Pakistan J. Zool., vol. 48(2), pp. 589-595, 2016.

Heavy Metal Concentration in *Sardinella longiceps* (Valeciennes, 1847) from Balochistan Coast

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Article Information

Received 13 January 2015
Revised 15 April 2015
Accepted 31 October 2015
Available online 1 March 2016

Authors' Contributions

QA and NE collected and analyzed samples. SB wrote the article.

Key words

Heavy metals, *Sardinella longiceps*.

A B S T R A C T

In this study, manganese (Mn) zinc (Zn), copper (Cu), cadmium (Cd) and lead (Pb) concentrations were determined in muscle and liver of Sind sardinella (*Sardinella longiceps*) collected from Balochistan coast, Pakistan, during autumn inter monsoon, north east monsoon, spring inter monsoon and south west monsoon during October 2005 – September 2006. Fish showed accumulation of Mn (1.73±0.96, 4.5±2.47 µg g^{-1}), Zn (3.10±2.11, 23.82±9.77 µg g^{-1}), Cu (3.18±1.50, 26.08±13.81 µg g^{-1}), Cd (0.17±1.88, 0.91±0.61 µg g^{-1}) and Pb (0.25±1.77, 1.33±0.53 µg g^{-1}) in muscle, and liver respectively. Mn was above the permissible limits.

INTRODUCTION

Fish has been considered good indicators for heavy metal contamination in aquatic systems because they occupy elevated trophic levels with different sizes and ages (Burger *et al.,* 2002). Meanwhile, fish are widely consumed in many parts of the world by humans, and polluted fish may endanger human health. The levels of toxic elements in fish are related to age, sex, season and habitat (Kagi and Schaffer, 1998). The ingestion of food is an obvious means of exposure to metals, not only because many metals are natural components of food stuffs, but also environmental contamination and contamination during processing (Voegborlo *et al.*, 1999). The distribution of metals varies between fish species, depending on age, development status and other physiological factors (Kagi and Schaffer, 1998). Industrial and agricultural activities have been reported to be the leading potential source of the accumulation of pollutants in the aquatic environment including the sea (Tarra-Wahlberg *et al*., 2001; Akif *et al*., 2002).

Fishes form an excellent source of digestible proteins, vitamins, minerals and polyunsaturated fatty acids (Daviglus *et al.*, 2002) but are also the source of heavy metals. Some of the metals found in the fish might be fundamental as they play vital role in biological system of the fish as well as in human beings but some of them may however, be toxic and may cause serious damage to the human health if present in excess to the permitted limits. The common heavy metals that are found in fish may include Cu, Fe, Zn, Ni, Mn, Hg, Pb, Cd, etc. from Pakistan waters or elsewhere (Connell, 1984, Rizvi *et al.,* 1988; Tariq *et al.,* 1991, 1998; Nair *et al.*, 1997; Zahra *et al.*, 2003; Agusa et al., 2005, 2007; Dalman *et al.*, 2006; Naidu *et al.*, 2008; Tabinda *et al.*, 2010; Shanthi and Ramanibal, 2011; Kumar *et al.*, 2012; Shivakumar *et al.*, 2014; El-Moselhy *et al.*, 2014; Al-Ghanim *et al.*, 2015). Heavy metals have the tendency to accumulate in various organs of marine organisms, especially fish, which in turn may enter into the human metabolism through consumption causing serious health hazards (Puel *et al*., 1987). Iron, copper, zinc and manganese are essential (physiological) metals.

The marine organisms accumulate contaminants such as metals from the environment and have been extensively used in marine pollution monitoring programmes (Linde *et al.*, 1998; Mora *et al.*, 2004). These metals accumulate in fish from water, food, sediment and some suspended particulate materials (Agusa *et al.*, 2005). In many countries, industrial wastes, geochemical structure and mining of metals create a potential source of heavy metals pollution in the aquatic environment due to their toxicity and accumulation behaviour. Under certain environmental conditions, these heavy metals might accumulate up to a toxic concentration and cause ecological damage (Sivaperumal *et al.*, 2007).

The objective of this study was to determine heavy metal (Mn, Zn, Cu, Cd and Pb) concentration in *Sardinella longiceps* (Valeciennes, 1847) fish during October 2005-September 2006.

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0030-9923/2016/0002-0589 $ 8.00/0

MATERIALS AND METHODS

Sardinella longiceps (Valeciennes, 1847) fish samples were collected during seasons (autumn inter monsoon, north east monsoon, spring inter monsoon and south west monsoon) from Balochistan coast. Fish samples were immediately transported to the laboratory and rinsed in distilled water to remove foreign particles. Fish length (cm) and weight (g) were measured. Fish were tagged for identification and then freeze until time for analysis. After biometric measurements, Approximately 2g of the epaxial muscle on the dorsal surface of the fish from each sample were dissected washed with distilled water, dried in filter paper, weighed, packed in polyethylene bags and kept at -20°C until analysis. AAnalyst 700 Atomic Absorption Spectrophotometer was used in present study in Centralized Science laboratory, University of Karachi. The absorption wavelengths (λ) used for the determination of various metals are as follows: Mn, 279.50 nm; Zn, 213.90 nm; Cu, 324.70 nm; Cd, 228.80 nm and Pb, 217.00 nm. Due to the lack of a reference standard material, accuracy of the analysis and the effect of the matrices in the media were controlled with the standard addition method. All studied elements were tested with standard addition method for 3 randomly selected samples. The samples taken from the muscles tissues were dried first and cut into pieces as small as possible. 3-20 mg portions were taken from the dried samples, placed into teflon cylindrical vessels and digested with 3 mL of H_2O_2/HNO_3 (1:2 v/v) at 250ºC. The organic part was discarded and the remaining part was diluted with demineralized water to 50 mL in a graduated flask (Bernhard, 1976).

For all heavy concentrations in *Sardinella longiceps* (Valeciennes, 1847), within muscle tissues among seasons were determined by analyses of variance Mann Witney U with the Tukey's HDS post-hoc comparisons. The results were evaluated on the basic of homogenous groups at a given significance level ($p<0.05$). The elements which were common in the muscle tissue of *Sardinella longiceps* are assessed by means of Pearson's correlation coefficients. Data collection and statistical calculations were performed by SPSS Ver.18 software.

RESULTS

Length and weight (min-max) of the fish was 13–22 cm and 40 - 92 g. The according to the seasons metal (Mn, Zn, Cu, Zn, Cd and Pb) contents of *Sardinella longiceps* are given in Table I and Figure 1.

In this study, the highest Mn contents were 3.64 μg g^{-1} in muscle; the lowest was 0.16 μg g^{-1} in muscle while

Table I.- According to seasons, heavy metal concentrations in *Sardinella longiceps* (Valeciennes, 1847) (μg g^{-1}).

	Mn		Zn		Cu		Cd		Pb	
	Range	Mean±SEM	Range	Mean±SEM	Range	Mean±SEM	Range	Mean±SEM	Range	Mean±SEM
Muscle										
Autumn inter monsoon	0.44-2.74	1.49±0.95	0.78-1.88	1.39±0.34	1.06-4.36	2.67±1.40	0.01-0.16	0.07±0.52	0.01-0.80	0.18±0.25
North east monsoon	0.18-2.46	1.26±0.83	1.42-3.98	2.61±0.87	1.02-5.26	2.87±1.31	0.01-0.52	0.25±0.16	0.15-0.54	0.33±0.12
Spring inter monsoon	0.16-2.87	1.58±0.96	1.21-7.61	3.26±2.16	2.29-7.34	3.89±1.37	0.02-0.69	0.25±0.25	0.04-0.41	0.15±0.11
South west monsoon	1.62-3.64	1.59±0.59	1.56-9.26	5.14±2.37	1.05-6.16	3.28±1.81	0.01-0.51	0.13±0.18	0.18-0.52	0.35±0.13
All Seasons	0.16-3.64	1.73±0.96	0.78-9.26	3.10±2.11	1.02-7.34	3.18±0.50	0.01-0.69	0.17±0.19	0.01-0.80	0.25±0.18
Liver										
Autumn inter monsoon		6.56±2.25		21.49±9.39		16.20±5.84		0.58±0.23		1.14±0.46
North east monsoon	1.14-7.62	2.77±2.11	13.64-29.87	21.74±6.28	13.95-67.31	27.47±10.38	0.23-1.78	0.73±0.50	0.24-1.94	1.02±0.63
Spring inter monsoon	2.41-8.77	4.68±2.29	11.60-37.84	22.60±7.41	10.92-68.46	30.90±16.04	0.34-2.98	1.12±0.78	1.17-2.73	1.67±0.46
South west monsoon	2.22-7.32	3.99±1.78	18.01-64.11	29.45±13.54	12.99-46.51	29.75±10.73	0.44-2.23	1.19±0.66	1.12-2.03	1.47±0.32
All Seasons	1.14-9.74	4.50±2.47	10.75-64.15	23.82±9.77	9.63-68.46	26.08±13.81	0.23-2.98	0.91±0.61	0.24-2.73	1.33±0.53

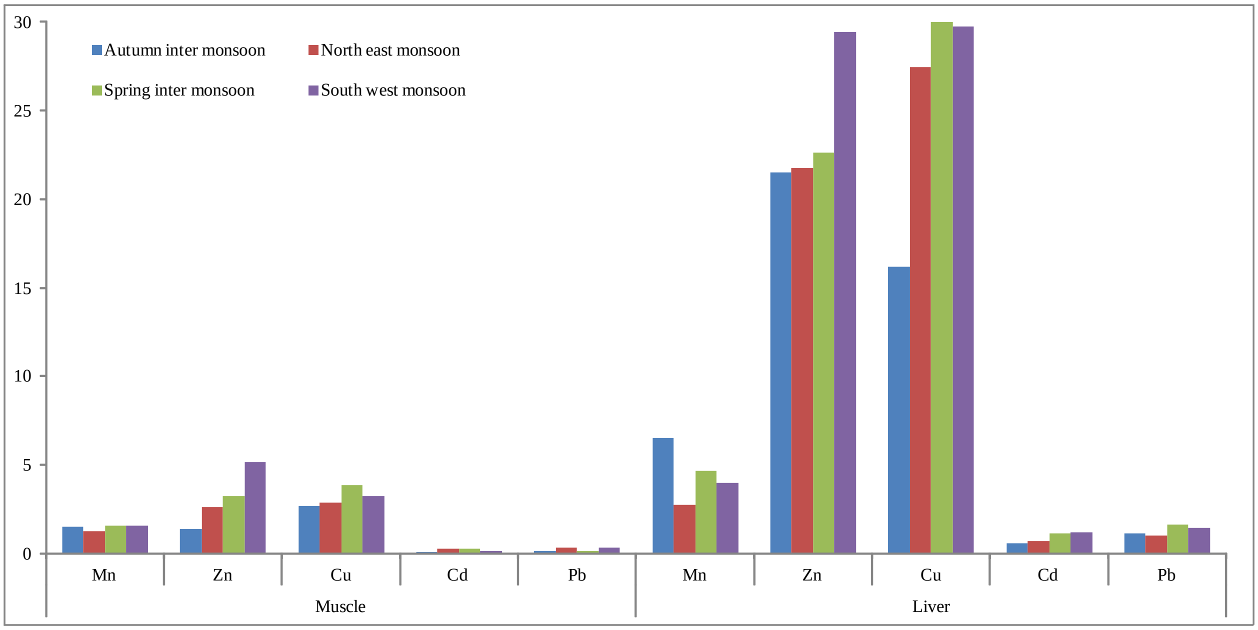

Fig. 1. According to seasons, heavy metal concentrations in *Sardinella longiceps* (Valeciennes, 1847) (µg g^{-1}).

Table II.- Comparison of concentration in fish tissues.

Locations/Limits	Fish	Metal and concentrations (µg g^{-1}) Mn	Zn	Cu	Cd	Pb	Studies
Port Blair (M)	*S. gibbosa*	0.70	45.6	0.00	0.00	0.0	Kaladharan *et al*., 2006
Kochi (M)		0.00	31.8	11.30	0.00	0.9	
Indian (M)	*R. kanagurta*	-	24.40	2.75	3.11	0.34	Rejomon *et al*., 2010
Keti Bunder Thatta (M)	*S. sindensis*	-	1.22	0.01	0.03	0.20	Tabinda *et al*., 2010
Kalpakkam (M)	*S. longiceps*	2.90	23.69	3.21	-	0.91	Biswas *et al*., 2012
Indian (M)	*R. kanagurta*	0.90	18.3	1.90	0.62	-	Bhupander *et al*., 2012
Persion Gulf							
(M)	*S. sindensis*	0.96	-	1.43	0.03	0.184	Khoshnood *et al*., 2012
(L)		-	-	0.85	0.18	0.87	
Bay of Bengal (M)	*R. kanagurta*	0.80	16	2.20	-	-	Bhupander *et al*., 2013
Kapar (M)	*A. thalasssinus*	0.62	20.54	1.21	0.06	-	Bashir *et al*., 2013
	J. belangeri	0.54	18.27	0.66	0.06	-	
Kapar (L)	*A. thalasssinus*	10.45	290.8	33.44	0.30	-	
	J. belangeri	9.09	51.55	7.87	0.58	-	
Mersing (M)	*A. thalasssinus*	0.92	30.21	1.56	0.03	-	
	J. belangeri	0.97	13.12	0.95	0.04	-	
Mersing (L)	*A. thalasssinus*	6.21	409.6	55.87	13.35	-	
	J. belangeri	5.69	56.30	13.50	0.47	-	
Kalpakkam region St 1 (M)	*R. kanagurta*	-	13.76	-	0.87	1.85	Saravanamurugan *et al*.,
Kalpakkam region St 2 (M)		-	22.87	-	1.12	2.97	2013
Balochistan							
(M)	*S. longiceps*	1.73	3.10	3.18	0.17	0.25	This study
(L)		4.50	23.82	26.08	0.91	1.33	
International Limits		-	40	10-100	0.50		FAO 1983
		1.00	50		1.00		WHO 1989

M, muscle; L, liver.

the highest Zn contents 9.26 µg g^{-1} in muscle; the lowest was 0.78 µg g^{-1} in muscle. It was found that the highest Cu contents 7.34 µg g^{-1} in muscle; the lowest was 1.02 µg g^{-1} in muscle while the highest Cd contents 0.69 µg g^{-1} in muscle; the lowest was 0.1 µg g^{-1} in muscle. It was found that the highest Pb contents 0.80 µg g^{-1} in muscle; the lowest was 0.1 µg g^{-1} in muscle. The highest Mn contents were 9.74 µg g^{-1} in liver; the lowest was 1.14 µg g^{-1} in liver while the highest Zn contents 64.15 µg g^{-1} in liver; the lowest was 10.75 µg g^{-1} in liver. It was found that the highest Cu contents 68.46 µg g^{-1} in liver; the lowest was 9.63 µg g^{-1} in liver while the highest Cd contents 2.98 µg g^{-1} in liver; the lowest was 0.23 µg g^{-1} in liver. It was found that the highest Pb contents 2.73 µg g^{-1} in liver; the lowest was 0.24 µg g^{-1} in liver (Table I).

Accumulation of metals in muscle was observed to follow the order of Cu > Zn > Mn > Pb >Cd. Respectively accumulation of metals in muscles at autumn inter monsoon, north east monsoon, spring inter monsoon and south west monsoon was observed to follow the order of Cu > Mn > Zn >Cd > Pb; Cu > Zn > Mn > Pb>Cd; Cu > Zn > Mn > Cd > Pb; Zn > Cu > Mn > Pb > Cd (Table I). Accumulation of metals in liver was observed to follow the order of Cu > Zn > Mn > Pb >Cd. Respectively accumulation of metals in muscles at autumn inter monsoon and other seasons (north east monsoon, spring inter monsoon and south west monsoon) was observed to follow the order of Zn > Cu > Mn > Cd > Pb and Cu > Zn > Mn > Pb > Cd (Table I).

Accumulation of Mn metals in liver was observed to follow the order of autumn inter monsoon, spring inter monsoon south west monsoon, north east monsoon. Accumulation of Zn and Cd metals in liver was observed to follow the order of south west monsoon, spring inter monsoon, north east monsoon and autumn inter monsoon. Accumulation of Cu metals in liver was observed to follow the order of spring inter monsoon, south west monsoon, north east monsoon and autumn inter monsoon. Accumulation of Pb metals in liver was observed to follow the order of spring inter monsoon, south west monsoon autumn inter monsoon and north east monsoon. Accumulation of Mn metals in muscle was observed to follow the order of south west monsoon, spring inter monsoon, autumn inter monsoon and north east monsoon. Accumulation of Zn metals in muscles was observed to follow the order of south west monsoon, spring inter monsoon, north east monsoon and autumn inter monsoon. Accumulation of Cu metals in muscle was observed to follow the order of spring inter monsoon, south west monsoon, north east monsoon and autumn inter monsoon. Accumulation of Cd metals in muscle was observed to follow the order of spring inter monsoon, north east monsoon, south west monsoon and autumn inter monsoon (Table I).

Mn values were reported as 1.14 - 9.74 µg g^{-1}, Zn values were reported 10.75 - 64.15 µg g^{-1}, Cu values were reported 9.63 - 68.46 µg g^{-1}, Cd values were reported 0.23 - 2.98 µg g^{-1}, Pb values were reported 0.24 – 2.73µg g^{-1} respectively at liver. Mn values were reported as 0.16 - 3.64 µg g^{-1}, Zn values were reported 0.78 - 9.26 µg g^{-1}, Cu values were reported 1.02 - 7.34 µg g^{-1}, Cd values were reported 0.1- 0.69 µg g^{-1}, Pb values were reported 0.1– 0.80 µg g^{-1} respectively at muscle (Table I).

DISCUSSION

Mn accumulation of muscle is higher than literature (Kaladharan *et al.*, 2006; Bhupander *et al.*, 2012; Khoshnood *et al.*, 2012; Bashir *et al.*, 2013; Bhupander *et al.*, 2013) and lower than reported data from literature (Biswas *et al.*, 2012). Zn accumulation of muscle is higher than literature (Tabinda *et al.*, 2010; Biswas *et al.*, 2012; Bhupander *et al.*, 2012, 2013; Bashir *et al.*, 2013; Saravanamurugan *et al.*, 2013) and lower than reported data from literature (Kaladharan *et al.*, 2006; Rejomon *et al.*, 2010). Cu accumulation of muscle is higher than literature (Rejomon *et al.*, 2010; Tabinda *et al.*, 2010; Bhupander *et al.*, 2012; Khoshnood *et al.*, 2012; Bashir *et al.*, 2013). Cd accumulation of muscle is higher than literature (Kaladharan *et al.*, 2006; Tabinda *et al.*, 2010; Khoshnood *et al.*, 2012; Bhupander *et al.*, 2013; Bashir *et al.*, 2013) and lower than reported data from literature (Rejomon *et al.*, 2010; Bhupander *et al.*, 2012; Saravanamurugan *et al.*, 2013). Pb accumulation of muscle is higher than literature (Tabinda *et al.*, 2010) and lower than reported data from literature (Rejomon *et al.*, 2010; Biswas *et al.*, 2012; Saravanamurugan *et al.*, 2013) (Table II).

Mn and Zn accumulation of liver are lower than Bashir *et al.* (2013) while Cu accumulation of liver is higher than Khoshnood *et al.* (2012). Pb accumulation of liver is lower than all literature while Cd accumulation of liver is higher than all literature (Table II). There is difference ($p < 0.05$) according to the seasons in terms of Mn (north east monsoon - south west monsoon), Zn (autumn inter monsoon - north east monsoon; north east monsoon - south west monsoon) and Pb (autumn inter monsoon - south east monsoon; north east monsoon - spring inter monsoon; spring inter monsoon - south west monsoon) metal accumulation in the muscle. There is difference ($p < 0.05$) according to the seasons in terms of Mn (autumn inter monsoon - north east monsoon), Cd and Cu (autumn inter monsoon - south west monsoon) and Pb (north east monsoon - spring inter monsoon) metal accumulation in the liver.

In the present study, Table III showed that there is only high correlation between Pb **-** Cu of muscle. There is a peak correlation between Zn – Mn; Cu – Mn; Pb – Mn (Table III).

Table III.- Pearson correlation coefficients between metal concentrations in the muscle tissue of *Sardinella longiceps.*

Metal	Mn	Zn	Cu	Cd	Pb
Mn	1.00				
Zn	0.44**	1.00			
Cu	0.44**	0.66**	1.00		
Cd	0.50**	0.59**	0.64**	1.00	
Pb	0.48**	0.68**	0.70**	0.52**	1.00

** $p < 0.01$; indicate significant levels.

To evaluate the health risk to Pakistan people through consumption of marine fish, daily intake of heavy metals was estimated on the basis of the concentrations of Mn, Zn, Cu, Cd and Pb in muscle of daily fish consumption. The average daily fish consumption in Pakistan is 33 g per person (Chughtai and Mahmood, 2012). The heavy metal accumulation in muscles of *Sardinella longiceps* was found to be below the nationally and internationally stipulated values and do not pose a serious health risk for Pakistan (Table IV). Cd accumulation was found to close nationally and internationally stipulated values. Therefore, Cd accumulations in *Sardinella longiceps* have to be monitored continuously especially in these regions.

The results of this study show that Zn, Cu and Cd accumulations of *Sardinella longiceps*, caught from Balochistan coast were generally below the international limits while Mn accumulations of *Sardinella longiceps* was above the international limits (FAO, 1983; WHO, 1989). Therefore, Mn bioaccumulations in *Sardinella longiceps* have to be monitored continuously especially in these regions. The present study shows that precautions are needed to be taken in order to obviate the metal pollution in future. Otherwise, these pollutions can be hazardous for fish and human health.

ACKNOWLEDGMENTS

The author would like to thank the editor and anonymous reviewers for their valuable comments and suggestions to improve the quality of the paper.

Table IV.- The estimated daily and weekly intakes for the economically important fish species consumed by adult people in Pakistan

Metal	PTWI	PTWI[*b]	PTDI[c]	*Sardinella longiceps* EWI[d] (EDI)[e]
Mn	980[a]	58800	8400	57.09 (8.16)
Zn	7000[a]	420000	60000	102.3 (14.61)
Cu	3500[a]	210000	30000	104.94 (14.99)
Cd	7[a]	420	60.00	5.61(0.80)
Pb	25[a]	1500	214.29	8.25 (1.18)

[*] Provisional Permissible Tolerable Weekly Intake (PTWI) in l g/week/kg body weight.
Mean weekly fish consumption in Pakistan is 33 g per person (Chughtai and Mahmood, 2012)
[a] (FAO/WHO, 2004).
[b] PTWI for 60 kg adult person (lg/week/60 kg body weight).
[c] PTDI, permissible tolerable daily intake (lg/day/60 kg body weight).
[d] EWI, estimated weekly intake in lg/week/60 kg body weight.
[e] EDI, estimated daily intake in lg/day/60 kg body weight.

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