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Toxic Impact Of Dimethoate On Glycogen, Protein And Lipid Level In Certain Tissues Of Freshwater Fish, *Rasbora Daniconius*.

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Abstract: The Dimethoate is one of the pesticide most widely used in agriculture sector in Maharashtra to control the varieties of pests and ectoparasites. The acute toxicity bioassay tests were conducted at various concentrations ranging from 5.0 and 13.0 mg/l freshwater fish, *Rasbora daniconius* for 24, 48, 72 and 96 h. The acute toxicity values were determined as 11.58, 10.25, 10.63 and 9.13 ppm respectively under laboratory conditions respectively. These LC50 values showed that the chemical is highly toxic to the fish. Then the fish were exposed to sub lethal concentrations 96 h LC50 9.13 ppm and studied the significant changes in glycogen, Protein and lipid levels of the fresh wet tissues. In the present investigation glycogen content of muscle, gill, liver and kidney as 12.78, 9.01, 42.30 and 11.12 mg/gm/wet.wt of tissues respectively and lipid content of muscle, gill, liver and kidney as 59.01, 49.23, 68.01 and 55.25 mg/gm/wet.wt of tissues respectively. In glycogen, protein and lipid content of muscle, gill, liver and kidney as 59.01, 49.23, 68.01 and 55.25 mg/gm/wet.wt of tissues respectively. In glycogen, protein and lipid content decreases in all tissues when compared with the control group.

Keywords: Glycogen, Protein, Lipid, Rasbora Daniconius, Dimethoate.

INTRODUCTION

Man has attempted to increase the world's food production to solve the problem of malnutrition. He achieved this by increased use of fertilizer to nourish the plant and by increased use of pesticides to protect them from pests. Recently a large quantity of pesticides and fertilizers are used to nourish the plants and food production. These chemicals have entered in to the aquatic system and create pollution which pose a great threat to aquatic organisms. There are several reports regarding the effects of pesticides (Arunachalam *et.al* 1985) on physiology of fish. The pesticides used in pest control programmes also produce many physiological and biochemical changes in freshwater organism particularly the fish (Girija 1984). The alteration of biochemical contents in different tissues of fish is due to toxic effects of different heavy metals and pesticides have been reported by many workers (Saxena *et.al.*, 1989, Khan *et.al.*, 1992, Virk and Sharma 1999, Rawat *et.al.*, 2002) Studied on effect of sevin on the Zebra fish Deirio sesio and stated that the organophosphorus (OPs) pesticides widely used in agriculture to prevent the disease causing pests, parasites due to their lower persistence in the environment. However, the intensive use of these chemicals in agriculture and public operations has changed the ecological balance of many non target

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organisms like fishes. Number of researchers observed that the Fishes are particularly very sensitive to the water contamination. Hence, pollutants such as insecticides, herbicides may significantly affect some physiological and biochemical processes when they enter into the organs of fishes M. Banaee et.al (2011), K.S. Tilak *et.al.* (2003), R. Mathivanan (2004), Susan, *et.al.* (1999) observed significant decrease in protein content in gills of *Catla catla* under sublethal concentration of pyrethroid fenvelerate. The alteration in biochemical contents in different tissues of fish due to toxic effects of different heavy metals and pesticides have been reported by number of workers Khan *et.al.* (1992); James & Sampath (1995); Das *et.al.* (1999); Khare and Singh (2002); Desai *et.al.* (2002); Remia *et.al.* (2008); Hadi *et.al.* (2009) and Ganeshwade, (2011) reported that the decline trend of glycogen content in liver and the intestine of *Ophiocephalus punctatus* exposed to sub lethal concentration of cypermethrin, these values of glycogen showed disturbance in carbohydrate metabolism due to toxic stress. Hence the present study was undertaken to evaluate the toxicity of Dimethoate on glycogen, protein and lipid levels of muscle, gill, liver and kidney in freshwater fish *Rasbora daniconius* at 96 hours exposure.

MATERIALS AND METHODS:

The test fish *Rasbora daniconius* were collected from Manjara River, Latur district and brought to laboratory. These fishes were observed for any pathological symptoms and then placed in 0.1% potassium permagnate (KMnO₄) for 2 minutes so as to avoid any dermal infection. fishes almost same size measuring 9 ± 2 cm and weighing about 7 ± 2 gms were selected for experimentation and divided into two groups of 10 fishes per aquarium. For the estimation of biochemical analysis the laboratory acclimatized Group 'A' served as control was kept in tap water. Group 'B' was exposed to lethal (9.136 ppm)

The physicochemical parameters were analyzed during the experimental period, by the standard methods suggested by APHA (1998) and IAAB (1998). For the biochemical analysis muscle, gill, liver and kidney were selected and the fishes were sacrificed immediately at the end of exposure period i.e 96 hours and tissue were excised rapidly and processed for the biochemical estimations after homogenizing the required media. The following standard methods were used for the determination of biochemical analysis. The protein contain analyzed by Lowery method (Lowery, *et.al* 1951). Glycogen estimation was done by Anthrone reagent (Seifer *et.al.* 1950). Lipid estimation was done by chloroform methanol method suggested by (Ramnik Sood, 2006). All values were expressed in mg/gm wet. wt. of tissues. **RESULTS:**

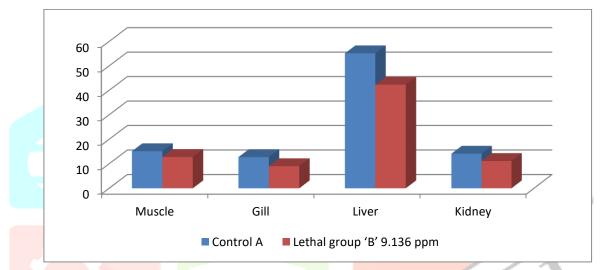
In the present investigation the biochemical changes in muscle, gill, liver and kidney of test fish *Rasbora daniconius* exposed to lethal (LC $_{50}$ of 96 hours i.e. 9.136ppm) of dimethoate. The effect of dimethoate on the different tissues at lethal concentration the glycogen content in control group were 15.12, 12.72, 55.14 and 14.11 mg/gm/wt respectively were as in experimental group were 14.08, 10.22, 48.28 and 12.08 mg/gm/wet.wt. of tissues in muscle, gill liver and kidney. Glycogen content in different tissues are represented in Table No. 1& fig. no.1. In protein content in control group were 18.04, 10.39, 15.85 and 10.02 mg/gm/wt respectively were as in experimental group were 19.10, 11.28, 16.11 and 10.90 mg/gm/wet.wt. of tissues in muscle, gill liver and kidney. Proetin content in different tissues are represented

in Table No. 2& fig. no.2. In lipid content in control group were 68.12, 65.30, 81.12 and 61.78 mg/gm/wt respectively were as in experimental group were 59.01, 49.23, 68.01 and 55.25 mg/gm/wet.wt. of tissues in muscle, gill liver and kidney. Proetin content in different tissues are represented in Table No. 3& fig. no.3

 Table No. 1.Effect of lethal concentration of dimethoate on glycogen content of muscle, gill, liver and kidney of fish at 96 hours exposure to *Rasbora daniconius*

Sr. No	Tissues	Control group 'A'	Lethal group 'B' (9.136ppm)
1	Muscle	15.12 <u>+</u> 0.47	12.78 <u>+</u> 0.28
2	Gill	12.72 <u>+</u> 0.31	9.01 <u>+</u> 0.09
3	Liver	55.14 <u>+</u> 0.41	42.30+ 0.76
4	Kidney	14.11 <u>+</u> 0.28	11.12 <u>+</u> 0.16

All values are expressed in mg/gm/wet.wt of tissues.



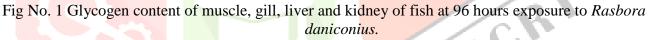


Table No. 2 Effect of lethal concentration of dimethoate on Protein content of muscle, gill, liver and kidney of fish at 96 hours exposure to *Rasbora daniconius*.

Sr. No	Tissues	Control group 'A'	Lethal group 'B' (9.136ppm)
1	Muscle	21.14 <u>+</u> 0.38	18.04 <u>+</u> 0.11
2	Gill	18.04 <u>+</u> 0.30	10.39 <u>+</u> 0.23
3	Liver	17.90 <u>+</u> 0.36	15.85 <u>+</u> 0.50
4	Kidney	11.20 <u>+</u> 0.23	10.02 ± 0.58

All values are expressed in mg/gm/wet.wt of tissues.

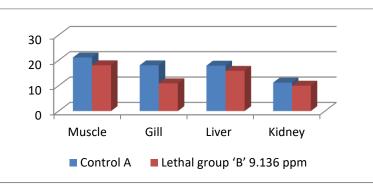


Fig No. 2 Protein content of muscle, gill, liver and kidney of fish at 96 hours exposure to *Rasbora daniconius*.

 Table No. 2 Effect of lethal concentration of dimethoate on Protein content of muscle, gill, liver and kidney of fish at 96 hours exposure to *Rasbora daniconius*.

Sr. No	Tissues	Control group 'A'	Lethal group 'B' (9.136ppm)
1	Muscle	68.12 <u>+</u> 0.23	59.01 <u>+</u> 0.47
2	Gill	65.30 <u>+</u> 0.31	49.23 <u>+</u> 0.37
3	Liver	81.12 <u>+</u> 0.55	68.01 <u>+</u> 0.37
4	Kidney	61.78 <u>+</u> 0.53	55.25 <u>+</u> 0.18

All values are expressed in mg/gm/wet.wt of tissues.

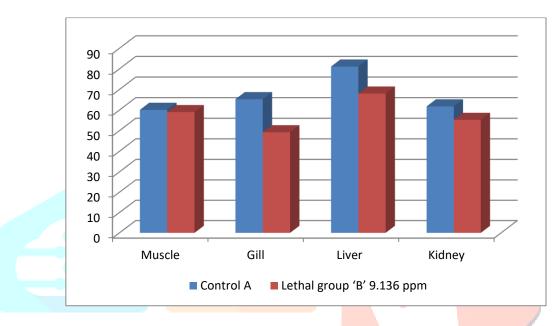


Fig No. 3 Lipid content of muscle, gill, liver and kidney of fish at 96 hours exposure to Rasbora daniconius.

DISCUSSION:

Kulkarni *et al.*, (2005) reported that the glycogen content in foot showed significant decrease after 24 hours of exposure. In hepato pancreas no significant decrease in glycogen content were observed after 24 hours but in gills the glycogen content showed significant decrease after 24, 48, 72 and 96 hours of exposure provide when compared with control. Abdul Naveed et al. (2006) worked on toxicity of lihocin on the activities of glycolytic and glycogenic enzymes of fish, Channa punctatus observed that the level of glycogen and pyruvate decreased while glucose and the lactic levels increased. Nagaraju Bantu and Rathnamma Vatika (2013) Studies effect of Dimethoate on mortality and biochemical changes of freshwater fish Labeo rohita (Hamilton) observed that the glycogen content in the different tissues in muscles 19.15, 7.18 and 5.29 at control, sublethal and lethal concentration respectively. In the kidney 9.18, 7.46 and 6.34 mg/g weight of tissue at control, sublethal and lethal concentraticon of dimethoate. In gills and liver 12.50, 8.15 and 3.38 mg/g weight of tissue at control, sublethal and lethal concentraticon of dimethoate. In liver 27.14, 8.29 and 5.39 mg/g weight of tissue at control, sublethal and lethal concentraticon of dimethoate. He stated that the changes in the amount of glycogen content the fish Labeo rohita exposed to sublethal and lethal concentration of dimethoate for 8 days. K. Satyavardhan (2013) studies on the effect of fenvalarate TM and Malathion TM on biochemical constituents of freshwater fish Ctenopharyngodon idella (Valenciennes) reported that glycogen content in gill, liver, kidney and muscle were 35.10, 82.22, 25.39 and 35.07 mg/g wet weight of tissue in control were as in lethal concentration

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19.95, 53.21, 16.93 and 15.12 m,g/g wet weight of tissue. He stated that depletion of glycogen content was noticed in exposed fish but significant decrease in glycogen content was noticed in muscles tissues in lethal exposure. A fall of glycogen level it might be due to the over activity of muscle under pesticides stress. The similar results were observed in the present investigation. In the present investigation the glycogen content in order of decreased in toxicant exposed fish observed in following manner Liver > Muscle > Kidney > Gill at sub lethal concentration. Decrease amount of glycogen shows in present study this may be due to the toxic stress such types opf results are reported by the number of researchres.

Aruna *et al.*, (2000) reported significant variations in the protein content of the kidney and *Clarias batrachus* exposed to sublethal concentration of malathion. Durga *et al.*, (2002) worked on effect of cypermethrin on protein metabolism of the fish, *Labeo rohita*, observed that the total protein level decreased in all the tissues tested where as the free amino acid levels were increased. Parate and Kulkarni (2003) reported that cypermethrin also decreases total protein content in the gills and muscles of freshwater crab, *Paratelphusa jacquinmtii*. It was due to alternative source of energy under the stress condition. Tripathi *et al.*, (2004) worked on endosulfan medicated biochemical changes in the freshwater fish, *Clarias batrachus* and observed that the high degree of change in protein metabolism of the organ of the fish exposed to the mixture of pesticide. Jagatheswari (2005) worked on biochemical changes induced by pesticide phosalone in *Cyprinus carpio* (Linn) and reported that the subacute period of exposure a significant depletion in protein contents were observed in different tissues.

Prashanth, (2006) worked on impact of cypermethrin on protein metabolism in freshwater fish, *Cirrhinus mrigala*, observed that the depletion of structural soluble and total protein was observed in all the tissues. Prabhakara Rao and Radhakrishnaiah (2006) worked on the pesticidal impact on protein metabolism of the freshwater fish, Cyprinus carpio, observed that the total protein content increased in the gills and decreased in the muscle of the freshwater fish. Ganeshwade (2011) reported that the decrease in the protein was observed throughout the exposure period. The two sublethal exposure results shows the protein content decreases are depend upon the concentration. There is progressive decrease in the protein content with increase in concentration. He stated that the toxicity of dimethoate showed a direct correlation with the concentration and time exposure while working on dimethoate toxicity to Puntius ticto. Ganeshwade (2012) observed that the decreased protein content in the gills after the exposure period of 60 days. As compared to lethal exposure, significant change in the protein content was observed during the sublethal exposure while working on biochemical changes induced by dimethoate (Roger 30%) in the gills of freshwater fish puntius ticto (Hamilton). In the present investigation the protein content decreased in all the tissues exposed to sub lethal concentration may be due to increase in proteolytic activity and toxicity stress. Similar results are reported by the researchres. Nagaraju Bantu and Rathnamma Vatika (2013) Studies effect of Dimethoate on mortality and biochemical changes of freshwater fish Labeo rohita (Hamilton) observed that the protein content in the different tissues in muscles 53.4, 52.0 and 51.15 mg/g weight of tissue at control, sublethal and lethal concentration respectively. In the kidney 55.4, 47.4 and 43.1 mg/gm weight of tissue at control, sublethal and lethal concentraticon of dimethoate. In gills 94.3, 69.4 and 56.5 mg/gm weight of tissue at control, sublethal and lethal concentration of dimethoate. In liver 51.3, 25.6

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and 49.4 mg/gm weight of tissue at control, sublethal and lethal concentration of dimethoate. He stated that the changes in the amount of glycogen content the fish *Labeo rohita* exposed to sublethal and lethal concentration of dimethoate for 8 days.

Amanulla Hameed *et al.* (2004) study on effect of butyllin toxicity of lipid metabolism in an easturine mussel, *Sunetta scripta* and reported that body tissue, significant decrease in the digestive gland was noticed and the percentage of lipid content decreased from 3.6 to 2.1, 3.7 to 1.6 and 3.2 to 1.0 at 24, 48, 72 and 96 hour respectively. Kulkarni *et al.*, (2005) reported that the no significant change in total lipid content in foot for each exposure period was observed when compared with control and no significant changes in total lipid content in hepatopancreas also at each exposure period was observed when compared with control and same result was observed in gills. The above researchers reported the similar results in his investigation. The lipid content after exposure of sub lethal concentration of dimethoate the decease trend in all the tissues were observed the decreasing trend as liver > muscle > kidney> gill. Sudhanshi Tiwari *et al.*, (2004) worked on toxic and sublethal effect of oleandrin on biochemical parameters of freshwater fish air breathing murrel, *Channa punctatus*, observed that the significant alteration in the level of total protein, total free amino acid, nucleic acid, glycogen, pyruvate, lactate and enzyme protease, phosphatases, alanine aminotransferase, asparate aminotransferase and acteyl- cholinestrase activity in liver and muscle tissues.

CONCLUSION

In the present investigation the effect of dimethoate on sub lethal concentration on the freshwater fish *Rasbora doniconius* showed variation in level of the biochemical components in muscle, gill, liver and kidney. The glycogen content in order of decreased in toxicant exposed fish observed in following manner Liver > Muscle > Kidney > Gill at sub lethal concentration. Decrease amount of glycogen shows in present study this may be due to the toxic stress in the stress condition of an organism needs sufficient energy which is supplied from reserved glycogen. The decrease in glycogen content may be due to the inhibition of hormones which contribute to glycogen synthesis. The protein content decreased manner were observed muscle > liver > gill > kidney. So the protein content decreased in all the tissues exposed to sub lethal concentration may be due to increase in proteolytic activity and toxicity stress. The lipid content after exposure of sub lethal concentration of dimethoate the decease trend in all the tissues were observed the decreasing trend as liver > muscle > kidney> gill.

In the present investigation, it has been found that decrease in biochemical components of all tissues. Decrease in biochemical component of tissues muscle, gill, liver and kidney might be due to meet energy demand of fish under pesticide stress.

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