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PROTECTIVE ROLE OF ASCORBIC ACID ON ENDOSULFAN INDUCED ALTERATIONS IN THE BEHAVIOR OF THE FRESHWATER FISH, CHANNA ORIENTALIS (SCHNEIDER)

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Abstract: Fresh water fishes, *Channa orientalis* were exposed to chronic dose of endosulfan without and with ascorbic acid. Behavioral changes were record. Remarkable behavioral changes were observed in pesticides. The groups exposed to pesticides along with ascorbic acid showed recovery than those exposed to only pesticide. Pre-exposed fishes to pesticides showed fast recovery than those, which were allowed to naturally. The probable role of ascorbic acid is discussed in the paper.

Keywords- Endosulfan, Behavior, Ascorbic acid, *Channa orientals*.

INTRODUCTION

Endosulfan is a cyclodiene insecticide. The symptoms of poisoning in insects and animals are hypersensitivity, hyperactivity with convulsive movement, disturbance in the ganglia of the central nervous system rather than in the peripheral nerves. In India, it has been identified as one of the main pesticides found in the waters of major rivers in a study conducted by central pollution control board of India [1]. In agricultural consumption of endosulfan was estimated to be 5,200 metric tons in 1994-95 [2]**.** Endosulfan may also be toxic to non target organisms like fish by altering the physiology, metabolism, behavior and fecundity of fish, ultimately affecting the survival of the population [3]**.** Pollution is an undesirable change in the physical, chemical or biological characteristics of air, land and waters that are harmful to human life. Rapid industrialization, spraying operation and constant discharge of effluents into water system made as an important pollutant of aquatic ecosystem is caused by the activities of organisms including man. Investigation of the effects on fish, *Channa orientalis* is one of the local fresh water edible fish of great economic importance as an experimental model has a diagnostic significance in evaluation of the adverse effects of endosulfan in the behavior.

Behavioral responses have been used as a tool in studies the effects of some pollutants on the biota of environmental stress. [4] studied the physical, morphological and behavioral changes in fish, *Channa punctatus* in exposure to pesticides. If toxic substance encountered in higher concentration they are bound to bring severe adverse effect on the organism. Adverse effect may be at cellular or even at molecular level but ultimately it would lead to behavioral, physiological, pathological and biochemical disorders that may prove fatal to the organism [5]**.** An opercular movements and surfacing behavior are the indicators of changes in respiratory mechanisms while irregular, erratic and jerky movements indicate improper co-ordination between nervous system and muscular tissue. Behavioral changes could causes ecological death by disrupting the normal function and life history of the exposed organism. [6] behavioral changes in the *Channa orientalis* were found more in cadmium chloride as compare to in the presence of

ascorbic acid. [7] reported the heavy metal cause metabolic disorders, morphological changes, behavioral stress, immuno-suppression and many other toxicological manifestations among fishes. However, studies on the vitamin C against heavy metals toxicity in animal system is limited and inconclusive, [8]**.** In animals ascorbic acid contents in the tissue increases in stress condition during metal toxicosis. The bio- regulatory role of ascorbic acid protects extra cellular protein function through gene expression highlighted by [9]**.** In the present investigation protective role of ascorbic acid is assessed in endosulfan induces changes in behavior in *Channa orientalis*.

MATERIALS AND METHODS

Medium sized fresh water fishes, *Channa orientalis* were collected from Shiven river area Nandurbar Dist. Nan durbar. The physico-chemical parameters of the water used for the maintenance of the fishes were analyzed as per the methods [10]**.** Fishes were divided in to three groups A, B and C. Group A fishes was maintained as a control. Group B fishes were exposed to $LC_{50/10}$ dose of endosulfan (0.07511ppm) for 30 days, while group C fishes were exposed to respective chronic concentration of pesticides with 50mg/l. of ascorbic acid for 30 days. Fishes from B groups were divided into two groups after 30 days exposure to endosulfan into D and E groups. D group were allowed to restore naturally while those of E groups were exposed to ascorbic acid (50mg/l.). Behavioral aspects such as opercular movement, surfacing phenomenon and jerky movement were recorded from A, B, and C group fishes after 15 and 30 days of exposure and from D and E groups after 35th and 40th days of recovery.

RESULTS AND DISCUSSION

The data obtained regarding the physico-chemical properties is given in Table. 1 while the behavioral changes in *Channa orientalis* after exposure to endosulfan with and without ascorbic acid and during recovery are given in the Table 1.1 to 1.3. The changes in the behavior due to pesticide stress indicate the changes in the activity of the organism. These alterations in body metabolism due to organochlorine intoxication have been extensively studied [11]. These studies indicate that all organochlorine pesticides are highly toxic to fish. The degree of toxicity produced by the poisonous substance is does independent upon environmental conditions [12].

Behavioral changes in the *Channa orientalis* were found more in endosulfan. In the presence of ascorbic acid (50 mg/1.) the behavioral changes is less as compared to those of pesticide stress fishes. The fishes pre- exposed to pesticide showed fast recovery in the presence of ascorbic acid than those allowed to cure naturally. Changes in behavior such as fin movement, opercular, jerky movement and surfacing phenomenon exposed to pollutants undergoes stress. As the period of exposure or the concentration of pollutants increases such as behavioral changes can be utilized as biological indicator or as biological early warning system of these test pesticide. Increased distance between gills and opercular movement may be utilized as an indication of low levels of dissolved oxygen or presence of such pollutants in water reported by [13]. Increase in the frequency of surfacing phenomenon in the fish either due to hypoxia condition difficulty to respire in the media or for the protection of the gill epithelium.

When pesticide is added a rapid opercular movements, gulping of air, increase in surfacing, followed by excited swimming and coughing was observed. This may be because of improper ventilation or inconvenience in breathing [14]. The substantial reduction in the growth of fish was observed in the sub-lethal concentrations as compared to the control group. This may be due to reduced amount of diet consumed by the fish due to toxicant stress, which was immediately utilized and was not stored in the body weight [15]. Lateral swimming and loss of equilibrium were probably due to the impairment of nervous and muscular system which may be due accumulation of acetylcholine in synaptic and neuromuscular junctions [16]. Impact of different toxicants on the behavior of *Labeo rohita* have been studied by various workers [17]. Any abnormal change in the behavior, physiology and the morphology of fish indicates the deterioration of water quality, as the fishes are the biological indicators. Increase in the secretion of mucus all over the body in the fish exposed to endosulfan may be an adaptive response providing additional protection against corrosive nature of the metal and to avoid the absorption of the toxicant by the general body surface. Similar observations were made by many workers [18]. The pale red color of the gills and their clumping increases with the increasing of concentration of toxicant. The fishes lost their natural coloration and become almost pale yellow in color.

The depletion of ascorbic acid in fishes under organochlorine stress [19]. Ascorbic acid play important role in the synthesis of collagen and thus can correct the altered situation due to heavy metals. Ascorbic acid plays an important role in distribution and excretion of toxic metals. Ascorbic acid has reversed dysfunction of cells lining blood vessels. The normalization of functioning of these cells may be link to prevention of heart diseases [20]. It has been realized that antioxidant can play significant role in the treatment of metal induced oxidative stress. Some antioxidants behave as efficient chelators [21]. The SH group of protein is mainly responsible metal interaction or bindings L-ascorbic acid is antioxidant and may extent in protective effects by chelating the metal and removing them from the system [22] . During toxicosis ascorbic acid indicate positive role in detoxification. It is necessary for the synthesis of collagen, growth and maintenance of epithelial tissue. It can acts as a hydrogen carrier it may have an essential role in the metabolism of carbohydrate or protein or both. It appears to function it maintaining strength in blood vessels.

CONCLUSIONS

The behavioral and morphological changes show direct response of the animals to the pollutants. This type of study can be useful to compare the sensitivity of various species of aquatic animals and to derive safe environmental concentration of the toxicants by which there is no lethality and stress to the animals. The current study evidenced that endosulfan is highly toxic and had a detrimental impact on the behavioral responses and morphology of the fish, *Channa orientalis*. These responses can be used as a tool in biomonitoring programme to monitor eco-toxicity risk of endosulfan to the fish. Ascorbic acid is necessary for normal functioning of cellular units and sub-cellular structures. It acts as a cofactor in the enzymes proly-hydroxylase, which are necessary for the synthesis of collagen. The antioxidant role of ascorbic acid acts like the scavengers, preventing cell damage, protects the tissue from the super-oxide radical generated due to different toxicological effects.

Table - 1:- Physico-chemical parameters of water used for experimentation

Temperature	25·0 ± 3·2^0
PH	7 · 6 1 ± 0 · 3
Conductivity	141 ± 15.7 µ mho/cm·
Free Co2	3·30 ±1· 3ml/l.
Dissolved O2	6·1 ± 1·1ml/l.
Total Hardness	203 ± 12·0mg /l.
Total Alkalinity	584·6 ± 32.8 mg /l.
Magnesium	31·61 ± 2.9 mg /l
Calcium	30·44 ± 3.06 mg /l
Chloride	107·90 ± 16.34 mg /l.

Table- 1.1

Table.1.1: Frequency of opercular movement in *Channa orientalis* after chronic exposure to Endosulfan without and with ascorbic acid (count/min.)

Group	Treatment	15d	30d	35d	40d
A	Control	31.0±1.63	33.0±1.41	34.0±2.72	32.0±2.20
B	Endosulfan (0.07511ppm)	23.0±1.61* (-22.58)	24.0±1.44* (-21.21)		--
C	Endosulfan (0.07511ppm)+AA	28.0±1.64* (-9.37)	29.0±2.95 ^{NS} (-12.12)		--
D	Recovery in Normal Water	--	--	24.0±1.63 $^{\Delta}$ [+11.53]	27.0±1.74 ^{NS} [+3.84]
E	Recovery in AA	--	--	27.0±1.41 $^{\Delta}$ [+3.84]	29.0±0.815 $^{\Delta\Delta}$ [+53.33]

Table- 1.2

Table.1.2: Frequency of surfacing phenomenon in *Channa orientalis* after chronic exposure to Endosulfan without and with ascorbic acid (count/hr.)

Group	Treatment	15d	30d	35d	40d
A	Control	111.0±2.95	100.0±3.55	112.0±4.31	109±3.50
B	Endosulfan (0.07511ppm)	141.0±3.40** (-21.27)	142.0±3.74** (-22.52)	--	--
C	Endosulfan (0.07511ppm)+AA	124.0±4.18 ^{NS} (-11.71)	129.0±4.39 * (-17.27)		--
D	Recovery in Normal Water	--	--	124.0±2.94 $^{\Delta}$ [+14.08]	119.0±2.00 $^{\Delta}$ [+3.84]
E	Recovery in AA	--	--	117.0±2.16 $^{\Delta}$ [+17.60]	115.0±2.58 $^{\Delta}$ [+19.01]

Table- 1.3

Table.1.3: Frequency of jerky movement in *Channa orientalis* after chronic exposure to Endosulfan without and with ascorbic acid (count/hr.)

Group	Treatment	15d	30d	35d	40d
A	Control	2.0±0.816	2.0±0.816	3.0±0.811	3.0±0.807
B	Endosulfan (0.07511ppm)	9.0±1.73* (-351.0)	15.0±2.56* (-233.32)	--	--
C	Endosulfan (0.07511ppm)+AA	7.0±1.28 * (-250.0)	10.0±1.61 * (-400.0)		--
D	Recovery in Normal Water	--	--	12.0±1.40 $^{\Delta}$ [+20.0]	1.00±2.94 $^{\Delta}$ [+26.64]
E	Recovery in AA	--	--	7.0±0.817 $^{\Delta}$ [+60.00]	4.0±0.814 $^{\Delta}$ [+73.30]

AA= Ascorbic acid (50 mg/l., ± indicates S.D. of three observations. Values in () indicates percent change over respective control. Values in [ ] indicates percent change over 40 days of respective B. *indicates significance with the respective control.$^{\#}$ indicates significance with 40 days of respective. $p<0.05$ =*& $^{\Delta}$, $p<0.01$ =** & $^{\Delta\Delta}$, $p<0.001$ = *** & $^{\Delta\Delta\Delta}$, ^{NS} and $^{\Delta NS}$ = Not significant.

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