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Toxicity of certain pesticides found in the habitat to the larvivorous fishes *Aplocheilus lineatus* (Cuv. & Val.) and *Macropodus cupanus* (Cuv. & Val.)

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Abstract. Bioassay studies reveal the toxicity levels of pesticides utilised in the area to the larvivorous fishes *Aplocheilus lineatus* and *Macropodus cupanus*. The resistance of both fishes decreases with increase in period of exposure to the pesticides. Comparing the major groups of synthetic organic pesticides, the chlorinated hydrocarbons, here exemplified by DDT, are more toxic to the fishes than ekalux and malathion, the organophosphates experimented with. The carbamate sevin is the least toxic. Nevertheless, all the pesticides are 'toxic' to 'very toxic' as defined by the Joint ICMO/FAO/UNESCO/WHO group of experts, having an acute lethal threshold of below 1 to 100 mg/l. *M. cupanus* is the more resistant of the two fishes, probably on account of its obligate air-breathing nature, and thus its tendency to absorb less toxicant across the gills. Contrasting the susceptibility of mosquito larvae and the fishes studied to the pesticides investigated, the closeness of the LC_{50} values obtained in *A. lineatus* to that recorded in certain species of mosquito larvae indicates that while *M. cupanus* could be employed in conjunction with pesticides for anti-larval work, *A. lineatus* should not be so utilised.

Keywords. Pesticides ; toxicity ; larvivorous fish ; *Aplocheilus lineatus* ; *Macropodus cupanus*.

1. Introduction

Larvivorous fishes such as *Gambusia affinis* and *Poecilia reticulata*, the primary biological control agents of mosquito larvae, have been extensively employed in certain regions in mosquito abatement programmes (Mallars and Fowler 1970; Bay and Self 1972). However, indiscriminate releases of these exotics into the aquatic environment has resulted in the alteration/eradication of valuable faunal components of the ecosystem (Myers 1965; Bay 1973; Menon 1977). This has renewed interest in the biocontrol potential of indigenous larvivorous fishes such as *Aplocheilus lineatus* (Cuv. & Val.) and *Macropodus cupanus* (Cuv. & Val.). An essential aspect of such assessments is information on the danger levels to the fishes of pesticide contaminants found in the aquatic ecosystem. This problem has assumed importance owing to the widespread and indiscriminate permeation

of pesticides in the aquatic environment (Muirhead-Thomson 1971; Edwards 1977) and the consequent risks to larvivorous fish populations. Such data are not available, leading to this study.

2. Materials and methods

In the present investigation, pesticides were chosen from each of the major groups of synthetic pesticide utiliseds in agricultural operations in the area—i.e., DDT (25 EC; manufactured by Bangalore Pesticides Limited) from the chlorinated hydrocarbons, malathion (50 EC; manufactured by Bangalore Pesticides Limited) and ekalux (25 EC; manufactured by Sandoz India Limited) from the organophosphates, and sevin (50% WP; manufactured by Union Carbide) from the carbamates and bioassay tests were conducted.

Healthy medium sized *A. lineatus* (mean standard length 25-40 mm) and *M. cupanus* (mean standard length 20-28 mm) collected from streams and water bodies in the Trivandrum (Kerala, South India) area were acclimated to laboratory conditions in well water at a temperature of 28 ± 2° C, pH of 7·1 and O_2 at near air saturation. The static test method (Doudoroff *et al* 1951) was used to directly estimate the toxicity levels, with certain modifications to guard against a depletion/alteration in the toxic material, as suggested by Muirhead-Thomson (1971) and Sprague (1973). Stock solutions of the different pesticides were diluted to the required parts by weight of active ingredient (= mg/l) by standard methods (Busvine 1977). However, since the water volume/weight of fish ratios utilised for bioassay tests vary greatly (Rita and Nair 1978), here, on the basis of preliminary trials, 1·8 gm/l solution and 1 gm/l solution were chosen as an adequate weight/volume ratio in *A. lineatus* and *M. cupanus*, respectively. Bioassays were carried out in 5 logarithmic concentrations. The period of exposure for each bioassay was 48 hr as subsequently the mortality curve flattened; neither the experimental nor control specimens were fed during this period. The lethal concentration 50 (LC_{50}) for 24 and 48 hr were calculated for each pesticide by the probit analysis method. The behavioural responses exhibited by the fishes during the exposure period were also recorded.

3. Results and discussion

A comparative statement of the results of the probit analysis, specifically regression equations and the LC_{50} values including the upper and lower limits (ULC_{50} and LLC_{50}) has been tabulated for both the 24 and 48 hr period of exposure in the case of each pesticide in tables 1 and 2.

Considering the physical reactions of the fish to the toxic solutions, in all cases undulation (mild to pronounced) of the body, increased oscillation of the pectoral, pelvic, anal and caudal fins, rapid and irregular movements of the opercular folds, loss of equilibrium (ranging from partial to complete) and excitation (mild to pronounced) were noted. At extremely toxic concentrations, the external body surface showed 'burnt' patches.

The lowering in the 48 h LC_{50} values when compared with the 24 hr ones suggests the decreasing resistance of the fish with increase in experimental time, a finding supported by Cairns and Scheier (1964) and Rita and Nair (1978).

Table 1. Acute toxicity levels of selected pesticides in *A. lineatus*.

Pesticide	Period of exposure (hrs)	LC_{50} values (mg/l)	Regression equation
DDT	24	0·1489±0·0212	$\log y = 9{\cdot}5405 \cdot \log x \times 100 - 6{\cdot}1893$
	48	0·1228±0·0182	$\log y = 8{\cdot}0885 \cdot \log x \times 100 - 3{\cdot}8103$
Ekalux	24	0·1939±0·0247	$\log y = 10{\cdot}2105 \cdot \log x \times 100 - 8{\cdot}1467$
	48	0·1699±0·0238	$\log y = 9{\cdot}6205 \cdot \log x \times 100 - 6{\cdot}8348$
Malathion	24	1·1500±0·3050	$\log y = 5{\cdot}0873 \cdot \log x \times 10 - 0{\cdot}3972$
	48	0·9750±0·2130	$\log y = 6{\cdot}1911 \cdot \log x \times 10 - 1{\cdot}1228$
Sevin	24	4·2070±0·3750	$\log y = 14{\cdot}3413 \log \cdot - 3{\cdot}9490$
	48	3·7470±0·3100	$\log y = 14{\cdot}6842 \log \cdot - 3{\cdot}4242$

Table 2. Acute toxicity levels of selected pesticides in *M. cupanus*.

Pesticide	Period of exposure (hrs)	$I C_{50}$ values (mg/l)	Regression equation
DDT	24	2·813±0·453	$\log y = 8{\cdot}6338 \cdot \log x + 1{\cdot}1219$
	48	2·277±0·310	$\log y = 9{\cdot}8746 \cdot \log x + 1{\cdot}4720$
Ekalux	24	3·659±0·434	$\log y = 11{\cdot}454 \cdot \log x - 1{\cdot}4533$
	48	3·453±0·584	$\log y = 7{\cdot}7358 \cdot \log x + 0{\cdot}8363$
Malathion	24	4·962±0·479	$\log y = 13{\cdot}2989 \cdot \log x - 4{\cdot}2607$
	48	4·594±0·557	$\log y = 10{\cdot}5503 \cdot \log x - 1{\cdot}9859$
Sevin	24	14·730±0·590	$\log y = 35{\cdot}2288 \cdot \log x - 36{\cdot}1552$
	48	13·910±0·380	$\log y = 44{\cdot}0285 \cdot \log x - 45{\cdot}3320$

The higher LC_{50} values in *M. cupanus* denote its greater resistance than *A. lineatus*. This may be because the principal route of entry of pesticides for non-feeding fish is through the gills (Johnson 1968); *M. cupanus*, being an obligate air-breather, naturally tends to absorb less toxicant across the gills. Comparing the main groups of synthetic organic pesticides, the results of the present study where DDT (a chlorinated hydrocarbon) is more toxic to the fish than ekalux, malathion (organophosphates) and sevin (a carbamate), are in agreement with the findings of Johnson (1968) and Rita and Nair (1978). However, all pesticides tested are 'toxic' to 'very toxic' as defined by the Joint ICMO/FAO/UNESCO/WHO group of experts (1964) since they have an acute lethal threshold of below 1 to 100 mg/l. A comparison of the acute toxicity levels of the pesticides in various species of fishes, given in table 3, reveals that wide variations in the

Table 3. Comparison of some acute toxicity levels of the pesticides investigated in different species of fishes.

Pesticide	Species investigated	Period of exposure (hrs)	LC_{50} (mg/l) (ppm)	Reference
DDT	*Lepomis macrochirus*	96	0·016	Edwards (1977)
	Salmo gairdneri	96	0·018	Edwards (1977)
	Salvelinus fontinalis	36	0·0323	Hatch (1957)
	Carassius auratus	72	0·1	Odum and Summerford (1946)
	Carassius auratus	96	0·027	Henderson *et al* (1959)
	Aplocheilus lineatus	24	0·1489	Present investigation
	Aplocheilus lineatus	48	0·1228	Present investigation
	Gambusia affinis	24	0·5	Mayhew (1955)
	Gambusia affinis	36	0·32	Hatch (1957)
	Gambusia affinis	72	0·01	Odum and Summerford (1946)
	Macropodus cupanus	24	2·813	Present investigation
	Macropodus cupanus	48	2·277	Present investigation
Malathion	*Puntius ticto*	24	0·0135	Bhatia (1971)
	Puntius ticto	48	0·011	Bhatia (1971)
	Puntius ticto	72	0·011	Bhatia (1971)
	Puntius ticto	96	0·0074	Bhatia (1971)
	Salmo gairdneri	96	0·1	Edwards (1977)
	Lepomis macrochirus	96	0·12	Edwards (1977)
	Aplocheilus blochii	48	1·3	VCRC Annual Report (1979)
	Aplocheilus lineatus	24	1·15	Present investigation
	Aplocheilus lineatus	48	0·975	Present investigation
	Cyprinus carpio	96	4·5	Nishiuchi and Hoshimoto (1967)
	Macropodus cupanus	24	4·962	Present investigation
	Macropodus cupanus	48	4·594	Present investigation
	Labeo rohita	24	7·15	Arora *et al* (1971)
	Labeo rohita	96	5·05	Arora *et al* (1971)
	Pimephales promelas	24	25	Tarzwell (1958)
	Pimephales promelas	96	12·5	Henderson *et al* (1959)
	Pimephales promelas	96	22	Tarzwell (1958)
	Lepidocephalus thermalis	24	22·69	Rita (1977)
	Lepidocephalus thermalis	48	20·61	Rita (1977)
Sevin	*Oncorhynchus kisutch*	96	0·7	Macek and McAllister (1970)
	Ameiurus melas	96	0·8	Macek and McAllister (1970)
	Fundulus similis	24	1·75	Butler (1963)
	Lepomis macrochirus	96	2·0	Henderson *et al* (1959)
	Lepomis macrochirus	96	3·4	Edwards (1977)
	Salmo gairdneri	96	3·5	Edwards (1977)
	Aplocheilus lineatus	24	4·207	Present investigation
	Aplocheilus lineatus	48	3·747	Present investigation
	Mugil curema	24	4·25	Butler (1963)
	Perca flavescens	96	5·6	Macek and McAllister (1970)
	Casterosteus aculeatus	24	6·7	Stewart *et al* (1967)
	Pimephales promelas	96	13	Henderson *et al* (1959)
	Macropodus cupanus	24	14·73	Present investigation
	Macropodus cupanus	48	13·91	Present investigation

pesticide concentrations that produce adverse effects have been recorded, depending on the species, environmental factors and even biological status and origin of the test organism. It must however be mentioned that the LC_{50} values obtained for *A. lineatus* exposed to malathion are comparable to those recorded in the related *Aplocheilus blochii* (VCRC Annual Report 1979). Again, in the case of specimens exposed to DDT, *M. cupanus* is even hardier than the 'resistant' mosquito fish *G. affinis* (Johnson 1968). Again, *M. cupanus* is the most resistant of all the species studied to sevin.

It may also be noted that Das and Rajagopalan (1976) working on the susceptibility of mosquito larvae to insecticides found that in *Anopheles stephensi, Culex fatigans, Anopheles culicifacies* and *Aedes aegypti*, the critical doses of malathion required were 0·8, 0·064, 0·08 and 0·48 mg/l respectively. In the case of sevin it was a uniform 4·0 mg/l. With DDT, the LC_{50} value was 0·2 mg/l for *A. stephensi* and *C. culicifacies* while in the case of *Ae. aegypti* and *C. fatigans* it was 0·02 mg/l and 0·03 mg/l, respectively (VCRC Annual Report 1979). These values are fairly close to those reported in *A. lineatus* in the present study. Therefore, while *M. cupanus* could be utilised in conjunction with such insecticides for anti-larval work, *A. lineatus* should not be so used under any circumstances.

It has thus been demonstrated that even 'safe' and often minute dosages of pesticides are highly toxic to fish life, as may be seen from the LC_{50} values. Therefore, studies of this nature are essential as they provide information on the concentrations of environmental contaminants that cannot be tolerated by fish populations and consequently aid not only in the effective control of mosquito larvae by the fish but also in the protection of the aquatic environment.

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