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Bioaccumulation of Zn, Cu and Cd in *Channa punctatus*

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Abstract: *Adult Channa punctatus were acutely exposed to LC_{50} of zinc (18.62 mg/l), cadmium (11.8 mg/l) and copper (0.56 mg/l) separately for 96 hr The concentration of metals was found maximum in liver and minimum in muscles. The degree of accumulation among the five tissues differed and it was in the order : gill>liver>kidney>blood>muscle in case of Zn, gills>kidney>blood> liver>muscle in case of Cd and gills>kidney> blood>liver>muscle in case of Cu exposure.*

Key words : *Copper, Cadmium, Zinc, Accumulation, Channa punctatus, LC_{50}*

Introduction

Heavy metals are available in small quantities in the water and are further added due to soil erosion and leaching of minerals. However in the recent past, fresh water pollution due to heavy metals has become a hazard due to discharge of industrial effluents. This wide spread problem has ultimate effect on aquatic fauna. Trace metals like Cr, Mn, Fe, Co, Ni, Cu, Zn, Se *etc.* are essential for the growth of organisms, while Pb, Cd, Hg and As are not only biologically non essential, but definitely toxic. Even the essential trace metals may be beyond certain optimum threshold levels, hazardous and toxic. After entering the water, metals may precipitate, get adsorbed on solid surface, remain soluble or suspended in water or taken up by fauna. A very important biological property of metals is, their tendency to bioaccumulation. Bioaccumulation is thus a response that is important in hazard evaluation strategies. Acute metal toxicity in fish is often characterized by gills damage and hypersecretion of mucus, ensuing mortalities are related to secondary physiological respiratory disturbance, resulting in ion regulatory and acid-base balance disturbances. The extent of physiological disturbances depends upon uptake and bio accumulation of metals.

Of the various heavy metals Zn, Cd and Cu are widely distributed and important as regard to their deleterious effects. Therefore in the present study the tissue uptake of Zn, Cd and Cu individually by the locally available poor men's fish, *Channa punctatus* has been examined. Sultana and Rao (1998), studied the bioaccumulation pattern of Zn, Cu, Pb and Cd in grey mullet, *Mugil cephalus* from harbour waters of Visakhapatanum. Kargin (1998) reported that the level of given metal showed significant difference between the tissue of *Capocta barroisi.* Muscles generally accumulate the lowest level of metals in every season. Ay *et al.* (1998) reported accumulation of copper and lead in the tissue of a fresh water fish, *Tilapia zilli*. Mazan and Fernandes (1999) studied the toxicity and differential tissue accumulation of copper in the tropical fresh water fish, *Prochilodus scrofa.* Karakoc (1999) observed an increase in the uptake of Cu in the liver, gill and muscle tissues of *Tilapia nilotica* at low salinities. Abreu (2000) studied accumulation of Hg in see bass. Karakoc and Dincer (2003) studied the effects of temperature on Zn accumulation in the different organs of *Oreochromis niloticus.* The present paper incorporates a part of our study aimed at understanding the physiological responses of fish following exposure to heavy metal mixture containing, Zn, Cu, Cd and includes the bioaccumulation and tissue distribution of individual metals from a metal mixture containing Zn, Cu and Cd in gill, liver, kidney, muscle and blood in the fresh water fish, *Channa punctatus.*

Materials and Methods

The live fish *Channa punctatus* caught from local ponds were brought to the laboratory and kept in aquaria (3''×3''×2'') for acclimation to laboratory condition for three week, before starting experimental studies. The fish were fed twice daily with pelleted diet (prawn powder, fish powder and minced liver in 2 : 2 : 1 ratio).

Eighty fish randomly divided into four groups of twenty each, were exposed to LC_{50} of zinc (18.62 mg/l), copper (0.56 mg/l) and cadmium (11.8 mg/l) separately. The fourth group of twenty fish maintained in metal free water, served as control for each experimental group. After 96 hr from each of the experimental and control groups surviving fish were sacrificed for the estimation of Cd, Cu and Zn in the muscle, liver, gills, kidney and blood.

Each tissue and blood were pooled separately in petridishes and dried at 60°C, until the weight became constant. One gram of each tissue from control and exposed groups were transferred to a 100 ml beaker and 1.0 ml H_2SO_4, 2 ml HNO_3 and 0.5 ml of perchloric acid was added (Topping, 1973). The beaker was gently heated on a hot plate, until the tissue dissolved. The content of the beaker was diluted to 10-15 ml with triple distilled water. The concentrations of the heavy metals were estimated with the help of Atomic Absorption Spectrophotometer with air-acetylene mixture as fuel.

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Results and Discussion

The data on metal concentration is summarized in Tables 1, 2 and 3. Irrespective of whether the metal is essential or non essential, the accumulation levels of all the three metals in the different organs was significantly greater. Metal uptake by aquatic organism is a two phased process, which involves initial rapid adsorption or binding to surface, followed by a slower transport into the cell interior. In epithelial tissues the last step is rate limiting factor in transepithelial movement of metals. Transport of metal into the intracellular compartment may be facilitated by either diffusion of the metal across the cell membrane or by active transport by a carrier protein (Brezonik *et al.,* 1991). A third process involved in determining metal uptake is the speciation of the metals in the medium before contact with gill epithelia. It is generally accepted that metal accumulation in tissues of aquatic animals is dependent upon the exposure concentration and period, as well as some other factors, such as salinity, temperature, interacting agents and metabolic activities of tissue concerned.

Karakoc and Dincer (2003) reported highest accumulation of Zn in kidney tissue at 15°C and 30°C for different concentrations which is followed by gills and liver. In all tissues Zn accumulation increased with increasing temperature. Karakoc (1999) observed an increase in the uptake of Cu in the liver, gill and muscle of *Tilapia nilotica* at low salinities since a decrease in salinity from 20 to 50 per cent caused an increase in the metal uptake. Dallinger and Kautzky (1985) reported the accumulation of class 'B' metals in metabolically active organs such as liver and kidney.

The accumulation of Zn, Cd and Cu was observed in the selected fish tissue in the order gill>liver>kidney>blood>muscles: gill>kidney>blood>liver>muscle and gill>kidney>blood>liver> muscle, respectively. Maximum accumulation of metals was recorded in gills as they were in direct contact with ambient medium and are the main site of water movement, while minimum accumulation was recorded in muscles.

It was also observed in the present study that adsorption of Cu by the gills of fresh water fish was greater followed by Zn and Cd. As copper has the highest covalent index value, it could be expected that it would be more competitive than Zn for available intracellular nitrogen and sulphur-rich ligands (Wepener *et al.*, 2001). Significant accumulation of Zn in gills was reported when rainbow trout exposed to high Zn concentrations (Hughes and Flos, 1978). Villegas Navarro and Villarreal Trevino (1989) reported that Zn uptake by the Texas cichlid was greater than the uptake of Pb and Cu.

The liver is the main organ for metal regulation in fish. Exposure to metals results in induction of metallothioneins production and subsequent binding of metals to the protein. The present study showed that uptake of Zn was maximum followed by Cu and Cd in liver.

The maximum Zn uptake in liver is reflected by its minimum concentration in blood stream. Stimulation of Zn uptake by liver tissue following Cd and Cu exposure was also recorded by Noel Lambot *et al.* (1978) in eel. The increased uptake of Zn in presence of other metals can be attributed to the proposed stress response mechanism, which liberates and depurates metals from metal-binding proteins in liver thus providing available proteins for the binding of excess metals. This is thought to be an intermediary response mechanism, which functions until sufficient MT has been produced to bind to the excess accumulated metals. Sultana and Rao (1998) noted that maximum accumulation of zinc occurred in gills followed by liver in grey mullet. Singh *et al.* (1990) during their study on the accumulation of copper, zinc, lead, iron and cadmium in some fresh water fishes of river Subernarekha reported that liver accumulated the highest quantity of these metals followed by kidney. Greater uptake of zinc by liver in comparison to kidney

Table - 1: Uptake of Zn by different tissues of *Channa punctatus* exposed to 18.62 mg/l of Zn for 96 hr

S. no.	Tissue	Control fish	Exposed fish	Percentage alteration
1.	Muscle	1.74±0.16	3.92±0.04***	125.3
2.	Liver	6.66±0.19	24.34±0.16***	265.4
3.	Gill	16.50±0.42	69.45±0.17***	320.9
4.	Kidney	18.20±0.09	49.72±0.29***	173.18
5.	Blood	16.52±0.80	42.14±0.16***	155.08

Units µg/g wet weight
Values are mean ± SD; n=6
***p < 0.001

Table - 2: Uptake of Cd by different tissues of *Channa punctatus* exposed to 11.80 mg/l of Cd for 96 hr

S. no.	Tissue	Control fish	Exposed fish	Percentage alteration
1.	Muscle	1.26±0.01	2.37±0.07***	88.1
2.	Liver	5.52±0.10	14.14±0.24***	156.2
3.	Gill	16.59±0.24	48.16±0.32***	190.3
4.	Kidney	5.34±0.34	4.19±0.18***	165.9
5.	Blood	10.96±0.14	28.42±0.12***	159.3

Units µg/g wet weight
Values are mean ± SD; n=6
***p < 0.001

Table - 3: Uptake of Cu by different tissues of *Channa punctatus* exposed to 0.56 mg/l of Cu for 96 hr

S. no.	Tissue	Control fish	Exposed fish	Percentage alteration
1.	Muscle	1.46±0.01	3.38±0.06***	131.5
2.	Liver	10.85±0.16	34.17±0.12***	214.9
3.	Gill	12.74±0.32	69.78±1.22***	447.7
4.	Kidney	9.49±0.32	42.56±0.18***	348.5
5.	Blood	15.72±0.88	66.43±0.72***	322.5

Units µg/g wet weight
Values are mean ± SD; n=6
***p < 0.001

after 96 hr exposure of fish may be due to greater synthesis of metallothioneins in liver in comparison to kidney.

Copper and cadmium accumulated preferentially in kidney of fish *Mylio macrocephalus* (Ooi and Law, 1989) and on prolonged exposure, cadmium is deposited initially both in liver and kidney in comparable concentrations. Its accumulation has been linked to renal hypertension. Highest copper content was bound to the relatively lower molecular weight protein fraction metallothionein in hepatopancreas and kidney as suggested by Muramoto (1981).

After gills, kidney is more important target organ, than the liver. Accumulation of Cu, Zn, Hg, Ni, Cd and Cr in liver, muscle and gills were studied in different fish species (white fish, perch, pipe, brown trout, burboat and vendance) and significant differences were found in between metal accumulation by these fish species (Amundsen *et al*., 1997). Adeyeye *et al*. (1996) studied the different accumulation rates and reported that Zn, Pb, Mg, Fe, Cu and Co accumulation in the organs (gill, liver, sex organs, intestine, eyes, head, scales, swim bladder and trunk muscles) of *Clarias gariepinus, Cyprinus carpio* and *Oreochromis niloticus.* In kidney Cu showed maximum accumulation followed by the Zn and Cd.

References

Abreu, S.N., E. Pereira, C. Vale and A.C. Duarta: Accumulation of mercury in seabass from a contaminated lagoon (Ria de Aveiro. Portugal). *Mar. Pollut. Bull.,* **40**, 293-297 (2000).

Adeyeye, E.E., N.J. Akinyugha, M.E. Fesobi and V.O. Tenabe: Determination of some metal in *Clarias gariepinus* (Cuvier and Vallencienner), *Cyprinus carpio* (L.) and *Oreochromis niloticus* (L) fishes in a poly culture fresh water pond and their environments. *Aquaculture*, **147**, 205-214 (1996).

Amundsen, P.A., F.J. Staldvik, A.A. Lukin, N.A. Kashulin, O.A. Popova, and Y.S. Reshetnikov: Heavy metal contamination in fresh water fish from the border region between Norway and Russia. *Sci. Tot. Environ*., **201**, 211-224 (1997).

Ay, O., M.Kalay, L. Tamer and M. Canli: Copper and lead accumulation in tissues of a fresh water fish *Tillapia zillii* and its effect on the branchial Na+K-ATPase activity. *Bull. Environ. Contam. Toxicol*., **60**, 160-168 (1998).

Brezonik, P.L., S.O. King and C.E. Mach: The influence of water chemistry on trace metal bioavailability and toxicity to aquatic organisms. *In*: Metal ecotoxicology concepts and applications (*Eds*: M.C. Newman and A.W. Mdntosh). Lewis Publishers Inc., Michigan. pp. 1-26 (1991).

Dallinger, R. and H. Kautzky: The importance of contaminated food for the uptake of heavy metals by rainbow trout (*Salmo gairdneri*) a field study. *Oceologia*, **67**, 82-89 (1985).

Hughes, G.M. and R. Flos: Zinc content of the gills of rainbow trout (*S. gairdneri*) after treatment with zinc solutions under normoxic and hypoxic conditions. *J. Fish Biol.,* **13**, 717-728 (1978).

Karakoc, M.: Effects of salinity on accumulation of copper in liver, gill and muscle tissue of *Tilapia nilotica*. *Tr. J. Zool*., **23**, 299-303 (1999).

Karakoc, M. and S. Dincer: Effect of temperature on zinc accumulation in the gill, liver and kidney of *Oreochromis niloticus*. *Bull. Environ. Contam. Toxicol*., **71**, 1077-1083 (2003).

Kargin, G.: Metal concentration in tissues of the fresh water fish *Capoeta barroisi* from the Seyhan river (Turkey). *Bull. Environ. Contam. Toxicol.,* **60**, 822-828 (1998).

Mazan, A.F. and M.N. Fernandes: Toxicity and differential tissue accumulation of copper in the tropical fresh water fish, *Prochilodus sarofa*. *Bull Environ. Contam. Toxicol*., **63**, 797-804 (1999).

Muramoto, S.: Influence of complexes (EDTA , DPTA) on metal toxicity of Cd, Cu, Zn and Pb at lethal level. *Bull. Environ. Contam. Toxicol.,* **25**, 878-883 (1981).

Noel-Lambot, F., C. Gierday and A. Distechae: Distribution of Cd, Zn, and Cu in liver and gills of the eel (*Anguilla anguilla)* with special reference to metallothionein. *Comp. Biochm*., **61C**, 171-187 (1978).

Ooi, V.E.C. and F.K. Law: Changes induced by cadmium in the kidney of back seabream *Mylio macrocephalus* (Telostei). *Bull. Environ. Contam. Toxicol.,* **43**, 769-775 (1989).

Singh, N.K., M.C. Verma and J.S. Datta Munshi: Accumulation of copper, zinc, lead, iron and cadmium in certain fresh water fishes of river Suberanrekha. *J. Freshwater Biol*., **2**, 189-193 (1990).

Sultana, R. and D.P. Rao: Bioaccumulation pattern of zinc, copper, lead and cadmium in grey mullet, *Mugil cephalus* (L.), from habour waters of Visakhapatnam, India. *Bull. Environ. Contam. Toxicol.,* **60**, 949-955 (1998).

Topping, G.: Heavy metals from Scottish water. *Aquaculture*, **1**, 379-384 (1973).

Villegas Navarro, A. and C.M. Villarreal Trevino: Differential uptake of zinc, copper and lead in Texas cichlid (*Cichlasoma cyanoguttatum*). *Bull. Environ. Contam. Toxicol*., **42**, 761-768 (1989).

Wepener, V., J.H.J. Van Vuren and H.H. du Preez: Uptake and distribution of a copper, iron and zinc mixture in gill liver and plasma of fresh water teleost, *Tilopia sparrmanii*. *Water,* **27(1)**, 99-108 (2001).