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Mosquito Larvicidal Effects of 33 Plants Aqueous Extracts of 14 Different Plants Against Larva of Culex Mosquito

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Abstract: ***In this study, the mosquito larvicidal activity of 33 plant's aqueous extracts of 14 different plants were studied against the 4th instar larva of Culex quinquefasciatus mosquito. Different concentration of these plant aqueous extracts for different time duration was assessed on C. quinquefasciatus but the larvae showed negligible effects. These observation suggest that a very negligible better to say no larvicidal activity seen by these plant's aqueous extract against Culex quinquefasciatus.***
Keywords: Mosquito larvicidal effect, Culex quinquefasciatus, Plant's aqueous extract, 4th instar mosquito, Mosquito Larva

I. INTRODUCTION

Every plant that we see around us is full of one or the other quality and each one has some or the other important medicinal properties. In addition to providing us with oxygen, trees and plants are essential to the meaningful growth of the entire planet and hence, their importance in our life is inexhaustible.

The 14 different plants chosen was *Alternanthera philoxeroides, Kalanchoe pinnata, Opuntia elatior, Centratherum punctatum, Colocasia esculenta, Cordyline fruticose, Cyperus eragrostis, Duranta, Euphorbia tithymeloides, Ixora coccinea, Jatropha gossypiifolia, Martynia annua, Persicaria longiseta, Stachyterpheta indica* from which different parts 33 aqueous extracts were formed to study mosquito larvicidal effects.

The most significant group of blood-sucking arthropods is the mosquito family. They not only irritate people by biting, but they can spread dangerous infections that affect a wide range of socioeconomic consequences. Global climatic changes are to blame for the situation's worsening during the past ten years.

This has helped them, along with other variables, to adapt to a variety of habitats and consequently grow their population in various regions of the world. Many harmful organisms producing diseases like Malaria, Filariasis, Japanese Encephalitis, Dengue fever, Yellow fever, etc. are transmitted by several mosquito species belonging to the genera Anopheles, Culex, and Aedes. These diseases are contagious on a global scale, leading to high rates of human death and obstructing the economic growth of the majority of developing nations worldwide. It has been attempted to reduce or completely eradicate mosquito populations by using a variety of insecticides and chemical compositions. These pesticides are threatened by mosquitoes developing resistance to chemical insecticides, which leads to rebounding vectorial capacity even though they are very effective against the target species. Many environmental and human health problems have been sparked by the long-term stability of many of these pesticides and their propensity to bioaccumulate in creatures that are not their intended targets. Due to their abundance in bioactive chemicals, ease of availability, environmental safety, etc., plant secondary metabolites are viewed as a promising alternative strategy against numerous mosquito species and their various juvenile stages.

The activity of crude aqueous extracts from 14 different plants of Jharkhand, Hazaribag were investigated in the current study as part of ongoing efforts to find plant extracts with mosquito larvicidal properties. The results of the investigations in this area will be helpful in encouraging research aimed at the creation of new mosquito-controlling substances derived from local plant sources.

II. MATERIAL AND METHODS

A. Collection of Plants

Plants collected from the area of Ranchi and Hazaribag of Jharkhand, India. All these were then brought to the Univesity Department of Biotechnology, Vinoba Bhave University, Hazaribag. All the plant parts were first washed by tap water and then by double distilled water three to four times and then kept for drying either in shaded area or in hot air oven at 40^{O}c till plat parts become dry. The details of collection of plants is summarised in Table-1.

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B. Plant's Extract Formation

The process of maceration was used for plant's extract formation. Details are given in Table-2.

C. Plant Extract Concentration

Concentration of all plant extract for the Mosquito Larvicidal assay was made 30mg/ml.

D. Mosquito Culture

For growing mosquito larvae, four old tyres were cut, placed in the garden at shaded area and was filled with water. It was then kept undisturbed for some days and wait for female mosquito to lay egg. After some days (3-4 days) small mosquito larvae were seen floating on the water surface. According to the floating pattern and resting position on the surface of water, the larvae of Culex mosquito was selected out at early 4th instar and taken for larvicidal assay. Difference between the larvae of Culex, Aedes and Anopheles mosquito is given in Table:3 below, which is used for screening of Culex mosquito larvae.

TABLE: 1
DETAILS OF PLANT COLLECTION AND IT'S FAMILY

Sl.No.	NAME OF PLANTS		Site of Collection / Place	Month-Year of Collecton	Lattitude	Longitude	Plant's Family
	SCIENTIFIC NAME	LOCAL NAME					
1	*Alternanthera philoxeroides*	Alligator weed	Hazaribag	Jul-22	24.02116512	85.38259052	Amaranthaceae
2	*Kalanchoe pinnata*	Pattharchatta	Ratu Road, Ranchi	Jul-22	23.379474	85.303406	Crassulaceae
3	*Opuntia elatior*	Nagfani, Red flower prickly pear	Hazaribag	Jul-22	24.02077681	85.38453244	Cactaceae
4	*Centratherum punctatum*	Lark Daisy	Hazaribag	Jul-22	24.02077681	85.38453244	Asteraceae
5	*Colocasia esculenta*	Arabi, Kachchu	Hazaribag	Jul-22	24.02077681	85.38453244	Araceae
6	*Cordyline fruticosa*	T - Plant	Ratu Road, Ranchi	Jul-22	23.379474	85.303406	Asparagaceae
7	*Cyperus eragrostis*	Nutgrass	Hazaribag	Jul-22	24.02219681	85.3743643	Cyperaceae
8	*Duranta*	Nilkanta	Hazaribag	Jul-22	24.02324405	85.37792767	Verbenaceae
9	*Euphorbia tithymeloides*	Agiya	Hazaribag	Jul-22	24.02808583	85.3742775	Euphorbiaceae
10	*Ixora javanica*	Rukmini	Hazaribag	Jul-22	24.02808583	85.3742775	Rubiaceae
11	*Jatropha gossypiifolia*	Ratanjoti, Bio-Diesel Plant	Hazaribag	Aug-22	24.01623186	85.38004435	Euphorbiaceae
12	*Martynia annua*	Hatha-Jodi, Devil's Claws	Hazaribag	Aug-22	24.01229203	85.3911435	Martyniaceae
13	*Persicaria longiseta*	Oriental lady's thumb	Hazaribag	Aug-22	24.02116512	85.38259052	Polygonaceae
14	*Stachyterpheta indica*	Indian Snakeweed	Hazaribag	Jul-22	24.02003846	85.36962373	Verbenaceae

TABLE: 2
DETAILS OF METHODS OF PLANT EXTRACT PREPARATION AND CONCENTRATION

Sl.No.	Scientific Name	Plant Part Used	Method of Drying	Plant Parts Dry Weight(grams)	Method of Drying of Filterate	Dry Pellet's Weight (mg)	Concentration of Stock Solution (mg/mL)
1	*Alternanthera philoxeroides*	Flower	Hot Air Oven, 40°C	1.21	Hot Air Oven, 40°C	123	30
2	*Alternanthera philoxeroides*	Leaf	"	3	"	468	30
3	*Alternanthera philoxeroides*	Stem	"	3	"	693	30
4	*Bryophyllum*	Leaf	Air Dry	10	"	1988	30
5	*Cactus Opuntia*	Spine	Hot Air Oven, 40°C	8.35	"	121	30
6	*Cactus Opuntia*	Flower	"	5	"	430	30
7	*Cactus Opuntia*	Leaf	"	10	"	2507	30
8	*Cactus Opuntia*	Green Ovary	"	5.955	"	292	30
9	*Centratherum punctatum*	Leaf	"	6	"	635	30
10	*Centratherum punctatum*	Flower	"	5	"	331	30
11	*Colocasia esculenta*	Leaf	"	15	"	1040	30
12	*Colocasia esculenta*	Stem	"	11.46	"	1216	30
13	*Cordyline fruticosa*	Leaf	Air Dry	10	"	1312	30
14	*Cyperus eragrostis*	Leaf	"	4.32	"	199	30
15	*Cyperus eragrostis*	Root	"	5.28	"	216	30
16	*Cyperus eragrostis*	Flower	"	2.13	"	117	30
17	*Duranta*	seed	"	5	"	687	30
18	*Euphorbia tithymeloides*	Leaf	"	7.5	"	1991	30
19	*Euphorbia tithymeloides*	Stem	Hot Air Oven, 40°C	5	"	651	30
20	*Ixora coccinea*	Flower	"	12.61	"	1251	30
21	*Jatropha gossypiifolia*	Leaf	"	10	"	981	30
22	*Jatropha gossypiifolia*	Flower	"	7	"	524	30
23	*Jatropha gossypiifolia*	Fruit Cover	"	5	"	243	30
24	*Jatropha gossypiifolia*	seed	"	8	"	455	30
25	*Jatropha gossypiifolia*	Stem	"	17.6	"	1305	30
26	*Martynia annua*	Flower	"	7.54	"	995	30
27	*Martynia annua*	Leaf	"	10	"	346	30
28	*Martynia annua*	Stem	"	20.1	"	1072	30
29	*Persicaria longiseta*	Leaf	"	5	"	402	30
30	*Persicaria longiseta*	Flower	"	1.149	"	102	30
31	*Stachyterpheta indica*	Leaf	Air Dry	2	"	995	30
32	*Stachyterpheta indica*	Flower Bud	"	20	"	1774	30
33	*Stachyterpheta indica*	Flower	"	1.8	"	355	30

TABLE: 3
DIFFERENCE BETWEEN CULEX, AEDES AND ANOPHELES MOSQUITO LARVAE:

Culex	Aedes	Anopheles
Larvae floats obliquely to the surface of water	Larvae floats horizontally to the surface of water	Larvae floats obliquely to the surface of water
It rests at a certain angle to the water surface	It rests parallel to the water surface	It rests at a certain angle to the water surface

III. LARVICIDAL ACTIVITY

With some modification, in "WHO/CDS/WHOPES/GCDPP/2005.13 GUIDELINES FOR LABORATORY AND FIELD TESTING OF MOSQUITO LARVICIDES", mosquito larvicidal experimentation was designed. The mosquito was feed with aquarium diet[12]. Five larvae, each were introduced into treatment container/beaker containing 25mL of their natural growth media. The efficacy was determined through[13] bioassay method. Different volume of plant extract (Stock concentration 30mg/mL) was added in beaker to get different concentration (20, 40, 80, 160, 320, 640, 1280, 2560 ug/ml) of plant extract. All these concentrations are maintained at triplicate so as to do statistical analysis. A control was also established for treatment set where the water on which larvae were grown was used. Larval mortality counts were checked at predetermined intervals, including 6, 12, 24, and 48 hours following treatment. Larvae were considered to be dead if they sank to the bottom of the treatment tray, stayed still, showed no response to light or sound, or failed to regain life functions even after being moved to a control water solution.

IV. RESULT AND DISCUSSION

Observation obtained after doing larvicidal assay for all the concentarion for 6, 12, 24, and 48 hours is summarised in Table:4. *Alternanthera philoxeroides* flower showed 40% larvicidal effect at 320µg/ml at 1st 6 hrs but after that no effect was observed even after increasing the concentration. *Opuntia elatior* flower and leaf, *Centratherum punctatum* leaf, *Colocasia esculenta* leaf, *Cyperus eragrostis* root and flower at 320µg/ml showed 20% larvicidal [13]effect at 24th hrs, 6th hrs, 24th hrs, and 48th hrs respectively and also larvicidal effects was not observed by increasing the concentration.

V. CONCLUSION

All the 33 plant's aqueous extract showed no significant mosquito larvicidal effect.

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TABLE: 4
Mosquito Larvicidal Effect by Plant Extracts in 48 Hrs Reading at Different Concentrations

Sl No.	Plant's Name	Plant Part Used	Volume of Water Used (mL)	Number of Larvae in each Plate	Number of Replica in each Concentration	Total Number of Larvae	Concentration (µg/mL)					Concentration (µg/mL)					Concentration (µg/mL)					Concentration (µg/mL)					Concentration (µg/mL)				
							20					40					80					160					320				
							Death of Larvae					Death of Larvae					Death of Larvae					Death of Larvae					Death of Larvae				
							1st 6 Hrs	12 Hrs	24 Hrs	48 Hrs		1st 6 Hrs	12 Hrs	24 Hrs	48 Hrs		1st 6 Hrs	12 Hrs	24 Hrs	48 Hrs		1st 6 Hrs	12 Hrs	24 Hrs	48 Hrs		1st 6 Hrs	12 Hrs	24 Hrs	48 Hrs	
1	*Alternanthera philoxeroides*	Flower	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		2	2	2	2	
2	*Alternanthera philoxeroides*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
3	*Alternanthera philoxeroides*	Stem	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
4	*Bryophyllum*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
5	*Cactus Opuntia*	Spine	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
6	*Cactus Opuntia*	Flower	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	1	1		0	0	0	0	
7	*Cactus Opuntia*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	1	1		0	0	0	0	
8	*Cactus Opuntia*	Green Ovary	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
9	*Centratherum punctatum*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	1	1		1	1	1	1	
10	*Centratherum punctatum*	Flower	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	1	1		1	1	2	2	
11	*Colocasia esculenta*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	1	1	
12	*Colocasia esculenta*	Stem	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	2	2		0	0	2	2	
13	*Cardiline futicosa*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
14	*Cyperus eragrostis*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	2	2	
15	*Cyperus eragrostis*	Root	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	1	
16	*Cyperus eragrostis*	Flower	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	1	1	
17	*Duranta*	seed	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
18	*Euphorbia tithymeloides*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
19	*Euphorbia tithymeloides*	Stem	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
20	*Ixora coccinea*	Flower	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
21	*Jatropha gossypiifolia*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
22	*Jatropha gossypiifolia*	Flower	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
23	*Jatropha gossypiifolia*	Fruit Cover	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
24	*Jatropha gossypiifolia*	seed	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
25	*Jatropha gossypiifolia*	Stem	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
26	*Martynia annua*	Flower	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
27	*Martynia annua*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
28	*Martynia annua*	Stem	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
29	*Persicaria longiseta*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
30	*Persicaria longiseta*	Flower	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
31	*Stachytarpheta*	Leaf	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
32	*Stachytarpheta*	Flower Bud	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	
33	*Stachytarpheta*	Flower	25	5	3	15	0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0		0	0	0	0	

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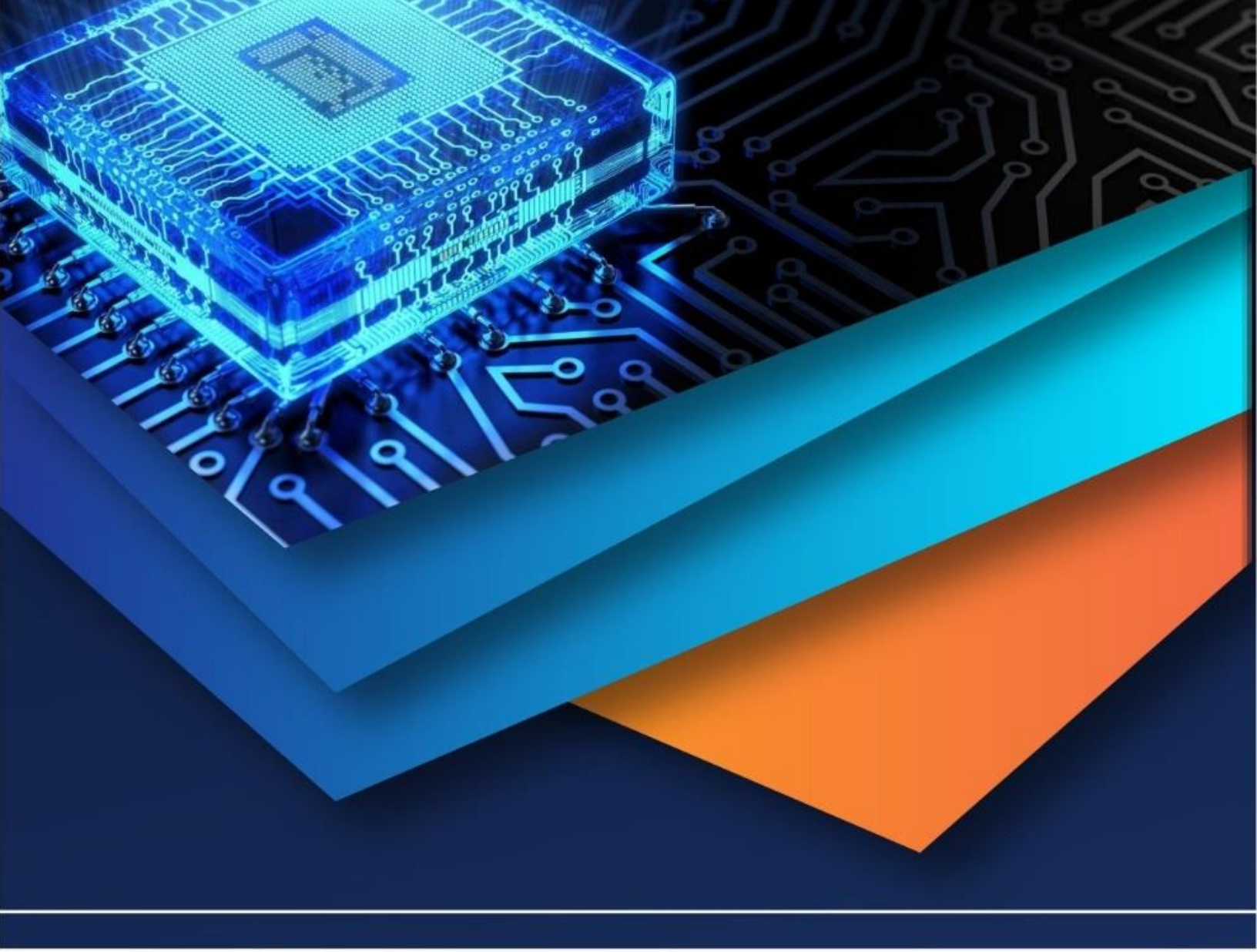

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