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Assessing toxicity of organophosphorus insecticide on local fish species of Bangladesh

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Abstract

Pesticide is a major factor for fish mortality along with water temperatures. Six fish species namely Climbing perch *(Anabas testudineus),* spotted snakehead *(Channa punctatus),* Thai shorputi (*Barbodes gonionotus),* Indian torrent catfish *(Amblyceps mangois),* pool barb *(Puntius sophore*) and tangra *(Batasio tengana*) were exposed to various concentrations of Confidor 70WG to assess the lethal concentrations (LC_{50}). For 96 hours of exposure, the median lethal concentration (LC_{50}) values of Confidor 70WG on these five species were 46.49, 40.80, 35.32, 49.25, 27.61 and 44.51 ppm, respectively. Mortality rate and behavioral changes of fish at different concentrations were determined. Some physicochemical parameters of water such as temperature, pH, dissolved oxygen, total dissolved solids and electrical conductivity were also measured and compared with the aquaculture water quality standard. Temperature remained within the range of 25-30 °C and the pH of water was 6.6-7.0 in presence of pesticide. Physiological responses like rapid opercular movement, hyper secretion of mucus and frequent gulping of air was observed during the initial stages of exposure after which it became occasional. By the cause of Confidor 70WG, the pectoral fins of the fishes were reversed. In stress, the unusual behavior of experimental fishes may be due to obstructed functions of neurotransmitters.

Keywords: Fish, Insecticide, Toxicity.

1. Introduction

Now a days, the mortality of fish due to the application of organophosphorus pesticides in crop field which has a great impact on aquatic system [12]. This has resulted from the agricultural contamination of waterways through fallout, drainage, or runoff erosion, and from the discharge of industrial effluents containing pesticides into waterways. Fish mortality occurs due to various natural and human-induced causes. Natural causes include extreme temperature fluctuations, starvation, and disease as for example Blue tilapia (*Tilapia aurea*) was died because of cold weather events due to low dissolved oxygen [33]. Naturally occurring fish mortality can be related to physical processes (e.g., rapid fluctuations in temperature), water chemistry changes (e.g., low dissolved oxygen and/or changes in pH), or they can be biological in nature (e.g., viruses, bacterial infection and/or parasites). Many of these processes are difficult to determine because by the time an investigator arrives at a fish mortality, fish have rotted or been partially eaten, and water chemistry has changed or moved down stream. Thus, many of the Florida fish mortality reports with unknown causes (13%) could have been of natural origin. Additionally, U.S. Fish and Wildlife Service [32] suggests it is easier to detect human-induced fish mortalities because there is generally a point source or obvious evidence of a spill, reducing the probability that the unknown causes are human induced.

Pesticide toxicity to fish has been investigated in several studies [34, 21]. However, fish are not usually target organisms for pesticides, and knowledge about effects of pesticides in the field is still sparse. Surprisingly, only a few studies have shown that fish, inhabiting natural freshwater ecosystems, may be affected by unintentional spreading of pesticides [6, 10]. The risk associated with contaminants was assessed using risk quotients based on the comparison of measured concentrations with original species sensitivity distribution-derived hazardous concentration values [1]. When croplands are treated, some impacts of pesticides occur on non-target terrestrial and aquatic ecosystems, as well as on adjoining agro ecosystems [9]. Unfortunately, farmers may not be aware of the source of the pesticides sold to them and unwittingly introduce dangerous chemicals into the environment [31].

Fish species are sensitive to enzymatic and hormone disruptors. Chronic exposure to low levels of pesticides may have a more significant effect on fish populations than acute poisoning. Doses of pesticides that are not high enough to mortality fish are associated with subtle changes in behavior and physiology that impair both survival and reproduction [15]. The fish show restlessness, rapid body movement, convulsions, difficulty in respiration, excess mucous secretion, change in color, and loss of balance when exposed to pesticides. Similar changes in behavior are also observed in several fishes exposed to different pesticides [5]. Organophosphorus pesticides are known to cause morphological damage to the fish testis and also affect delayed oocyte development and inhibition of steroid hormone synthesis [16].

Bangladesh possesses much more open waters than closed ones, due attention must be paid to these water bodies to increase the fish production significantly. Unfortunately, open water fish production has decreased because of various natural and man-made interventions. Indiscriminate use of pesticides is one of the causes of decreased fish production. Pesticides are useful tools in agriculture and forestry, but the gradual degradation of aquatic ecosystem and consequent disaster cannot be ignored. Therefore**,** the objectives of this study are to evaluate the toxic effects of insecticides on local fish population and to assess the cause of the extinction of certain fish species and the species are going to be extinct.

2. Materials and Methods

The experiments were conducted in the laboratory of the Department of Agricultural Chemistry, Hajee Mohammad Danesh Science and Technology University Dinajpur, Bangladesh. Six fish species of an average size of *Anabas testudineus*, averaging 11.45±0.94 cm, *Channa punctatus*, averaging 13.29 ± 1.08 cm, *Barbodes gonionotus* averaging 13.0 ± 1.55 cm, *Amblyceps mangois* averaging size 11.04±1.24 cm, *Puntius sophore* averaging size 9.33±0.61 cm and *Batasio tengana* averaging size 11.01±0.60 cm were collected in their maturing stage from unpolluted water body without any injury. Eighteen plastic dishes of size 60 cm × 30 cm × 30 cm with 50 litres of tap water were used in the experiments. The treatments were selected based on range finding test and the concentrations were 20, 40, 60, 80 and 100 ppm of Confidor 70WG including a control with three replications. In the test dishes desired concentrations of pesticides were poured carefully and mixed gently with a glass rod. Fifteen fishes each were released after proper acclimatization in dishes containing different concentrations of pesticide as well as in the control. All tests were done at ambient temperature. The behavior and other external changes in the body of fishes were observed. Dead fishes were removed and mortality was recorded at 6, 12, 24, 48, 72 and 96 hours of exposure time. Temperature, dissolved oxygen, electrical conductivity, total dissolved solids (TDS) and pH of the test media were recorded daily. The temperature was measured by a thermometer, dissolved oxygen and pH were measured by DO meter (Model 2020, UK) and pH meter (Model YSI 58, USA) respectively. The TDS was determined following the methods given by APHA (2005). The EC values of water samples were measured by the conductivity bridge (model WTW LF521). During the experimental period, the fishes were in starved condition. All the precautions laid by committee on toxicity tests to aquatic organisms were followed [4]. The LC_{50} values for different fish species were calculated for 96 hours of exposure time by probit analysis of the computer program SPSS.

2.1 Probit analysis

Probit analysis is used to analyze many kinds of dose-response or binomial (e.g. death/no death) response experiments in a variety of fields. By plotting the response of the fishes to various concentrations of pesticides it is observed that each pesticide affected the different fish species at different concentrations, i.e. the rate of mortality of the fish species is not equal. The most logical approach would be to fit a regression of the response versus the concentration, or dose and compare between the different pesticides. Yet, the relationship of response to dose was sigmoid in nature and at the time regression was only used on linear data. The probit model may be expressed mathematically as follows:

$$P = \alpha + \beta\,[\log_{10}(\text{dose})]$$

Where P is five plus the inverse normal transform of the response rate (called the probit). The five is added to reduce the possibility of negative probits, a situation that caused confusion when solving the problem by hand. Probit analysis is a specialized regression model of binomial response variables and a binomial response variable refers to a response variable with only two outcomes. And regression is a method of fitting a line to your data to compare the relationship of the response variable or dependent variable (Y) to the independent variable (X).

$Y = a + b X + e$

Where a = y-intercept

b = the slope of the line

e = error term

3. Results and Discussion

3.1 Mortality studies

An increase in the number of the mortalities with increase in concentration of the toxicity of Confidor 70WG was observed. There was no mortality at low concentration and as well as control group. The probit analysis for acute toxic test at 96 h estimated lethal concentration values and their confidence intervals were shown in Table 1-6. It is clear from Figure 1 that as the concentration of the pesticide chemical increased, fish mortality also increased, which indicates a direct proportional relationship between mortality and concentration of test chemical. No mortality was observed during the experimental period in controls.

The mortality rate of all experimental fish species increased together with the increment of the Confidor 70WG concentrations in water (Table 1-6). In this experiment the results from the six local fish species were different in different concentrations but the trend is similar i.e. the limit of probit is increasing with the increasing rate of concentration. For *Anabas testudineus*, the lethal concentration (LC_{50}) was 46.49 ppm at 95% confidence limit. The lower limit was 34.91 ppm and the upper limit was 58.41 ppm (Table 1). For *Channa punctatus*, the lethal concentration (LC_{50}) was 40.80 ppm at 95% confidence limit where the lower limit and the upper limit were 29.84 ppm and 51.15 ppm, respectively (Table 2) while for *Barbodes gonionotus* the lethal concentration (LC_{50}) was 35.32 ppm at 95% confidence limit and the lower limit and the upper limit was 25.37 ppm and 44.03 ppm, respectively (Table 3). At 95% confidence limit*,* the lethal concentration (LC_{50}) for *Amblyceps mangois* was 49.25 ppm, and the lower limit and the upper limit were 39.15 ppm and 59.57 ppm, respectively (Table 4). In case of *Puntius sophore*, the lethal concentration (LC_{50}) was 27.61 ppm at 95% confidence limit.

The lower and upper limits were 16.33 ppm and 36.27 ppm, respectively (Table 5). For *Batasio tengana,* the lethal concentration (LC_{50}) was 44.51 ppm at 95% confidence limit while the lower limit was 32.85 ppm and the upper limit was 56.37 ppm (Table 6).

Linear relationship between the mortality and the insecticide concentrations indicated a positive correlation and showed a significant difference at $P<0.05$. The present study indicates that the mortality rate of exposed fish increased concomitantly with the increase in the concentration of Confidor 70WG ($r^2 \geq 0.932$, 95% confidence interval). Steep slope functions of the toxicity curve of 96 h mortality concentration data for Confidor 70 WG indicate a large increase in the concentration of the insecticide. Steep slope functions of the toxicity curves are due to rapid absorption of the insecticides and rapid onset of effects. Flat slope function indicates slow absorption, rapid excretion, detoxification or delayed toxicity of the toxicant [9]. Compared to test organisms, *Labeo rohita* proved to be a particularly sensitive and reliable indicator as it appears from the high factor of determination LC_{50} for Confidor 70WG. Decreased swimming behavior and increased respiration rate were other effects of pesticides in the present study, found that contaminants such as pesticides disturb normal fish behaviour after exposure [29].

Exposure to toxic substances may not result in immediate fish mortalities, but may affect fish populations by decreasing fecundity (number of eggs produced), reducing the viability of sperm, eggs and larvae, decreasing life expectancy, increasing the incidence of abnormalities and increasing natural mortality [2]. Fish mortality occurs when pesticides are improperly applied to or otherwise end up in bodies of water through either misapplication or drift [35]. Disruption of schooling behaviour of the fish, due to the lethal and sub lethal stress at the toxicant, results increased swimming activity and entails increased expenditure of energy [20]. The erratic swimming of the treated fish indicates loss of equilibrium. An alteration in the levels in tissues of fresh water was observed in *Labeo rohita* exposed to Fenvalerate [11].

Several studies demonstrate the values of LC_{50} and the effects of the Atrazine on teleosts fish. The LC_{50}-96h for trout embryos and larvae exposed to Atrazine ranged from 0.87 to 1.11 mg/L [23]. In *Tilapia mossambicus*, the Atrazine LC_{50}-96h was 8.8 ppm [24]; in *Rhamdia quelen*, it was 10.2 ppm [17] and in *Cyprinus carpio* it was 18.8 ppm [22]. Figure 1 shows different lethal concentration (LC_{50}) levels on different fish species in this experiment. Among these results it is clear that lethal concentration is varying species to species. Because the shape, size, physiological adaptability and body strength in these six species was different. Among the study species of fishes, the higher lethal concentration of *Amblyceps mangois* was 49.25 ppm and the lower was 27.61 ppm in *Puntius sophore*. LC_{50} value for 96 hours was 95.03 ppm for Basudin (Diazinon) 10G on *Clarias gariepinus*[18]. The LC_{50} for *Anabas testudineus* was lower than both of the above findings, however, the test species was different and Malathion was also another organophosphate. LC_{50} value for Malathion on fathead minnows to be 23 ppm which was lower than the LC_{50} value of the present experiment for *Anabas testudineus* while the test fish and test chemical was different [3]. The LC_{50} value of 40.80 ppm of the present work for *Channa punctatus* was higher than *Channa striatus*. LC_{50} value of Basudin 10G on *Barbodes gonionotus* fry to be 380.14 ppm for 24 hours of exposure [18]. The LC_{50} value of Dipterex on *Oreochromis niloticus* was reported to be 253 ppm at 12 hours [28]. However, the LC_{50} value of the present experiment on *Barbodes gonionotus* was higher than the result of Dipterex on *Oreochromis niloticus* [28].

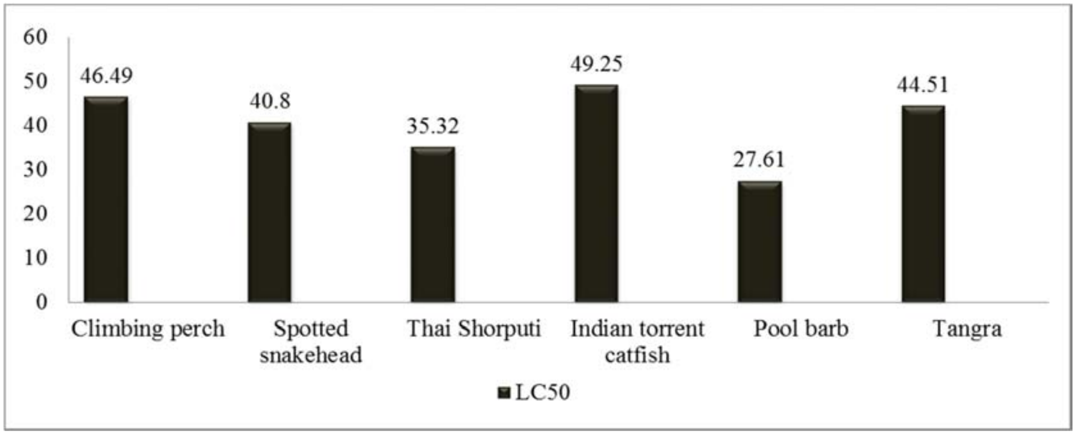

Fig 1: Value of LC_{50} on six experimental fish species

Table 1: Probit analysis on the effect of Confidor 70WG to *Anabas testudineus* at 96 hours of exposure.

Concentration (ppm)	Log concentration	No. of organism	No. of organism dead	Percent mortality	Probit	LC50 (ppm)	95% confi. limit Lower (ppm)	Upper (ppm)
00	-	15	00	00	-	46.49	34.91	58.41
20	1.30	15	02	13	3.87			
40	1.60	15	07	46	4.90			
60	1.78	15	08	53	5.08			
80	1.90	15	10	66	5.41			
100	2.00	15	15	100	7.33			

Intercept (a) = - 5.478, Regression co-efficient (b) = 3.286, Heterogenicity (χ^2) = 4.514 (Not significant)

* Probit = N.E.D. increased by 5, N.E.D. - Normal equivalent deviate, confi.= confidence

Table 2: Probit analysis on the effect of Confidor 70WG to *Channa punctatus* at 96 hours of exposure.

Concentration (ppm)	Log concentration	No. of organism	No. of organism dead	Percent mortality	Probit	LC50 (ppm)	95% confi. limit	
							Lower (ppm)	Upper (ppm)
00	-	15	00	00	-	40.80	29.84	51.15
20	1.30	15	03	20	4.16			
40	1.60	15	07	46	4.90			
60	1.78	15	09	60	5.25			
80	1.90	15	12	80	5.84			
100	2.00	15	15	100	7.33			

Intercept (a) = - 5.336, Regression co-efficient (b) = 3.313, Heterogenicity (χ^2) = 2.944 (Not significant)
* Probit = N.E.D. increased by 5, N.E.D. - Normal equivalent deviate, confi.= confidence

Table 3: Probit analysis on the effect of Confidor 70WG to *Barbodes gonionotus* at 96 hours of exposure.

Concentration (ppm)	Log concentration	No. of organism	No. of organism dead	Percent mortality	Probit	LC50 (ppm)	95% confi. limit	
							Lower (ppm)	Upper (ppm)
00	-	15	00	00	-	35.32	25.37	44.03
20	1.30	15	03	20	4.16			
40	1.60	15	09	60	5.25			
60	1.78	15	11	73	5.61			
80	1.90	15	13	86	6.08			
100	2.00	15	15	100	7.33			

Intercept (a) = - 5.562, Regression co-efficient (b) = 3.593, Heterogenicity (χ^2) = 1.407 (Not significant)
* Probit = N.E.D. increased by 5, N.E.D. - Normal equivalent deviate, confi.= confidence

Table 4: Probit analysis on the effect of Confidor 70WG to *Amblyceps mangois* at 96 hours of exposure.

Concentration (ppm)	Log concentration	No. of organism	No. of organism dead	Percent mortality	Probit	LC50 (ppm)	95% confi. limit	
							Lower (ppm)	Upper (ppm)
00	-	15	00	00	-	49.25	39.15	59.57
20	1.30	15	01	07	3.52			
40	1.60	15	06	40	4.75			
60	1.78	15	08	53	5.08			
80	1.90	15	11	73	5.61			
100	2.00	15	15	100	7.33			

Intercept (a) = - 7.000, Regression co-efficient (b) = 4.136, Heterogenicity (χ^2) = 3.154(Not significant)
* Probit = N.E.D. increased by 5, N.E.D. - Normal equivalent deviate, confi. = confidence

Table 5: Probit analysis on the effect of Confidor 70WG to *Puntius sophore* at 96 hours of exposure.

Concentration (ppm)	Log concentration	No. of organism	No. of organism dead	Percent mortality	Probit	LC50 (ppm)	95% confi. limit	
							Lower (ppm)	Upper (ppm)
00	-	15	00	00	-	27.61	16.33	36.27
20	1.30	15	06	40	4.75			
40	1.60	15	09	60	5.25			
60	1.78	15	12	80	5.84			
80	1.90	15	14	93	6.48			
100	2.00	15	15	100	7.33			

Intercept (a) = - 4.384, Regression co-efficient (b) = 3.042, Heterogenicity (χ^2) = 1.819 (Not significant)
* Probit = N.E.D. increased by 5, N.E.D. - Normal equivalent deviate, confi.= confidence

Table 6: Probit analysis on the effect of Confidor 70WG to *Batasiotengana* at 96 hours of exposure.

Concentration (ppm)	Log concentration	No. of organism	No. of organism dead	Percent mortality	Probit	LC50 (ppm)	95% confi. limit	
							Lower (ppm)	Upper (ppm)
00	-	15	00	00	-	44.51	32.85	56.37
20	1.30	15	03	20	4.16			
40	1.60	15	06	40	4.75			
60	1.78	15	08	53	5.08			
80	1.90	15	11	73	5.61			
100	2.00	15	15	100	7.33			

Intercept (a) = - 5.183, Regression co-efficient (b) = 3.144, Heterogenicity (χ^2) = 4.248 (Not significant)
* Probit = N.E.D. increased by 5, N.E.D. - Normal equivalent deviate, confi.= confidence

3.2 Behavioral studies

The behavioral changes were also seen in the present investigation, over the duration of 96 h of exposure to Confidor 70WG, overall statistically significant changes in almost all behavioral patterns were observed in six fish species. The surfacing phenomenon of fish observed under Confidor 70WG, overall exposure. The control group shows the normal behavior during the whole experiment and also normal responses was observed at the low concentration (20 mg/L). After 24 h of exposure to Confidor 70 WG, significantly increased hyperactivity in terms of surfacing and scraping moments and schooling were observed in comparison to control. At 48 h of exposure, the surfacing as well as scraping moments decreased in fish species, other behaviors increased significantly hyper secretion of mucus, opening mouth for gasping, losing scales, hyperactivity were observed. After 72 h of exposure, all the fishes showed decreased surfacing and jerky movements and increased grasping movements, sank to bottom of the test chamber and independency in swimming. Subsequently fish moved to the corners of the test chambers, which can be viewed as avoidance behavior of the fish to the toxicant. During the 96 h of exposure, all body activities were nearly ceased. Complete loss of body balance, exhibited irregular, erratic, darting swimming movements and loss of equilibrium were followed by hanging vertically in water. Hyper excitation, loss of equilibrium, increased cough rate, flaring of gills, increase in production of mucus from the gills, darting movements and hitting against the walls of test tanks were noticed in all the species tested. A film of mucus was also observed all over the body and also on the gills. The increase ventilation rate by rapid, repeated opening closing mouth and opercula coverings accompanied by partially extended fins (coughing) was observed. This could be due to clearance of the accumulated mucus debris in the gill region for proper breathing. This may be due to the activity of organophosphorus insecticides acting on the enzyme acetyl cholinesterase especially when dealing with neurotoxic compounds. Neurotransmitters are acetyl choline profoundly important in the brain's development, in the absence of acetyl cholinesterase; acetylcholine level increased resulting in the failure of transmission of stimuli to the nerves or organs. Although carbmate/ organophosphorus pesticides tend to undergo fairly rapid degradation in the environment, repeat input into the aquatic environment may result in harmful exposures. The loss of balance in swimming and walking in both prawn species with increasing metal concentration may due to impairments either in nervous tissues or in muscle fibres [30]. This leads to the abnormal functioning of the body including loss of balance, moving in circular form (convulsions) and at higher concentrations of insecticides resulting in death of the organism [13]. Fish in the experimental group applied with highest concentration of the pesticide were lying laterally at bottom with loss of balance, swimming down in a spiral movement with jerks.

No marked instant behavioral responses were observed. After 48h of insecticide exposition, hemorrhages was observed in the eyes, lips or even in the whole body of fish exposed at the concentrations of 20, 40 and 60 ppm. These concentrations induced loss of equilibrium and lethargic behavior. The frequency of opercular movements and the thickness of the inferior lips were either notably increased. This casual lips development is already well document as an adaptation of fish to certain situations where the increase of the inferior lips surface can facilitate the oxygen uptake by the process called as aquatic surface respiration. Changes in fish behavior can be used as indicator of acute changes in the environmental chemical composition [26]. For example, goldfish exposed to a 5 µg L^{-1} Atrazine solution affected directly and indirectly fish behavior, altering the chemical perception of natural substances of eco-ethological importance [26]. The median lethal concentration values (LC_{50}) of Dimecron 100SCW were estimated to be 46.75, 22.95 and 375.26 ppm for *Anabas testudineus, Channa punctatus* and *Barbodes gonionotus* for 96 hours exposure respectively [14]. At the higher doses, slight increase in opercular movement and restlessness were found. With the increase of exposure time the fishes became sluggish, settled at the bottom, opercular activities became slower, ultimately the fish became completely paralyzed and died in an increasing order at the higher doses. Normal shiny color and behavior of test fishes were observed in the control groups, while the color became light yellowish-red in an increasing order towards the higher doses at the end of 96 hours exposure time. Dimecron was very little toxic to *Heteropneustes fossilis* and treated fishes swam freely in the aquarium containing the chemical, which were similar to the findings of the present work. However, at the higher doses slight increase in opercular movement and restlessness were observed [25].

The water quality parameters viz., temperature, dissolved oxygen; pH, total dissolved solids and electrical conductivity of the test medium are presented in Table 7-12. The average dissolved oxygen was higher in the lower concentrated media. Metabolites and pesticidal effects might be the probable reasons of declining oxygen concentration in the lower to higher concentrated test media during the present study. However, the water quality parameters of the test media varied little and were within the desirable range fish for *Anabas testudineus, Channa punctatus*, *Barbodes gonionotus, Amblyceps mangois, Puntius sophore and Batasio tengana* [8].

Table 7: Water quality parameters of the test media (Confidor 70WG) on *Anabas testudineus* during the experimental period.

Conc. (ppm)	Temperature (°C)	Dissolved oxygen (ppm)	pH	Total dissolved solids (mg/L)	Electrical conductivity (µS/cm)
00	30.1±0.06	7.8±0.05	6.8±0.01	152	260
20	30.6±0.08	7.7± 0.06	6.6± 0.10	160	265
40	30.6 ±0.10	7.6± 0.08	6.7 ± 0.13	161	273
60	30.6±0.12	7.4±0.08	6.7 ±0.01	163	273
80	30.7±0.41	7.2±0.03	6.8± 0.02	165	274
100	30.7±0.89	7.0±0.05	6.9± 0.02	165	277

Table 8: Water quality parameters of the test media (Confidor 70WG) on *Channa punctatus* during the experimental period.

Conc. (ppm)	Temperature (°C)	Dissolved oxygen (ppm)	pH	Total dissolved solids (mg/L)	Electrical conductivity (µS/cm)
00	27.5±0.58	7.6± 0.08	6.8± 0.01	156	254
20	27.7±0.79	7.6± 0.04	6.5± 0.10	162	261
40	28.6±0.87	7.5± 0.06	6.6 ±0.10	163	263
60	28.7±0.96	7.5± 0.08	6.7 ±0.01	164	263
80	28.7±1.1	7.4±0.05	6.9 ±0.01	165	264
100	28.7±1.16	7.1±0.05	6.9± 0.01	166	267

Table 9: Water quality parameters of the test media (Confidor 70WG) on *Barbodes gonionotus* during the experimental period.

Conc. (ppm)	Temperature (°C)	Dissolved oxygen (ppm)	pH	Total dissolved solids (mg/L)	Electrical conductivity (µS/cm)
00	27.2±0.48	8.0 ±0.13	6.8± 0.01	142	234
20	27.3±0.48	7.9± 0.08	6.4± 0.14	154	241
40	27.6±0.50	7.8± 0.06	6.6 ±0.05	153	243
60	28.0±0.82	7.6± 0.08	6.6± 0.10	158	243
80	28.7±0.91	7.7 ±0.24	6.8± 0.07	159	244
100	29.2±0.93	7.6 ±0.14	6.8± 0.01	156	248

Table 10: Water quality parameters of the test media (Confidor 70WG) on *Amblyceps mangois* during the experimental period.

Conc. (ppm)	Temperature (°C)	Dissolved oxygen (ppm)	pH	Total dissolved solids (mg/L)	Electrical conductivity (µS/cm)
00	27.8± 0.75	8.0± 0.56	6.8± 0.01	142	234
20	28.1± 0.81	7.9± 0.10	6.3 ±0.19	143	241
40	28.8± 0.82	7.7 ±0.10	6.6± 0.07	144	242
60	28.5± 1.05	7.6± 0.39	6.6 ±0.16	144	243
80	29.0± 1.14	7.5± 0.36	6.6± 0.09	145	244
100	30.4± 0.24	7.5 ±0.37	6.7± 0.08	146	245

Table 11: Water quality parameters of the test media (Confidor 70WG) on *Puntius sophore* during the experimental period.

Conc. (ppm)	Temperature (°C)	Dissolved oxygen (ppm)	pH	Total dissolved solids (mg/L)	Electrical conductivity (µS/cm)
00	27.9± 0.64	7.8± 0.15	6.8± 0.01	142	234
20	28.0± 0.70	7.6± 0.37	6.4± 0.10	139	231
40	28.3± 0.74	7.5± 0.31	6.5± 0.08	143	233
60	28.3± 1.10	7.5± 0.31	6.7± 0.06	144	233
80	28.5± 0.58	7.3± 0.20	6.8 ±0.08	145	234
100	28.6 ± 1.10	7.2± 0.28	6.9± 0.01	147	236

Table 12: Water quality parameters of the test media (Confidor 70WG) on *Batasio tengana* during the experimental period.

Conc. (ppm)	Temperature (°C)	Dissolved oxygen (ppm)	pH	Total dissolved solids (mg/L)	Electrical conductivity (µS/cm)
00	28.1± 0.64	7.8± 0.19	6.8± 0.01	132	234
20	28.3± 0.88	7.6± 0.24	6.7 ±0.08	133	231
40	28.3 ±1.08	7.6 ±0.40	6.7± 0.09	133	230
60	28.7± 1.28	7.6± 0.26	6.7 ±0.09	138	233
80	28.8± 0.83	7.4 ±0.16	6.7± 0.02	135	234
100	29.5 ±0.90	7.4± 0.16	6.8± 0.04	134	232

4. Conclusions

Usually Confidor 70WG is used at a dose of 3458 ppm in the crop field by the farmers. But in this experiment six treatments were used including a control dose where the doses were lower than the recommended dose (3458 ppm) used by farmers in the field. In spite of using this lower doses all fish species used in this experiment were badly affected and their percentages of mortality were high. In this study, test organisms showed jerky movements, hyper secretion of mucus, opening mouth for gasping, losing scales, hyperactivity were observed with the increase in insecticidal concentration. Behavioral characteristics are obviously sensitive indicators of toxicant effect. All the pesticides induced oxidative stress in the form of behavioral responses in fish. Acute toxicity studies have been recognized as the very first step in determining the water quality management of fish and reveal toxicant concentrations that cause fish mortality even at short period of exposure. Additional research work is on the test species needed to arrive at more definite general conclusions. It should remember that only insect mortality should not be the target for sustainable environment. For sustainability we should try to use the minimal concentration that can control the insects as well as can save the aquatic and terrestrial environment.

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