (ml)
5	3	15
10	5 1	18
15	7 1	19
25	10	17
30	15	20

Table 4

Different concentrations prepared for *Mentha* sp.

Dilutions (%)	Stock solution (ml)	Distilled water (ml)
10	5	18
15	8l	19
20	10	17
30	15	18
40	20	20

Table 5

Different concentrations prepared for *Azadirachta indica.*

Dilutions (%)	Stock solution (ml)	Distilled water (ml)
5	1	19
15	3	17
25	5	15
35	7	18
40	10	19

SUSCEPTIBILITY BIOASSAY

Material required

- 2-3 White papers
- 10 Disposable plastic cup
- Permanent black marker
- Larvae of *Aedes aegypti*

Procedure of bioassay

Disposable plastic cups were taken, washed and dried properly to check the mortality rate of larvae of *Ae. aegypti*. For this purpose, two white papers were taken and cut into pieces according to the bottom of plastic cups. Different dilutions of each extracts were made from stock solution. Pre-determined number of larvae (20-25 larvae) were dipped in solution in the cup for different time hours and mortality of larvae was evaluated after different intervals of time.

RESULTS AND DISCUSSION

The results of the percent mortality data after 24 and 48 hours of exposure to different plant extracts revealed that the extracts of *A. indica* proved the most toxic to the mosquito larvae at all the concentrations (Table 6-7). After 24 hrs of exposure, 15-45% mortality was observed at different concentrations of *A. indica* followed by *C. sativum* (10-35%) and *C. gigentea* (5-30%) (Table 6). After 48 hrs of exposure, 35-70 % mortality was observed at different concentrations of *A. indica* followed by *C. sativum* (20-60%) and *E. globulus* (20 -30%) (Table 7).

The use of synthetic insecticides for the management of insect pests of agricultural and public health importance has created many problems like the development of insecticide resistance, environmental pollution (Khan *et al.,* 2016a, b, 2017). To overcome these issues, researchers around the world have recommended many tools like integrated vector management, use of protective measures and the replacement of synthetic insecticides with bio-rational or plant based insecticide for the management of different insect pests (Khan *et al.,* 2016c). In the present study, larvicidal potential of different plant extracts have been evaluated and *A. indica* proved the most toxic against the tested larvae of *Ae. Albopictus*. Currently, mosquito control all over the world focuses on targeting larval habitats by using different plant based insecticides. Since the larval stages are confined to the water habitats and not able to fly, therefore, targeting this

Table 6

Percent mortality of *Aedes aegypti* larvae (4th instars) after 24 hours of exposure to different plant extracts.

Conc. %	*Calotropis gigantean*	*Eucalyptus globulus*	*Coriandrum sativum*	*Mentha*	*Azadirachta indica*
10	5b	5b	10b	0a	15a
20	10b	5b	15b	5b	25a
30	20b	15c	25b	15c	30a
40	25b	20c	30b	20c	40a
50	30b	25c	35b	20c	45a
Control	0a	0a	0a	0a	0a

Mean percent mortality values within a row with different letters are significantly different by applying one-way ANOVA and LSD test.

Table 7

Percent mortality of *Aedes aegypti* larvae (4th instars) after 48 hours of exposure to different plant extracts.

Conc. %	*Calotropis gigantean*	*Eucalyptus globules*	*Coriandrum sativum*	*Mentha*	*Azadirachta indica*
10	15b	20b	20b	10b	35b
20	25c	20b	25b	20b	45b
30	30d	35c	35b	25bc	50b
40	35e	40d	45b	25bc	60b
50	45f	50e	60b	35c	70b
Control	0b	0b	0a	5a	0a

Mean percent mortality values within a row with different letters are significantly different by applying one-way ANOVA and LSD test.

stage is very helpful in the management of mosquitoes. Moreover, using chemicals in the water habitats also reduces air-borne residues and ultimately reduces the chances of inhalation. Keeping in view environmental concerns and resistance issues, plant based insecticides are very helpful in managing mosquitoes and environmental health (Din *et al.,* 2011).

There are some previous reports from Pakistan which also proved the potential of the different plant based extracts against larvae of *Ae. Albopictus*. For example, citrus based plant extracts have a strong larvicidal potential against *Ae. Albopictus* (Akram *et al.,* 2010, Singh *et al.,* 2003). Neem oil has also been reported as very effective against mosquitoes from the genera *Aedes*, *Anopheles* and *Culex* (Dua *et al.,* 2009).

In short, the results of the present investigation revealed the potential of some plant extracts which could be helpful in devising an effective and environment friendly approach for the management of dengue mosquitoes. On the basis of the present investigation, it can be concluded that *A. indica* followed by *C. sativum* and *C. gigentea* contains potent larvicidal bioactive molecules which need further purifications to have its synthetic analogues.

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