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Muneesh kumar
Department of Zoology Govt.
S.S.L. Jain P.G. College,
Vidisha, Barkatullah
University Bhopal (M.P.)
India.

Parvinder kumar
Department of Zoology
University of Jammu, Jammu,
India.

Sangeeta Devi
Department of Zoology and
Applied Aquaculture
Barkatullah University Bhopal
(M.P.) India.

To study the DNA and RNA content in some tissues of freshwater fish *Clarias batrachus* exposed to heavy metal copper sulphate

Muneesh kumar, Parvinder kumar, Sangeeta Devi

Abstract

Industries are the major sources of heavy metal pollution and it is released into water and soil. Heavy metals cause several ill effects to aquatic living organisms and environment. The nucleic acid (DNA and RNA) content in different tissues such as brain, liver, kidney and ovary of copper sulphate exposed freshwater fish, *Clarias batrachus* has been studied. The changes in the nucleic acid content of the tissues have been observed. The DNA content in the ovary is higher in comparison to other tissues. The variation of DNA content in control and copper sulphate exposed fish is ovary >liver>brain>kidney. The RNA content also exhibited similar to that of DNA, having higher amount in the ovary and the variation is ovary>liver>brain>kidney. Although the degree of variation between the tissues remains same in both control and experimental groups, the nucleic acid content reduced under copper sulphate exposed freshwater fish, *Clarias batrachus* indicating copper sulphate as a pollutant effect the nucleic acid content in the tissue.

Keywords: DNA, RNA, *Clarias batrachus*, Copper Sulphate.

1. Introduction

The problem of pollution of the water where the wastes are usually discharged has increased to a great extent in recent years. Aquatic environments are loaded with several types of organic and inorganic pollutants. Huge amounts of agriculture pesticides, used for crop protection, eventually enter into the aquatic system. Similarly, heavy metals, which are released as industrial effluents form the major constituents of aquatic pollution. The presence of excess quantities of these toxic pollutants in water bodies has caused mass mortality of fishes in the past (Wanganeo *et al.*, 1994) [8]. Copper sulphate is a fungicide used to control bacterial and fungal diseases of fruit vegetable, nuts and field crops. Some of the diseases that are controlled by this fungicide include mildew, leaf spots, blights and apple scab. It is also used as an algacide, an herbicide in irrigation and municipal water treatment system. Copper sulphate is a naturally occurring inorganic salt and copper is an essential trace element in plant and animal nutrition. It is available in dusts wet table powders and fluid concentrates. Copper sulphate is also widely used as an algacide for controlling phytoplankton in fish ponds and lakes as well as a herbicide used in aquatic weed control since 1982 (Carbonell and Tarazona, 1993) [1]. As copper sulphate found to be a pollutant causing deleterious effect on aquatic organisms at different levels, in the present study effect of copper sulphate on nucleic acid content of some important tissues of the freshwater fish, *Clarias batrachus* has been undertaken.

2. Materials and Methods

Adult and live fish *Clarias batrachus* were collected from the farm Patra and Bhadbhada Bhopal M.P.) brought to the laboratory, cleaned by using 0.1% KMnO₄ to avoid dermal infection. Only healthy fishes (Length: 12-15cm, Weight: 50-60g) were taken for experiment. Fishes were acclimatized in glass aquaria for 15 days and were fed with fish food (earthworms) and water in the aquaria was replaced by freshwater at every 24h. Determination of LC50 50 fishes were used for the determination of LC50. The concentration that kills 50 per cent of the fish in 96 h duration (LC50/96h) was determine by Static Bioassay method (Doudoroff *et al.*, 1951) [2] by using the mortality values for 96h the

Correspondence:
Muneesh kumar
Govt. S.S.L. Jain PG College
Vidisha, Bhopal. (M.P)

LC50 was determined by plotting the graph. The exposed fish were sacrificed after 96h (LC50) and the tissues such as brain, liver, kidney and ovary were dissected out and processed for determination of DNA/RNA content. In all the cases six observations were made and the results (DATA) were expressed as arithmetic mean with their Standard Deviation, Standard Error and Student 't' were made as described by Suedecur (1946) and Fisher (1963) [3]. The nucleic acid (DNA and RNA) content of the tissues was estimated by following the Diphenylamine method of Schneider (1940) [6] using DNA as standard. The RNA content of the tissues was also estimated by Orcinol method using RNA as standard.

Table 1: Showing DNA and RNA content (mg/g) in different tissues of the freshwater fish, *Clarias batrachus* on exposure to Copper sulphate.

	Brain		Liver		Gonads		Kidney	
	DNA	RNA	DNA	RNA	DNA	RNA	DNA	RNA
Control	133.75±0.81	49.33± 0.25	145± 0.80	51.5±0.31	87.91± 0.40	49± 0.13	170± 0.79	58± 0.81
CuSo4 exposed	135.20±0.79	46.76± 0.49	137.5± 0.86	46.76±0.42	85.41± 0.76	45.95± 0.29	142.5± 0.86	55.51± 0.31

DNA/RNA ratio in different tissues: - Control:-brain has 2:1, liver 2:1, ovary 2:1 and only in kidney it is 1:1. Copper sulphate (CuSO₄):- brain has 2:1, liver 2:1, ovary 2:1 and only in kidney it is 1:1. Total RNA content comprised of m, r and t RNA are variable. Miglavs and Jobling (1989) [5] The RNA/DNA ratio indicates the protein synthetic potential of a cell and it is an index of fish growth. Wilder and Stanley (1989) reported the fall of RNA/DNA ratio of salmonid fishes by the treatment of carbaryl. Significant loss of metachromasia has been observed in mercury treated fishes after 9 and 30 days of exposure and moderate loss was found after 22 days. The effects of sub-lethal concentrations (3% and 15 % v/v) of untreated nickel chrome electroplating effluent on the reproduction of female *Clarias batrachus* were studied during preparatory, prespawning and spawning phases of reproductive cycle. Both GSI and HSI of the exposed fish were lower as compared to control fish in all the phases. An irregular pattern of deposition of macromolecules (DNA, RNA and Proteins) in liver as well as in ovaries indicated that exposed fish were not cycling properly due to stress. Alterations in the contents of macromolecules were greater in treatment T2 (15 % v/v) than T1 (3 % v/v) during all the phases (Kaur and Kaur, 2005). The present study clearly indicates that a short term exposure to copper sulphate, the DNA and RNA content of tissues get reduced in the freshwater fish *Clarias batrachus* indicating copper sulphate as pollutant effect the nucleic acid content in the tissues.

4. Conclusion

After the above discussion it had been concluded that copper sulphate causes deleterious effects on fishes and much alters the DNA and RNA contents of certain tissues. In sub lethal concentration it may not be fatal for an individual organism but it does affect the growth rate and reproduction resulting in disturbance to whole community and tropic levels of food chains, ultimately the ecosystem.

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3. Results and Discussion

The nucleic acid content in different tissues such as brain, liver, kidney and ovary of both control and copper sulphate exposed fish *Clarias batrachus* has been studied. The following results were observed in both control and copper sulphate exposed fish, ovary contain large amount of DNA in comparison to other tissues (Table 1). The degree of DNA content in control and copper sulphate exposed fish, ovary >liver>brain>kidney. The RNA content also exhibited similar to that of DNA having higher amount in the ovary. The degree of RNA content in control and copper sulphate exposed fish (Table.1) ovary >liver>brain>kidney.

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