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Concentration of Some Heavy Metals in Tissues of Stellate Sturgeon (*Acipenser stellatus*) in the South Caspian Sea

Behrooz Abtahi*[1] , Mehdi Ghodrati Shojaii[2], Abbas Esmaili Sari[1]
Masoomeh Rahnema[3], Isa Sharif Pour[4], Mahmood Bahmni[5]
Rezvanallah Kazemi[5], Ali Hallajian[5]

1- Faculty of Natural Resources and Marine Sciences, Tarbiat Modares University
2- Persian Gulf and Oman Sea Ecology Institute
3- Ministry of Education &Culture
4- Iranian Fisheries Research Organization
5- International Research Institute of Sturgeon Fishes, Iran

Abstract
The concentration of five metals, including Pb, Cu, Cd, Ni and V, in the blood serum and tissues of Stellate sturgeon was evaluated in spring and autumn during 2001-2, using Shimatsu AA 670 G Flameless and Shimatsu AA G70 Flame, according to AOAC, 1990. The specimens were caught in two important sturgeon fishery zones located in Guilan and Golestan provinces in the Southern Caspian Sea. Concentrations of the tested elements followed the sequence $Cu>Pb>Cd>Ni,V$ in tissues, as well as $Cu>Ni>Pb>V>Cd$ in serum. There were significant differences in the levels of these elements in the blood serum and tissues of the Stellate sturgeon ($P<0.05$). In this study, the relationship between the concentration of the elements and biological characteristics has been examined. The significant length- and weight-dependent relationships were observed for Cd in blood serum. Only in the case of Pb in the liver were age related differences found. Only in the case of Ni in blood serum could significant differences between two selected sampling areas be detected. To our knowledge, this study provides the first extensive data of elemental accumulation in serum, liver and gill of Sturgeons of the Southern Caspian Sea.

*Keywords***:** stellate sturgeon, heavy metal, liver, gill, Caspian Sea.

غلظت برخی از فلزات سنگین در بافت‌های ماهی اوزن برون (*Acipenser stellatus*) صید شده در خزر جنوبی

بهروز ابطحی*[۱]، مهدی قدرتی شجاعی[۲]، عباس اسماعیلی ساری[۱]، معصومه رهنما[۳]، عیسی شریف‌پور[۴]، محمود بهمنی[۵]، رضوان‌ا... کاظمی[۵]، علی حلاجیان[۵]

۱- دانشکده علوم دریایی، دانشکده تربیت مدرس
۲- پژوهشکده اکولوژی خلیج فارس و دریای عمان، بندر عباس
۳- وزارت آموزش و پرو رش
۴- مؤسسه تحقیقات شیلات ایران، وزارت جهاد کشاورزی
۵- انستیتو تحقیقات بین‌المللی ماهیان خاویاری، رشت

چکیده
غلظت پنج فلز شامل: سرب (Pb)، مس (Cu)، کادمیوم (Cd)، نیکل (Ni) و وانادیوم (V) در بافتهای ماهی اوزن برون (*Acipenser stellatus*) صید شده در سال ۱۳۸۰ در سواحل جنوبی دریای خزر با دستگاه جذب اتمی کوره ای و شعله ای، طبق روش استاندارد AOAC مورد بررسی قرار گرفت. نمونه ها از مراکز مهم صید ماهیان خاویاری خزر جنوبی در استان های گیلان و گلستان در فصول بهار و پاییز جمع آوری شدند. نتایج حاصله نشان داد که تجمع فلزات در بافتهای کبد و آبشش از ترتیب مس> سرب> کادمیوم> نیکل و وانادیوم پیروی می کند، در حالیکه این روند در سرم ماهی ها بترتیب مس> نیکل> سرب> وانادیوم> و کادمیوم بود. اختلاف معنی داری در غلظت فلزات در بافتها و سرم ازون برون مشاهده گردید (p<۰/۰۵). بعلاوه در این تحقیق رابطه غلظت فلزات و شاخصهای زیست سنجی مورد توجه قرار گرفت، که در آن همبستگی مثبت و معنی داری بین طول و وزن ماهی و غلظت کادمیوم مشاهده شد (p<۰/۰۵). همبستگی غلظت فلز با سن فقط در مورد سرب کبد یافت شد. در بررسی تفاوت های منطقه ای تجمع فلزات سنگین، تنها در مورد نیکل تفاوت معنی داری بین دو ناحیه‌ی گیلان و گلستان مشاهده گردید (P<۰/۰۵). یافته های این تحقیق برای اولین بار بیانگر آلودگی های کبد، آبشش و سرم خون و ارتباط آنها با برخی ویژگیهای زیستی در ازون برون خزر جنوبی می باشد.

واژه های کلیدی: ازون برون، فلزات سنگین، کبد، آبشش، دریای خزر.

* Corresponding author. *E-mail Address:* Behroozabt@yahoo.com

Introduction

The Caspian Sea, bounded by the five littoral states of Azerbaijan, the Russian Federation, Iran, Kazakhstan and Turkmenistan, is the largest inland water body in the world (De Mora *et al*, 2004). During the past forty years, especially in the last decade, the levels of pollutants (including heavy metals, pesticides, petroleum hydrocarbons etc.) in the Caspian Sea have increased, subsequently the anthropogenic pressures on the coastal and marine ecosystems have grown progressively (Pourang *et al.*, 2003; Kajiwara *et al.*, 2003). Because it is a closed environment without any outlet, various environmental pollutants released from coastal catchment areas have accumulated in the Caspian Sea (Anan *et al.*, 2002) Furthermore, since there is no outlet, various pollutants arising out of fuel leakage from oil wells and wastewater from the river basin and urban area accumulated in the Caspian Sea (Karpinsky, 1992). Heavy metals are important for the ecology of the Caspian because they do not decompose, only change chemical bonds. Metals thus gradually accumulate in the sea, in sediments and in living marine organisms (DHI, 2001).

Metal levels in the water of Volga River, which contributes > 80% of the total water inflow to the Caspian Sea (Anan *et al.*, 2002), have increased from 1977 to 1992: Cu by 11.5 times, Zn by 9.8 times, Pb by 5.6 times, and Cd by 4.9 times (Karpinsky, 1992). Mainly four commercial species of sturgeon inhabit the Caspian Sea: the northern part of the Caspian Sea and Volga River are the habitats for the beluga (*Huso huso*), Russian sturgeon (*Acipenser gueldenstaedtii*) and Stellate sturgeon (*Acipenser stellatus*), while the southern part of the Caspian Sea is the habitat for the Persian sturgeon (*Acipenser persicus*) (Khodorevskaya *et al*, 1997). Catches of sturgeons in the Azov and Caspian Seas, which represented 90% of the world landings, totaling about 24,000–25,000 t annually during 1970–1985, have fallen to less than 2000 t in 1999 (Billard and Lecointre, 2001). This decline results from over-fishing and environmental degradation such as construction of dams across the river and contamination of water and sediment by pollutants, which are disrupting for the migration and reproduction of the sturgeons (Billard and Lecointre, 2001). Among these, chemical contamination seems to be one of the most significant factors influencing the population of sturgeons in the Caspian Sea. Furthermore some other studies also reported elevated levels of heavy metals in some fishes (Anan *et al.*, 2005), seals (Anan *et al.*, 2002) and sediment of Caspian Sea (De Mora *et al.*, 2004). However, a few studies in this field have been conducted on Sturgeons (Gapeova *et al.*, 1990; Pourang *et al.*, 2003; Agusa *et al.*, 2004). In the present study, we examined the accumulation of five trace elements in the blood serum, liver and gill of Stellate sturgeons in the Caspian Sea and discussed the results in relation to location, and growth.

Materials and Methods

The samples were collected from two sturgeon fishery zones in Guilan and Golestan provinces were located in southern part of the Caspian Sea (Figure 1) during 2001-2002. The total number of samples collected was 79. The biological characteristics including total length and weight were recorded. The age of specimens determined using pectoral fin sections. The biological data of specimens are given in Table 1.

Tissue samples in the weight of 20 gr and blood samples in the minimal valium of 10 ml obtained from fishes for measuring of metal concentrations. Tissues and sera after remove of hematocrite, were saved in a freezer until analysis at -20 t° C. The preparation of samples for instrumental analysis was done according to AOAC (1990). The concentrations of four trace elements (Pb, Cd, V, and Ni) were measured with flameless atomic absorption spectrometry (Shimatsu AA G70 G flameless). Cu concentration was determined by Flame atomic absorption spectrometry (FAAS) (Shimatsu AA G70 Flame). In this study, trace element concentration was expressed on the wet weight basis (µg/g wet wt.). All data were tested for goodness of fit with Kolmogorov–Smirnov's one sample test. Because most of the variables were not normally distributed, the data were log transformed and subjected to parametric statistics. Pearson's correlation coefficients were used to examine relationships between the elements as well as between elements and the biological characteristics. The concentrations of the elements in blood serum and tissues were compared statistically by means of one way ANOVA and Duncan's new multiple range tests. Regional differences in trace element concentration of sturgeons were tested by independent samples t-test. The level of significance was set at $P < 0.05$. These statistical analyses were performed using SPSS program.

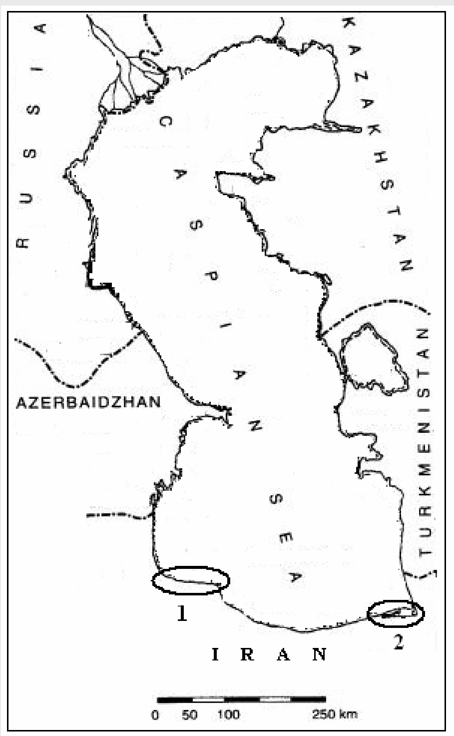

Figure 1- Map of the Caspian Sea showing area of sample collection. 1. Guilan catch area, 2.Golestan catch area

Results

The biometrical characteristics of specimens are shown in table 1.

The concentration of Cu, Cd were found to follow the order liver> gill> serum while the Pb level follows the sequence gill> liver> serum. In liver and gill of fish the average concentration of the tested elements follows the sequence Cu>Pb>Cd. Ni and V values in liver and gill in all the cases were less than the detection limits (<0.001± g/g wet weight). Thus, these data could not be considered in the statistical analyses. Concentrations of the tested elements followed the sequence Cu>Ni>Pb>V>Cd in serum.

The elements concentration in serum and tissues of sturgeons are shown in Table 2.

Discussion

In this study, a significant positive correlation between body length and weight was found ($P<0.01$). Growth of sturgeons is known to be continuous with age and growth rate dose not be reduce appreciably even after reproduction (Billard and Lecontre, 2001). The relationships between trace element levels in the tissues and the biological characteristics (especially length) have been documented by several investigators. The results varied between the studies. Sometimes even contradictory results have been obtained from different researches. For example, Canli and Atli (2003) reported that lead in the liver was negatively correlated to length of *Scomberesox saurus* specimens (Henry *et al.*, 2004), in contrast, reported no clear relationship between lead levels in liver of several fish species. Windom *et al* (1987) reported that copper in muscle was positively correlated to length of *Coryphaenoides armatus* specimens, but Canli and Atli (2003) observed a negative correlation between the size and the muscle copper content in *Atherina hepsetus*. There was positive correlation between the concentration of Cu in the muscle of *Acipenser persicus* from the Caspian Sea and body length, whereas a significant negative correlation was found in the case of Vanadium (Agusa *et al*, 2004). Pourang *et al* (2003) found that accumulation of Cd decreased with an increase in the weight of sturgeons. As mentioned before, in the present study a length and

Table 1- Some characteristics of Stellate sturgeons from South Caspian Sea

Location		Fork length (Cm)	Weight (Kg)	Age (year)
Guilan	Mean±SD	126.63±10.79	10.04±2.89	12.45±1.51
	Min	106	5.50	10
	Max	143	15	15
Golestan	Mean±SD	123.83±17.54	10.70±5.30	11.66±2.06
	Min	111	7.50	9
	Max	159	21.30	14

Table 2- Heavy metal concentrations (±g/g Wet Wt.) in Blood serum and tissues of Stellate sturgeons from Caspian Sea

(Mean ±SD, Min, Max)

Location	Tissue	Pb	Cu	Cd	Ni	V
Guilan	Serum	0.0066±0.0025 0.0023 0.0106	0.2275±0.1089 0.0880 0.4620	0.0015±0.0009 0.0008 0.0037	0.0922±0.0242 0.0698 0.1395	0.0263±0.0032 0.0234 0.0333
	Liver	0.4527±0.5873 N.D 2.125	18.2028±14.9805 N.D 66.50	0.3234±0.2457 N.D 1.250	*N.D	N.D
	Gill	0.6007±0.8966 N.D 4.750	1.0873±0.4652 0.125 2.875	0.1214±0.1376 N.D 0.375	N.D	N.D
Golestan	Serum	0.0058±0.0019 0.0040 0.0103	0.4130±0.1283 0.1660 0.6670	0.0018±0.0018 0.0002 0.0074	0.0502±0.0271 0.0184 0.0935	0.0284±0.0039 0.0245 0.0380
	Liver	0.6250±0.6175 N.D 2.50	24.41±17.09 1.50 67.50	0.5446±1.2168 N.D 6.50	N.D	N.D
	Gill	0.7317±0.4711 N.D 1.50	1.2625±0.6825 N.D 2.88	0.1750±0.2236 N.D 0.75	N.D	N.D

weight positive relationship was observed only for Cd in blood serum. The lack of any relationship between the other metals concentrations and the length may be explained in part due to the body capacity to regulate the concentrations of these metals and the fact that body size and biochemical factors associated have a small or null influence on variability (Paez-Osuna & Ruiz-Fernandez, 1995).

In the present study, an age dependent relationship was observed for Pb in liver. Pourang *et al* (2003) reported no clear relationship between the heavy metal levels in muscle of sturgeons and fish age. According to Rashed (2001) different trace elements can be classified into three groups from relationship between the concentrations in the muscle of *Tilapia nilotica* and the fish ages: (a) Cu showed increasing tendency with fish age, (b) Cr and Sr were independent of fish ages, (c) Co, Fe, Mn, Ni and Zn increased slightly with fish age. Except the temperature, salinity, dissolved oxygen, and pH all exhibit unique patterns of fluctuation, as their amount decrease from the west coast (Port of Astara) toward the east coast (Torkaman Port) (Anan, 1998). Regional differences in trace element concentrations of sturgeons might reflect the contamination in the respective areas. The difference between the specimens from the two sites in accumulation of Ni in blood serum can be attributed to the differences in ambient environmental conditions (especially salinity and temperature). Moreover, the possible differences in bioavailability of Ni between the two sites can be cited as another reason for the significant differences in the levels of Ni in the specimens collected from the sites. Pb and Cd concentrations were higher in *Rutilus rutilus caspicus* from eastern stations than those from western stations (Anan *et al*, 2005). This condition is not seen in stellate sturgeon. These inconsistencies might be due to the migration of sturgeons for feeding and reproduction in the Caspian Sea (Bemis and Kynard, 1997).

The relationships between some of the metal pairs may be attributable to similar physicochemical properties of the metals involved; also it has been regarded as indicative of similar biochemical pathways or, at its simplest, as demonstrating that the binding of certain metals in animals indicates the occurrence of particular ligands (Paez-Osuna *et al*., 1995). In the present study, there were no significant ($P>0.05$) correlation between element concentration pairs in blood serum and tissues. Hence, relatively few similar trends can be expected for them. Mean concentrations of heavy metal in the tissues and blood serum of *A. stellatus* do show variations. Statistical comparisons revealed that metal concentrations were significantly different in liver, gill and blood serum. The differences in metal concentration of the tissues and serum might be as a result of their capacity to induce metal binding proteins such as metallothioneins.

To understand the magnitude of heavy metal contamination in Stellate sturgeons of the Caspian Sea, the levels noticed in this study were compared to those reported for fish in other regions (Table 3). The present data showed that metal concentration of the liver and gill in the present study were higher than in fish caught from marine stations. To understand the magnitude of heavy metal contamination in sturgeons of the southern Caspian Sea, the levels noticed in this study were compared to those reported for fish species in this and other regions. There have been very few studies on metals in sturgeons. Gapeova *et al* (1990) examined the levels of heavy metals in four chosen tissues and blood serum of stellate sturgeon from the South Volga River. Comparison with the data obtained from our study with the mentioned research shows that in case of Cu in serum, Pb in liver and Cu and Cd in gill the results of the two studies are comparable while the levels of Pb and Cd in serum and Pb in the gills of fish from the southern Caspian Sea are well below the concentrations in the stellate sturgeons from the Volga River (Table. 3).

Based on the results, it can be concluded that:

- Concentrations of the tested elements followed the sequence Cu>Pb>Cd>Ni,V in tissues, and Cu>Ni>Pb>V>Cd in serum

- In the case of Cd, length and weight related differences (positive) could be observed in blood serum.
- An age dependent relationship was observed for Pb in liver.
- Concentrations of Ni in the blood serum showed significant variation between the selected sites ($P<0.05$).
- There were no significant correlation between element concentration pairs in serum and tissues ($P>0.05$).

Table 3- Comparison of Heavy metals concentrations in Serum and tissues of fish from various waters

References	Serum or Tissues	Species	V	Ni	Cd	Cu	Pb
		Caspian Sea					
This study	Serum	Stellate sturgeon	0.0481	0.139	0.007	0.387	0.006
This study	Liver	Stellate sturgeon	-	-	0.428	20.89	0.524
This study	Gill	Stellate sturgeon	-	-	0.144	1.159	0.655
		Caspian Sea					
Gapeova *et al*, 1990	Seum	Stellate sturgeon	-	0.21	0.06	0.54	0.55
Gapeova *et al*, 1990	Liver	Stellate sturgeon	-	0.63	0.18	7.52	0.68
Gapeova *et al*, 1990	Gill	Stellate sturgeon	-	0.50	0.11	0.87	1.09
		Caspian Sea					
Agusa *et a*l, 2004	Muscle	Stellate sturgeon	0.019	-	<0.001	1.50	0.013
		Caspian Sea					
Fazeli *et al*, 2005	Liver	*Liza aurata*	-	6.14	-	-	17.51
		Mediterranean Sea					
Canli and Atli, 2003	Gill	*Sparus auratus*	-	-	1.79	5.01	13.31
Canli and Atli, 2003	Gill	*Atherinahepsetsu*	-	-	1.85	14.64	12.37
Canli and Atli, 2003	Liver	*Mugil cephalus*	-	-	1.64	202.8	12.59
Canli and Atli, 2003	Liver	*Scomberesox saurus*	-	-	1.72	18.18	17.54
		North French Costal Waters					
Henry *et al*, 2004	Liver	*Limanda limanda*	-	-	0.13	16.8	0.04
Henry *et al*, 2004	Liver	*Pleuronectes platessa*	-	-	0.12	11.1	0.09

References

Agusa, T., T. Kunito, S. Tanabe, M. Pourkazemi, and D.G. Aubrey (2004). Concentrations of trace elements in muscle of sturgeons in the Caspian Sea. *Marine Pollution Bulletin*, 49: 789-800.

Al-Yousuf, M.H., M.S. El-Shahawi and S.M. Al-Ghais (2000). Trace metals in liver, skin and muscle of *Lethrinus lentjan* fish species in relation to body length and sex. *The Science of the Total Environment*, 256: 87-94.

Anan, Y. (1998). *Caspian Sea Environment National Report of I. R. Iran*. Tehran: Department of the Environment: 129p.

Anan, Y., T. Kunito, S. Tanabe, I. Mitrofanov, and D. Aubery (2005). Trace element accumulation in fishes collected from coastal waters of the Caspian Sea. *Marine pollution Bulletin* (Article in press).

Anan, Y., T. Kunito, T. Ikemoto, R. Kubota, I. Watanabe, S. Tanabe, N. Miyazaki, and E.A. Petrov (2002). Elevated concentrations of trace elements in Caspian seals (*Phoca caspica*) found stranded during the mass mortality events in 2000. *Archives of Environmental Contamination and Toxicology*, 42: 354-362.

AOAC (1990). Official method of analysis, Association of Analytical Chemistry. 15th edition, AOAC Inc. USA: 1298 p.

Bemis, W.E., and B. Kynard (1997). Sturgeon rivers: an introduction to Acipenseriform biogeography and life history. *Environmental Biology of Fishes*, 48: 167-183.

Billard, R., G. Lecointre (2001). Biology and conservation of sturgeon and paddle Fish. Reviews in Fish Biology and Fisheries, 10: 355-392.

Canli, M., and G. Atli (2003). The relationship between heavy metal (Cd, Cr, Cu, Fe, Pb, Zn) levels and the size of six Mediterranean fish species. *Environmental Pollution*, 121: 129-136.

De Mora, S., M.R. Sheikholeslami, E. Wyse, S. Azemard, and R. Cassi (2004). An assessment of metal contamination in coastal sediments of the Caspian Sea. *Marine Pollution Bulletin*, 48: 61-77.

DHI (2001). Bioresources, 37pp; http://www.dhi.dk/News/TacisCaspianSea/ TDA_3_ Bioresources & Biodiversity. pdf file

Fazeli, M., B. Abtahi, and A. Sabbagh Kashani (2005). Determination of Pb, Ni and Zn Accumulation in the Tissues of *Lizza aurata* in the South Caspian Sea. (in Persian) *Iranian Journal of Fisheries Sciences*, V.14,1: 65-78.

Gapeova, A.L., M.A. Tselmovich and V. Shirakov (1990). Heavy metals in sturgeons of Volga River down stream, Research Report, GosNIORKh. (In Russian)

Henry, F., R. Amara, L. Courcot, D. Lacouture, and M. Bertho (2004). Heavy metals in four fish species from the French coast of the Eastern English Channel and Southern Bight of the North Sea. *Environmental International*, Vol 30: 675-683.

Kajiwara, N., D. Ueno, I. Monirith, S. Tanabe, M. Pourkazemi, and D.G. Aubrey (2003). Contamination by organochlorine compounds in sturgeons from Caspian Sea during 2001 and 2002. *Marine Pollution Bulletin*, 46: 741-747.

Karpinsky, M.G. (1992). Aspects of the Caspian Sea benthic ecosystems. *Marine Pollution Bulletin*, 24: 384-389.

Khodorevskaya, R. P., O. L. Zhuravleva and A.D. Vlasenko (1997). Present status of commercial stocks of sturgeons in the Caspian Sea basin. *Environmental Biology of Fishes*, 48: 209-219.

Paez-Osuna, F. and C. Ruiz-Fernandez (1995). Trace metals in the Mexican shrimp *Penaeus vannamei* from estuarine and marine environments. *Environmental Pollution*, 87: 243-247.

Paez-Osuna, F.R., G. Perez-Gonzalez, H.M. Izaguirre-Fierro, Zaazueta-Padilla and L.M. Flores-Campana (1995). Trace metal concentrations and their distribution in the lobster *Panulirus inflatus* (Bouvier, 1895) from the Mexican pacific coast. *Environmental Pollution*, 90(2): 163-70.

Pourang, N., S. Tanabe, S. Rezvani and J.H. Dennis (2003). Trace elements accumulation in edible tissues of five sturgeon species from the Caspian Sea. Environmental Monitoring and assessment, Vol. 100, No1-3: 89-108.

Rashed, M.N. (2001). Monitoring of environmental heavy metals in fish from Nasser Lake. *Environmental International*, 27: 27-33.

Windom, H., D. Stein, R. Sheldon and J.R. Smith (1987). Comparison of trace metal concentrations in fish muscle tissue of a benthopelagic fish (*Coryphaenoides armatus*) from the Atlantic and Pacific Oceans. *Deep Sea Research*, 34: 213-220.