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Studies on the insecticidal properties of *Chromolaena odorata* (Asteraceae) against the life cycle of the mosquito, *Aedes aegypti* (Diptera: culicidae).

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ABSTRACT:

Leaf extract of the species, *Chromolaena odorata* was evaluated for the egg hatchability, larvicidal and pupicidal activity of mosquito, *Aedes aegypti* under the room temperature in the laboratory. Dosage value as expressed in ppm was 10 to 140 for *Aedes aegypti*. A relationship was observed between the plant extract doses and percentage mortality. The percentage of egg hatchability, larval and pupal mortality were found to be increased with increase in the dosage. Based on the probit analysis, the L_c50 value of egg (99.15), I instar (42.24), IV instar (101.49) and pupae (121.57) were hence assumed.

Keywords:
Aedes aegypti, Chromolaena odorata, L_c50.

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INTRODUCTION

Vector control is a serious concern in developing countries like India due to lack of general awareness, development of resistance and socio-economic reason. The role of mosquitoes is becoming increasingly important in the recent years because of change in ecology caused by human intervention.

Mosquitoes constitute a major public health menace as vectors of serious human diseases (EI Hag *et al.,* 1999). Of the various mosquito spread diseases, dengue fever transmitted by *Aedes aegypti* is dangerous and is spreading into dengue shock syndrome have taken epidemic form and have been reported from Tamil Nadu, West Bengal, Uttar Pradesh, Gujarat and Delhi (Kebra *et al.,* 1992). In recent years, scientists try a variety of botanical derivatives to eradicate many harmful insect pests including mosquitoes. Insecticidal activity of neem has been reported. Vector control is facing a threat due to the emergence of resistance to synthetic insecticides. Insecticides of botanical origin may serve as suitable alternative biocontrol techniques in the future. (Nandita Chowdhury *et al., 2008).*

Aedes are vectors for the pathogens of various diseases like dengue fever , dengue haemorrhagic fever and yellow fever (Rajmohan and Ramaswamy, 2007). Many authors world wide started large screening activity for using extracts of medicinal and herbaceous plants to control mosquitoes (Halawa, 2001; Das *et al.,* 2003; Choochote *et al.,* 2004; Logankumar *et al.,* 2008). The plant species, *Chromolaena odorata* is a widely distributed neotropical shrub introduced to many parts of the tropics. It is commonly seen in wastelands and hedges on weed.

For the present study, this species was screened against egg hatchability, larval and pupal mortality of the mosquito *Aedes aegypt.* So as to control the population of *Aedes aegypt* by an eco-friendly approach.

MATERIALS AND METHODS

Fresh leaves of *Chromolaena odorata* were collected from the plants growing in agricultural lands. Leaves were washed, shade, air dried and ground in a mixture to form a fine powder. The 25g of the powder was then used for extraction in acetone in soxhlet apparatus. The extract was concentrated on water bath to evaporate the acetone. The filterate was considered as pure material and redissolved in acetone to form standard formulation. By further dilutions with required amount of water, different doses (ppm) were prepared.

Eggs of *Aedes aegypti* were procured from the Research Laboratory of National Institute of Communicable Diseases (NICD), Mettupalayam, Coimbatore and brought to the laboratory and cultured. Eggs, first and fourth larval instars and pupae were harvested from the colony and were placed in different concentrations of biocide. Twenty individuals were used for each concentration. Eggs, larval instars and pupae were checked for mortality every 24 hours. In the case of control only carrier solvent was added. Food was provided in all the test beakers. Each test was replicated for five times.The effect of leaf *Chromolaena odorata* on the egg hatchability, mortality of first and fourth larval instars and pupal mortality of *Aedes aegypti* was studied. Following 24 hours were corrected for natural response by Abbott's formula (Abbott, 1925) as follows:

$$\text{Corrected percentage kill} = \frac{\text{Proportion of test mortality- Proportion of control mortality}}{\text{1- Proportion of control mortality}} \times 100$$

Busvin (1971) suggested that the critical doses of susceptibility can be estimated with sufficient accuracy from a probit / log concentration graph. Based on the log concentration and the probit mortality percentage values, regression equation was obtained. Using the regression, a straight line was fitted. Fitting of regression line and homogeneity of population were also tested employing chi-square (χ^2) test. By graphical interpolation, LC_{50} values of the leaf extract for 24 hours of exposure of egg, first and fourth instar larvae and pupae of *Aedes aegypti* and their fiducial limits (95% upper fiducial limit and lower fiducial limit) were calculated.

RESULTS AND DISCUSSION

Mortality values of egg, larvae and pupae treated with different concentrations (ranging from 10ppm to 140 ppm) of the leaf extract of *Chromolaena odorata* at the end of 24 hrs are given in Table 1-4 for egg, I instar, IV instar larvae and pupae of *Aedes aegypti*. The L_C50 values and their 95% upper and lower fiducial limits, and chi-square value of the leaf extract of *Chromolaena odorata* for 24h exposure of *Aedes aegypti* are given in Table 5. Based on the probit analysis the 24 hr L_C50 value of the leaf extract of *Chromolaena odorata* for egg, I instar and IV instar larvae and pupae of

Table 1. Effect of crude sample of *Chromolaena odorata* against the egg hatchability of *Aedes aegypti.*

No of eggs exposed	Concentration (ppm)									
	70		80		90		100		110	
	h	uh	h	uh	h	uh	h	uh	h	uh
20	17	3	14	6	11	9	6	14	3	17
20	16	4	12	8	10	10	7	13	2	18
20	17	3	11	9	10	10	5	15	3	17
20	18	2	13	7	11	9	4	16	1	19
20	15	5	11	9	10	10	5	15	4	16
Mean	**16.6**	**3.4**	**12.2**	**7.8**	**10.4**	**9.6**	**5.4**	**14.6**	**2.6**	**17.4**
SD	**1.14**	**1.14**	**1.30**	**1.30**	**0.55**	**0.55**	**1.14**	**1.14**	**1.1**	**1.14**
Mean %	**83**	**17**	**61**	**39**	**52**	**48**	**27**	**73**	**4**	**87**
									13	

h - hatched, un - unhatched.

Table 2. Effect of crude sample of *Chromolaena odorata* against the I Instar larvae of *Aedes aegypti.*

No of larvae exposed	Concentration (ppm)									
	10		20		30		40		50	
	Alive	dead	Alive	dead	Alive	dead	Alive	dead	Alive	dead
20	19	1	17	3	12	8	11	9	2	18
20	18	2	15	5	11	9	10	10	4	16
20	19	1	16	4	12	8	10	10	3	17
20	16	4	17	3	13	7	12	8	4	16
20	17	3	15	5	11	9	10	10	5	15
Mean	**17.8**	**2.2**	**16**	**4**	**11.8**	**8.2**	**10.6**	**9.4**	**6.4**	**13.6**
SD	**1.30**	**1.30**	**1.00**	**1.00**	**0.84**	**0.84**	**0.89**	**0.89**	**1.14**	**1.14**
Mean %	**89**	**11**	**80**	**20**	**59**	**41**	**53**	**47**	**32**	**68**

Table 3. Effect of crude sample of *Chromolaena odorata* against the IV instar larvae of *Aedes aegypti.*

No of larvae exposed	Concentration (ppm)									
	80		90		100		110		120	
	Alive	dead	Alive	dead	Alive	dead	Alive	dead	Alive	dead
20	19	1	15	5	11	9	8	12	3	17
20	18	2	12	8	10	10	9	11	2	18
20	16	4	11	9	10	10	8	12	1	19
20	19	1	13	7	11	9	7	13	2	18
20	17	3	12	8	12	8	6	14	4	16
Mean	**17.8**	**2.2**	**12.6**	**7.4**	**10.8**	**9.2**	**7.6**	**2.4**	**2.4**	**17.6**
SD	**1.30**	**1.30**	**1.52**	**1.52**	**0.84**	**0.84**	**1.14**	**1.14**	**1.14**	**1.14**
Mean %	**89**	**11**	**63**	**37**	**54**	**46**	**38**	**62**	**12**	**88**

Aedes aegypti was found to be 99.15, 42.24, 101.49 and 121.57 respectively (Fig.1). These results are in agreeing with the earlier findings made by many workers with botanicals for various properties (for oviposition avoidance, larvicidal, Halawa, 2001; Saleh, 1995, adulticidal, Choochote *et al*., 2004 and repellent activities, Choochote *et al.,* 2004; Prakash *et al*., 2000). As the botanical insecticides for including the extract of *C. odarata* are biodegradable and harmless to the environment, pest – specific and relatively harmless to non-target organisms (Su and Mulla 1998; Sivagnaname and Kalyana Sundaram, 2004; Sun *et al*., 2006) they are more eco-friendly. The results of the present study, indicate that the leaf extract of the weed species, *Chromolaena odorata* caused low percentage of egg hatchability and high percentage of larval and pupal mortality. Hence the large biomass of *C. odarata* available in Southern India can be used by the pharmacological industries to obtain effective repellent to control mosquito population is an ecofriendly manner.

Table 4. Effect of crude sample of *Chromolaena odorata* against the pupae of *Aedes aegypti.*

No of larvae exposed	Concentration (ppm)									
	100		110		120		130		140	
	Alive	dead	Alive	dead	Alive	dead	Alive	dead	Alive	dead
20	18	2	15	5	12	8	9	11	5	15
20	19	1	14	3	10	10	8	12	3	17
20	18	2	11	9	10	10	9	11	2	18
20	17	3	12	8	10	10	6	14	3	17
20	16	4	11	9	11	9	7	13	4	16
Mean	**17.6**	**2.4**	**12.6**	**6.8**	**10.6**	**9.4**	**7.8**	**12.2**	**3.4**	**16.6**
SD	**1.14**	**1.14**	**1.82**	**2.68**	**0.89**	**0.89**	**1.30**	**1.30**	**1.14**	**1.14**
Mean %	**88**	**12**	**63**	**34**	**53**	**47**	**39**	**61**	**17**	**83**

Table 5. 24 hours LC_{50} values (ppm) and their 95 % Fiducial (upper and lower) regression equation and Chi-square (c^2) values of the crude extract of *Chromolaena odorata* for the different developmental stages of *Aedes aegypi.*

Stages	LC_{50} (ppm)	95% Fiducial limit (ppm)		c^2	$\overline{X}$	SD	SE
		Upper	Lower				
Egg	99.15	103.74	95.65	2.57	52.8	24.79	3.88
I instar	42.24	45.58	39.75	6.66	30.2	13.22	11.52
IV instar	101.49	105.38	107.61	4.58	48.8	25.63	5.25
Pupa	121.57	125.43	117.14	4.25	47.4	24.02	3.11

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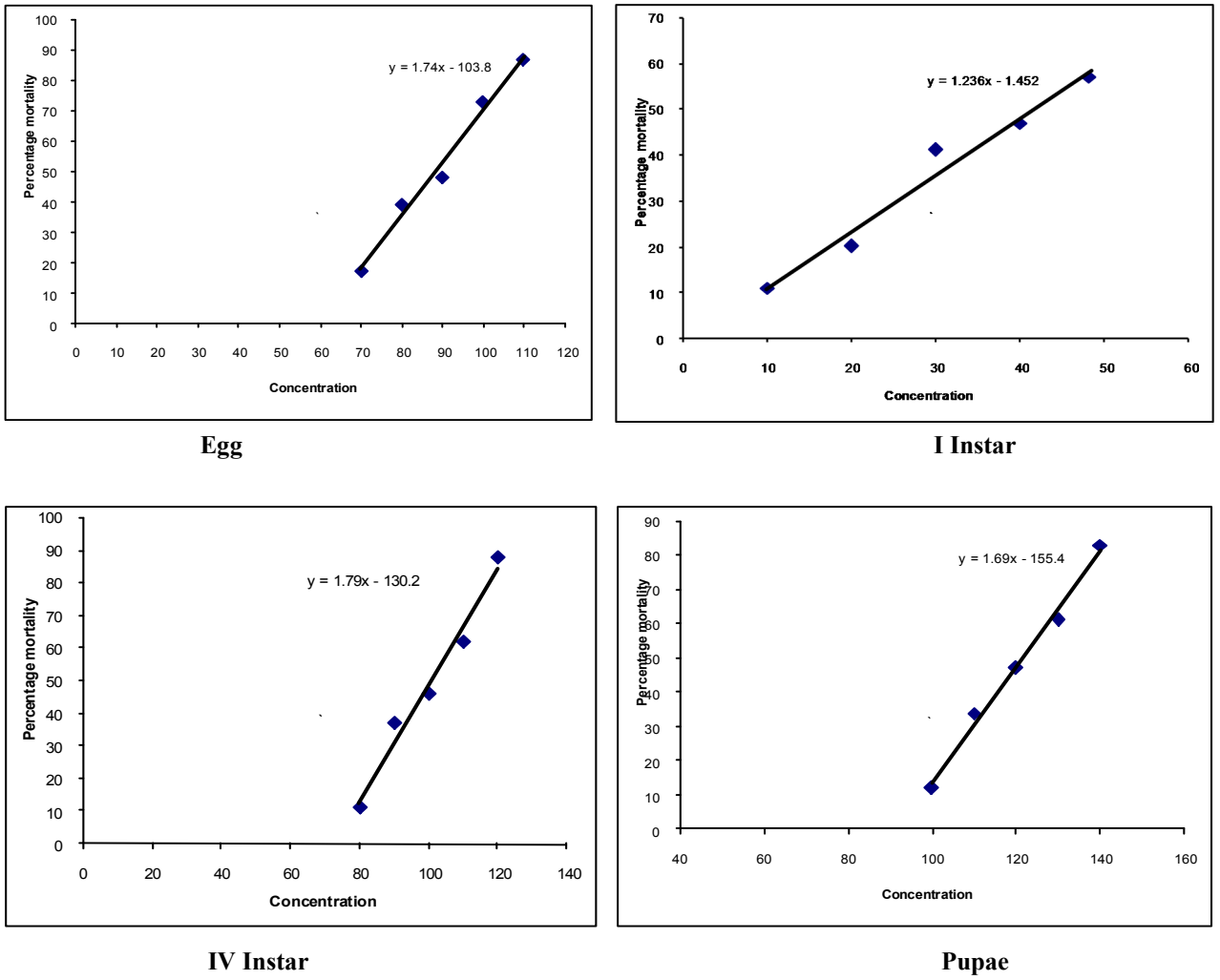

Fig 1. Regression line of log concentration of *Chromolaena odorata* crude extract vs. percent egg hatchability, mortality of larvae and puape of *Aedes aegypti.*

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