

## ACUTE TOXICITY OF FIVE CHLORINATED HYDROCARBON INSECTICIDES TO THE FISH, *CHANNA PUNCTATUTS*

M. YAQUB JAVAID and ABDUL WAIZ

Department of Zoology, University of the Punjab, Lahore

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**Abstract.** A study of the acute toxicity of five chlorinated hydrocarbon insecticides to the fish, *Channa punctatus*, was carried out. The insecticides in decreasing order of toxicity to the fish were Endrin, Dieldrin, DDT, Aldrin and BHC, respectively. The behaviour of fish during exposure to insecticides was also observed.

Pakistan is basically an agricultural country. The use of insecticides, especially the chlorinated hydrocarbon insecticides, have been continuously mounting over the years for the control of the agricultural crop pests. With the increasing population, the rise in the requirements of food, will result in further increase in the use of these pesticides.

The chlorinated hydrocarbon insecticides are in fact 'biocides' which apart from controlling the pest insects, in very small doses, cause mass mortalities of fish and wildlife.<sup>2</sup> A great deal of work has been done in other countries on the toxic effects of these pesticides<sup>1,4,6,9-11,12,15-18</sup> and recently, in some countries their use has been banned, but unfortunately no attention has been paid to this problem in Pakistan.

This paper presents the results of laboratory experiments on the acute toxicity of five chlorinated hydrocarbon insecticides to a common fish, *Channa punctatus*, and is the first report of its kind in Pakistan.

### Material and Methods

#### Chemicals

The insecticides tested were as follows:

**Endrin.** 1,2,3,4,10,10-Hexachloro-6,7-epoxy-1,4,4a,5,6,7,8,8a-octahydro-1,4-endo-endo-5,8-dime-thanonaphthalene.

**Dieldrin** 1,2,3,4,10,10-Hexachloro-6,7-epoxy-1,4,4a-,5,6,7,8,8a-octahydro-1,4-endo,exo-5,8-dimethanaonphthalene.

**DDT.** 1,1,1-Trichloro-2,2-bis(*p*-chlorophenyl) ethane.

**Aldrin.** 1,2,3,4,10,10-Hexachloro-1,4,4a,5,8,8a-hexachloro-1,4-endo-exo-5, 8-dimethanonaphthalene.

**BHC.** 1,2,3,4,5,6-Hexachlorocyclohexane.

#### Animals

The *Channa punctatus*, used in this study, were caught by means of cast nets from (i) Khori fish sanctuary about 22 miles from Lahore on Lahore-Gujranwala road, (ii) Malam Klan near Kasur about 38 miles from Lahore, and (iii) Methasuja near Narowal about 35 miles from Lahore.

The fish were maintained in the laboratory in marble chips tanks measuring 4×2×1½ ft. Fish (100-150), were kept in each tank which contained approxi-

mately 22 gallons of water. The tanks were kept in a verandah and were not exposed to direct sunlight. The water in each tank was changed twice a week in winter and thrice a week in summer. All the tanks were scrubbed clean with a strong detergent and then thoroughly rinsed with clean water once a week.

Different types of foods such as fish-food tablets prepared by the Punjab Fisheries Department, minced beef liver, and minced beef were initially fed to *C. punctatus*, but only the minced beef was taken. The fish were then regularly fed on this 5 days a week, from Thursday to Monday in the morning. The unconsumed food was removed each morning before feeding. Feeding was discontinued once the fish were transferred to the experimental aquaria. The fish apparently remained healthy under these conditions and there was no mortality throughout the study.

No attempt was made to control water temperature. However, a temperature record was kept of all the tanks and aquaria containing the fish. The temperature was taken twice daily at 8 a.m. and 5 p.m. The months and average water temperatures during treatment with the insecticides are presented in Table 1.

The pH of the soft water used in the toxicity experiments was 7.2±0.1. The water in the experimental aquaria was always brought to equilibrium with the atmospheric oxygen by passing air through it for 12 hr before every experiment.

### Experimental Procedure

The toxicity experiments were carried out in aquaria measuring 2×1×1 ft. The front of each aquarium was made of ¼ in thick plexiglass while the other sides

TABLE 1. THE MONTHS AND AVERAGE WATER TEMPERATURES DURING TREATMENT WITH VARIOUS INSECTICIDES.

Insecticide	Period of treatment	Average water temperature during experiments (°C)	
		Mean	Range
DDT	December, 1969-January, 1970	13.0	12.0-14.5
BHC	January-February, 1970	12.0	10.3-11.7
Dieldrin	April, 1970	23.6	22.2-24.8
Endrin	May, 1970	25.8	23.3-26.5
Aldrin	May-June, 1970	26.1	24.0-27.2



were of wood which was coated on the inside with a layer of nontoxic plastic about  $\frac{1}{8}$  in thick.

Preliminary tests were carried out with each insecticide using two fish in 20 litres of water, to determine the range of toxicity. Once this was known, test concentrations of the insecticides were prepared in a logarithmic series as suggested by Doudoroff *et al.*<sup>3</sup> and Florin and Muller.<sup>7</sup> All concentrations were in p.p.m. by weight (mg/litre), and were prepared by adding 1 ml acetone containing the appropriate amount of insecticide to 20 litres of water. Control aquaria received 1 ml of acetone without any insecticide. Each concentration was tested in duplicate experiments and 10 fish measuring 10–15 cm length were used in each experimental aquarium. The fish of these lengths were selected as it has been suggested, that in toxicity experiments, the largest fish should not exceed the smallest by more than one and half times in length.<sup>3</sup>

During the course of an experiment with an insecticide, the number of dead fish in each aquarium was recorded at the end of each 24 hr period and the remaining live fish were transferred to other aquaria containing freshly prepared concentrations of the insecticide. Each experiment was terminated at the end of 96 hr. The criteria used to determine the death of the fish, as suggested by Doudoroff *et al.*<sup>3</sup> were the cessation of gill movements and lack of any response to mild mechanical stimulus with a glass rod.

The data were plotted on semilogarithmic paper with concentrations on the logarithmic scale and survival and mortality percentages on the arithmetic scale. From these plots the median tolerance limit (TLM), that is, the concentration at which 50 per cent of the fish survived for a specific period of exposure, was determined by straight line graphical interpolation for 24, 48, 72 and 96 hr respectively.

The behaviour of fish during the toxicity experiments was noted carefully to determine the differences if any, between the effects of various insecticides.

### Results

The toxicity of the five insecticides has been presented in Fig. 1 (A–E). The curves were drawn as lines of best fit by visual examination which represent percentage survival and mortality at 24, 48, 72, and

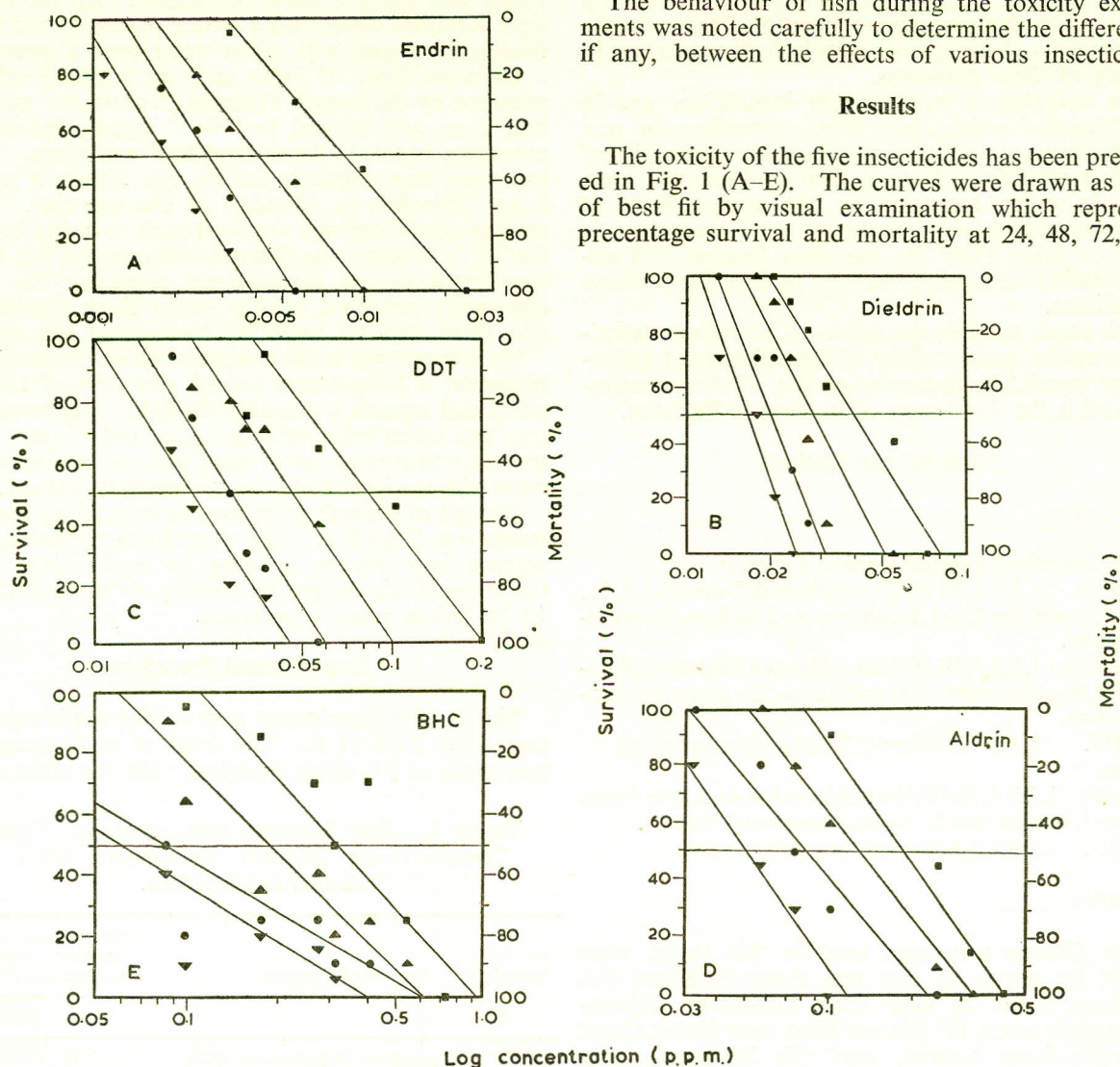


Fig. 1. Toxicity of five chlorinated hydrocarbon insecticides to *Channa punctatus*. The curves which were drawn as lines of best fit by visual examination represent the per cent survival and mortality for 24 hr (squares), 48 hr (triangles), 72 hr (circles), and 96 hr (inverted triangles), respectively. The line of 50% survival and mortality is also drawn for each insecticide.



TABLE 2. ESTIMATED TLM VALUES OF FIVE CHLORINATED HYDROCARBON INSECTICIDES FOR *Channa punctatus*.

Insecticide	TLM p.p.m. (mg/litre) active ingredient			
	24 hr	48 hr	72 hr	96 hr
Endrin	0.0085	0.0041	0.0027	0.0019
Dieldrin	0.0402	0.0290	0.0204	0.0168
DDT	0.0830	0.0460	0.0290	0.0214
Aldrin	0.1850	0.1280	0.0840	0.0510
BHC	0.3200	0.1900	0.0870	0.0630

96 hr, respectively. The median tolerance limit for each 24 hr period for each insecticide was computed by straight line graphical interpolation, has been given in Table 2. It can be surmised that for *C. punctatus*, the insecticides tested in the order of their toxicity are Endrin > Dieldrin > DDT > Aldrin > BHC.

The behaviour of *C. punctatus* during treatment with the insecticides was observed and found to be similar to that already described.<sup>8,15</sup> The fish became restless and highly sensitive to noise in the initial stages of insecticide-treatment as any tapping on the side of the aquarium resulted in violent movements. This was followed by a loss of sense of direction and the fish often bumped with the sides of the aquarium. Next, there was a partial upsetting of the equilibrium resulting in vertical or upside down swimming for short intervals. This was followed by a complete loss of equilibrium as the fish floated in water vertically or upside down. After this, the fish exhibited jerky movements and opened and closed its mouth violently. At this stage if it settled down it landed on its side on the bottom. Finally, the fish simply laid at the bottom of the aquarium and the only signs of life at this time were the slight movements of the operculum and the pectoral fins. Usually, the fish died soon after this.

These stages always followed the same sequence. The time interval that the fish remained in a particular stage depended upon the concentration and the toxicity of the insecticide. The stages were shortened during treatment with higher doses of insecticides or with more toxic insecticides.

The *C. punctatus* became lighter in colour and its abdomen was swollen at the time of death with all the insecticides. In addition, the fish exposed to higher concentrations of Endrin had bulging of eyes.

#### Discussion

It is obvious that *C. punctatus* is highly sensitive to the chlorinated hydrocarbon insecticides and minute doses prove to be lethal to the fish. Of the five insecticides tested, Endrin is the most toxic. This is in conformity with the observations made on other species.<sup>5,8,13</sup> The 96-hr TLM for Endrin for the present species, 0.0019 p.p.m. at 25.8°C, comes nearest to that reported for *Carassius auratus* (0.0021 p.p.m. at 25°C).<sup>8</sup> Dieldrin is the next in order of toxicity for which *C. auratus* has been observed to be most resistant of the species studied.<sup>8</sup> The *C. punctatus* (96-hr TLM 0.0168 p.p.m. at 23.6°C) fell very close to *Pimephales promelas* for which the 96-hr

TLM at 25°C was found to be 0.0180 p.p.m.<sup>8</sup> Aldrin has been found to be comparatively more toxic to fish than DDT by some authors<sup>8,13</sup> but others have found it less toxic.<sup>5</sup> In the species under study the latter has been found to be true. The 96-hr TLM value of Aldrin at 25°C for *Oncorhynchus kisutch* being 0.0459 p.p.m. is closer to *C. punctatus* (96-hr TLM 0.0510 p.p.m. at 26.1°C) than any other species examined.<sup>13</sup>

Our experiments with DDT and BHC were carried out at the temperatures 13 and 12°C respectively. The 96-hr TLM of these insecticides for *C. punctatus*, therefore, could not be compared with those reported in literature at 20–25°C for other species. Nevertheless, from the point of view of comparative toxicity of the insecticides to the present species, it can be stated that DDT is more toxic than Aldrin, and BHC is the least toxic of the insecticides studied.

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