LARVICIDAL ACTIVITY OF METHANOLIC EXTRACT OF *Gossypium hirsutum* Linn. (Bt cotton) ROOTS AGAINST THE LARVAE OF *Culex Spp.* MOSQUITOES

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ABSTRACT

Mosquitoes have developed resistance to various synthetic pesticides and insecticides, making its control increasingly difficult. Insecticides of plant origin may serve as suitable alternative biocontrol techniques in the feature. The present study was to evaluate the larvicidal activity of methanolic extract of Gossypium hirsutum L. (Bt cotton) against the fourth instar larvae of Culex spp. The methanolic extract shows the effective larvicidal activity after 48 hour exposure at 10000 ppm. Thus, G. hirsutum L. (Bt cotton) root extract hasthe potential to be used as an eco-friendly approach for the control of the Culex Spp. Therefore, this study provides information on the mosquito larvicidal activity of plant extract.

Keywords: *Larvicidal, cotton, Culex spp., extract, insecticide.*

Introduction

Mosquitoes are responsible for the transmission of more diseases than any other group of arthropods and play an important role as etiologic agents for malaria, filariasis, dengue, yellow fever, Japanese encephalitis and other viral diseases (James 1992). Mosquitoes alone spread the disease to more than 700 million people yearly (Jang et al. 2002; Taubes 2000). Japanese encephalitis (JE), a mosquito-borne viral disease, is a severe public health problem in Asia. JE is highly prevalent in a few districts of Tamil Nadu, Kerala, Southern India (Libraty et al. 2002; Ravi et al. 1989; Reuben and Gajanana 1997). In 2001, resistance to insecticides concerned 540 species of arthropods, of which 198 were of medical and veterinary importance (Bills 2001).

In recent years, however, mosquito control programs have unsuccessful because of the ever-increasing insecticide resistance (WHO 1992). It has also resulted in the development of resistance, undesirable effects on non-target organisms, and fostered environmental and human health concerns (Brown 1986), which initiated a search for alternative control measures. The plant-based insecticides are generally pest-specific, readily decomposable, and usually lack toxicity to higher animals (Bowers 1992).

Gossypiumhirsutum L. (belongs to the plant family Malvaceae) is commonly known as upland cotton or Mexican cotton. It is native from Central America and Mexico and an important fiber producing crop and most commonly cultivated species. Worldwide around 35 mha land is under cotton crop cultivation. India alone constitutes 9.5 mha about one-quarter of the global area under cultivation of this crop. The country reported a little over 21% of the global cotton production in 2008–2009 and stands second in cotton production having 4.9 million tons behind China having 7.8 million tons (Karihaloo and Kumar 2009). Advancement in plant biotechnology with the introduction of genetically modified (GM) variety of cotton known as 'Bt- cotton' by MAHYCO seeds in collaboration with Monsanto has improved the situation of *H. armigera* attack and production of cotton in India. The Bt cotton variety involves the integration of the Cry1Ac gene from common soil bacterium *Bacillus thuringiensis*, which gives defense against bollworm.

In the present study, we have tried to extract the ingredients from roots of *G. hirsutum* L. (Bt cotton) using methanol solvent and evaluate for potential use to control different larval stages of *Culex spp.*

Materials and Methods

Collection of plant material:

The required plant material *Gossypiumhirsutum* L. (Bt cotton) was collected from the farm of Asarjan village, (19.128143 N, 77.286267 E) Nanded, Maharashtra, India. The collected plant was washed and plant roots separated. The plant roots were kept for shade drying at room temperature till they were completely dried.

Collection of Mosquito larvae:

The mosquito larvae were collected from the water tank of the School of Life Sciences SRTMU Nanded. The batches of 25 identified *Culex spp*. mosquito larvae (third and fourth Instar), as per guidelines of WHO, were procured to test the effect of plant extract on the third and fourth instar larvae.

Preparation of extract:

The dried plant roots were then powdered with the help of an electric blender. The powdered roots were weighed. The extraction was done in a Soxhlet extraction apparatus. A total of 25gm of powdered roots were taken in the extractor. Methanol is used as a solvent for extraction. The 1gm of the extract was dissolved in 100 ml of methanol (Stock solution) and considered as 1% stock solution, from this stock solution different concentrations were prepared.

Larval toxicity test:

Culex spp. mosquito larvae were used for the larvicidal activity. Twenty-five fourth instar larvae were introduced into 100 ml glass beaker. The *Culex spp*. larvae thus obtained were kept in 100 ml distilled water along with different solution grades ranging from 0.1 ppm to 10000 ppm (Table: 1) and spinosad were used as standard. Larvae were exposed for 48 hours. After 48 hours results were recorded in percent (%) mortality. The experiment was repeated thrice. The control was set up by mixing 1 ml of methanol with 100 ml of distilled water. The larvae which were exposed to distilled water served as a control. The larvicidal activity was assessed by the procedure of WHO (1996) with some modifications and as per the previous method (Patil et al. 2012).

$$\text{Percentage Mortality} = \frac{X-Y}{X} \times 100$$

Where, X= Control survival, Y = Treated sample survival

Result and Discussion

Larval mortality after treatment of methanolic extract of *Gossypium hirsutum* L. (Bt cotton) roots was observed. Table: 1 shows the larval mortality of *Culex spp.* (fourth instar) after the treatment of *G. hirsutum* L. (Bt cotton) at different concentrations (0.1 ppm to 10000 ppm), 12% mortality was noted at fourth instar larvae by the treatment of *G. hirsutum* L. (Bt cotton) at 0.1 ml extract having a 0.1 ppm concentration whereas it has been increased to 82% at 1.0 ml extract having a 10000 ppm concentration. This experiment was performed in triplicates and mean of 79.33 % mortality rate was observedagainst *Culex spp.* mosquito larvae by using a methanolic extract of roots of *G. hirsutum* L. (Bt cotton) after 48 hours of incubation.

Sr. No.	Methanolic root extract concentrations (ppm)	% Mortality after 48 hours exposure		
		0.1 ml	0.5 ml	1 ml
1	10000	20	24	82
2	1000	24	16	20
3	100	08	08	12
4	10	12	12	12
5	01	20	12	20
6	0.1	12	08	28

Table 1: Set first for the larvicidal activity of methanolic extract of *G. hirsutum* L. (Bt cotton) roots against the fourth instar larvae of *Culex spp*.

Sr. No.	Methanolic root extract concentrations (ppm)	% Mortality after 48 hours exposure		
		0.1 ml	0.5 ml	1 ml
1	10000	20	24	78
2	1000	24	16	24
3	100	12	08	16
4	10	12	08	12
5	01	24	16	20
6	0.1	20	08	24

Table 2: Set second for the larvicidal activity of methanolic extract of *G. hirsutum* L. (Bt cotton) roots against the fourth instar larvae of *Culex spp*.

Sr. No.	Methanolic root extract concentrations (ppm)	% Mortality after 48 hours exposure		
		0.1 ml	0.5 ml	1 ml
1	10000	24	20	78
2	1000	20	16	24
3	100	08	08	08
4	10	12	16	12
5	01	20	12	24
6	0.1	08	12	28

Table 3: Set third for the larvicidal activity of methanolic extract of *G. hirsutum* L. (Bt cotton) roots against the fourth instar larvae of *Culex spp*.

Sr. No.	Methanolic root extract concentrations (ppm)	% Mortality after 48 hours exposure		
		0.1 ml	0.5 ml	1 ml
1	10000	21.33	22.66	79.33
2	1000	21.33	16.00	22.66
3	100	09.33	08.00	12.00
4	10	12.00	12.00	12.00
5	01	21.33	13.33	21.33
6	0.1	13.33	09.33	26.66

Table 4: Mean of table No.1, 2 and 3.

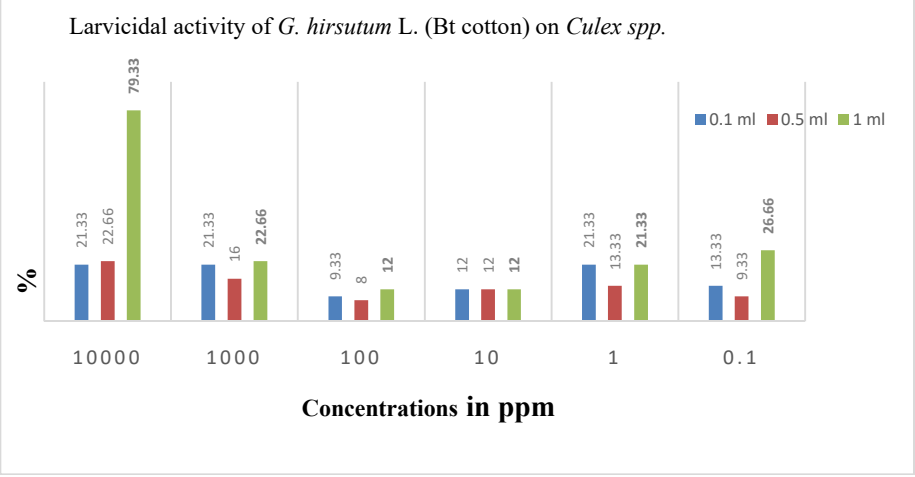

Fig. 1: Graph showing the larvicidal activity of methanolic root extract of *G. hirsutum* L. on *Culex spp.*

Conclusion

In the present study, it is concluded that the methanolic root extract of *Gossypiumhirsutum* L. (Bt cotton) displayed considerable larvicidal activity against *Culex spp.* mosquitoes. This result may be useful for developing newer and possibly safer and more effective larvicidal products against the *Culex spp.* mosquitoes. A similar kind of research work done by (Patil et al., 2014). Against the *Aedesaegypti* and *Anopheles stephensi* larvae. The current research provides an alternative to synthetic chemical pesticides by plant-based bio-pesticide which shows 82% mortality in 10000 ppm concentration. This study also throws light on the use of the waste plant body part of *G. hirsutum* L. as a potential mosquito larvicide. Plants can be a good and safer alternative for modern deadly poisonous synthetic chemicals.

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