

Effects of Three Organophosphorus Insecticides in the Reproductive Potential of *Culex quinquefasciatus*

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A Culex quinquefasciatus Say 1823 strain with resistant genes to organophosphates was tested in the laboratory to know the reproductive potential after exposure, as larvae, at the LC³⁰ and LC⁷⁰ (mg/l) of three organophosphorus insecticides: malathion, chlorpyrifos and methyl-pirimiphos. Data showed that fecundity was decreased significantly by malathion at LC³⁰ = 0.0025 and LC⁷⁰ = 0.0075, whereas fertility had a no significant decrement by chlorpyrifos and methyl-pirimiphos at the LC⁷⁰ (0.000016, 0.00043). The sexual index was affected by chlorpyrifos and methyl-pirimiphos showing a greater number of adult females.

Key words: insecticide - malathion - chlorpyrifos - methyl-pirimiphos - reproduction - *Culex quinquefasciatus*

In 1981 Cuba started an intensive campaign for the control of mosquito *Aedes aegypti* (Linnaeus) (Diptera: Culicidae). The insecticide more used between 1981 and 1986 was malathion, in addition, temephos and fenthion were also used. In 1986 began the use of other organophosphorus insecticides like methyl-pirimiphos and chlorpyrifos. This campaign reached success in the control of *A. aegypti*, but at the same time *Culex quinquefasciatus*, as a secondary effect, began the colonization of the typical urban breeding-sites of that species (Bisset et al. 1987), increasing their populations in spite of the continuous use of insecticides in those areas, suggesting the presence of resistance in this mosquito (Bisset & Marquetti 1983). The treatment of a determinate species with sublethal doses of insecticides may affect their survival, their reproduction or the genetic composition of future generations (Moriarty 1969). Hunter (1958), working with the malathion and diazinon found a reduction of *Musca domestica* descent, while El-Khatib and Geroghiou (1985) reported that the treatment of *C. quinquefasciatus* larvae with temephos decreased the fecundity of survivors. These reports indicate that the sublethal effects of the pesticides can be important for the reduction of the number of injurious insects in pest control programs.

The present work describes the effect of the sublethal doses of three organophosphorus insecticides on the reproductive potential of *C. quinquefasciatus*.

MATERIALS AND METHODS

A strain of *C. quinquefasciatus* denominated "quibú" was used. It was maintained in standard conditions of laboratory (25 ± 2°C and 75 ± 10% of relative humidity) and free of insecticides for ten successive generations. In one liter of distilled water five rafts of eggs were placed. Larvae were fed daily with fishmeal. Pupae were transferred to a beaker containing water and placed in a breeding box.

Adults mosquitoes were blood fed on chicks three days after emergence and offered a 10% sugar solution on cotton pads as a carbohydrate source.

Insecticides tested were malathion (95% TG), chlorpyrifos and methyl-pirimiphos (90% TG) and ethanol was used as solvent. Three tests were done for each insecticide and, each test consisted of five concentrations of chemical, using five replicates for concentration. Controls for each insecticide were used and treated with ethanol. Twenty five larvae were placed into glass flasks containing 200 ml of distilled water and were exposed to the insecticide for 24 hr without food. Survivor larvae were washed and put in distilled water, offered daily fishmeal and were maintained in trays until they reached the pupal stage.

Pupae were sexed for each insecticide and concentration, and 100 male pupae and 100 female were placed in breeding boxes, for each dose of each insecticide and for the control. After the coupling of the adults, rafts of each box were collected up to a number of fifty.

Eggs were counted in the rafts obtained per insecticide and concentration, and the percent eclosion was recorded. The emergent larvae were placed into trays until they reached the pupal instar and these were finally sexed to establish the

sexual index for each insecticide and concentration.

Data of bioassays were submitted to a computer program called probit-log (Raymond 1985). The ANOVA test of Linear Regression, the Kruskal-Wallis and Student-Newman Keul tests and the test for the comparison of two proportions were also used. It was considered significant at the level of $p \leq 0.05$.

RESULTS AND DISCUSSION

The toxicity of the three tested insecticides on larvae of early fourth stage of *C. quinquefasciatus*, is shown in Table I. From this Table is observed that chlorpyrifos was the most toxic ($LC^{50}=0.000009$ mg/l), followed by methyl-pirimiphos ($LC^{50}=0.00026$ mg/l) and malathion ($LC^{50}=0.0035$ mg/l). These values of LC^{50} are in accordance to those previously reported by Villani et al. (1983) and El-Khatib and Georghiou (1985), who worked with malathion and chlorpyrifos insecticides.

In Table II the effect of these insecticides on the fecundity of *C. quinquefasciatus* is shown. In the survivors at LC^{30} and LC^{70} of malathion, the production of eggs was reduced to 57% ($p \leq 0.05$) and 59% ($p \leq 0.05$), respectively, as compared with the control. However, the fecundity was not affected in the survivors at the LC^{30} and LC^{70} of chlorpyrifos and methyl-pirimiphos. Similar results were reported by Wijeyaratne (1976), El-Khatib and Georghiou (1985) and Robert and Olson (1989) for *C. pipiens* and *C. quinquefasciatus* larvae exposed to different organophosphorus insecticides. On the other hand, Ferrari and Georghiou (1981) reported a significant reduction in the fecundity of *C. quinquefasciatus* when the larvae were exposed to the organophosphate temephos.

The fertility of the eggs was not affected in the treated larvae with malathion, whereas it was observed a slightly reduction, $p > 0.05$, in larvae exposed at the LC^{70} of chlorpyrifos and methyl-pirimiphos. This is consistent with previous studies which reported that expositions to malathion of larvae of *A. aegypti* did not result in a reduction of the fertility of eggs (Firstenberg & Sutherland 1981, Ferrari & Georghiou 1981, Robert & Olson 1989).

The pupation time had a significant increase ($p \leq 0.05$) for all concentrations of the insecticides tested. The male mosquitoes exposed to the insecticides had an emergence time slightly longer than the controls, however it was increased significantly ($p \leq 0.05$) at the LC^{30} of malathion. The females exposed as larvae at the LC^{70} of malathion and methyl-pirimiphos also had a significant longer time of emergence ($p \leq 0.05$) than the controls. The increase of the larval and pupal periods that resulted from treatments with insecticides are in accordance to the results obtained by Wijeyaratne (1976) and Robert and Olson (1989). Larval prolonged development has been reported by Farghal (1979) with various doses of other insecticides in *C. pipiens* autogenic.

The sexual proportion in adults was affected by exposition to chlorpyrifos and methyl-pirimiphos. So, in larvae treated with these insecticides the quantity of adult females had a significant increase ($p < 0.01$). However this was not observed in larvae treated with malathion where the sexual proportion was not affected. Robert and Olson (1989), working with the same species, found a reduction of the proportion of females coming from larvae exposed to methoprene, propoxur and resmethrine, whereas Farghal and Temerak (1981) found gradual increments of female proportion, accompanied by gradual increments of concentrations of methoprene. Martinez-Mole (1991) working with the same species found that sublethal doses of cipermethrine did not produce any change in the female-male proportion.

CONCLUSIONS

From three tested insecticides in "quibú" strain of *C. quinquefasciatus*, it was demonstrated that chlorpyrifos was the most toxic, followed by methyl-pirimiphos and malathion.

A reduction of the fecundity in the studied species was found when it was pressed with malathion doses, whereas the fertility decreased with doses of chlorpyrifos and methyl-pirimiphos.

The sexual index in the studied species was affected with exposure periods of methyl-pirimiphos and chlorpyrifos, but not with malathion.

TABLE I

Values of LC_{50} and LC_{90} (mg/l) of *Culex quinquefasciatus* larvae exposed to three organophosphorus insecticides

Insecticide	LC_{50}	Fiducial limit	LC_{90}	Fiducial limit	Slope
Malathion	0.0035	0.003-0.004	0.0075	0.007-0.008	9.8
Chlorpyrifos	0.000009	0.000007-0.00001	0.00004	0.00003-0.00005	1.97
Methyl-pirimiphos	0.00026	0.00023-0.00028	0.00089	0.00079-0.00103	2.39

TABLE II
Effect of three organophosphorus insecticides on the fecundity of *Culex quinquefasciatus*

Insecticide	Total egg production	Egg proportions in relation to the control
Malathion		
LC ₃₀ (0.0025)	7.036	0.57 ^a
LC ₇₀ (0.0075)	6.281	0.51 ^a
Control	12.181	1.00
Chlorpyrifos		
LC ₃₀ (0.000004)	12.109	0.90
LC ₇₀ (0.000016)	11.458	0.94
Control	12.181	1.00
Methyl-pirimiphos		
LC ₃₀ (0.00015)	11.492	0.94
LC ₇₀ (0.00043)	10.005	0.82
Control	12.181	1.00

^a: values that are different for p<0.05, in relation to the control

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