



LARVICIDAL EFFICACY OF *CATHARANTHUS ROSEUS* LINN. LEAVES AND FLOWERS AGAINST LARVAE OF IMPORTANT HUMAN MALARIA VECTOR *ANOPHELES STEPHENSI* LISTON (INSECTA:DIPTERA:CULICIDAE)

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ABSTRACT

The development of resistance by *malaria parasite* to conventional drugs poses a threat to malaria control. Hence there is a need to find new, effective, and ecofriendly remedies for malaria, including those derived from plants. Herbal products have been used traditionally by humans in many parts of the world. In the present study leaves and flowers of *Catharanthus roseus* Linn. were evaluated for larvicidal activity against *Anopheles stephensi* (L.). The larval mortality was observed after 24 and 48 hrs of time exposure. Both plant extracts showed significant larvicidal activity against *Anopheles stephensi* mosquito larvae. *Catharanthus roseus* Linn. flowers extract was found to be more effective mosquito larvicide, which was manifested by the highest percentage mortality on the larvae after 24 and 48 hours as compared to *Catharanthus roseus* Linn. leaves extract. LC50 value was calculated against different concentrations. It was recorded that lowest LC50 value was observed in the *Catharanthus roseus* Linn. flowers followed by leaves i.e. 72.10mg/l and 87.16 mg/l respectively after 24 h of exposure time and 41.02mg/l, and 62.05mg/l, respectively at 48 h of exposure time. The present study indicates that the larvicidal activities of the plants differ according to the plant species and plant parts used, which is supported by the presence of several bioactive chemicals. Further *Catharanthus roseus* Linn. plant extracts are effective mosquito vector control agents and deserves further investigation and may be useful for further integrated pest management programme.

Key words: *Catharanthus roseus* Linn., *Anopheles stephensi*, larvicidal, plant extract

INTRODUCTION

Vector-borne diseases have become a challenging problem to public health in these days as it has social and economical impact. Insect-transmitted diseases becomes a chief cause of illness and death worldwide (Pavella, 2009). Mosquitoes are vectors of several diseases affecting humans and domestic animals around the world and are the major vectors for the transmission of malaria, dengue, yellow fever, filariasis, Japanese encephalitis, etc. causing millions of deaths every year (James, 1992). Malaria is responsible for major cause of childhood mortality and adult morbidity in several parts of the world. In the last 50 years, these insect have mainly been controlled with the help of synthetic insecticides. However, one major drawback with the use of these chemical insecticides is that these are non-selective and could be harmful and adversely affect the other non target organisms in the environment (Omena *et al.*, 2007). Simultaneously the development of resistance by *malaria parasite* to conventional drugs poses a threat to malaria control. Hence there is a need to find new, effective, and ecofriendly

remedies for malaria, including those derived from plants. Herbal products have been used traditionally by humans in many parts of the world against these vectors. Phytochemicals explored from plant sources can act as larvicides and can be responsible for the interruption of the transmission of mosquito borne diseases at the individual as well as at the community level (Govindrajana *et al.*, 2008). In the present study *Catharanthus roseus* Linn. (leaves and flowers) were selected as a biopesticide against *Anopheles stephensi* (L.) - the primary vector of malaria in India and other west Asian countries.

MATERIALS AND METHODS

Collection of plants: Fully developed leaves and flowers of the *Catharanthus roseus* Linn. were collected from in and around the Mohan Lal Sukhadia University campus, Udaipur, Rajasthan, India. The shade dried plant materials were powdered by an electrical blender.

Extraction: The powders of plant materials (10g/ solvent) were loaded in Soxhlet apparatus (Vogel, 1978). The solvents from the extracts were removed using a rotary

vacuum evaporator to collect the crude extract. Standard stock solutions were prepared at 1% by dissolving the residues in methanol. From this stock solution, different concentrations were prepared and these solutions were used for larvicidal bioassays.

Test organisms: The *Anopheles stephensi* (L) mosquitoes were reared in the Insect Microbial and Herbal Control laboratory, Department of Zoology, Mohan Lal Sukhadia University. The larvae were fed on dog biscuits and yeast powder in the 3:1 ratio. Adults were provided with 10% sucrose solution and rabbit for blood meal. Mosquitoes were held at 28±2°C, 70–85% relative humidity, with a photo period of 12-h light and 12-h dark.

Larvicidal bioassay: The larvicidal activity of the plants crude extracts were evaluated as per the method recommended by World Health Organization (2005). Batches of 30 early fourth instar larvae were transferred to small disposable test beakers, each containing 100 ml of water. The appropriate volume of dilution was added to 100 ml water in the beakers to obtain the desired target dosage, starting with the lowest concentration. Three replicates were set up for each concentration, and an equal number of controls were set up simultaneously using tap water.

Statistical analysis: The control mortalities were corrected by using Abbott's formula (Abbott, 1925). The LC50 and LC90 were calculated after 24hrs and 48 hrs by Probit analysis (Finney 1979).

$$Pr = \frac{Po - Pc}{100 - Pc} \times 100$$

Where,

Pr = Corrected mortality (%)

Po = Observed mortality (%)

Pc = Control mortality (%)

RESULTS AND DISCUSSION

The extracts of different plant materials of *Catharanthus roseus* Linn. have been studied for use as ecofriendly insecticide instead of eco enemy synthetic insecticide. The effect of larvicidal activity was demonstrated in the present study confirm their potential for control of larval population. When different doses of leaves and flowers of *Catharanthus roseus* Linn. were applied on *Anopheles stephensi* larvae, significant increase in mean mortality with increase in dose as well as time was observed. Mean mortality was directly proportionate to time and doses. At lower doses of 20 ppm, 4.1 ± 1.53 and 10.3 ± 2.23 mean mortality was observed at 24 hrs and 48 hrs of time exposure respectively against *Catharanthus roseus* Linn. leaves extract. At higher dose of 100 ppm,

15.8 ± 2.09 and 27.5 ± 3.08 per cent mean mortality were reported after 24 hrs and 48 hrs respectively, as compared to 0.00 ± 0.00 and 0.3 ± 0.58 in control group (Table 1).

Table 1. Efficacy of leaves extract of *Catharanthus roseus* Linn. against *Anopheles stephensi* (L) at 24hrs and 48 hrs

Dose (ppm)	Time (Hrs)	
	24 hrs	48 hrs
20	4.1 ± 1.53	10.3 ± 2.23
40	8.9 ± 3.11	12.8 ± 3.73
60	11.3 ± 2.12	17.5 ± 3.64
80	13.9 ± 2.56	22.1 ± 5.14
100	15.8 ± 2.09	27.5 ± 3.08
C	0	0.3 ± 0.58

In case of *Catharanthus roseus* Linn. flowers extract at lower dose of 20 ppm, mortality was 4.9–1.18 whereas at highest concentration of 100 ppm, mortality reached to 17.4–3.46 at 24 hrs of exposure. Further at 48 hrs of exposure time the mortality increased from 11.4 ± 1.03 to 28.1 ± 1.25 from 20 ppm to 100 ppm respectively against control mean mortality of 0.00 ± 0.00 at 24 hrs and 0.3 ± 0.58 at 48 hrs of exposure respectively (Table 2).

Table 2. Efficacy of flower extract of *Catharanthus roseus* Linn. against *Anopheles stephensi* (L) at 24hrs and 48 hrs

Dose (ppm)	Time (Hrs)	
	24 hrs	48 hrs
20	4.9 ± 1.18	11.4 ± 1.04
40	8.8 ± 1.18	14.5 ± 1.62
60	12.6 ± 1.64	18.7 ± 2.74
80	15.1 ± 3.03	24.2 ± 2.42
100	17.4 ± 3.46	28.1 ± 1.25
C	0.0	0.3 ± 0.58

LC50 value was calculated against different concentrations. It was recorded that lowest LC50 value was observed in the *Catharanthus roseus* Linn. flowers followed by leaves i.e. 72.10 mg/l and 87.16 mg/l respectively after 24 hrs of exposure time and 41.02 mg/l, and 62.05 mg/l, respectively at 48 hrs of exposure time. LC90 values obtained against *Catharanthus roseus* Linn. flowers followed by leaves were 141.27 mg/l and 173.90 mg/l respectively after 24 hrs of exposure time and 84.71 mg/l, and 112.83 mg/l, respectively at 48 hrs of exposure time (Table 3).

The 24 hrs and 48 hrs bioassay is the major tool for evaluating the toxicity of phytotoxins and a number of

researchers have been applying this method to evaluate the toxic effect of different plant extraction against mosquitoes. Plants produce a wide spectrum of allelochemicals, however, many of such chemicals have not been explored for their physiological significance (Norduland and Sauls, 1981). These phytochemicals specifically inhibit growth, morphogenesis, metamorphosis and reproduction (Ahmad, 2007). Currently there is resurgence of interest in plant derived compounds for developing them commercially as ecofriendly insecticides. Tropical plants are more promising for the development of new insecticides (Jacobson and Crosby, 1971). The mosquito larvae exposed to plant extracts showed significant behavioural changes. The changes were observed within 30 min of exposure. The most prominent sign was inability to come to the surface. The larvae also showed loss of equilibrium, restlessness and finally led to death. The behavioural effects were more pronounced against flower extracts of *Catharanthus roseus* Linn. as compared to its leaves after exposure. These effects may be due to presence of neurotoxin compounds in *Catharanthus roseus* Linn.. No such behavioural changes were obtained in the control sets.

Table 3. Toxicity of leaves and flowers extracts of *Catharanthus roseus* Linn. against *Anopheles stephensi* (L) after 24 hrs and 48 hrs exposure time

	Dose Concentration at 24 hrs	Dose Concentration at 48 hrs
<i>Catharanthus roseus</i> Linn. leaves		
LC50	87.16	62.05
LC90	173.90	112.83
<i>Catharanthus roseus</i> Linn. flowers		
LC50	72.10	41.02
LC90	141.27	84.71

In the present study *Catharanthus roseus* Linn. leaves and flowers extract showed good effects on *Anopheles stephensi* and it is also non toxic to human beings. The present observation also revealed that leaves and flowers extracts of *Catharanthus roseus* Linn. has a promising larvicidal efficacy. Plants are rich sources of bioactive organic chemicals and offer a promising advantage over synthetic pesticide as naturals are non toxic, less prone to development of resistance and easily biodegradable. The leaves and flowers extract of *Catharanthus roseus* Linn. therefore plays an important role in the mosquito control. Many compounds of plant origin are reported as larvicidal (Jeyabalan and Murugan 1996; Curtis *et al.*, 1989; Curtis *et al.*, 1990; Foid *et al.*, 2001; Murugan *et al.*, 1996; Pushpanathan and

Muthukrishnan 1995; Saxena *et al.* 1993; Sharma and Saxena, 1994; Vatandoost *et al.*, 2010). There is wide scope for the discovery of more effective plant products (Saxena and Yadav, 1986). Ahmad (2007) reported that plant compounds "with combined behavioural and toxic effect are more likely to have successful practical application than the synthetic insecticides.

CONCLUSION

The bioactivity of tested phytochemical extracts varied significantly with solvents used for the extraction and instar stage of the larvae. Briefly, considering the information available in literature on insecticidal of plant extracts, the present study has shown that there is a wide scope for application of *Catharanthus roseus* Linn. as larvicidal agent in integrated pest management programs.

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