

Effect of Insect Growth Regulators, Hydroprene and Methoprene Treatment on Oviposition of selected Three Mosquitoes

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ABSTRACT

Mosquitoes are medically and veterinary important vectors, responsible for the transmission of many human and animal diseases, such as malaria, yellow fever, dengue and West Nile Fever. The present study was conducted under laboratory conditions and consisted to evaluate the oviposition attractancy of Insect Growth Regulator (IGR) Hydroprene and Methoprene, on the oviposition of three species of mosquitoes, viz., *Culex quinquefasciatus* (Say), *Aedes aegypti* (L.) and *Anopheles stephensi* (Liston).

Keywords: Hydroprene, Methoprene, oviposition attractancy, *Culex quinquefasciatus*, *Aedes aegypti* and *Anopheles stephensi*.

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INTRODUCTION

For many viral, bacterial and protozoans' diseases mosquitoes act as vector (Kalita *et al.*, 2013). In term of disease transmission and public health importance mosquito are considered as very important group of insects. Population of mosquito's increases exponentially that is major problem for many countries because mosquito spread the different diseases such as filarial, Japanese encephalitis, Lyme disease, Yellow fever, encephalitis, malaria, chikungunya, dengue, and epidemic poly-arthritis (Kamareddine.2012). In tropical and subtropical countries mosquito borne diseases are main problem (Kalita *et al.*, 2013).

Mosquito has approximately 3500 species and present in tropical and subtropical regions (Ghosh *et al.*, 2011). Major genera of mosquitoes that act as vector for various diseases are *Culex* (Japanese encephalitis, west Nile, chikungunya, *Anopheles* (filariasis, malaria) and *Aedes* (chikungunya, dengue, Yellow fever) (Ghosh *et al.*, 2012).

Major cause for the chikungunya and dengue is *Aedes aegypti* that act as vector for the disease and affect the 2.5 million people every year (Mendki *et al.*, 2015). Feletti, vivax Grassi, protozoal parasites, *Plasmodium ovale* Stephens and *Plasmodium falciparum* Welch are the major cause of malaria that are spread by *Anopheles* mosquito (Gaddaguti *et al.*, 2016). Most important reason for the increase of dengue fever are increased breeding places for the *Aedes* mosquitoes, less effective control of mosquito, more urbanization and enhanced growth of population (Soonwera.2015).

Annually worldwide the 200 million-450 million infections are caused by the *Anopheles* mosquito that leads towards 2.7 million deaths. In more than 100 rising countries it remains endemic disease (Koech and Mwangi, 2014). Virus of Japanese encephalitis occurs in the children with malnutrition and present in the areas that are linked with animal reservoirs particularly with pigs. On the other hand main cause for the encephalopathy is encephalitis (Yoon *et al.*, 2015). According to the BBC world service health program the mosquito is considered as the world's most dangerous animal (Mendkiet *al.*, 2015).

Mosquito is vector and it causes severe diseases which can lead toward death so accurate action is necessary to get away from the disease as soon as possible (Gaddagutiet *al.*, 2016). There are various methods that can be adopted to protect itself from the bite of mosquitoes.

Insect growth regulators (IGRs), the third generation insecticides, are diverse groups of chemical compounds that are highly active against larvae of mosquitoes and other insects. The IGRs in general have a good margin of safety to most non-target biota including invertebrates, fish, birds and other wildlife. They are also relatively safe to man and domestic animals. The IGR compounds do not induce quick mortality in the pre-imaginal stages treated and occur many days post treatment. This is indeed a desirable feature of a control agent because larvae of mosquitoes and other vectors are an important source of food for fish and wildlife. On account of these advantages of IGRs and the high level of activity against target species, it is likely that IGRs could play an important role in vector control programs in the future (Mulla, 1995). They are more specific for mosquitoes than conventional insecticides. The IGRs interfere with the hormonal mechanisms of target organisms and result in various kinds of morphological, anatomical and physiological abnormalities so that the target species does not reach the final stage of development (Amalraj *et al.*, 1988a). There is no likelihood of resistance development

against these IGRs. A large number of IGRs, both juvenoids and chitin synthesis inhibitors, have been evaluated for the vector control but only very few of these are found effective and commercially feasible, e.g., diflubenzuron, methoprene, fenoxycarb (Tyagi *et al.*, 1985, 1987; Amalraj *et al.*, 1988b, Vasuki, 1988). The IGRs have an added advantage of being used at a relatively very low dose compared to the conventional insecticides. Ecdysone agonists are hormonally active insect growth regulators that disrupt development of larvae and are found to be active against *Ae. aegypti*, *An. gambiae*, and *Cx. quinquefasciatus* (Beckage *et al.*, 2004). The present work has been designed to study the oviposition attractancy of IGR, Hydroprene and Methoprene against the mosquitoes *Cx. quinquefasciatus*, *Ae. aegypti* and *An. stephensi*.

MATERIALS AND METHODS

Test insects

Cx. quinquefasciatus, *Ae. aegypti* and *An. stephensi* mosquitoes were obtained from a stock colony being maintained in the insectary at $27 \pm 1^\circ\text{C}$

and $75 \pm 5\%$ relative humidity at laboratory, Department of Zoology, Jamal Mohamed College, Trichirappalli. 10% sucrose was provided to females. Female mosquitoes were fed on rabbit blood for 4-5 days. Five days after blood feeding, the gravid female mosquitoes were used for bio assay experiments.

Test chemicals

IGR compounds namely Hydroprene chemically known as Ethyl (2E, 4E)-3, 7, 11-Trimethyldodeca-2, 4-dienoate were received as gratis (15% EC formulation Malwa Colony, Patiala, Punjab). Methoprene 20% EC chemically known as isopropyl Propan-2-yl (2E, 4E)-11-methoxy-3, 7, 11-trimethyldodeca-2, 4-dienoate. Supplied by Chemi International, Lokmanya Nagar, Mumbai, India.

Bioassay

Oviposition behaviour tests were carried out (Xue *et al.*, 2001). Fifteen gravid female mosquitoes (10 days old 5 days after blood feeding) were transferred to each mosquito cage (45x38x38 cm). Concentrations of 0.0015, 0.0020 and 0.0025 mg/L were made from each compound in 100 ml of water. Two enamel bowls holding 100 ml of water were placed in opposite corners of each cage, one treated with the test material and the other one was ethanol control. The position of the bowls were alternated between the different replicates, so as to nullify any effect of position on oviposition. Five replicates for each concentration were run, with cages placed side by side for each bioassay. The percent effective attractancy (% EA) for each oviposition concentration was calculated (Kramer and Mulla, 1979) and Oviposition Active Index (OAI) were assessed (Graham-Bryce *et al.*, 1987).

$$\text{OAI} = \frac{\text{NT} - \text{NS}}{\text{NT} + \text{NS}}$$

Where NT - Total number of eggs in the treated water and NS - Total number of eggs laid in the control water.

RESULTS

The Hydroprene ovipositional responses of the normal gravid females of the three vector mosquito species to Novaluron treated water medium are presented in Table 1. The results indicate that the number of eggs laid by the females was higher in control than in treated water. The total number of eggs laid in the treated water was also found to vary at different dosages. The OAI values calculated from the standard formula revealed that this IGR compound has repelling activity at higher dosages.

On comparing the OAI values of the three species *Cx. quinquefasciatus* (-0.40), *Ae. aegypti* (-0.32) and *An. stephensi* (-0.30) showed considerable negative response at

the dose 0.010 mg/l. At the dose of 0.020 mg/l *Cx. quinquefasciatus*(-0.52), *Ae. aegypti*(-0.45) and *An. stephensi*(-0.55), showed a higher negative response.

The ovipositional responses of the normal gravid females of the three vector mosquito species to Methoprenetreated water medium are presented in Table 2. The results indicate that the number of eggs laid by the females was higher in control than in treated water. The total number of eggs laid in the treated water was also found to vary at different dosages. The OAI values calculated from the standard formula revealed that this IGR compound has repelling activity at higher dosages.

On comparing the OAI values of the three species *Cx. Quinquefasciatus* (- 0.37), *Ae. aegypti*(-0.35) and *An. stephensi*(- 0.31) showed considerable negative response at the dose of 0.010 mg/l. At the dose of 0.020 mg/l. *Cx. quinquefasciatus*(-0.55), *Ae. aegypti* (-0.50) and *An. stephensi*(-0.65), showed higher negative response. On comparison of ovipositional responses of three vector mosquitoes to two IGRs compounds, Methoprene exhibited more ovipositional negative responses.

Table 1 Oviposition response of mosquitoes to Hydroprenetreated water

Concentration (mg/l)	<i>Cx. quinquefasciatus</i>			<i>Ae. aegypti</i>			<i>An. stephensi</i>		
	No. of egg rafts		O AI	No. of egg rafts		O AI	No. of egg rafts		O AI
	Treated	Control		Treated	Control		Treated	Control	
0.015	45.66 ±1.21	101.16 ±3.97	- 0.40	52.0±2.52	95.83±1.4 7	- 0.32	39.33±1.8 7	93.83±2.13	- 0.30
0.020	41.16 ±1.16	100.16 ±2.92	- 0.43	48.66±1.2 1	95.55±1.5 1	- 0.40	36.83±1.1 6	94.5±1.04	- 0.38
0.025	37.66 ±1.03	102.21 ±3.04	- 0.52	45.66±1.2 1	94.32±1.2 8	- 0.45	33.53±1.9 4	94.16±1.63	- 0.55

OAI - Oviposition Active Index

Table 2 Oviposition response of mosquitoes to Methoprene treated water

Concentration (mg/l)	<i>Cx. quinquefasciatus</i>			<i>Ae. aegypti</i>			<i>An. stephensi</i>		
	No. of egg rafts		O AI	No. of egg rafts		O AI	No. of egg rafts		O AI
	Treated	Control		Treated	Control		Treated	Control	
0.015	92.16±3 .04	207.53± 6.71	- 0.3 7	105.5±4 .03	205.71± 3.67	- 0.3 5	98.14±2 .44	203.83± 3.43	- 0.3 1
0.020	81.33±3 .65	210.13± 5.94	- 0.4 4	90.16±3 .94	208.16± 4.08	- 0.3 9	81.83±2 .94	208.56± 4.32	- 0.3 3
0.025	70.5±2. 86	212.64± 6.48	- 0.5 5	79.5±4. 08	209.7±4. 32	- 0.5 0	54.83±3 .71	216.16± 4.07	- 0.6 5

DISCUSSION

Mosquitoes are the most important single group of insects in terms of public health importance, which transmit a number of diseases, such as malaria, filariasis, dengue, Japanese encephalitis etc., causing millions of deaths every year (Rahuman *et al.*, 2009). Human beings are compelled to fight against them using available technical ornaments. There was initial success in controlling vectors by using synthetic insecticides. Since 1900, the World Health Organization has warned about the possible emergence and reemergence of arthropod-borne disease due to combined human, biological, environmental and climatic factors (WHO, 1990).

One of the approaches for control of these mosquito-borne diseases in the interruption of disease transmission is by killing or preventing mosquitoes to bite human beings. Although numerous synthetic pesticides are presently available for vector control programmes operating in many areas of the world (Walker, 2000; Curtis and Davis, 2001), the intense and wide spread use of these products has caused concerns regarding their impact on both human and environmental health, and has led to a buildup of resistance in pest population (Tremblay, 1982; Schmutter, 1990).

The most effective way to combat with this mosquito infestation is the prevention of mosquito breeding through the use of larvicides, synthetic insecticides such as organophosphates have

been used as larvicide in several countries for the last 30 years (Chavasse and Yap, 1997). However, one major drawback with the use of these chemical insecticides is that they are non-selective and could be harmful to other organisms in the environment (Omenaet *al.*,2007).The comparison of OAI values indicated that the IGRs compounds eliciting repelling activity. It is quite possible that this IGR acts more as an ovipositiondeterrent inhibiting oviposition at higher concentrations in the oviposition medium than as an oviposition repellent that causes insects to make oriented movements away from the source as proposed by Rajasekar and Jebanesan, (2011).

The oviposition deterrent activity exhibited by this IGRs relatively higher when compared to that of few insecticides like Pyriproxyfen (0.3 or 0.4mg/cm²)against *Ae. aegypti*mosquitoes Chism and Apperson(2003).

Ganesanet *al.* (2006) have found *Ae. aegypti* females not responding to oviposition substrates treated with 100 ppm of certain C12-C18 fatty acids and their methyl esters identified from the eggs, synthetic pyrethroids like cypermethrin, fentaletrate, decamethrin and permethrin have also shown repellency against *Cx. quiquefasciatus*, *Ae. aegypti* and *An. stephensi* in laboratory oviposition experiments (Verma, 1986). However, another IGRs triflumuron has been reported to elicit increased oviposition by *Ae. aegypti* in field and laboratory experimetnts (Ziechner and Peric, 1999).

The results of present study are interesting. The findings indicate the importance of traditional knowledge in science. The selected two IGR compounds Hydroprene and Methoprene were tested against the mosquito in laboratory condition for oviposition deterrent activity. On comparison of ovipositional responses of three vector mosquitoes to two IGRs compounds, Methoprene exhibited more ovipositional negative responses.

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