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Research Paper

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Observation of Biochemical Property in Brain Tissue of Fish *Channa punctatus* and Exposed to Deltamethrin

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Abstract- The concept of pesticides is known from ancient time and available in literature around 1000 BCE. Homer referred to the use of sulfur to fumigate homes and by 900 C.E. Synthetic pesticides emerged between 1930 & 1940 as the result of research aimed at developing chemical weapons that were originally tested in insects. The fishes are directly exposed with water containing pesticides & other water soluble pollutants; in hepatocytes detoxification of pesticides takes place and being a neurotoxic nature of deltamethrin, braius, gills & liver tissues were selected to assess the deltamethrin toxicity.

Keywords **:** Deltamethrin, D.O., LC 50

I. INTRODUCTION

Water provides a good medium for growth of aquatic flora and fauna with great biodiversity. Major problem associated with the natural and man made water bodies across the globe is its pollution. Pollutants are ranging from natural to synthetic, nontoxic to highly toxic, degradable to non biodegradable. i.e. Xenobiotic compounds. These pollutants also destroy the quality of the aquatic media and render it unifit various aquatic, particularly fishes. These aquatic ecosystem comprising greater part of the natural environment is also faced with the. threat of a shrinking genetic base & biodiversity (Ramamurthy et.al., 2009). A variety of organophosphate, organochloride, organometallic & carbamide pesticides are extensively used in agriculture for the control of pests. The toxicity level were influenced by the sex and the nutrients supply (Arunachalam 1980; Sharma & Ansari 2010). Pesticides exert many physiological & biochemical changes by influencing the activities of various enzymes of organisms. Altered chemical environment of the natural aquatic bodies has profound effect on behavioural and physiological system of inhabitants, specially the fishes (Khan & Law, 2005).

The oxidative stress is an inescaple component of aerobic life. In the aerobic organism, a balance between the reactive oxygen species (ROS) production & the system to protect. Cells from reactive oxygen species exists (Davies, 1995). The most important enzymes for detoxification A ROS in all organisms are SOD, CAT, GPx, Xanthine oxidase & G6 PD. (Mishra D.B. et.al. 2008-09, Dubey, J.K. et.al.,2016), N. Liyana et.al. 2017. Deltamethrin belongs to the synthetic pyrethroids that are most popular & widely used insecticides all over the world.

II. MATERIALS AND METHODS

Glasswares used in present investigation were a "Borosil make" Glasswares rinsed with double distilled water and dried in a hot air oven at 100^0C for over night. Most of the chemical used from Hi Media, Sisco, SRL India. All the general chemicals used were AR grade.

Water Analysis of Experimental Site – The examination of physio-chemical characteristics of water carried out by standard methods as APHA 2005.

Temperature – At the time and sampling on the site in degree centigrade.

pH – pH was ensured in laboratory with the help of systronic pH meter (Model 324).

D.O. – Winkler's method was used for the estimation of dissolved oxygen in water.

Total Alkalinity –

The alkalinity of water can be determined by titrating the water samples with sulphuric acid of known values of pH, Volume and concentration. Based on stoichiometry of the reaction and number of moles of sulphuric acid needed to reach the end point, the concentration of alkalinity in water is calculated.

Collection & Acclimatization of Fish –

The adult & healthy fishes, irrespective of sex (17.80 + 0.50 cm length & 47.85 + weight) of *Channa punctatus* were

collected from Gujartal Jaunpur. The collected fishes were airbreathing teleost cat fish. The fish were transported in plastic containers to the laboratory & washed with 0.1 $KMNO_4$ solution. Prior to the starts of experiments the fish were acclimatized to the laboratory condition for 15 days in dichlorinated tap water.

They were maintain glass aquaria containing dichlorinated tap water. The commercial grade pesticide deltamethrin was dissolved in acetone and required volume of the desired concentration of pesticides in added in each aquarium. The aquarium was replaced every 24 hrs with fresh water in control & deltamethrin solution, were mixed into the water aquarium. Feeding of fish was stopped 48 hrs prior to the commencement of the experiment with a view to avoid and possible change in situ in the toxicity of pesticide.

Determination of LC_{50} –

Bioassay or toxicity tests were carried out for the determination of LC_{50} values by following FAO procedure for short term bio-assays (Reish & Oshida, 1987).

Biochemical Studies –

The fishes were sacrificed by decapitation & tissues, where taken out s thoroughly washed & cleaned in chilled KC 10.15 M to remove the blood & adhering tissues.

The specimen were sacrificed, the brain removed, cleaned & weighed rapidly. A ten percent (w/v) homogenate of different tissues were prepared in cold fish saline with the aid of potter Elvehjem type homogenizer fitted with the teflon pestle. The homogenate was fish centrifuged at 2000 rpm for 15 minutes in a refrigerated centrifuge, the pellet consisting of a nuclear fraction & cell debris is discarded. The supernatent again centrifuge at 8000 rpm for 20 minutes for the post mitochondrial supernatent, and taken for the biochemical studies & crude enzyme studies.

Estimate the total carbohydrate by added 4 ml of the anthrone reagent to 1 ml of a protein free carbohydrate solution and rapidly mixed. Protein estimation in post mitochondrial and cystosolic supernatent was carried out by the Lowry et.al. 1951. Briefly, the lipid samples & standards are placed in a heating block set at 100^0C to allow the solvent to evaporate. Once the solvent is gone (about 10 min.) 0.1 ml of concentrated sulfuric acid is added to each tube; vortexed, the heated 100^0C for 10 min. Samples are then removed from the heat block and allowed to cool to room temperature before adding 2.4 ml of Vanillin reagent & vortexed.

III. RESULT AND DISCUSSION

The physical and chemical conditions of a water bodies gives it to living and setting the hydrodynamics of the water bodies. Several parameters are taken into account & considered to the prime and are temperature, change in acid & based strength, dissolved oxygen concentration with play critical role and directly affect the aquatic flora and fauna. and its diversity, physico-chemical parameters of water was recorded & presented in following tables 1.

Physico-chemical Properties of Water Bodies –

S.No.		
1.	Temperature	**26.5^0C**
2	pH	7.3
3	D.O.	5.8 ppm
4	Total Alkalinity	methyl orange– 265 ppm phenolphthalein - Nil

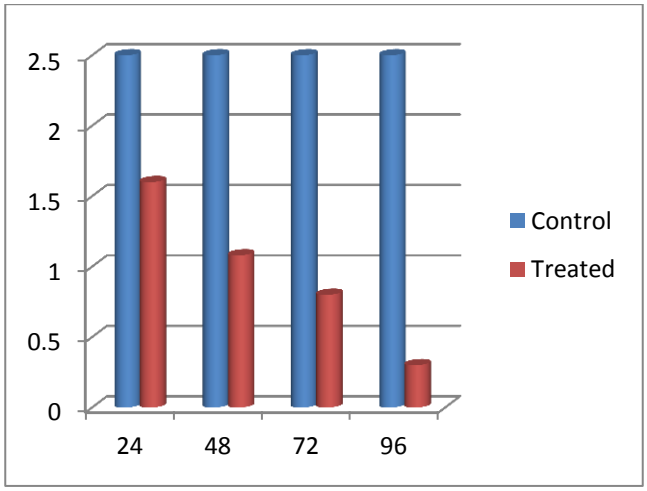
Bioassay or toxicity tests were carried out for the determination of LC_{50} values by following FAO procedure for short term bio assays (Reish & Oshida, 1987). The duration of the tests was 96 hrs stock solution of deltamethrin 10% effective concentration (EC) was prepared by diluting 1 ml insecticides in 100 ml of distilled water, and was prepared by diluting concentration of 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.08 ppm which were used as experimental waters for toxicity study of selected model animal channa. The experiment was set in duplicate & healthy fishes (n=10) were maintained in 10 litre of experimental water having different concentration of deltamethrin. Similarly a control was set up with water devoid of deltamethrin. Feeding was stopped one day prior to the experiment and also during the experimental period, as recommended by Ward & Parrish (1982) and Reish & Oshida (1987). The LC_{50} values were calculated as average from the two replicates for each experimental concentration of water by arithmetic graph method as showing graph-1 (Reish & Oshida, 1987). LC_{50} at 96 hrs were four at 1.3 ppm.

Carbohydrate Contents –

Carbohydrate content in different tissue are different and are showing progressive decreasing profile. Carbohydrate content in brain tissues was evaluated in test animal in *Channa punctatus* was found 1.6, 1.08, 0.8 & 0.3 mg/gm of wet weight of tissues as compared with control (2.5 mg/gm of tissue) at 24, 48, 72 & 96 hrs respectively as shown in graph-1. The decreased level of carbohydrate contents in organs of the fish after 96 hrs exposure with deltamethrin in brain tissues when compared to control.

Decreasment in carbohydrates level indicates in rapid utilization to meet and the increased energy demand to cope up with stress due to deltamethrin toxicity. The demand is channelized either glycolytic pathway or oxidative reaction of pentose phosphate pathway (Cappon et.al. 1975, Prusty et.al. 2015 & Amin et.al. 2012, Dubey J.K. et.al. 2016). The carbohydrate content reduction is more prevalent under hypxic condition due to stress produced by a pesticides or heavy metals. (Dezwann et.al. 1972, Chandrawathy &

Reddy, 1995, Winkle et.al. 2007, Singh A.K. et.al. 2009 & Yunsus et.al. 2015).



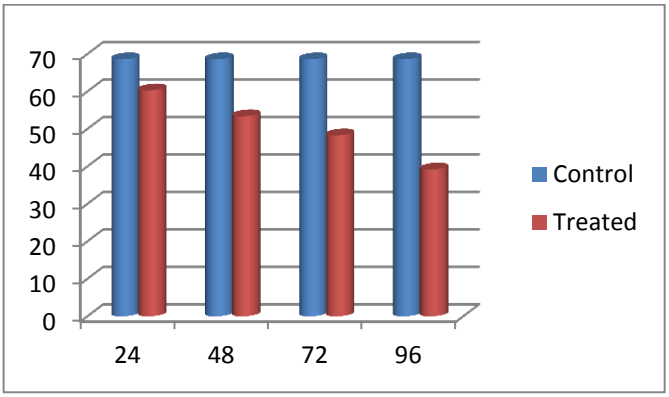
Time in hours

Graph -1 : Carbohydrate content in brain tissues

Protein Content –

Proteins are very important biomolecules and building block of organisms and considered as final class of biomolecules as for as energy requirement is concerned. Protein content in brain tissue were found to be 60.3, 53.4, 48.3 & 39.2 mg/gm of we weight of tissues as compared with control 68.6 mg/gm of tissue at 24, 48, 72 & 96 hrs respectively is shown in graph-2.

In present study the protein content was found to be decreased in all organs after the treatment with deltamethrin upto 96 hrs when compared with the control. The decreased level of proteins might be due to inhibition of translational process are increased rate of metabolism of protein which may be entered Krebs cycle. Carried out due to deltamethrin stress (Ganeswade 2011, Binu Kumari & Vasanthi 2013, Dubey J.K. et.al. 2016).

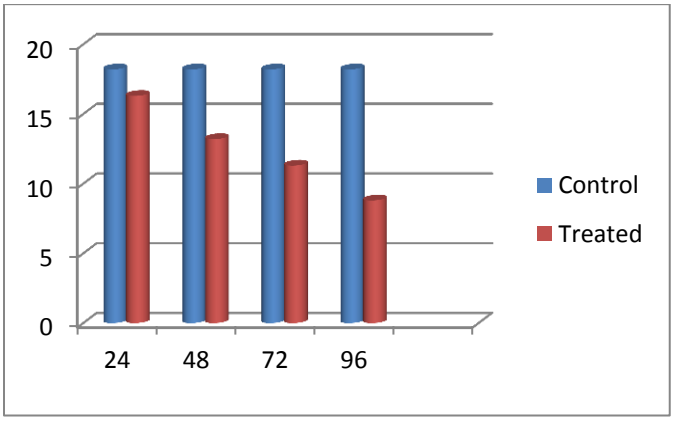


Time in hours

Graph-2: Protein content in brain tissues

Lipid Contents –

Lipids are important constituent of cell membrane and it also provide buoyancy to aquatic organism. Lipid content in brain tissue was found to be 18.2 mg/gm of wet weight of tissue in *Channa punctatus* was observed at 0, 24, 48, 72 & 96 hrs respectively. In treated brain tissue of *Channa punctatus*, the lipid content was found to be 16.3, 13.2, 11.3 & 8.8 mg/gm observed at set time intervals shown as graph-3.



Time in hours

Graph-3: Lipid content in brain tissues

Lipid are important Biochemical constituent & upon oxidation contributes a significant amount of energy via beta oxidation & structural components for reproductive growth (Surgent 1995, Kylie et.al. 2013). It is due to exposure of deltamethrin that in term resulting into exhaust of energy during stress leading, to oxidation of lipids to meet out the organisms high energy demand.

IV. CONCLUSION

The carbohydrate content in brain tissues was evaluated in *Channa punctatus*. The decreased level of carbohydrates contents in organs of fish after 96 hours exposure with deltamethrin in all tissues when compared to control. Decreasment in carbohydrates level indicates in rapid utilization to meet and the increased energy demand to cope up with the stress due to deltamethrin toxicity. this demand is channelized either glycolitic pathway of oxidative reaction of pentose phosphate pathway.(Cappon et.al.1975,Prusty et.al.2015).

The protein contents found to be destroyed in all organs after the treatment with deltamethrin upto 96 hours when compared with control . this decreased level of protein might be due to inhibition of translational process are increased high rate of catabolism of protein which may be entered into Krab's cycle via transmination process carried out by amino transferase and it might due to meet out organism high energy demand due to deltamethrin stress.

Reduced level of lipid contents in brain tissues after 96 hours exposure to deltamethrin . this reduction in absorption

of carbohydrates and protein which in turn resulting into exhaust of energy during stress leading to oxidation of lipid to meet out the organic high energy demand.

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