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INFLUENCE OF QUINALPHOS (25% EC) ON DNA AND RNA IN GILLS AND MUSCLE OF THE FRESHWATER FISH *CHANNA GACHUA* (HAMILTON, 1822)

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KEYWORDS

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ABSTRACT

The changes in DNA, RNA and RNA/DNA ratio in gill and muscle of freshwater fish *Channa gachua* have been studied. The DNA, RNA and RNA/DNA ratio were estimated in gill and muscle tissues after exposure to the Quinalphos 25% EC sub lethal concentration (0.0125μl/L^{-1}) for 24, 48, 72 and 96 hours.The amount of DNA in gill of experimental fish reduces as 10.84, 10.55, 9.60 and 8.85 (mg/g wet weight of tissue) for 24, 48, 72 and 96 hours,The RNA in the gill of experimental fish also reduced as 12.46, 12.20, 11.86 and 10.66 for 24, 48, 72 and 96 hours. The DNA and RNA in the muscle for experimentalfish also showsreduction of DNA level 4.70, 4.56, 4.20 and 3.89and reduction inRNA 5.92, 5.77, 5.32 and 4.32at different exposure period. Whereas the RNA/DNA ratio observed changed in gill (1.14 1.15 1.23 and 1.20) and in muscle (1.25, 1.26, 1.26 and 1.11) respectively at different exposure period as compares to control. DNA and RNA show alteration in their levels under the exposure of Quinalphos. The freshwater fish *Channa gachua* indicates the Quinalphos affects the nucleic acid in the gill and muscle tissue.

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INTRODUCTION

Aquatic environments are loaded with several types of organic and inorganic pollutants. Human population growth and industrial development have been the major causes of coastal contamination around the world during recent years (Caussy *et al.,* 2003). Pesticides play an important role in sustaining the agricultural production byprotecting all kinds of crops from pest attack and vector-bornediseases (Adhikari *et al.*, 2004; Abhilash and Singh, 2009) Organophosphate (OP) compounds are one of the most commonly used insecticides in agriculture and public health, accounting for 50% of the global insecticidal use (Casida and Quistad 2004). Organophosphate pesticides constitute a large proportion of the total synthetic chemicals employed for the control of pests in the field of agriculture, veterinary practices and public health. Insecticides are among the most hazardous chemicals to human and all other environment, such pesticides used for crop protection in agriculture and horticulture may enter ditches, ponds, lakes and rivers in numerous ways such as direct overspray, spray drift, leaching to surface and ground water, run off from land and or accidental spills (Capri and Trevisan, 1998). Water pollution is a major problem of this century and addition of pollutants changes the natural qualities of water (Voltz *et al*., 2005). Contamination of surface waters has been well recognized worldwide and constitutes a major concern at local, regional, national and global levels (Cerejeira *et al*., 2003, Spalding *et al*., 2003). Environmental contamination by pesticides has been documented in both biotic and abiotic components.The pollution of freshwater ecosystem by chemical pesticides has become one of the most critical environmental problems (Northoffand William, 2004). This causes extensive damage to the activities of the living resources of food-web due to their toxicity, persistency with half-lives of decades and tendency to accumulate in the organisms (Joseph and Raj, 2010, Joseph and Raj, 2011). The pesticide causes various problems affecting to the ecosystem and effect on growth of aquatic organism (Chinni, 2001) and histopathological and physiological changes in fishes (Holden A. V., 1973).

Quinalphos 25% emulsified concentration is an organophosphate insecticide, the widespread use of quinalphos results in extensive contamination in freshwater. Quinalphos is highly active against biting and sucking insects and due to its short life with easy detoxification in animal tissue they are still widely used in agriculture and aquaculture (Vidyarani *et al*., 2010). Increase use of pesticides not only helped in controlling insects and pests but also created great environmental pollution especially hazardous influences in media as well as aquatic fauna (Bradbury and Coats, 1989). The toxicity study is essential to find out toxicants limit and safe concentration, so that there will be minimum harm to aquatic fauna in the near future (Nikam and Shejule, 2015). Hence the present study has been focused to investigate the impact of sub-lethal concentration of Quinalphos 25% EC on DNA, RNA and RNA/DNA ratio in gill and muscle of fresh water teleost fish *Channa gachua*.

MATERIALS AND METHODS

The freshwater fish *Channa gachua* (15-18 cm and 45-50 gm) was obtained from

Godavari River near Aurangabad. These fishes were acclimatized to the laboratory conditions with de-chlorinated tap water for 10-15 days at a room temperature 27 + 2ºC prior to the experimental condition.

Quinalphos 25% EC an organophosphate pesticide manufactured by Syngenta India Ltd. was used as test chemical. The LC_{50} of Quinalphos 0.0125μl/L^{-1} for 96 hours was determined to decide its sub-lethal concentration for experimentation. Control and experimental groups of fishes were sacrificed after 24, 48, 72 and 96 hrs.

Gills and muscles tissue were removed washed in saline and lyophilized at -5ºC with the Mini-Lyodel freeze dryer manufactured by Delvac Pumps Pvt. Ltd. Chennai. The nucleic acid (DNA and RNA) content of the tissues wa sestimated by the following method.

DNA estimated by DPA method Searchy and Maclinnis (1970, a) and absorbance was recorded at 595 nm.

RNA estimated by Dische-Orcinol method Searchy and Maclinnis (1970, b) and absorbance was recorded at 655 nm.

Student's t-test was used to calculate the significance of the differences between control and experimental means. P values of 0.05 or less were considered statistically significant (Fisher, 1950).

RESULTS AND DISCUSSION

The results of the present investigation revealed that Quinalphos induced reduction in DNA and RNA contents in both gill and muscle tissues of *Channa gachua*. Sub-lethal concentration of Quinalphos shown significant increased level of DNA of experimental fish gill 10.84, 10.55, 9.60 and 8.85 (mg/g wet weight of the tissue) and decreased level of RNA 12.46, 12.20, 11.86 and 10.66 (mg/g wet weight of the tissue). While the RNA/DNA ratio significantly changed (1.14, 1.15, 1.23 and 1.20) respectively at different exposure periods as compared to control group (Table 1).

In the muscle tissue shows the similar results to that of gill tissues. The concentration shows the increased in the level of DNA 4.70, 4.56, 4.20 and 3.89 (mg/g wet weight of the tissue) and RNA 6.30, 5.95, 5.32 and 4.32 (mg/g wet weight of the tissue). While the RNA/DNA ratio significantly changed (1.34, 1.30, 1.26 and 1.11) respectively at different exposure periods as compared to control group (Table 2).

The heterogeneous levels of DNA and RNA in the tissue of brain, liver, muscle, gill and kidney were observed after exposure to sub-lethal and lethal concentrations of cadmium chloride in freshwater fish *Cirrhinusmrigala*(Veeraiah, 2013) and in exposure of Quinalphos to sub-lethal and lethal concentration to the freshwater fish *Labeorohita* (Venkata Rathnamma and Nagaraju, 2013). According to Holbrook (1980) Thymidine incorporation into hepatic tissue, the DNA is markedly increased after 1-3 days of administration of various toxicants. Maruthanayagam and Sharmila (2004) studied the effect of monocrotophos on *Cyprinus carpio* tounderstand the toxic effects of toxicant on the nucleic acids and concluded that the pesticide lead to several changes in thebiochemical markers like DNA and RNA which may be due to the increased activity of the enzyme DNase and theinhibition of RNA polymerase function. But during recovery period, the DNA and RNA levels increased progressi velyindicating a probable from the disruption of internal organs. The increase of DNA in gill region may be due to hypertrophic nature of chloride cells. These results are in agreement with the works of Natarajan (1981), which reveal the enlargement of nuclei in the chloride secreting cells in *Channa striatus* exposed to metasystox. Organophosphorus compounds exhibit strong mutagenic and clastogenic potentiality (Patankar and Vaidya, 1980) which may be responsible for the alteration of DNA level. Ravikiran and Kulkarni (2012) observed that a short term exposure to copper sulphate, the DNA and RNA content of tissues get reduced in the freshwater fish *Notopterus notopterus* indicatingcopper sulphate as pollutant effect the nucleic acid content inthe tissues.

In the present study decrease in level of RNA was observed in the gill and muscle tissues of *Channa gachua* exposed to sublethal concentration of Quinalphos. These results are in

Table 1: Change in the Amount of DNA and RNA (mg/g wet weight of the tissue) and Ratio of RNA/DNA Change over the Control in Gill of Freshwater Fish *Channa gachua* Exposed to Sub-lethal Concentration of Quinalphos 25% EC

Hrs	DNA Control	DNA (Expt.)	RNA Control	RNA (Expt.)	RNA/DNA ratio (Control)	RNA/DNA ratio (Expt.)
24	8.86 ± 0.012	10.84 ± 0.022	15.97 ± 0.015	12.46 ± 0.012	1.80 ± 0.020	1.14 ± 0.022
48	8.60 ± 0.015	10.55 ± 0.013	15.72 ± 0.026	12.20 ± 0.015	1.82 ± 0.026	1.15±0.026
72	8.42 ± 0.019	9.60 ± 0.021	15.66 ± 0.020	11.86 ± 0.020	1.85 ± 0.026	1.23 ± 0.025
96	7.78 ± 0.026	8.85 ± 0.015	15.36 ± 0.022	10.66 ± 0.014	1.97 ± 0.016	1.20 ±0.017

Table 2: Change in the Amount of DNA and RNA (mg/g wet weight of the tissue) and Ratio of RNA/DNA Change over the Control in Muscle of Freshwater Fish *Channa gachua* Exposed to Sub-lethal Concentration of Quinalphos 25% EC

Hrs.	DNA Control	DNA (Expt.)	RNA Control	RNA (Expt.)	RNA/DNA ratio (Control)	RNA/DNA ratio (Expt.)
24	3.96 ± 0.019	4.70 ± 0.010	6.30 ± 0.024	5.92 ± 0.012	1.59 ± 0.022	1.25 ± 0.015
48	3.62 ± 0.015	4.56 ± 0.013	5.95 ± 0.030	5.77 ± 0.017	1.64 ± 0.020	1.26 ±0.018
72	3.24 ± 0.020	4.20 ± 0.025	5.56 ± 0.026	5.32 ± 0.025	1.71 ± 0.016	1.26 ± 0.015
96	3.12 ± 0.036	3.89 ± 0.026	5.26 ± 0.012	4.32 ± 0.016	1.68 ± 0.026	1.11 ±0.012

accordance with Durairaj and Selvarajan (1992), they observed a significant decline in RNA content of liver, muscle and gill and DNA content of brain of fish *Oreochromis mossambicus* after exposure to Quinalphos. Dawood (1986) and Benjamin (1990) have suggested that the decline in the RNA may also be due to the non-coding for the process of protein synthesis, thereby decrease in the RNA content, which turn would have reduced the concentration of RNA. As per the Samyuktha Rani and Veeraiah (2014), the effects of sub-lethal and lethal concentrations of Methyl parathion 50% EC on DNA and RNA contents show moderate toxicity on the main biochemical machinery of the freshwater fish *Channa punctatus*. The role of nucleic acids particularly RNA/DNA and protein/DNA ratios, which are used as an index of protein synthesis and cell size, are considered to be important and form an treatment with the pesticides causes variability in the nucleic acid contentin different tissues and the degree of variability or extent of alterations caused by the pesticides is found to be dose dependent(Malla Reddy and Bashamohideen, 1988). The sub-lethal exposure of Cypermethrin can alter the nucleic acid content in liver, muscle, kidney, brain and gill tissue of *Cyprinus carpio* and the variation in the nucleic acid content are tissue specific (Neelima *et al.,* 2015).

The results obtained in the present study shows that the effect of Quinalphos 25% EC sub lethal concentration altered the DNA, RNA and RNA/DNA ratio in the gill and muscle tissue of the fish. The variation in the nucleic acid content are tissue specific and dose dependent, so very lower concentration of organophosphate insecticides may affects fishes.

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REFERENCES

Abhilash, P. C. and Singh, N. 2009. Pesticide use and application: an Indian scenario. *J. Hazard Mater*.**165:** 1-12.

Adhikari, S., Sarkar, B., Chatterjee, A., Mahapatra, C. T. and Ayyappan, S. 2004. Effects of cypermethrinand carbofuran on certain hematologicalparameters and prediction of their recovery in a freshwater teleost, *Labeorohita* (Hamilton). *Ecotoxicol Environ Saf*. **58:** 220-226.

Benjamin, N. 1990. Biochemical changes due to learning of the brain of rat exposed to an Organophosphate pesticide, M. Phil thesis of University of Madras, India.

Bradbury, S. P. and Coats, J. R. 1989. Toxilogical and toxicodynamics of pyrethroids insecticides in fish Environ. *Toxical. Chem*. **8:** 373-386.

Capri, E. and Trevisan, M. 1998. Prediction of environmental concentrations (PECs) by mathematical model application in Europe. *Pestic outlook*. **9:** 26-30.

Casida, J. E. and Quistad, G. B. 2004. Organophosphate toxicology: safety aspects of Non acetylcholine esterase secondary targets, *Chemical Research in Toxicology*. **17:** 983-99.

Caussy, D., Gochfeld, M. and Gurzau, E. 2003. Lesions from case studies of metals investigating exposure, bioavailability and risk. *Ecotoxicol. Environ. Safety*. **56:** 45-51.

Cerejeira, M. J., Viana, P., Batista, S., Pereira, T., Silva, E. V., Alerio, M. J., Silva, A., Ferreira, M. and Silva Fernandes, A. M. 2003. Pesticides in Portuguese surface and ground waters.*Water Research*. **37(5):** 1055-1063.

Chinni, C. S., Khan, R. N. and Yallpragada, P. R. 2001. Larval growth in post larval of *Paenus indicus* an exposure to lead, Bull *Environ. contam Toxical*. **67:** 24-34.

Dawood, N. 1986. Studies on the functional orgnization of the brain of rabbit, *Oryctala guscuniculus* Ph.D thesis of University of Madras, India.

Durairaj and Selvarajan, V. R. 1992. Influence of quinalphos an organophosphorus pesticide,on the biochemical constituents of the tissues of fish, *Oreochromis mossambicus*. *J. Environ. Biol*. **13(3):** 181-185.

Fisher, R. A. 1950. Statistical Methods for research workers, 11th edition, (Edinburgh UK: Oliver and Boyd Ltd.).

Holbrook, (Jr.) D. J. 1980. Effect of toxicants on nucleic acid and protein metabolism in: Introduction go biochemical toxicology (Eds: E. Hodgson and F. E. Guthrie). *Blackwell Scientific Publications, Oxford*. pp. 261-284.

Holden, A. V.1973. Effect of pesticides on fishes in environmental pollution by pesticides Ed. *C-A Edward. Plenum press NY*. pp. 213-253.

Joseph, B. and Raj, S. J. 2010. Effect of curacron toxicity on the total serum protein content of *Cyprinus carpio*. *Toxicol. Environ. Chem*. **92:** 1889-1893.

Joseph, B. and Raj, S. J. 2011. Impact of Pesticide Toxicity on Selected Biomarkers in Fishes. *Int. J. Zoology. Res*. **7:** 212-222.

Malla Reddy, P. and Bashamohideen, Md. 1988. Toxic impact of fenvalerate on the protein metabolism in the branchial tissue of a fish, *Cyprinus carpio*. *Curr. Sci*. **57:** 211-212.

Maruthanayagam, C. and Sharmila, G. 2004. Biochemical variations induced by monocrotophos in *Cyprinus carpio* during the exposure and recovery period. *Nat. Environ. Poll*. **3(1):** 1-9.

Natrajan, G. M. 1981. Effect of lethal concentration of metasystox on selected oxidative enzymes, tissue respiration and haematology of the freshwater air breathing fish *Channa striatus* (Bleeker). *Pest. Biochem and Physiol*. **21:** 194-198.

Neelima, P., Govinda Rao, K., Gopala Rao, N. and Chandra Sekhara Rao, J. 2015. Acute toxicity of cypermethrin (25% EC) on nucleic acids (DNA and RNA) in *Cyprinus carpio* (Linn.).*International J. Recent Scientific Research*. **6(7):** 5219- 5224.

Nikam, S. M. and Shejule, K. B. 2015. Study of acute toxicity of Bis (Tributyltin) oxide (TBTO) on the freshwater fish, *Nemacheilus botia,* from Nandur Madhmeshwar dam at Maharashtra, India. *The Bioscan*. **10(2):** 517-519.

Northoff, E. and William, M. 2004. Farm workers need to be better protected against pesticides; FAO, UNEP call for stronger safety measures. Press release SAG/296.FAO-UNEP.*http://www.un.org/news/press/docs/2004/sag 296.doc.htm*.

Patankar, N. and Vaidya, V. G. 1980. Evaluation of genetic toxicity of the insecticide phosphamidon using *in vitro* & *in vivo* mammalian test systems. *Indian J. Exp Biol*. **18:** 1145-1147.

Ravikiran, K. and Kulkarni, R. S. 2012. DNA and RNA content in some tissues of freshwater fish *Notopterus notopteru* sex posed t o copper sulphate. *The Bioscan*. **7(2):** 309-310.

Samyuktha Rani, A. and Veeraiah, K. 2014. Methyl parathion 50% EC an Organophosphorus insecticide induced toxicity, biochemical and histopathological changes in the freshwater murrel *Channa punctatus* (Bloch) Ph.D thesis of Acharya Nagarjuna University, AP,

India.

Searcy, D. G. and Maclnnis, A. J. 1970a. Biochemical estimation of DNA by Burton's Diphenylamine method .*J. Biochem*. **62:** 315-323.

Searcy, D. G. and Maclnnis, A. J. 1970b. Biochemical estimation of RNA by Dische-0rcinal method. *J. Biochem*. **62:** 315-323.

Spalding, R. F., Exner, M. E., Snow, D. D., Cassada, D. A., Burbach, M. E. and Monson, S. J. 2003. Herbicides in ground water beneath Nebraska's management systems evaluation area. *J. Environmental Quality*. **32(1):** 92-98.

Veeraiah, Vivek, C. H., Srinivasa Rao, P. and Venkata Rao, G. 2013. Biochemical changes induced by Cypermethrin (10% EC), A pyrethroid compound in sub-lethal and lethal concentrations to the freshwater fish *Cirrhinus mrigala* (Hamilton). *J. Atoms and Molecules*. **3(6):** 625-634.

Venkata Rathnamma, V. and Nagaraju, B. 2013. Impact of Quinolphos On DNA And RNA Content of the Freshwater Fish *Labeo Rohita* (Hamilton). *International J. Research in Applied. Natural and Social Sciences (IJRANSS)*. **1(2):** 53-60.

Vidyarani, W., Sunita, H. and Sanayaima, S. 2010. Toxic effect of selected pesticides on an endemic loach *Lepidocephalichthys irrorata* (fam. cobitidae). *The Bioscan*. **3:** 635-641.

Voltz, M., Louchart, X., Andrieux, P. and Lennartz, B. 2005. Process of water contamination by pesticides at catchment scale in mediteranean areas. *Geophysical Research*. **7:** 10634.