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Effect of Temperatures on Zinc Accumulation in the Gill, Liver, and Kidney of *Oreochromis niloticus* (L. 1758)

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In recent years, different factors such as increases in pesticide usage, industrial development and mining have led to an increase in the levels of Zn, Cu, Cd and the other metals in the aquatic environments (Guerrero and Kesten 1993).

Cu, Zn and Hg are the most harmful polluting agents in the aquatic environments. The accumulation of these class B metals in the tissues of fish and other water organisms may cause various physiological defects and mortality (Brown and Ahsanullah 1971, Coello and Khan 1996).

It is well known that temperature plays an important role in the toxicity of aquatic metals (Yang and Chen 1996). In addition, temperature affects mitochondrial activity, ATP synthesis (Blier and Guderly 1993), enzyme activity (Overnell and Batty 2000) and oxygen consumption (Espina et al. 2000).

The purpose of present study is to evaluate the accumulation of Zn in gill, liver and kidney of freshwater fish, *Oreochromis niloticus,* (L. 1758) at various temperatures.

MATERIALS AND METHODS

O. niloticus (L. 1758) were brought to the laboratory from pools and acclimatized to laboratory conditions at 25±1°C for three months in 5 stock aquaria, each containing 120 liters of fresh water. *O. niloticus* (L.1758) tested had an average length of 10.82±1.19 cm and weighted 15.34±1.172 g. They were then acclimatized to experimental medium temperatures (15 °C and 30°C) for three weeks.

Experiments were conducted at two test temperatures (15°C and 30°C) for 10, 20 and 30 days and at 1.0, 5.0 and 10.0 ppm test concentrations. Four glass aquaria with sizes of 40cmx120cmx40 cm were used for each experiment. The first three aquaria were filled with 1.0, 5.0 and 10.0 ppm of Zn solution and the fourth aquarium was filled with tap water and used as a control. Each experiments had three replicates.

Stock solution of Zn - sodium citrate was prepared by mixing zinc sulphate

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($ZnSO_4;7H_2O$) and tri - sodium citrate ($C_6 H_5 O_7 Na_3; 5H_2 O$) solutions (Brown and Ahsanullah 1971). Test solutions were prepared by dilution of this stock solution. Zn solutions in the experimental tanks were changed once every three days.

Tissue samples were dried at 105 °C for 48 hours. Dried samples were digested in a nitric-perchloric acids mixture (2:1) at 120 °C for 3 hours and made up to 5 ml constant volume with distilled water (Muramato1983). Zn concentrations were determined using a Perkin –Elmer 3100 Atomic Absorbtion Spectrophotometer.

Student-Newman Keul's Test (SNK) was used for the statistical analysis of the data.

RESULTS AND DISCUSSION

Results were analysed by using SNK test. Mean and standard errors of Zn concentrations in the tissues for each exposure temperature, metal concentration and period are given in tables (1-3). Different letters to indicate the statistical differences at P assigned the data points<0.05 level. No mortality was observed in *O. niloticus* at any Zn concentrations and temperatures.

Accumulation of Zn in the kidney, liver and gill tissues *of O. niloticus* increased with metal concentrations and exposure time. After 30 days, the highest Zn accumulation was observed in the kidney tissue at 15°C and 30°C for three test concentrations (1.0, 5.0 and 10.0 ppm) and followed by gills and liver (Table 1-3, $P<0.05$). In all tissues, the Zn accumulation increased with increasing temperature. In other word, Zn accumulation was always higher at 30°C than at 15°C ($P<0.05$).

After 30 days, Zn accumulation in the kidney of fish exposed to 10 ppm Zn concentration at 15°C was 1.8 and 3.7 times higher than that in gill and liver respectively while it was 2.1 and 2.8 times higher in the same tissues at 30°C. Zn accumulation in the kidney, gill and liver tissues of control organisms were always lower than those exposed to 1.0, 5.0 and 10.0 ppm Zn concentrations.

In the present study, the accumulation of Zn in kidney, liver and gill tissues of *O. niloticus* increased with increasing Zn concentrations and exposure time at all temperatures as was observed also by Brotheridge et al. (1998) for the accumulation of Cu, Zn, Ni and Co in the liver, kidney and gill tissues of *Salmo trutta.*

Metal accumulation in fish tissues is dependent upon targets organ and species of fish as well as other factors such as exposure time, temperature, salinity and type of metal (Bradley and Morris 1986, Yang and Chen 1996, Abreu 2000). In present studies carried out with *Clarias lazera* the highest Zn and Cu accumulation occurred in the liver and followed by the kidney and gills (Hilmy et al. 1987a).

Karaoke (1999) observed an increase in the uptake of Cu, in the liver, gill and muscle tissues of *Tilapia nilotica* at low salinities since a decrease in salinity from

Table 1. Effect of temperatures on the accumulation of Zn (μg Zn/g dry. wt.) in the kidney of *Oreochromis niloticus* at different concentrations and times.

		Temperature (°C)			
		15°C		30°C	
Time (Day)	Zn Concentrations (ppm)	$\overline{X} \pm s_{\overline{x}}$	*	$\overline{X} \pm s_{\overline{x}}$	*
	Control	80.03± 3.14	ae	119.25±2.83	af
10	1.0	119.81±1.48	be	372.48±6.40	bf
	5.0	131.81±3.06	ce	449.11±16.48	cf
	10.0	137.44±0.88	ce	582.25±21.62	df
	Control	82.11±2.41	ae	118.65±3.70	af
20	1.0	124.57±1.26	be	542.66±2.70	bf
	5.0	134.70±0.72	ce	590.33±14.12	cf
	10.0	150.94±1.42	de	692.79±10.06	df
	Control	81.69±1.32	ae	122.05±1.34	af
30	1.0	135.70±0.79	be	619.18±1.13	bf
	5.0	143.06 ±1.97	be	683.73±8.51	cf
	10.0	200.81±3.86	ce	800.94±12.25	df

* = SNK: Letters *a, b, c* and *d* show differences among zinc concentration; *e* and f among temperatures. The letters denote statistically significant differences at the P<0.05 level.

$\overline{X} \pm s_{\overline{x}}$: Mean Standard error

20%0 to 5%0 caused an increase in the metal uptake. Accumulation of Cu, Zn, Hg, Ni, Cd and Cr in liver, muscle and gills were studied in different fish species (whitefish, perch, pike, brown trout, burbot and vendace) and significant differences were found between in metal accumulation by these fish species (Amundsen et al. 1997). On the other hand, different accumulation rates were also observed by (Adeyeye et al. 1996) who studied the accumulations of Zn, Pb, Mg, Fe, Cu and Co accumulations in the organs (gill, liver, sex organs, intestine, eyes, head, scales, swim bladder and trunk muscles) of *Clarias gariepinus, Cyprinus carpio* and *Oreochromis niloticus.* Zn accumulation was found in gill to be higher than that in liver in *Anguilla anguilla* and *S. trutta,* (Legorburu et al. 1988). In the present study, the acumulation of Zn was highest in the kidney but lowest in liver.

In fish and other aquatic animals exposed to class B metals, accumulation dominantly occurs in metabolicaly active organs, such as liver and kidney (Dallinger and Kautzky 1985). Fish respond to class B metal exposure by producing Metallothioneins (MTs). MT bind metals and thus protect cellular proteins from metal poisoning (Carginale et al. 1998). They are accumulated and some times stored specially by liver tissue accumulates and at the some time stores them (Hogstrand et al. 1996). An increase was observed in liver MT synthesis in *Oncorhynhus kisutch* and *Pleuronectes platessa* parallel to the increase of metal concentration in the medium (Overnell et al. 1987). It was observed that the MT synthesis under the effects of Cu, Zn and Cd increased for four weeks and then remained at a steady state level in *Salmo gairdneri* (Roch and

Table 2. Effect of temperatures on the accumulation of Zn (μg Zn/g dry. wt.) in the gill tissue of *Oreochromis niloticus* at different concentrations and times.

		Temperature (°C)			
		15°C		30°C	
Time (Day)	Zn Concentrations (ppm)	$\overline{X} \pm s_{\overline{x}}$	*	$\overline{X} \pm s_{\overline{x}}$	*
	Control	34.82±0.75	ae	84.64±3.03	af
10	1.0	44.81±0.89	be	150.19±1.47	bf
	5.0	53.09±0.28	ce	200.51±3.53	cf
	10.0	71.39±1.47	de	255.21±1.35	df
	Control	35.09±0.98	ae	86.26±4.03	af
20	1.0	56.19±1.03	be	194.46±0.51	bf
	5.0	68.16±2.14	ce	253.05±0.10	cf
	10.0	92.34±0.72	de	318.98±18.31	df
	Control	35.34±0.68	ae	85.21±2.99	af
30	1.0	75.72±1.68	be	253.57±5.58	bf
	5.0	91.40 ±0.79	ce	336.06±3.59	cf
	10.0	110.15±2.90	de	371.24±2.18	df

* = SNK : Letters *a, b, c* and *d* show differences among zinc concentration; *e* and *f* among temperatures. The letters denote statistically significant differences at the P<0.05 level.

$\overline{X} \pm s_{\overline{x}}$: Mean±Standard error

McCarter 1984). Zn accumulation in all tissues of *O. niloticus* at 30°C temperature was observed higher than 15°C temperatures. Accumulation of Zn in gill, kidney and liver tissues of fish increased with increasing exposure temperatures.

It is well known that temperature plays an important role in the toxicity of metal ions in aquatic environments (Poleo et al. 1991). Also, Temperature affects growth, swimming performance, activity, metabolic rate, metal accumulation and metal toxicity (Larson et al. 1985, Foster et al. 2000).

Another study was carried out with bluefish *Potatomus saltatrix* temperature caused a change on swimming speed (Olla and Stuholme 1971). Gupta and Rajbanshi (1988) found in *Channa punctatus* that Cd toxicity was the lowest at 22°C and the highest at 31.1°C. The gill accumulate the highest amount of Cd, the liver accumulated a slightly smaller amount than the gill, while the kidney accumulated the least. Hilmy et al. (1987b) showed that higher temperature increased the accumulation of Zn in *Tilapia zilli* and *C. lazera*. It appears that *Tilapia zilli* is more susceptible to Zn than *Clarias lazera*. After a 96 hr exposure period, Zn was decreased in the following order: gill > liver > muscle. Similarly, Köck et al. (1996) showed, in *Salvelinus alpinus*, that the Cd and Pb accumulation increased in liver, kidney and stomach with increasing temperature.

At the end of the 30th day, Zn accumulation in the tissues of *O. niloticus* exposed

Table 3. Effect of temperatures on the accumulation of Zn (μg Zn/g dry. wt.) in the liver tissue of *Oreochromis niloticus* at different concentrations and times.

		Temperature (°C)			
		15°C		30°C	
Time (Day)	Zn Concentrations (ppm)	$\overline{X} \pm s_{\overline{x}}$	*	$\overline{X} \pm s_{\overline{x}}$	*
	Control	21.36±0.79	ae	65.85±0.40	af
10	1.0	26.06±1.14	be	105.52±1.75	bf
	5.0	29.21±0.18	ce	171.82±3.23	cf
	10.0	36.41±0.60	de	242.70±1.17	df
	Control	20.90±0.97	ae	63.96±1.83	af
20	1.0	32.13±0.43	be	123.90±6.72	bf
	5.0	35.62±0.37	be	193.25±0.31	cf
	10.0	41.72±3.29	ce	253.13±0.31	df
	Control	20.40±0.33	ae	64.52±1.23	af
30	1.0	34.61±2.95	be	174.71±4.25	bf
	5.0	37.72±0.76	be	226.90±8.43	cf
	10.0	54.03±0.19	ce	279.04±4.03	df

* = SNK : Letters *a, b, c* and *d* show differences among zinc concentration; *e* and *f* among temperatures. The letters denote statistically significant differences at the $P<0.05$ level.

$\overline{X} \pm s_{\overline{x}}$: Mean±Standard error

to 10.0 ppm Zn concentration at 30°C increased five times, four times and around three times in the liver, kidney and gills, respectively, compared with those exposed to the same concentration at 15°C. The highest Zn accumulation was found in the liver tissue. It was probably due to the fact that, temperature affects the metabolic activity of fish. Felts and Heath (1984) showed that temperature and sublethal environmental Cu effected on the energy metabolism of bluegill, *Lepomis macrochirus*. As a result, the increase in temperature might lead to the changes in the metabolic activity of *O. niloticus* and on the other hand, decreasing of temperature lowered the uptake rate of Zn from the medium.

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