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Malathion Induced Changes in the Optomotor Behavior of an Indian Carp, *Labeo rohita*

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The organophosphates such as malathion and carbamates are esterase inhibiting insecticides which enter the aquatic environment in large quantities (Coppage and Matthews 1974). Malathion reaches the aquatic environment by aerial spraying, runoff from agricultural lands and in factory effluents. Malathion has a low toxicity to mammals and a relatively high toxicity to fish (Mount and Stephan 1967). It is an anticholinesterase insecticide which blocks the synaptic transmission in the cholinergic parts of the nervous system (O'Brien 1960).

Aquatic organisms exhibit a broad range of responses to organophosphate insecticides depending on the compound, exposure time, water conditions and species (Eisler 1970; Coppage and Matthews 1974). It has been shown that some fish behaviors (e.g., locomotor activity and avoidance) are extremely sensitive to pollutants (Heath 1987). The change in the locomotor activity of bluegills exposed to sublethal doses of DDT, cadmium, and zinc was observed by Ellgaard et al. (1977, 1978). Macmillan (1987) recorded the changes in the optomotor responses of fathead minnows exposed to the herbicides alachlor and atrazine. The optomotor response is widespread throughout the animal kingdom. It is considered to be essential for maintenance of position within the habitat and for schooling in fish. This response is defined as a movement of the animal in the direction of moving reference points in the field of vision (Scherer and Harrison 1979).

The purpose of this study was to investigate the changes that occur in the optomotor behavior of malathion exposed carp, Labeo rohita (Ham), which is one of the most expensive edible fish of India. Malathion was selected because it is an organophosphorus insecticide used in large quantities in India.

MATERIALS AND METHODS

Labeo rohita (10-15 g) were obtained from ponds in and around Bhagalpur, India. They were fed twice daily with commercial fish food. Procedures for holding and acclimation were according to the

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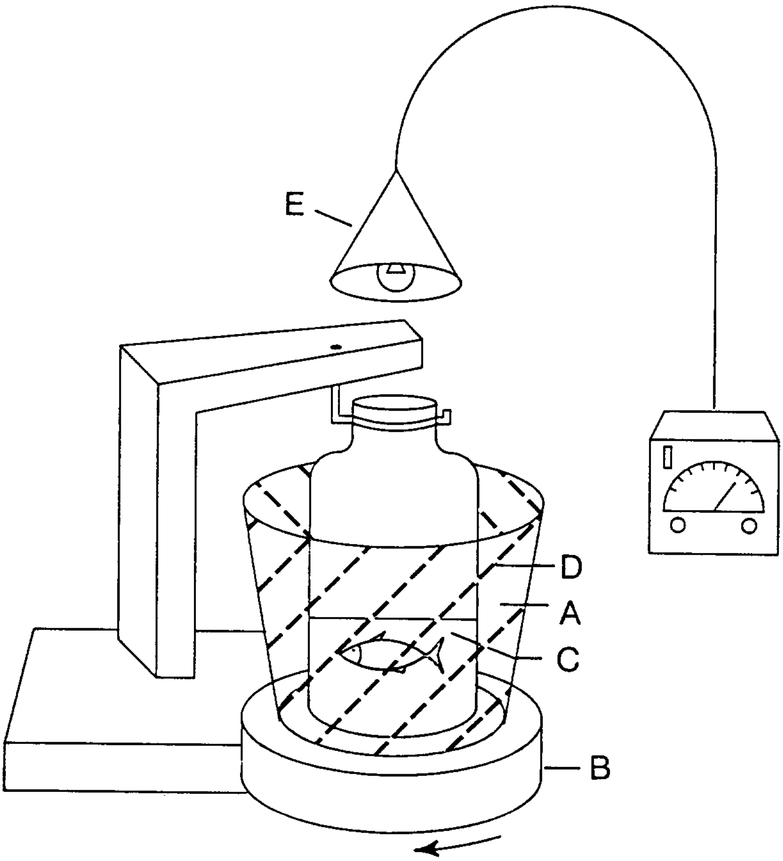

Figure 1. Optomotor response test apparatus
A-plastic bucket B-turntable
C-glass jar D-stripes
E-light

guidelines of the American Public Health Association (1975). The average values for water quality data for the water from the holding and exposure tanks were as follows: temperature 29^{0}C, pH 7.35, DO 7.5 mg/L, alkalinity 115 mg/L as $CaCo_3$, hardness 140 mg/L as $CaCo_3$.

Commercial grade malathion containing 50% active ingredient (North Minerals Ltd., Haryana, India) was used in this study. The fish were exposed to 0.2 mg/L, 0.4 mg/L, 0.6 mg/L, 0.8 mg/L and 1 mg/L malathion concentrations in large plexiglass aquaria using the exposure method as described by APHA et al. (1975). The duration of exposure was 24 hr. Ten fish were used for each trial. Two trials with controls were run for each concentration. Feeding was stopped 24 hr before the start of the exposure.

The optomotor response of experimental and control fish were measured using the apparatus shown in (Fig. 1). The apparatus consisted of a plastic bucket (A), a turntable (B) and a 2.2 L mason jar (C). The inside of the bucket wall contained 1.2 cm-wide stripes of black electrical tape placed 7 cm apart at an angle of 55^0 (D). The jar was suspended inside a plastic bucket. The glass jar was filled with water containing the same concentration of pesticide to which the fish were exposed. The bucket was made to rotate at 15 rpm with the direction of the movement being in the downward slope of the stripes.

An illumination of 20 lux (E) was used for the experiment. The bucket was made to rotate and the fish was allowed to settle down with the bucket rotating for 6 min. Rotation of the bucket was stopped for 6 min. Then fish behavior and movement were observed for 3 min during "off" period. This was followed by an observation and recording period of 3 min with the bucket rotating.

Every 90^0 turn or movement of the fish, referred as "quarter turn" was recorded as one movement. Quarter turns in the direction of the drum movements were recorded as "followings" and quarter turns in the opposite direction of the rotations were recorded as "reversals."

The mean scores of 20 readings of 10 fish, each with two trials for control and different concentrations of malathion exposed fish, were calculated and the standard deviation of the means was also derived. Based on mean scores, graphs were drawn for the "following" and "reversal" movements of the fish (control and at different concentrations of malathion) during bucket rotating and bucket stopped. Data were analyzed using students T test for the overall effect of the malathion dose on the optomotor behavior.

RESULTS AND DISCUSSION

Table 1 shows the mean scores of behavioral responses at different concentrations of malathion. The mean score for "following" during "on" position (bucket rotating) was significantly lower at 0.6, 0.8 and 1 mg/L concentrations compared with the scores for 0.0, 0.2 and 0.4 mg/L. At a concentration of 0.2 mg/L and 0.4 mg/L, the fish showed more activity compared with the control fish. Mean "following" and "reversal" scores were higher during the bucket rotating compared with the bucket stopped condition.

During the bucket stopped condition the scores for "following" were significantly ($p \leq 0.01$) higher at 0.2 and 0.4 mg/L exposure concentrations and lower at 1.0 mg/L exposure concentration.

At 0.2 mg/L and 0.4 mg/L exposure concentrations, the fish showed restlessness, both during "on" and "off" periods. In both "on" and "off" positions, another interesting feature was that the exposed fish changed directions in quick successions. In case of the highest concentration (1.0 mg/L) the "following" during "on" and "off" periods were occasionally performed on their backs. Sometimes the fish did not make "following" or "reversal" motions but rotated around an axis. The 1.0 mg/L concentration induced a heavy breathing in the fish and a remarkable reduction in the "following" and "reversal" movements.

Table 1. Mean (±S.D.) of "Following" and "Reversal" scores of fish at different concentrations of malathion exposure.

Concentrations (mg/L)	Rotations: Following, Bucket Rotating	Following, Bucket Stopped	Reversals, Bucket Rotating	Reversals, Bucket Stopped
0.0	79.35±16.03	10.80±3.16	12.05±3.82	9.80±2.27
0.2	97.50±21.87^{ns}	22.45±6.77^{++}	16.55±5.26^{ns}	12.50±2.64^{ns}
0.4	108.30±20.46^{ns}	29.15±10.45^{++}	13.40±3.69^{ns}	18.95±2.17^{++}
0.6	33.05±14.13^{++}	16.80±5.81^{ns}	16.55±5.93^{ns}	12.50±3.46^{ns}
0.8	28.25±5.13^{+++}	9.40±2.87^{ns}	7.20±1.49^{+}	6.25±1.31^{ns}
1.0	14.50±7.00^{+++}	5.70±1.36^{++}	8.70±2.38^{ns}	3.55±0.74^{++}

ns = not significant ++ = $p \leq 0.01$
+ = $p \leq 0.01$ +++ = $p \leq 0.001$

The score for "following" during "off" period was significantly higher at 0.2 mg/L and 0.4 mg/L exposure concentrations and significantly lower at the exposure concentration of 1.0 mg/L. "Reversal" score during "on" was significantly lower at 0.8 mg/L exposure concentration compared to other exposure concentrations. Reversal score during "off" was significantly higher at 0.4 mg/L and significantly lower at 1.0 mg/L.

Figure 2 (A-D) represents the four types of movements namely "following" during "on" (A), "following" during "off" (B), "reversal" during "on" (C) and "reversal" during "off" (D) periods. Except for the "reversal" during "on", all other scores exhibited a similar pattern. The "following" during "on", "following" during "off" and "reversal" during "off" rotations indicated an ascending trend in activity with an increase in the malathion concentration up to 0.4 mg/L. Above 0.4 mg/L exposure concentration the scores showed a negative relationship with increase in the concentration. The fish became hypoactive in 1.0 mg/L of malathion. "Reversal" scores during rotation (Fig. 2C) exhibited an erratic trend.

The optomotor function is essential for behaviors such as searching for food, orientation toward food odor, location of a mate, escaping from a predator and avoidance of an adverse situation. Three major behavioral patterns such as "hyperactivity," "hypoactivity" and "lethargy" have been noticed in the optomotor response of *Labeo rohita*.

At an exposure concentration of 0.2-0.4 mg/L the fish exhibited hyperactivity. Henry (1984) and Henry and Atchison (1986) have observed hyperactivity in bluegills exposed to methylparathion and copper. The hyperactivity has been defined as an almost continuous swimming combined with numerous S-jerks, partial jerks and flickering of the fins. These responses were observed by the aforementioned authors to all toxicant concentrations with 10-14 hr of exposure.

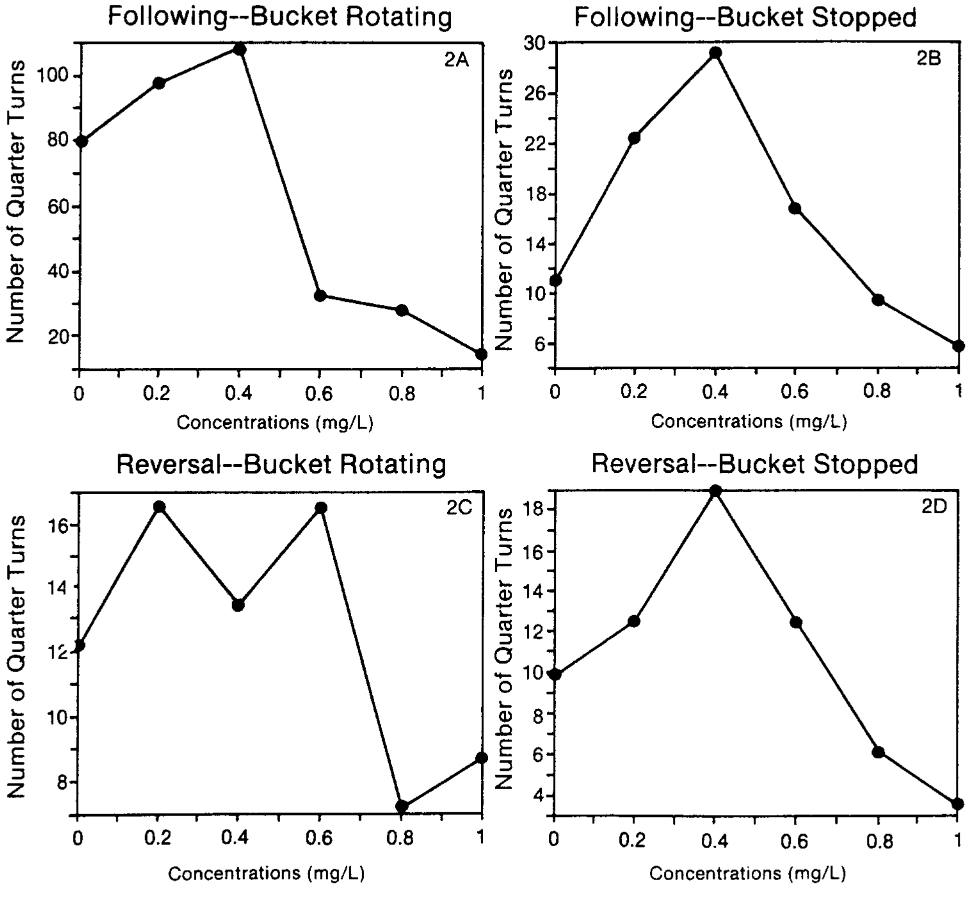

Figure 2. Changes in four types of movements: control and exposed fish at different concentrations of malathion

Labeo rohita (Ham) showed signs of lethargy to a considerable extent above an exposure concentration of 0.6 mg/L. In natural habitat this lethargy would make these fish an easy prey to any predator. A similar observation was made by Matton and LaHam (1969) after an exposure to the organophosphate pesticide Dylox in rainbow trout larvae. According to them, after exposure, the larvae became hyperactive. In the case of Lebeo rohita, hyperactivity led into hypoactivity until they turned on their sides.

The loss of orientation is expected to be more in the fish inhabiting water having currents or waves and affected by organophosphate or any other kind of insecticide pollutants. A partial loss of orientation was observed in carp exposed to concentrations of 0.6 mg/L, 0.8 mg/L and 1.0 mg/L. There was considerable hypoactivity in such exposures. According to Bull and McInerney (1974) many fish (juvenile coho salmon) were unable to maintain position and were swept downstream after being exposed to an organophosphorus insecticide (sumithion) in a stream aquarium.

Occasional somersaults noticed in 0.2 mg/L and 0.4 mg/L exposure concentrations may be signs of stress shown by the fish. Singh and Sahai (1984) have also noticed erratic jumping, swimming on one side of the body in the freshwater teleosts, Rasbora daniconius and Puntius ticto exposed to lethal and higher concentrations of malathion respectively. At all concentrations of malathion exposure, a partial dysfunction of the pectoral fins was observed. Erratic swimming movements followed by lethargy in Clarias batrachus exposed to 0.25 to 2.00 mg/l malathion was also reported by Sharma et al. (1983).

The results of this study indicate that exposure of fish to malathion in the aquatic environment may not cause immediate death, but certainly would bring about some behavioral changes or inability to continue their normal activity.

A behavioral bioassay is quick and sensitive and can be easily performed. An optomotor response is a behavior which helps the fish to differentiate useful objects and predators in the aquatic environment. This behavioral index provides an excellent measure of the effect of a toxicant on the fish. A rapid and objective quantification of normal behavior and pollutant induced changes in fish may be used to assess and evaluate the effects of sublethal doses of pollutants that are found in very small quantities in the aquatic environment.

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