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Heavy Metals Contamination in Sediment and Sole Fish (*Euryglossa orientalis*) from Musa Estuary (Persian Gulf)

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Abstract: Musa estuary is an industrialized area, which receive different types of contaminants. Heavy metals concentration in sediment and fish collected from five different creeks along Khor-Musa were investigated to determine the level of heavy metals in the area. The maximum concentration of the metals in sediment ranged as fallows: Cd 0.55, Co 23.55, Cu 18.7, Ni 68.86 and Pb 4.7 and in fish ranked as: Cd 6.66, Co 1.59, Cu 350, Ni 3.5 and Pb 4.11 dry weight respectively. The metal concentrations in the edible part of fish were under the permissible limits proposed by FAO, WHO, MAFF, NHMRC and TEG.

Key words: Khor-Musa · Khor-Jafari · Petrochemical units · Imam port

INTRODUCTION

Heavy metals are a common group of marine pollutants, which is not biologically degradable. In seawater, heavy metals appear in two different forms including dissolved cations and metals bound to suspended organic materials. The second form always deposit in the sediment, so that sediment is known as the final destination for discharged metals into the seawater [1]. All forms of heavy metals in aquatic ecosystem may enter aquatic food chains and accumulate in various types of organisms [2-4] such as marine macrophytes [5, 6], invertebrates [7] and fishes [8, 9]. Accumulated metals in aquatic organisms could be potentially harmful to them [10]. However, the worst impacts of these elements appear when they transfer to the human body through food chains or trophic relations [11].

Khor-Musa is a complex waterway system located in the north Persian Gulf that is consisted of several estuaries, creeks and a main canal. This Khor is situated closed to the Imam port one of the biggest ports in Iran and connects this port to the Persian Gulf through a 60 Km length canal. There are several sources of anthropogenic pollutants including petrochemical industries, oil transportations and agriculture activities, which produce and release substantial amount of contaminant such as heavy metal into the seawater [12]. Like many other estuaries, Khor-Musa is an important place for fisheries and aquaculture activities. Considerable amount of fish and shrimp are caught from this Khor annually, which are introduced to the markets for human consumption. The presence of high concentrations of heavy metals in aquatic environment could result in metal accumulation by marine organisms [13, 14] and increase the risk of metal toxicity in the people who consume contaminated seafood. Information on heavy metals concentration in sediment and organisms such as fish is therefore useful to estimate the level of metals contamination and bioavailability in marine environment [15]. So far, such valuable information about this region has not been published. This study aimed to determine the level of heavy metals in the sediment and fish from Khor-Musa in order to have an estimate of heavy metal pollution in the area.

MATERIALS AND METHODS

Samples of fish and sediment were taken from five different stations including Khor_Gazaleh, Khor_Ahmadi, Khor_Jafari, Khor_Zangi and Khor_Ghanam. Each Khor is a branch of Musa estuary (Fig 1). Sediment samples were taken from three parts of each khor including upper, middle and lower parts. Three sediment samples were taken from each part (9 samples from each khor) using stainless steel Van Veen Grab. Fish sampling was performed by trawling all over the Khors and About 25 to

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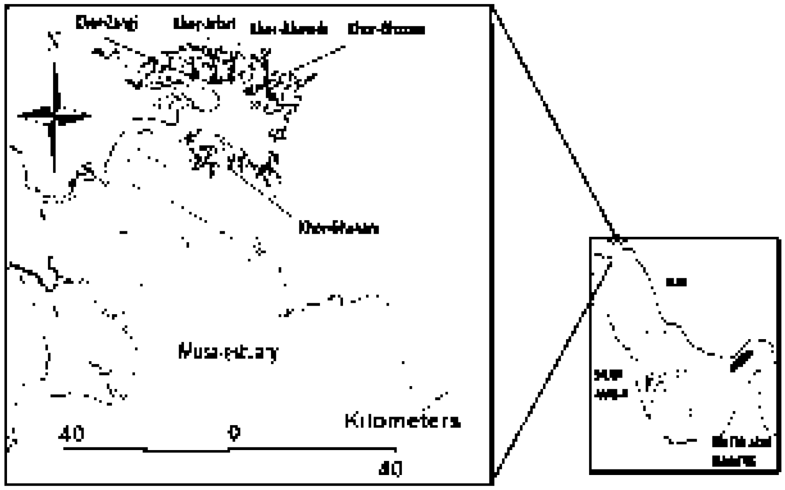

Fig. 1: A map showing of Khor-musa

Table 1: The operation condition of The AAs

Metals	Wavelengh (nm)	Silt Width (nm)	Lamp Current (MA)	Gas	Support
Cd	228.8	0.05	3	Acetylene	Air
Co	240.7	0.2	6	Acetylene	Air
Cu	324.7	0.5	3	Acetylene	Air
Ni	232	0.2	4	Acetylene	Air
Pb	217	1	5	Acetylene	Air

Table 2: Concentration of heavy metals observed in dogfish muscle certified reference material Dorm-2 and Dorm-3 (National Research Council, Canada)

	Observed values	Certified values
Dorm-2 (muscle)		
Cd	0.04	0.043
Co	0.208	0.182
Cu	2.61	2.34
Ni	17.63	19.4
Pb	0.071	0.065

Each value obtained by the average of 5 detection (µg/g d. w)

30 fishes of the same size (19 to 21 cm total length) were collected from each station. The samples were transferred to the laboratory immediately using icebox. They were deep-frozen at-20 °C until analysis. To determine the metals concentration in the samples, they were thawed in the room temperature firstly. Sediment samples were dried in the oven at 105 °C for 24 hour. Fishes were dissected, their muscle and liver tissues were obtained and oven dried in 80 °C for 24 hour until constant weigh was obtained. Ten muscle samples from each station were obtained for metal analysis. Additionally sufficient amount of fish liver tissues were pooled together to obtain 10 samples. The sediment samples were digested in a mixture of nitric and perchloric acids in the ratio of 4:1. Muscle and liver samples were also digested in concentrated nitric acid with the same procedure followed Ismail and Safahieh [16]. After digestion of samples, they were made to certain volume and filtered by 42-µm filter paper. Heavy metal analysis performed using flame Atomic absorption spectro meter (model Savant AA?). The operation conditions of the AAs are presented in table 1. All chemical reagents were analytical reagent grade (Merk, Germany). The glassware and plastic containers were soaked in nitric acid solution (10%) for 24 hour and washed with double distilled water before use. Instrument calibration was performed using calibration solution prepared from 1000 mg/l stock solutions. Blank samples were digested and analysed in the same way in order to avoid samples contamination. These blank samples were also use to auto zero AAs. To check the accuracy of method used for heavy metal analysis, CRM (Certified Reference Material) Dorm 2 were analyzed and the results are presented in table 2. The agreement between actual metal concentration in Dorm 2 and metal levels analysed in the laboratory was 93%, 114%, 111%, 109% and 90% for Cd, Co, Cu, Ni and Pb respectively.

All data were subjected to Shapiro-wilk test in order to check normal distribution. In order to compare heavy metals concentration in the sediment or fish samples collected from different stations, one way analysis of variance was used. If significant difference was observed, the data were submitted to tukey post hoc test for separating of different groups. Comparison of heavy metals accumulation in muscle and liver tissues was performed using t-test. Correlation and regression analysis were used to determine relationships between metals level in fishes and sediment.

RESULTS AND DISCUSSION

Heavy Metals in Sediment: Heavy metals level (expressed as µg/g d. w) in the analyzed sediment samples are listed in table 3. The highest levels of Cd (0.55 µg/g), Co (23.55 µg/g) and Ni (68.86 µg/g) were found in Khor-Jafari while the highest levels of Cu (18.7 µg/g) and Pb (4.7 µg/g) were found in Khor-Ghazale. Concentration of Co, Cu and Ni in Khor-Ghanam were significantly less than other Khors ($P<0.05$). The extent of heavy metals input, physicochemical factors and physical characteristics of sediment could play core role in the inter-sites accumulation of heavy metals [17-20]. The significant differences, which were observed between metals concentration in the sediment samples collected from various sampling sites indicated that, heavy metals do not originate from the same sources. For example, the elevated levels of metals in Khor-Jafari could be related to inputs from several petrochemical units, which surround this Khor. Metals contamination in Khor-Ghazale on the other hand might be due to heavy traffic of oil tankers in this Khor. Among different Khors studied here, Khor-Ghanam was found to be less contaminated compared to others. Khor-Ghanam is located far from petrochemical units and port of Imam, the major sources of metals in the studied area. This Khor is located at the end of Khor-Musa where it joins to the Persian Gulf. Since there is no traffic of ships or tankers in this place, dissolved metals in seawater and those bounded to suspended organic matter, which is transported by tidal currents and waves are the main sources of heavy metals in Khor-Ghanam. The fact that tidal current is responsible for transporting and spreading of contaminant in the coastal waters have frequently confirmed by previous studies [21-23].

Comparison of heavy metals level in the sediment of the northwest Persian Gulf found in the present work with the result of the previous studies performed by other researchers in other parts of the Persian Gulf is given in table 3. According to the results, the level of Co, Cd and Cu in Khor-Musa was higher than those reported by de Mora *et al.* [24], with exceptions of Co in Akhak Beach and Cu in Bapco Refinery in Bahrain. Industrial or refinery complexes and local mineralogy have been reported as the major sources of metals contamination in the mentioned locations. Comparison of metals in the sediment with data that reported by ROPME [25] showed that Cd, Ni and Pb in our findings were lower than northeastern coasts and Ni concentration was higher than Bahrin coasts of the Persian Gulf. The concentrations of Cd and Pb in the sediment collected from north Persian Gulf by Pourang *et al.* [26] were found to be higher than the present results, whilst Ni concentration was well agreed with our results.

Table 3: Heavy metals concentration (µg/g d.w.) in sediment in different branches of Khor-Musa and Persian Gulf and compared to some available standards

	Cd	Co	Cu	Ni	Pb	References
Khor-Ghazale	0.49	21.63	18.7	64.89	4.70	This study
Khor-Ahmadi	0.46	21.34	12.93	51.22	2.59	This study
Khor-Jafari	0.55	23.55	18.01	68.86	3.16	This study
Khor-Zangi	0.32	21.34	15.46	55.28	3.81	This study
Khor-Ghanam	0.42	17.24	9.24	18.96	2.14	This study
Northeastern Persian Gulf	1.25		103	25	[25]	
Persian Gulf-Qatar	0.08	2.20	8.02	20.8	3.16	[24]
Persian Gulf-UAE	0.09	45.2	3.31	1010	1.30	[24]
Persian Gulf-Bahrin	0.182	2.43	48.3	23.2	99	[24]
Persian Gulf-Oman	0.14	6.92	6.66	77.8	0.44	[24]
North Persian Gulf	2.89		64.89	90.47	[26]	
Persian Gulf-Bahrain coast	0.4		15	12.3	[25]	
ERL	1.2		21	47	[49]	
ERM	9.6		52	220	[49]	
Southwestern Turkey	0.8	_	13	_	83	[27]

ERL=Effects Range Low. ERM=Effects Range Medium

Table 4: Heavy Metals concentration (μg/g d.w.) in liver and muscle and comparison of different Khors within same species (*Euryglossa orientalis*)

Tissues	Location	Cd	Co	Cu	Ni	Pb
	Khor_Ghazale	3.25 ± 0.19^{a}	0.84 ± 0.09^{a}	285.32 ± 9.29^{ab}	1.97 ± 0.24^{a}	3.05 ± 0.51^{b}
Liver	Khor_Ahmadi	2.27 ± 037^{a}	1.14 ± 0.16^{a}	309.84 ± 23.53^{ab}	2.38 ± 0.43^{a}	3.49 ± 0.24^{b}
	Khor_Jafari	1.34 ± 037^{a}	1.04 ± 0.32^{a}	350.55 ± 53.18^{b}	3.50 ± 0.89^{a}	4.11 ± 0.35^{b}
	Khor_Zangi	6.66 ± 1.87^{b}	1.29 ± 0.14^{a}	311.92 ± 49.27^{ab}	1.11 ± 0.26^{a}	1.05 ± 0.46^{a}
	Khor_Ghanam	1.97 ± 0.44^{a}	1.59 ± 0.25^{a}	182.01 ± 25.38^{a}	2.06 ± 0.55^{a}	1.32 ± 0.14^{a}
	Khor_Ghazale	0.14 ± 0.01^{ab}	ND	1.34 ± 0.10^{a}	2.49 ± 0.27^{b}	1.61 ± 0.28^{a}
Muscle	Khor_Ahmadi	0.07 ± 0.02^{a}	ND	2.44 ± 0.26^{a}	2.14 ± 0.27^{b}	0.88 ± 0.11^{a}
	Khor_Jafari	0.15 ± 0.04^{abc}	ND	8.78 ± 1.72^{b}	2.25 ± 0.18^{b}	1.2 ± 0.34^{a}
	Khor_Zangi	0.32 ± 0.06^{c}	ND	1.20 ± 0.18^{a}	$.72 \pm 0.14^{a}$	1.47 ± 0.19^{a}
	Khor_Ghanam	0.26 ± 0.05^{bc}	ND	1.13 ± 0.20^{a}	2.71 ± 0.33^{b}	1.02 ± 0.33^{a}

[a,b,c] Show differences among Khors (P<0.05). ND means Not determined

According to the mentioned researches, heavy metals contamination in the Iranian coasts of the Persian Gulf had been increasing from year 1999 to 2005. Pourang *et al.* [26] concluded that oil tankers, oil drilling in the region and current transportation were possible sources of metals contamination. The concentrations of Pb and Cd in the sediment from southeastern coasts of Turkey [27] were higher than present study. On the other hand, the level of Cu in their finding was lower than current study. The highest levels of measured metals in the sediment in the present study did not exceed the Effects Range Low (ERL) and Effects Range Medium (ERM) proposed by NOAA (National Oceanic and Atmospheric Administration), except for Ni concentration.

Heavy Metals Concentration in Fish: The concentration of heavy metals in fish tissues are given in table 4. Metal concentrations in the liver ranked as Cu > Cd = Pb =Ni > Co. The highest Concentration of Cu (350.55 μg/g) was obtained in the liver samples taken from Khor_Jafari. The highest Cd (6.66 μg/g) concentration and the lowest level of Pb (1.05 μg/g) were detected in the liver samples from Khor_Zangi. Concentration of heavy metals in the muscle ranked as Cu = Ni > Pb > Cd > Co. The highest level of Cu (8.78 μg/g) was detected in the muscle samples in Khor_Jafari. The maximum level of Cd (0.32 μg/g) and the lowest concentration of Ni (0.72 μg/g) were measured in muscles from Khor_Zangi. No significant variation was detected for Ni and Co concentration in the liver as same as Pb in the muscle. The concentration of Co in the muscle was below the detection limit of AAS. This finding is also confirmed by several investigations that metals accumulation in liver tissues is higher than muscle [28, 29].

The results of the present study indicate that Cd, Co, Cu, Ni and Pb concentrations in liver were higher than muscle. Some metals such as Cu are essential element for organisms [30, 31]. Because of their vital role in physiological and biochemical processes, many organisms accumulate them in high concentrations within their tissues [32]. In addition, liver plays an important role in storage and detoxification of uptaken metals for fish [33]. Therefore, it is a target organ for most of heavy metals [34, 35]. While low Cd concentration was found in the sediment samples of Khor-Zangi, surprisingly high concentration of Cd was measured in fish samples from this Khor. Heavy metals accumulation by aquatic organisms depends on the metals concentration in ambient environment, however; this is not the only factor affecting metals level in fish. Many other factors such as salinity, temperature, pH and suspended organic carbon may affect speciation and bioavailability of metals [36, 37], which finally affect metal accumulation. The concentration of some metals such as Co and Ni in fish was found to be lower than those of their ambient sediment. Even though in the presence of high metals concentration in the environment, low bioavailability of metals may also result in low uptake rate and low metals accumulation [38, 39].

Heavy metals concentration in *E. orientalis* was higher than *Epinephelus coioides* and *Lethrinus nebulosus* from south Persian Gulf [31], except Cu in muscle of fish from Dahannak site. According to the results Pb and Ni in *E. orientalis* was closed to what was determined by Pourang *et al.* [26] for *Solea elongata, Psettodes erumel* and *Epinephelus coioides* from north Persian Gulf. Unlikely Pb concentration in *Solea Bleekeri* caught in Kuwait coastal waters [40] was more than Pb level in *E. orientalis* Henry *et al.* [8] studied heavy metals in liver and muscle tissues of some fish species from North Sea. The maximum concentrations of Cd, Cu and Pb in their results were lower than results of current study. These results confirmed that difference species in the same region have various capacities on metals accumulation. In addition, this fact pointed out by many

Table 5: Comparison between average metals concentration in *E. orientalis* with some other fish species from other locations

Location	Species	Tissues	Cd	Co	Cu	Ni	Pb	References
Persian Gulf-Qatar-Doha	*Epinephelus coioides*	L	0.417	0.517	90.9	0.08	0.074	[31]
		M	0.003	0.013	0.54	0.09	0.209	
Persian Gulf-UAE-Dhannah	*Epinephelus coioides*	L	9.94	0.228	9.25	<. 01	0.308	[31]
		M	<. 001	0.012	0.88	<. 01	20.01	
Persian Gulf-Bahrain-Badaiya	*Epinephelus coioides*	L	0.369	0.423	159	0.085	0.012	[31]
		M	<. 001	<. 01	0.294	<. 01	0.005	
Persian Gulf-Oman-Raysut Port Area	*Lethrinus nebulosus*	L	109	0.256	98.8	<. 05	0.175	[31]
		M	0.011	<. 05	0.519	<. 05	0.014	
Persian Gulf-Kuwait	*Solea Bleekeri*					3.6	0.4	[6]
North Persian Gulf	*Solea elongata*		0.07			6.69	2.43	[35]
North Persian Gulf	*Psettodes erumei*		0.10			1.09	2.09	[35]
North Persian Gulf	*Epinephelus coioides*		0.11			1.56	2.32	[35]
North sea-Dunkirk	*Platichthys flesus*	L	0.26		52.2		0.09	[21]
		M	0.003		0.78		0.02	
North sea-Dunkirk	*Limanda limanda*	L	0.13		16.8		0.04	[21]
		M	0.003		0.94		0.07	
North sea-Calais	*Platichthys flesus*	L	0.42		49.6		0.08	[21]
		M	0.02		1.8		0.04	
North sea-Calais	*Limanda limanda*	L	1.1		11.9		ND	[21]
		M	0.02		0.85		0.12	

L= Liver, M=Muscle

Table 6: Permissible upper limits of heavy metals in various standards

Standard	Cd	Cu	Pb	Reference
FAO	0.5(mg/kg)	30(mg/kg)	2(mg/kg)	[32]
WHO	30(mg/kg)	0.5(mg/kg)	[32]	
MAFF	20(μg /g W.W)	1 (μg /g W.W)	[45]	
NHMRC	2(μg /g W.W)	30(μg /g W.W)	5.5(μg /g W.W)	[45]
TEG	0.1(μg /g)	20(μg /g)	1(μg /g)	[27]
Germany	0.5(μg /g W.W)	0.5(μg /g W.W)		
Saudi Arabia	0.5(μg /g)	2(μg /g)	[50]	
EC	0.05(μg /g W.W)	0.5(μg /g W.W)	[50]	

FAO (1983), WHO (1996), MAFF= Ministry of Agriculture Fisheries and Food (United Kingdom), NHMRC= National Health Research Council (Australia), TEG= Turkish Environmental Guidelines, EC= European Communities

authors that fish habitats plays a significant role in accumulation of metals in fish [41, 42]. Generally, metals accumulation by fish depends on many factors such as bioavailability, physiology, behavior and environmental conditions [43]. Metals concentration in the edible tissue of *E. orientalis* measured in this study were compared with some available standards for human consumption in table 6. To make comparison easy, the values have been converted into μg/g w. weight with a wet wt. dry wt ratio of 0.2. Accordingly, the highest level of Cd, Cu and Pb in muscle was below the permissible limits provided by FAO (1983), WHO (1996), MAFF (Ministry of Agriculture Fisheries and Food, United Kingdom), NHMRC (National Health Research Council, Australia) and TEG (Turkish Environmental Guidelines). The maximum level of Pb was close to European Communities (1997) limit and in Khor-Zangi was even above Limit of EC. The maximum concentrations of studied metals also do not exceed the legal limits of heavy metals regulated via other countries such as Germany and Saudi Arabia.

Correlation Between Metals Concentration in Sediment and Fish: The studied species, *E. orientalis* is a demersal fish, which feeds on benthic invertebrates in the sediment [44]. In the other word, it is non-migrate fish that does not leave its habitat. No significant correlation was found between metals concentration in sediment and fish tissues ($P>0.05$). Therefore, in spite of being demersal species, *E. orientalis* could not be considered as a suitable biomonitor agent for heavy metals in the study area.

It is suggested that the lack of significant correlation between metals concentration in sediment and fish may be related to low variability between metals level in the sediment of different stations. Although the metal concentrations in liver of some fish species may reflect the level of metals in the seawater [45]. However, metals concentration in seawater was not measured in this study because of the detection limits of AAs. The results also showed significant correlations between, Co-Cu ($r=0.64$, $P<0.05$), Co-Ni ($r=0.67$, $P<0.05$) in the sediment. The inter-element relations that are observed in the sediment could be a result of similarity in physicochemical conditions [46] or in the sources of metals input [47]. In addition, no significant correlation was found between elements in tissues that strongly depend on physiological properties such as biochemical pathways, binding properties and detoxification [48].

CONCLUSION

Heavy metals concentration in Khor-Musa was found to be moderate and within the range of some previous studies in the Persian Gulf. Therefore, no pollution threat is aspect about metals contamination in the area. Although the level of metals in the liver of fish samples was much higher than muscles, metals concentration in edible tissues (muscles) was less than maximum permissible levels proposed by different food standards. The low variations of heavy metal concentration caused the lack of significant correlations between metals level in the sediment and fish. Since Khor-Musa is receiving various types of wastewaters, regular monitoring of this estuary is recommended.

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