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## Accumulation of Heavy Metals (Lead and Zinc) in the Liver of Some Edible Fishes in Zayandeh-rood

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### ABSTRACT

The aim of the study is measuring and studying the amount and variation of some important heavy metal on fish body. Zayandeh-rood flows from origin in Chahar-Mahal-Bakhtyari to the East of Iran and ends in Gavkhooni swamp in Isfahan province. Waste waters from farming, homes and industry are responsible for its pollution with heavy metals which can be absorbed from the liver of fishes. Therefore a study on environmental varieties is important. In order to determine the amount of lead and zinc in the liver of fishes, 5 stations have been organized as follow: Zarinshahr, Nekooabad, Falavarjan, Wahid, Choom. Edible fishes belong to the Cyprinidae group and *Carrassius*, *C. aculeata*, *C. damasciana*, *C. carpio* species. In order to assess the amount of lead and zinc, the liver tissue of fishes separates, burns, digests their ashes then the amount of heavy metal is calculated. The A.A.S. average of lead was  $3.13 \pm 0.9$  mg kg<sup>-1</sup> dry weight,  $p < 0.05$  and zinc was  $146.5 \pm 36.9$ ,  $p > 0.05$ . The zinc level is high but lead is normal. There is a high level of zinc in Zayandehrood. These results support with other studies by other researchers. A high level of zinc could cause metabolic disorders and a detoxification process could create many clinical damages in human.

**Key words:** Amount, waste water, pollution, Cyprinidae, tissue

### INTRODUCTION

Zayandeh-rood is the most important river in central of Iran and flows from Zagros Mountain in Charmahal-Bakhtyari to Zayandeh-rood collective water plan in Isfahan province. Of course this is changed to the receptor source of sewage waters of farming and industrial and home human activities. So studying on this changes and measuring the input pollutant into the river is necessary (Sadeghi-Rad, 1997).

The information of Isfahan fishery office and Isfahan environment protection organization and other research sources on fish of river especially in Zayandeh-rood are from Cyprinidae family that they are Teleost, without teeth on their mandibles and with gullet teeth (Vosooghi and Mostagir, 2003).

In this research two heavy metals lead and zinc in the Zayandeh-rood fish liver is studied. Lead and Zinc are trace heavy metal. These two metals have expanded effect on environment because of their uses in his life (Stanley, 1993). Lead uses for making tubs, wires, print industry and many kinds of lead compound are used in petrol. Zinc uses in metal paint and pigments and uses for making color and pigment in aliaje types (Pajoomand and Shariat, 1999).

On of most important internal for metal is atmosphere then metals inter to the aquatic ecosystem lead and zinc have the most amount intrance level in aquatic ecosystem (about 100-9000 ppm). Other entrance for metal is waste water of industry, farming and home actually (Stanley, 1993). Heavy metal stability in environment produced some special problems. One of the important results of stability is metal expansion in food chain. Plants and animals are different in metal content regulating ability (Jalali, 2003).

Heavy metals are absorbed from the body organ surface and usually absorbed with passive transition or with between transmitter in gill. But metals in water are eaten in combination form with organic matter and then are absorbed by endocytose in testine (Jalali, 2003). In usual studies metal transmittion by chloride cells from gill epithellium is the best found mechanisms in the early vertebrates (Saltes and Bailey, 2001). Many of this metals act in limited range and not exertion metals remain in body and always increase the amount of them in life-hood of organisms; this is called bioaccumulation (Farkos *et al.*, 2003). The accumulation of heavy metals has toxic effect on fish. Lead enters into the body with gill cells and especially is accumulated in gills; the latter aim organs are liver and muscle (Sadeghi-Rad, 1997). Standard level of lead for common carp is  $0.07 \text{ mg L}^{-1}$  and lethal amount of it is  $10\text{-}100 \text{ mg L}^{-1}$ . When blood lead concentration in human arrive to  $1000 \text{ } \mu\text{g L}^{-1}$  appear chromosomal disorders and it can damage brain children (Jalali, 2003). Although, zinc usually is accumulated in bone and skin, liver, gill and kidney are accumulated the great amount of them (Celik and Oehlenshlager, 2004). Standard level of zinc for common carp is  $0\text{-}0.05 \text{ mg L}^{-1}$  and lethal amount is  $0.5\text{-}1 \text{ mg L}^{-1}$  (Jalali, 2003). In human Zinc is one of the important matter in metabolic enzymes like anhydrase carbonic, otherwise it can bind with proteins as carrier or transition factor in human body so it is necessary for him; but high accumulation of zinc can damage skin, digestive system and nervous system (Pajoomand and Shariat, 1999). Many of the fish organs are sensitive to metal but liver is detoxification organ in fish body and muscle is eaten with other organisms like human (Jalali, 2003).

Sometimes in aim tissues one metal affects the other kind of metal that accumulates there so they can increase or decrease their toxic effect; of course lead and zinc don't have this effect to each other (Mansour and Sidky, 2003). In fish, toxic level of metals depend on the type of its compound (organic or inorganic), existence the other metals, temperature, dissolved oxygen, salinity, pH, feeding behavior, mating season, sex and biologic period of fish (zygote, larva, juvenile or mating fish) (Jalali, 2003).

Because of stability of metals, the organism can't protect itself with metabolism activities from toxic effects (Farkos *et al.*, 2003).

Some heavy metal is very harmful for creatures and aquatic ecosystems such rivers become pollutant so measuring and studying on level and progression of some hazard metals in ecosystem seem to be important.

In this study the amount of some heavy metals in Zayandeh rood measured and compared with the other information in last.

## **MATERIALS AND METHODS**

**Experienced animals:** All of the four species are belong on Cyprinidae family; original characteristics in this family are:

The number of them is high and diverse. They have spindle form of their body, single dorsal fin and none teeth on jaws but they have gully teeth. Their head is without scale and some of them don't have adipose fin. The type of their scale is cycloid. The pear-like points present on the

head and skin especially in male sex when they are breeding and spawning and then these points disappeared (Vosooghi and Mostagir, 2003). The species are:

***Carrassius auratus***: This fish is like with common carp. The ventral and lateral sides of its body are silverfish and caudal fin does not have black spots. The skin usually is black.

Black fish have stretch body and large sharp and U-form mouth and under lip is swelling and its edge is stip. These fish have one-two small bristle (Vosooghi and Mostagir, 2003).

***Cyprinus carpio***: Common carp have two small bristle that one of them is tall and the other one is short. Wild carp has body that is compacted form lateral side (Vosooghi and Mostagir, 2003).

**Sampling zone and time**: According to the necessity in this research five stations in Isfahan river (Zayandeh-rood) are determined and sampling from 2004 til 2005. These stations from the west to the east are:

- Zarinshahr in the North; there is Isfahan Iron Molting Factory construction unit
- After 20 km is Nekoabad that the factorial waste waters enter to this station and is determined for the amount of self refined power
- Falavarjan for self refined power
- Wahid bridge is in river entrance to the town and the home and city pollutant enters it
- Choom Bridge in down stream of the river and water velocity usually is low and has higher pollution probability

From these water stations sampling in 2003 February and 2004 June and 2002 July-August is done accidentally. Hunting fish way is waiting way. After biometry the fish was operated and after biological study on digestive system, their liver was taken (Jalali, 2003). The liver of cyprinidae is distribution in digestive fish tubule; that its color is pink and under the liver is spleen that it is darker from liver (Bozcaarmutlu and Arnic, 2004).

After separating the liver tissue of all fish was weighted and kept in coded polyethylene dishes. After dehydration and drying by oven; provided ashes from samples by electric furnace then each of them digested separately in a 1:3 ratio of HCl/HNO<sub>3</sub> combinant. (Stanley, 1993). Then injected the solutions in Atomic absorption set and measured the amount of lead and zinc by the comparison with standard curve (Table 1, 2).

For statistical accounting was used Excel software and for comparison among accumulation in different species and various stations used the SPSS software. For earning the equal groups used One-way ANOVA, Tukey and Man-Whitney. Of course the used confidence level in statistical analysis was 95% (Afshinnia, 2000).

Table 1: Lead (Pb) and zinc (Zn) levels in liver tissues of 5 species of Cyprinidae fish

Metal	Species				
	Amur (1)	Cyprinus (3)	<i>C. damascina</i> (24)	<i>C. aculeata</i> (42)	<i>Carrassius</i> (21)
Lead	3.63	1.30	3.63	3.65	2.40
SD	-	0.05	2.10	1.80	0.43
Zinc	138.60	170.40	138.10	154.70	138.20
SD	-	12.00	45.20	35.50	6.10

Table 2: Lead and zinc amounts in fish liver from 5 station of Zayandeh-rood river

Metal	Station				
	Choom (28)	Wahid (33)	Falavarjan (10)	Nekooabad (10)	Zarinshahr (10)*
Lead	4.67	2.3	3.82	4.61	2.81
SD	2.60	0.7	0.82	0.33	0.25
Zinc	166.50	104.7	174.10	179.30	107.90
SD	33.40	40.4	39.00	17.00	15.80

## RESULTS AND DISCUSSION

From 5 stations in the river (Zarinshahr, Nekooabad, Falavarjan, Wahid and Choom Bridge) was hunted 90 fish with special net. The species are: 21 of Carrassius (20 from Choom, 1 from Wahid) with the weight between 22-187 g. Forty two of aculeata (2 from Choom, 15 from Wahid, 5 from Falavarjan, 10 from Nekooabad, 10 from Zarinshahr) with the weight between 36-189 g. Twenty four of damascina (6 from choom, 13 from Wahid, 5 from Falavarjan) with the weight between 43-351 g. Three of common carp (all of them from Wahid) with the weight between 350-2550 g.

Table 1 show that the range amount of lead is 1.3-3.65 mg kg<sup>-1</sup> of dry fish weight otherwise it show standard deviation.

Lead level is highest in capoeta species and common carp has the lowest amount of it (Table 1).

Lead metal in Carassius has significant difference with aculeate and common carp, damascina with common carp and common carp with other three species are so (Table 1).

The range amount of zinc in fish liver are between 138.13-170.4 mg kg<sup>-1</sup> of dry weight of fish (Table 1).

Zinc level is high in all of the species but the common carp has the highest level of it (Table 1).

Zinc metal has no significant difference in every species. The most pollutant fish is capoeta that accumulates great amount of lead and zinc on this liver (Table 1).

The range of lead in liver of different station is 2.3-4.67 mg kg<sup>-1</sup> of dry weight (Table 2).

The fish of Choom station have the most level of lead metal and Wahid station has the lowest amount (Table 2).

Comparison among the metal concentration averages in different station shows that lead levels in Zarinshahr have significant difference with Nekooabad and Choom and Nekooabad with Zarinshahr and Wahid, Falavarjan with Wahid, Wahid with Nekooabad and Falavarjan and Choom are so with p<0.05 (Table 2).

Zinc levels are between 104.7-179.3 mg kg<sup>-1</sup> of dry weight (Table 2).

Nekooabad fish have the highest level of zinc in their liver tissues (Table 2).

The number of hunted fish and the average of humidity percentage of each specie are shown in Table 3 and 4.

Zinc levels in Zarinshahr with Nekooabad and Falavarjan and choom have significant difference and Nekooabad with Zarinshahr and Wahid, Falavarjan with Zarinshahr and Wahid, Wahid with other stations are so with p<0.05 (Table 2).

The liver tissues are burned and putted all of the water in them. According to the results *C. damascina* has the most amount of water and Amur has the lest amount of it.

The statistical analysis from Zn and Pb concentration is taken in Table 5.

Table 3: The number of hunted fish in each station. Because of happening hunting (Waiting hunting) the number of hunted fish from each station is different and it is undetectable

Humidity	Species				
	Amur	Cyprinus	<i>C. damascina</i>	<i>C. aculeata</i>	Carrassius
Zarinshahr	-	1	3	5	1
Nekooabad	-	2	2	4	2
Falavarjan	-	2	3	3	2
Wahid	1	3	15	13	1
Choom	-	-	2	6	20

Table 4: The average of humidity percentage in each species of all over the river

Humidity	Species				
	Amur	Cyprinus	<i>C. damascina</i>	<i>C. aculeata</i>	Carrassius
Average	66	69.7	82.4	80.2	81.00
SD	-	1.3	2.9	4.3	1.02

Table 5: Analysis variance of Pb and Zn concentration in the liver tissue of different species and stations

Metals	SS	F-value	P-value
<b>Pb</b>			
Species	25.64	3.41	0.022
Station	77.84	10.99	0.0
<b>Zn</b>			
Species	6093.98	1.74	0167
Station	41938.93	12.74	0.0

The data analyze (Confidence level 95%) in species are show: lead level have great difference with  $p < 0.05$  (Table 5) but zinc level don't have significant difference because of  $p > 0.05$  so the amount of lead depends on type of species but the zinc independent on it; and the amount of lead and zinc depends on location of station (Table 5).

The results were analyzed (Confidence level 95%) in different station and show: the zinc and lead level in liver tissue have significant difference because of  $p < 0.05$  in 95% of confidence level (Table 5).

In this research the metals level of lead and zinc in some edible fish of Zayandeh-Rood were measured in accidentally way. The results were analyzed in different species and show (Table 1) the zinc and lead level in liver tissue have significant difference because of  $p < 0.05$  in 95% of confidence level (Table 5) (Afshinnia, 2000).

The data analyze in species are show: lead level have great difference with  $p < 0.05$  but zinc level don't have significant difference because of  $p > 0.05$  (Table 5) so the amount of lead depends on type of species but the zinc independent on it and the amount of lead and zinc depends on location of station. These results support with Celik and Oehlenshlager (2004) results.

Lead metal in Carassius has significant difference with aculeata and common carp, damascina with common carp and common carp with other three species are so (Table 1).

Zinc metal has no significant difference in every species. The most pollutant fish is capoeta that accumulates great amount of lead and zinc on this liver (Table 1).

The amount of zinc is higher than lead (Table 1). In the years 1998-1999 the level of lead and zinc on Zyandeh-Rood were measured and the zinc level is higher than this research, by the way, it is higher than standard level. Although, the lead level shows medium increasing, it isn't as high as the zinc level and the lead level stays in standard level for Cyprinidae. Of course this accumulation is a warning results for healthy of society. these results support with Jalali (2003).

The zinc metal is the trace and necessary element in animal body so a little of it is necessary for respiratory and enzymatic activities. Great increasing in zinc level explains that many pollutant materials enter the river and lead and zinc enter the river from factories and industrial sewage so are the origin source of pollution (Jalali, 2003; Pajoomand and Shariat, 1999). Lead metal isn't a necessary metal (Sadeghi-Rad, 1997) and the high level of lead is because of entering pollutant in Zayandeh-Rood. High amount of heavy metal accumulation in fish liver have direct ratio with liver detoxification task, these results support with Henry *et al.* (2004) and Mansour and Sidky (2003) and report with Jalali (2003). Metalothyonin is one of the intercellular protein against with metal toxic effect in different organs but liver is the most important organ and this activities (Jalali, 2003). Otherwise because of increasing the metals level, consumption of metalothyonin can be one of the biological factors in metal toxification study (Saltes and Bailey, 2001).

Comparison among the metal concentration averages in different station shows (Table 2) that lead levels in Zarinshahr have significant difference with Nekooabad and Choom and Nekooabad with Zarinshahr and Wahid, Falavarjan with Wahid, Wahid with Nekooabad and Falavarjan and Choom are so with  $p < 0.05$  (Table 5).

Zinc levels in Zarinshahr with Nekooabad and Falavarjan and choom have significant difference and Nekooabad with Zarinshahr and Wahid, Falavarjan with Zarinshahr and Wahid, Wahid with other stations are so with  $p < 0.05$  (Table 5). The most pollutate region in all over the river is Nekooabad station in up stream. And this result is because of being in industrial sewages entrance place that this result accorded with Isfahan protection environment organization reports. These results support with Sadeghi-Rad (1997) that sampling in Iran; by the way sampling season of this station is the late of spring that has high temperature, So that hot season and hot water affect the level of heavy metals. This result support with Farkos *et al.* (2003). Choom station in down stream has great amounts of metal for material sedimentation process in entrance unit of the river that support with Jalali (2003) research.

So the amount of heavy metals depend on fish species and sampling area that these results support with Farkos *et al.* (2003) and Mansour and Sidky (2003).

In average zinc level is higher than lead (Table 1, 2) that support with Farkos *et al.* (2003) and Mansour and Sidky (2003). Although, high level of zinc metal use in metabolic active enzyme, it can affect circulatory and heart system of fish body badly and long-term accumulate of lead can damage nerve, blood and respiratory systems.

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