BIOMEDICAL AND ENVIRONMENTAL SCIENCES **20**, 313-316 (2007)

Comparative Larvicidal Property of Leaf Extract of *Chromolaena odorata L* (Composidae) and Chlopyrifos (Organophosphorus Compound) on *Simulium* Larvae

B. M. MATUR[1] AND B. J. DAVOU

University of Abuja, Department of Biological Sciences, P. M. B, 117, Abuja, Nigeria

Objective To assess the phytochemical properties of *Chromoleana odorata* in the control of blackflies to help check the problem of environmental pollution from the use of chemical insecticides. **Methods** Dried pulverized leaves of *Chromolaena odorata* (100 g) were soaked in distilled water for 24 hours and the extract was filtered, marc pressed and evaporated over water bath. Stock solution of the dried mass (1 g) was concentrated by dissolving in 100 mL of water. Serial dilutions (100 mg/mL, 10 mg/mL, 1 mg/mL, 0.1 mg/mL, 0.01 mg/L, and 0.001 mg/mL) of both the extract and Chlopyrifos (organo phosphorus) were prepared and toxicity was tested on the larvae of *Simulium* species. **Results** The chemical compound recorded 100% larval mortality at all concentrations while the plant extract recorded 100% larval mortality in three (100, 10, and 1 mg/mL) of its diluent concentration. The percentage of larval mortality by chemical compound and plant extract was statistically insignificant (t = 0.2456, P>0.05), but the mortality rate was significant within 30 min interval of exposure time (t= 3.756, P<0.05). The LC_{50} of the plant extract was determined at about 0.001 mg/mL concentration. This showed that the plant extract had an appreciable larvicidal property close to that of the chemical compound. **Conclusion** This result indicates the usefulness of plant materials in the control of obnoxious insect vectors of mn. It also opens additional approach to the management of environment from toxic chemicals that are non-biodegradable and cause environmental imbalance in the ecosystem.

Key words: *Chromolaena odorata;* Chlopyrifos; Larvicidal property; *Simulium* species

INTRODUCTION

Onchocerciasis control program in West Africa is primarily concerned with vector (*Simulium damnosum complex)* control by chemical compounds. These chemicals are known to be effective up to a distance of 50 km or more down stream from the point of larvicide's application[1]. Although, synthetic chemicals form an important component of Simulidae control, it is important to take into account the safety of our environment and the aquatic habitat[2].

Early chemical control schemes in West Africa used DDT at 0.1 ppm/30 min. Later temephos (Abate), chlorphoxin, carbosulfan or permetherin were adopted in the control programmed[1]. It was reported that treatment of river with 0.1 ppm of Abate for 30 min could yield 100% larval reduction[2]. The problem of development of resistance to chemical insecticides (larvicide's) results in the promulgation of the Onchocerciasis Control Program to fight the resistance of *Simulium damnosum* to chemical larvicides. There is yet a problem of the control scheme (operational gap) at discharged points between 5 and 70 m/s for the treatment of river where resistance to organophosphates occurs. Another problem is distortion of our environment by some chemicals such as permethrin and carbosulfan as a result of their non-biodegradability[1].

Among the possible complementary groups of larvicides tested, pseudo-pyrethroids hold a high promise because the mode of action is similar to that of pyrethroids usually with low toxicity to fish. Also Etofenprox, one of the pseudo-pyrethriods tested, shows a detachment of non-target insects in 24 hours close to that of chlopyrifos, an organo-phosphorus compound[3].

Laboratory experiments on the operational doses

[1]Correspondence should be addressed to Dr. B. M. MATUR, E-mail: malaumatur@yahoo.com

Biographical note of the first author: Dr. B. M. MATUR, a senior lecturer in the Department of Biological Sciences, University of Abuja, P. M. B 117 Abuja, Nigeria. He is a research fellow and a member of the American Society for Parasitology, Nigerian Society for Parasitology, Entomological Society of Nigeria and the Zoological Society of Nigeria. His areas of research and teaching interest are applied entomology and parasitology, research orientation and biostatistics and biodiversity conservation. He has well over 21 publications in the fields of parasitology, entomology, public health, and biodiversity conservation.

0895-3988/2007
CN 11-2816/Q

(0.03 mg/mL) of clopyrifos for 10 minutes caused a 50% mortality of *cardina* sp. (small shrimp's species) and thirty (30) times this same dose caused a 95% mortality of fish species. A safety margin of 400-800 times the operational dose has been observed for *Oreochromis niloticus* and 200-400 times for *Tilapia zillii*[3].

In recent years, use of environment-friendly and biodegradable natural insecticides of plant origin has received renewed importance for disease vector control. Interest in this field has increased, as they are least phytotoxic and do not accumulate chemical residues in flora, fauna, and soil[4].

The larvicidal properties of extracts from fruit of *piper guneense, Mondora myristica, Eugenia aromatica*, and *Dennettia tripetala* were evaluated on larvae of mosquito, *Aedes acgypti* (L). At different concentrations (2.8 ppm, 91.2 ppm, and 109 ppm) the extracts were found to be effective for 96 hours. Thereafter, the action became weak[5]. An effort to avoid the problem associated with synthetic chemical pesticides such as pest resurgence and detrimental effect of pesticides on non-target organism has been reported[6]. For example Ivbijaro and Agbaje[7] reported that the extract from *P. giuneense* is toxic to adult bull weevil. Hassanali *et al.*[8] have reported the insecticidal property of *D. tripetala* on the adult and nymphs of *Periplaneta americana* and *Zonocerus caryophylla.* Anil *et al.*[9] have reported the larvicidal activity of oils from 10 plants, including *Z. caryophllata*.

The problem of development of resistance to chemical insecticides and environmental pollution necessitates the search for a possible alternative for the control of obnoxious insects by using medical plants which are biodegradable, if applied into the environment.

MATERIALS AND METHODS

Larvae of *Simulium* species were collected from submerged vegetation in fast flowing streams of Shetuko River in Kuje Area Council, southeast of the FCT, Abuja, Nigeria (lat. 8°25′ and 9°25′N and long. 6°45′ and 7°45′E). Identification of the fourth and fifth instar larvae of *Simuliun* species was carried out as previously described[10]. The larvae were identified and selected for larvicidal treatment with crude extract form *C. odorata* (Plate 1) and synthetic chemical compounds (chlopyrifos). One hundred grams of dried pulverized leaves of *C. odorata* was soaked in 500 mL distilled water for 24 hours and periodically shaken. The extract was filtered, marc pressed and evaporated over a water bath. About 1 g of the residue (an equivalent of 10 g of the dried leaves) was dissolved in 100 mL distill water to produce a stock solution of crude extract with various solutuions of 100 mg/mL, 10 mg/mL, 1 mg/mL, 0.1 mg/mL, 0.01 mg/mL, 0.001 mg/mL for bioassay[11]. The same serial solutions from the chlopyrifos synthetic chemical compound were also prepared for bioassey. The two results were recorded and compared.

The fourth and fifth instar larvae (n=150) were collected for larvicidal test. Twenty-five were placed in six enamel glass containers (15 cm in diameter and 7 cm height) filled at 150 mL with river water. The river water was gently discarded and immediately replaced with 100 mL of the various test solutions. A control experiment was set-up (n=25) with river water as control treatment. The test experiment was kept at 25℃. Larval mortality in each of the concentrations was recorded at 30 min intervals for 3 hours and the result was presented as mean triplicate determination of the larval mortality as previously described[12]. The data obtained were expressed as percentage mortality. The Student t-test statistics for determination of mean difference between two means was used to determine the mean difference and ascertain the level of significance between the two treatments.

Plate 1: *Chromolaena odorata* Leave and flower.

RESULTS

The results of larvicidal properties of chlopyrifos (an organophosphorus) insecticide and crude extract from *Chromolaena odorata* against the larvae of *Simulium* species were recorded at 30 min interval for 3 hours.

The results (Table 1) showed that a 100% larval mortality could be achieved with the chemical compound at all concentrations, while the plant extract recorded 100% larval mortality only at the concentrations of 100 mg/mL, 10 mg/mL, and 1 mg/mL. The chemical compound at concentration of 0.001 mg/mL recorded 100% larval mortality while the plant extract recorded 58%.

TABLE 1

Effect of *C. odorata* and Chlopyrifos Chemical at 30-minute Exposure Rate Per 3 Hours of Treatment on *Simulium* Larvae

	Concentration (mg/mL)											
Exposure Rate (min)	100		10		1		0.1		0.01		0.001	
	A	B	A	B	A	B	A	B	A	B	A	B
:30	14.7	25	8.0	25	1.0	25	0	25	0	25	0	25
1:00	10.3	0	15.3	0	2.7	0	0.3	0	0.5	0	0.3	0
1:30	0	0	1.7	0	13.3	0	5.7	0	1.3	0	1.0	0
2:00	0	0	0	0	0	0	3.3	0	3.3	0	3.0	0
2:30	0	0	0	0	1.0	0	6.3	0	4.7	0	5.0	0
3:00	0	0	0	0	0	0	5.0	0	4.7	0	5.2	0
Total	25	25	25	25	25	25	20.6	25	14.5	25	14.5	25
%mort	100	100	100	100	100	100	82.4	100	58	100	58	100
Tmort A = 122.5 (82%)												
Tmort B = 150 (100%)												

Note. All values are mean of triplicate determination; A = plant extract (*C. odorata*); B = chemical compound (Chlopyrifos), Tmort = total mortality.

The recorded rate of larval mortality showed that chlopyrifos could achieve a 100% larval mortality (n=150) after 3-hour exposure while the plant extract recorded a 82% mortality (n=122.5). The plant extract at 0.001 mg/mL/3 hours recorded a 58% larval mortality while the chemical compound recorded a 100% mortality at 0.001 mg/mL, suggesting that the LC_{50} of the plant extract was within 0.001 mg/mL (linear determination gave its LC_{50} at 0.0008 mg/mL).

DISCUSSION

The chemical extract recorded a 100% mortality rate and the plant extract recorded a 82% mortality rate. This might be due to the fact that the chemical compound is synthetic highly concentrated as compared to the plant extract which is still in the crude form. Nevertheless, the mortality rates recorded with chemical compound and plant extract were statistically insignificant (t=0.2456, P>0.005). This concurs that they are both highly toxic to the target organism.

Worthy of mention is that the plant extract was significantly toxic at the concentration of 0.001 mg/mL. Death of about 14.5 (58%) of the total larvae (n=25) subjected to treatment was ascertained. The lethal concentration dose required to give a 50% mortality (12.5) of *Simulium* larvae as compared to the 100% mortality produced by the chlopyrifos compound at 0.0008 mg/mL obtained by linear determination. At the lowest concentration (0.001 mg/mL), the extract had no significant effect on the pH and conductivity of the river water since there was an insignificant difference in the readings at the end of the treatment (t=0.5674, P>0.005). Another important fact is that as an organic compound, its high rate of biodegradability would add advantages to biodiversity conservation.

The chemical compound and plant extract (at 0.001 mg/mL) which recorded 100% and 52% larval mortality after 3 hours of treatment might be the causal factors for mortality. Since the estimated average treatment time of the larvae in the laboratory was 18 hours, the effects of the chemical compound and the plant extract are completely responsible for the death of the larvae (t= 0.3467, 0.3268 at P<0.005, respectively).

The chemical compound recorded a 100% mortality at 0.001 mg/mL, the crude plant extract recorded a 58% mortality rate revealing its high potency though not yet concentrated or in purified form. The crude extract's potency is an acceptable standard[12] since more than half of the larvae died within 3 hours of treatment. This finding is in concurrence with the report of many plant pathologists who work on the effect of different plant extracts on mosquito larvae and other aquatic insects[3-4,13].

In conclusion, crude extract at 0.0008 mg/mL and 100 mg/mL could result in a 50% and a 100% larval mortality respectively. Thus, the plant extract is a promising substitute for chemical larvicides used for the control of black flies. More efforts should be made on the large-scale vector control using these environment-friendly compounds since other studies indicate that they are a possible alternative to synthetic chemical insecticides[13].

ACKNOWLEDGEMENT

This investigation received material and human support from Department of. Biological Sciences, University of Abuja, Mrs. R. W. NDANA, Dr. B. A. AZARE, and all laboratory staff of the Department Biological Sciences University of Abuja.

REFERENCES

1. WHO (1986). Geographical Distribution of River blindness or Onchocerciasis in Africa and the Arabian Peninsula, *WHO* 86 1085.
2. Singh H, Tripathi V N (2003). Field Trial of Relative Efficacy of Abate and Bacillus Thuringiensis against Simulium himalayense Larvae (Diptera simulidae). *MJAFI* **59**, 111-113.
3. Green M M, Singer D M, Sutherland D J, *et al*. (1991). *J. A. M. Mosquitoes Control Association* 1992, **2**, 282-286.
4. Thomas T G, Sharma S K, Jalees S, *et al*. (1994). *J Basic Appl Bio-Med* **2**, 53-55.
5. Okorie T G, Lawal L A (1998). The toxicity of Ethanotic Extracts of Fruits of *piper guineese* (African Black Paper), *Monodara Myristica* (NUTMEG), *Eugenia aromatica* and *Dennettra tripetala* (Pepper fruit) on Larvae of *Aedes aegypti* (L). *Nigerian Journal of Science* **32**, 79-82.
6. Sieber K P, Rembold H (1983). The effect of Azadirachtin on the endocrine control of moulting locusta migratoma D. *Ins Phy* **29**, 523-527.
7. Ivbijaro M F, Agbaje M (1986). Insecticidal activities of *Piper guineense* and *Calsicum species* on the cowpea *Bruchid callosobrukhus maculates. Insect science and its Application* **7**(4), 521-524.
8. Hassanali A W, Wande N, Moreka S, *et al*. (1990). Weevil repellent constituent of *Oeimum Suare* Leaves and *Eugenia caripphyllata* used as grain Protestants in parts of Eastern Africa. *Discovery and Innovation* **2**(2), 91-95.
9. Anil K, Dutta G P (1987). Indigenous Plant Oils as Larvicidal agent against *Anopheles stephensi*. *Current Science* **56**(15), 959-960.
10. Davis J B, Crosskey R W, Johnson M R L, *et al*. (1962). *Bulletin World Health Organisation* pp. 27-491.
11. Wonang D L, Onuwulii F C, Ogbonna C I C (2000). The Effect of Extracts of Some Nigerian Plants on Bacteria and Fungi Implicated in Skin Diseases. *Nigeria Journal Experimental Applied Biology* 1(1), 19-23.
12. WHO (1981). Instruction for determining the susceptibility or resistance of Blackfly larvae to insecticide WHO/VBC/81.811 Geneva 5pp.
13. Yameogo L, Traore K, Back C, *et al*. (2003). Risk Assessment of etofenprox (vectron) on non-target aquatic fauna compared with other pesticides used as *simulium larvicide* in a tropical environment *PMID: 11272920 (Pub. Med. - Index for MEDLINE).*

(*Received April* 29, 2006 *Accepted March* 28, 2007)