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**ABSTRACT:** A laboratory bioassay of insecticides against laboratory-reared late third/early fourth-instar larvae of *Mansonia uniformis* indicated that the synthetic pyrethroids tested (cyfluthrin and permethrin) were more active than the organophosphate compounds tested (chlorpyrifos, fenthion, and temephos). The  $LC_{50}$  values ranged from 0.09 (cyfluthrin) to 7.68  $\mu\text{g/l}$  (temephos). Field trials of fenthion (Baytex GR2) and cyfluthrin (Baythroid H10WP) against laboratory-cultured late third/early fourth-instar larvae of *Mansonia uniformis* in floating screened cages were conducted in small plots on Penang Island, Malaysia. At respective doses of 0.50 and 0.30  $\text{g/m}^2$  (5.0 and 3.0  $\text{kg/ha}$ ), both fenthion and cyfluthrin caused more than 90% mortality of the *Mansonia* larvae exposed at 0 hour post-treatment. The use of these larvicides for the control of *Mansonia* larvae in open field environments is discussed.

**Keyword Index:** *Mansonia uniformis*, larvicide, fenthion, cyfluthrin.

### INTRODUCTION

*Mansonia uniformis* is one of the major vectors of Brugian filariasis in Southeast Asia (Mak 1981, Yap 1985). Approximately one billion people lived in endemic areas with approximately 5.8 million cases of infection in this region (World Health Organization 1994). Currently, Brugian filariasis is controlled through mass drug treatment using diethylcarbamazine citrate, and new drugs such as ivermectin are still under experimentation (Mak 1981, 1990, Ramachandran 1993, World Health Organization 1994).

In comparison with other mosquito genera, there have been few larvicidal tests against *Mansonia* (Yap 1985, Yap et al. 1995a). Chapman (1955) conducted laboratory and field tests with 15 insecticides against *Mansonia dubitans* and concluded that parathion and EPN were most effective. Further laboratory studies on *Mansonia perturbans* (Yap et al. 1968) and *Ma. uniformis* (Yap and Sulaiman 1976) indicated good efficacy of temephos and chlorpyrifos. Field studies further confirmed the efficacy of temephos against *Mansonia* larvae (Gass et al. 1985). More recently, etofenprox, a relatively new insecticide with low mammalian toxicity, was also found to provide good larvicidal activity in the

laboratory (Yap et al. 1995b).

We report here the efficacy of two synthetic insecticides, namely fenthion and cyfluthrin, as tested in the laboratory (compared with chlorpyrifos, temephos and permethrin) and field against *Ma. uniformis* larvae.

### MATERIALS AND METHODS

The *Ma. uniformis* larvae used in both laboratory and field assessments were from well-established laboratory colonies at the Vector Control Research Unit, Universiti Sains Malaysia. The colony was initiated from collections in Permatang Damar Laut, Penang Island in 1990. The mosquitoes used in this study were late third and early fourth-instar larvae.

Technical grade fenthion (*o,o*-dimethyl-*o*-[4-(methylthio)-*m*-tolyl] phosphorothioate, temephos (*o,o*-[thiodi-4-1-phenylene] *o,o,o,o*-tetramethyl phosphorothioate), chlorpyrifos (*o,o*-diethyl *o*-3,5,6-trichloro-2-pyridyl phosphorothioate), cyfluthrin [ $\alpha$ -cyano (4-fluoro-3-phenoxyphenyl) methyl 3-(2,2-dichloroethenyl)-2,2-dimethyl-cyclopropanecarboxylate] and permethrin [3-phenoxybenzyl-3-2,2-dichlorovinyl-2,2-dimethylcyclopropane carboxylate] were used in the laboratory bioassay. The organophosphates chosen for

this study are currently being used as mosquito larvicides in Malaysia. Temephos is used for clear water breeders, such as the *Aedes* species, whereas fenthion and chlorpyrifos are used for the polluted water breeding *Culex* species. The choice of the two residual pyrethroids (permethrin and cyfluthrin) takes into consideration the greater use of pyrethroids for household and public health insect control in recent years based on their selective toxicity.

The bioassay method was essentially that established by the World Health Organization for larvicidal susceptibility (World Health Organization 1981) with the following modifications: (1) technical grades of test insecticides were diluted to test concentrations using analytical grade acetone as solvent; (2) twenty late third/early fourth-instar larvae in three replicates were used for each concentration of insecticides per test. The experiment was then repeated three times using different batches of larvae and insecticide preparations; (3) total volume of test solution was 100 ml instead of 250 ml per beaker and; (4) a small piece of styrofoam was placed in each beaker for attachment of *Mansonia* larvae after the addition of insecticide.

For field efficacy tests, only fenthion granule (Baytex GR2) and cyfluthrin wettable powder (Baythroid H10WP) formulations were used. The field trials were conducted by exposing mosquito larvae to insecticide applications in floating screened cages in small plots (15 - 18 m<sup>2</sup>). Test plots had a vegetative cover of water hyacinth (*Eichhornia crassipes* Solm), which is the natural host plant of *Mansonia*. Six treated and three untreated control plots were used. The nine plots totaled 145 m<sup>2</sup>. The field plots were located in a swampy ditch of an abandoned coconut plantation in Permatang Damar Laut on the southern coastal alluvial plain of Penang Island, Malaysia. The detailed protocols for tests in

small plot trials followed essentially those of Yap et al. (1991). Efficacy and residual effects of the insecticides against *Mansonia* larvae were determined by introducing batches of larvae into additional cages at intervals of 0, 24, 48, 72, and 168 hours post-treatment. The mortality readings were conducted 24 hours after the introduction of each batch of larvae at the above designated intervals.

For both formulations tested, application rates ranged from 0.01 to 2.00 g/m<sup>2</sup>. The fenthion granule formulation (Baytex GR2) was mixed thoroughly with fine sand and dispersed by hand. The cyfluthrin wettable powder formulation (Baythroid H10WP) was mixed with seasoned tap water (local tap water kept for more than 72 hours) and applied using a Geizhal ES10 pressurized knapsack sprayer (Dr Stahl and Sohm GmbH and Co, Uberlinger, Germany).

Rainfall, temperature, pH, dissolved oxygen, and water conductivity at the field sites were recorded daily. Measurements were made using portable meters including a membrane pH meter (Hanna HI 8314, Italy), dissolved oxygen meter (Yellow Spring Instrument, YS IM67, USA) and conductivity meter (WIW LF 91 with probe KLEI/T, USA). Data were subjected to probit analysis (Finney 1971) using a computer program by Daum (1970).

## RESULTS AND DISCUSSION

Based on the laboratory bioassays, the *Ma. uniformis* larvae appeared to be more susceptible to the two pyrethroids (cyfluthrin and permethrin) than to the organophosphates (chlorpyrifos, temephos, and fenthion) (TABLE 1). The LC<sub>50</sub> values for these insecticides ranged from 0.09 to 7.68 µg/l. Cyfluthrin seemed to be the most effective of all the insecticides tested. Among the organophosphates, chlorpyrifos and

TABLE 1. Activity of five insecticides against late third/early fourth-instar *Mansonia uniformis* larvae in the laboratory.<sup>1</sup>

Insecticide	LC (95% fiducial limit) [in µg/l]		Slope ± SE
	50	90	
fenthion	4.96 (4.81-5.10)	7.54 ( 7.21- 7.95)	7.04 ± 0.25
temephos	7.68 (7.32-8.05)	18.54 (17.17-20.22)	3.35 ± 0.09
chlorpyrifos	3.85 (3.36-4.27)	6.70 ( 5.87- 8.31)	5.33 ± 0.59
cyfluthrin	0.09 (0.08-0.10)	0.31 ( 0.26- 0.37)	2.34 ± 0.10
permethrin	1.74 (1.63-1.84)	3.93 ( 3.61- 4.36)	3.61 ± 0.15

<sup>1</sup>Based on pooled data of three experiments.

fenthion were more effective than temephos. Dose-response values of some of the insecticides tested here agree with those from earlier publications (Yap et al. 1968, Yap and Sulaiman 1976). Some minor discrepancies occurred due to species differences (Yap et al. 1968) and the use of laboratory-cultured versus field collected mosquito larvae (Yap and Sulaiman 1976).

Both fenthion and cyfluthrin proved to be effective in the simulated field control of *Ma. uniformis*. The larvicidal efficacy of fenthion and cyfluthrin against sentinel *Ma. uniformis* in small plots in swampy ditches showed that the latter had a higher efficacy than fenthion at the initial introduction of *Mansonia* larvae (immediately after insecticidal application) (TABLE 2). When new *Mansonia* larvae were introduced at 24 and 48 hours post-treatment, some significant mortalities of larvae occurred at the higher dosages used. For the cyfluthrin formulation, introduction of new batches of larvae at 24, 48, 72, and 168 hours post-treatment indicated significant residual effects of up to seven days at the two highest dosages used (1.00 and 2.00 g/m<sup>2</sup>, TABLE 2). The effective dosages for both insecticides for *Mansonia* control in such habitats appeared to be lower than those of microbial insecticides such as *Bacillus thuringiensis* H-14 (Foo and Yap 1983) and *Bacillus sphaericus* (Yap 1990, Yap et al. 1991) tested under the

same environmental conditions.

The meteorological and water quality conditions for the field site in Permatang Damar Laut, Penang Island were similar to those recorded in the same plots in earlier publication (Yap et al. 1991). The mean values ( $\pm$  S.E.M) for temperature, pH, dissolved oxygen, and conductivity of the field water were  $27.4 \pm 0.3^\circ\text{C}$ ,  $6.7 \pm 0.1$ ,  $1.8 \pm 0.1$  mg/l, and  $10.1 \pm 2.1$  mmho/cm, respectively.

Field trials indicated that at respective application rates of 0.50 and 0.30 g/m<sup>2</sup> (equivalent to 5.0 and 3.0 kg/ha), fenthion and cyfluthrin caused more than 90% mortality of the *Mansonia* larvae at 0 hour post-treatment in natural larval habitats with high organic and ion contents. At subsequent intervals (24, 48, 72, and 168 hours post-treatment), there was a decrease in efficacy of insecticides against the introduced larvae (TABLE 2). Thus, both fenthion and cyfluthrin showed only slight residual effects against *Mansonia* larvae in the natural polluted habitats except at considerably higher dosages. However, such lack of persistence at lower dosages should still provide reasonable effective control if the application routine was conducted monthly. This is because *Mansonia* mosquitoes in general need a much longer time (20-25 days) to complete their larval development (Wharton 1962) as compared with other mosquito species.

Results from the present field studies indicated that

TABLE 2. Small-plot field trials on fenthion and cyfluthrin against *Mansonia uniformis* larvae placed in floating cages.

Insecticide	Dosage (g/m <sup>2</sup> )	No. live larvae over 50 introduced at following intervals after 24 hours exposure <sup>1</sup>				
		0 hr	24 hr	48 hr	72 hr	168 hr
fenthion	control	44.0 $\pm$ 2.6	44.3 $\pm$ 2.1	41.6 $\pm$ 1.5	-	-
	0.01	43.6 $\pm$ 1.2	41.0 $\pm$ 2.0	38.3 $\pm$ 7.0	-	-
	0.10	26.0 $\pm$ 10.5	43.7 $\pm$ 4.6	37.3 $\pm$ 1.5	-	-
	0.30	16.3 $\pm$ 18.0	28.3 $\pm$ 16.1	42.0 $\pm$ 2.6	-	-
	0.50	2.3 $\pm$ 3.2	18.7 $\pm$ 12.7	42.3 $\pm$ 6.4	-	-
	1.00	0.7 $\pm$ 0.6	7.3 $\pm$ 0.6	7.3 $\pm$ 3.8	-	-
	2.00	0	5.0 $\pm$ 4.6	30.6 $\pm$ 16.3	-	-
cyfluthrin	control	46.0 $\pm$ 1.0	41.3 $\pm$ 1.2	43.7 $\pm$ 0.6	44.7 $\pm$ 1.5	46.3 $\pm$ 2.5
	0.01	37.3 $\pm$ 1.2	38.0 $\pm$ 1.7	37.7 $\pm$ 0.6	42.7 $\pm$ 4.0	41.7 $\pm$ 2.9
	0.10	8.3 $\pm$ 4.6	32.6 $\pm$ 2.1	35.7 $\pm$ 2.9	42.0 $\pm$ 1.0	45.3 $\pm$ 1.5
	0.30	3.3 $\pm$ 4.0	19.3 $\pm$ 0.6	25.7 $\pm$ 6.0	36.0 $\pm$ 5.0	42.3 $\pm$ 3.1
	0.50	1.3 $\pm$ 1.2	7.0 $\pm$ 5.6	18.7 $\pm$ 8.1	31.7 $\pm$ 4.2	40.0 $\pm$ 2.0
	1.00	1.0 $\pm$ 1.7	6.7 $\pm$ 3.1	13.0 $\pm$ 6.9	26.3 $\pm$ 1.2	31.0 $\pm$ 3.5
	2.00	0.3 $\pm$ 0.6	0	0	2.0 $\pm$ 1.7	6.7 $\pm$ 3.1

<sup>1</sup>All values are mean  $\pm$  S.E of three experiments of live larvae recovered after 24 hours exposure.

both fenthion and cyfluthrin have the potential to be effective larvicides for *Mansonia* control in natural breeding habitats. However, the choice of any control agents, whether chemical or biological, should take into consideration the environmental impacts of such agents under operational usage.

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