RESEARCH ARTICLE



Determination of nickel and thallium concentration in *Cynoglossus arel* fish in Musa estuary, Persian Gulf, Iran

Neamat Jaafarzadeh Haghighi Fard¹ • Ahmad Zare Javid^{2,3} • Maryam Ravanbakhsh⁴ • Zahra Ramezani^{5,6} • Mehdi Ahmadi¹ • Kambiz Ahmadi Angali⁷ • Shirin Ardeshirzadeh⁸

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Abstract Heavy metals with high bioaccumulation capacity are considered as important contaminants and may be available in high concentrations in environment and biota samples. The main aim of this study was to determine the concentration of nickel and thallium in *Cynoglossus arel* fish in Musa estuary. Sixty-seven fish samples were collected from Musa estuary during five intervals of 15 days in summer 2013. After biometric measurements, the concentrations of nickel and thallium were measured

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Highlights • Ni and Tl was determined in *Cynoglossus arel* fish in Musa estuary, Persian Gulf.

- · Concentration of Ni was higher than threshold limits.
- Ni concentration had significant reverse relationship with length.
- Tl concentration had significant direct relationship with length.

Maryam Ravanbakhsh mapseh@gmail.com

Neamat Jaafarzadeh Haghighi Fard n.jaafarzade@yahoo.com

Ahmad Zare Javid

ahmaddjavid@gmail.com

Zahra Ramezani zramezani@ajums.ac.ir

Mehdi Ahmadi ahmadi241@gmail.com

Kambiz Ahmadi Angali kzfir@yahoo.com

Shirin Ardeshirzadeh Sh.ardeshirzadeh@yahoo.com

¹ Environmental Technologies Research Center, Department of Environmental Health Engineering, School of Public Health, Ahvaz Jundishapur University of Medical Sciences, Ahvaz 15794-61357, Iran

by graphite furnace atomic absorption spectrophotometer. The

mean concentration of nickel and thallium in muscle tissue of fish samples was 2.458 ± 0.910 and 0.781 ± 1.754 mg kg⁻¹/ww,

respectively. The GLM analysis showed a significant negative

relationship between nickel concentration and length. In addi-

tion, there was a significant positive relationship between thalli-

um concentration and fish length. Nickel concentration exceeded

the allowable standards of WHO and FDA in Cynoglossus arel.

- ² Nutrition and Metabolic Diseases Research Center, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran
- ³ Hyperlipidemia Research Center, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran
- ⁴ Student Research Committee, Department of Environmental Health Engineering, School of Public Health, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran
- ⁵ Nanotechnology Research Center, Faculty of Pharmacy, Jundishapur University of Medical Sciences, Ahvaz, Iran
- ⁶ Medicinal Chemistry Department, Faculty of Pharmacy, Jundishapur University of Medical Sciences, Ahvaz, Iran
- ⁷ Department of Biostatistics, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran
- ⁸ Family Medicine Department, Cleveland Clinic Foundation, Cleveland, OH, USA

Therefore, regarding with high consumption of seafood in this region, it is recommended that these fishes should be consumed under a nutritionist counseling.

Keywords Heavy metal \cdot Nickel \cdot Thallium \cdot Cynoglossus arel fish \cdot Persian Gulf \cdot Musa estuary

Introduction

The Persian Gulf, which is located in the south of Iran, is a shallow basin with depth of approximately 35-40 m (Mortazavi and Sharifian 2011) and 240,000 km² (Elahi et al. 2012). It is joined to free international waters via Hormuz Strait (Banat et al. 1998). The Persian Gulf, as a part of ROPME Sea, is polluted by several human activities. The oil pollution in the Persian Gulf constitute about 4.7% of total oil pollution in the world (Hosseini et al. 2013). Several factors such as low turnover and flushing time (3–5 y), shallow depth, limited circulation, high salinity, high temperature, and evaporation may lead to longer residence of different pollutants in the Persian Gulf. So, it is presumed that these pollutants may have some effects on marine environment (Sheppard 1993).

Musa estuary with several branches and creeks is located in northern coastal area of the Persian Gulf (latitude of 30°, 15' to 30°, 32' and longitude of 49° to 49°, 20'). It is a coastal ecosystem containing several unique marine fauna and flora. Some economically important fish and shrimp species migrate to this area and make it as a commercial region for fisheries. The self-purification capacity in Musa estuary is low and the concentrations of suspended solids are high (Javid and Sanmadyar 2006). A massive contamination is transferred to this estuary by the Persian Gulf. In addition, because of specific geographical situation of Musa estuary, it is used as a waterway by many crude oil tankers and ships. Furthermore, a huge quantity of effluents from several huge petrochemical plants and large ports urban and agricultural resources enter to the Musa estuary every day (Malmasi et al. 2010). In 1993, about 1.22 km² of Musa estuary (an area of Jafari creek) was drained off for Petrochemical Special Economic Zone (PETZONE). Thus, it is likely that the load of contamination in this region has turned it as a sensate area accumulating heavy metals within harmful and toxic levels for human being and other organisms (Abdolahpur Monikh et al. 2013a).

Regarding with nature management and the issue of human consumption, paying attention to heavy metal content of fishes would be required (Karadede et al. 2004). Heavy metals with high bioaccumulation capacity are considered as the main and persistent contaminants and may exist in high concentrations in environment and biota samples. It can be originated from natural and anthropogenic sources (Jaafarzadeh et al. 2011).

The accumulation of heavy metals in fish depends on several factors including different chemical interactions (Wang et al. 2005), bioavailability of heavy metals and pollutant source (Abdolahpur Monikh et al. 2013b), the region and time of fishing, requirements, gender, size, species, nutrition, concentration of heavy metals in water, exposure time against heavy metals, and other environmental factors such as salinity, pH, and water temperature (Zhang and Wong 2007). It is suggested that fishes may considerably reflect the ecosystem contamination. In several studies, fish samples were selected to evaluate the bioaccumulation of heavy metals in the environment (Fard et al. 2015; Javed et al. 2016; Javed and Usmani 2014; Mohammadnabizadeh et al. 2013; Mortazavi and Sharifian 2012). Fishes are nutritionally important in human diet and considered as important sources of human exposure against environmental pollutants (Guisan et al. 2002), so they can reserve considerable amount of heavy metals in the body (Fard et al. 2015; Ikem and Egiebor 2005). However, there are limited studies on nickel (Ni) and thallium (Tl) and therefore, it may be needed to address the accumulation of heavy metals as components in oil pollutants and industrial wastewater among the myriad of toxic organic and inorganic substances. Heavy metals can be released into aquatic ecosystems and accumulated in the sediments and organisms and finally transferred to human body through the food chain (Fard et al. 2015; Sheppard 1993).

According to the statistics (Statistics 2010), fish consumption has been rapidly increased in Iran during the last decade. The fishes specifically *Cynoglossus arel* caught in Musa estuary constitute the main source of animal protein in the diet of local residents (Mortazavi and Sharifian 2011). *Cynoglossus arel* are flatfish species and benthophagus; adults feed primarily on polychaetes, while juveniles more often consume smaller prey such as hyperiid amphipods and copepods (Rajaguru 1992). Flatfishes may be particularly sensitive to pollution and other types of habitat degradation (Abdolahpur Monikh et al. 2013b). Therefore, the evaluation of nickel and thallium in *Cynoglossus arel fish* may be an appropriate indicator of assessing heavy metal bioaccumulation.

Regarding with important pollutant sources releasing nickel and thallium in marine environment in Musa estuary, along with their usage as indicators of marine pollution, the necessity of this study would be claimed. The main aim of this study was to determine the concentrations of nickel and thallium in muscle tissue of *Cynoglossus arel* fish collected from Musa estuary, in Persian Gulf, Iran. The other aims of current study were (a) to evaluate the relationship between metal concentration, length, and weight of fish samples, and (b) to determine the relationship between nickel and thallium.

Material and methods

Totally, 67 *Cynoglossus arel* fish samples were collected from Musa estuary, located in the north west of the Persian Gulf

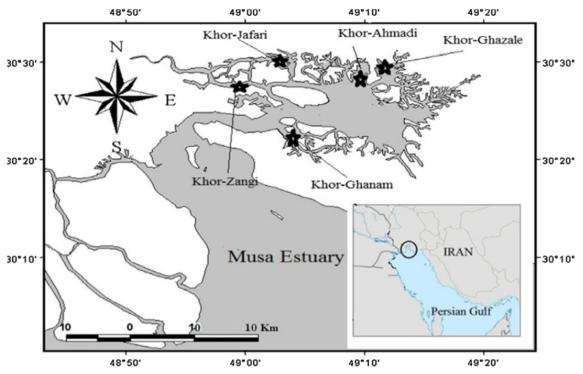


Fig. 1 Musa estuary and creeks around it (Abdolahpur Monikh et al. 2013a)

(latitude of 30° , 15' to 30° , 32' and longitude of 49° to 49° , 20') (Fig. 1), during 2 months (in five time points with 15-day intervals) in summer 2013 (from 22 May until 22 July 2014). Usually, summer is the best fishing time for *Cynoglossus arel* fish in this estuary.

All caught fish samples were transferred to the lab in a cool box. Biometric indices including weight and length were measured by a biometric board and a digital scale. All extra parts and skin of fish samples were dispatched using a stainless steel knife. Then, the muscle filet (without any bone) was separated and maintained in a -70 °C freezer.

All containers were rinsed with a diluted nitric acid solution and dried in an oven at 105 $^{\circ}$ C.

Nitric acid (65% (v/v)) was added to 2.5-g wet muscle sample and maintained at room temperature for 24 h. Then, the hydrogen peroxide 30% (v/v) was added. After then, samples were placed in a high-pressure microwave (Milestone Ethos 900) equipped with a probe section to control the pressure and temperature. Digested samples were diluted with deionized distilled water (Milli-Q Millipore 18.2 M Ω cm⁻¹) and brought to the volume of 100 ml.

Nickel and thallium concentrations were measured by an atomic absorption (Carl Zeiss Jena GmbH - AAS 5 EA, Germany) equipped with a graphite oven and an automatic sampler. After calibration, nickel and thallium concentrations were measured at wavelength of 232 and 276.8 nm and the slit width of monochromator of 0.8 and 0.5 nm, respectively. There was no need to matrix correction (Table 1). The background correction was D_2 . Twenty microliters of digested samples were

injected to the device in all steps. The recovery percent of added nickel and thallium in digested samples is shown in Table 2. The standard addition method was also used in the procedure.

Statistical analysis was performed using SPSS software (version 16). In this study, "generalized linear model" (GLM) was used for analyzing the effect of an independent variable on a dependent variable. GLMs are mathematical extensions of linear models that do not force data into unnatural scales and thereby allow for nonlinearity and nonconstant variance structures in data. They are based on an assumed relationship (called a link function) between the mean of response variable and the linear combination of explanatory variables. Data are assumed to be from several families of probability distributions, including the normal,

 Table 1. The condition used in atomic absorption spectrophotometer for determination of nickel and thallium

Step	Tempera	ture (°C)	Ramp (°C/S)	Hold (S)		
	Ni	T1	Ni	T1	Ni	Tl	
Drying	90	90	5	5	20	20	
Drying	105	105	3	3	20	20	
Drying	110	110	2	2	10	10	
Pyrolysis	950	500	250	250	10	10	
Az	950	500	0	0	6	6	
Atomization	2100	1250	1500	1500	5	5	
Cleanout	2400	2400	500	500	4	4	

 Table 2
 The recovery percent of added nickel and thallium to digested samples

Sample	Concentration (Rsd (%)		Recovery (%)		
	Tl	Ni	T1	Ni	Tl	Ni
Real sample 1	11.08772	38.5	4.8	0.6		
Spike sample 1(100 ppb)	115	138.8846	4.5	0.7	103.9123	100.3846
Real sample 2	11.66667	42.73077	4.2	2.5		
Spike sample 2(100 ppb)	113.9123	139.5769	1.8	2.7	102.2456	96.84615
Real sample 3	11.49123	35.53846	1.1	0.7		
Spike sample 3(100 ppb)	116.1404	136.7308	0.5	3.4	104.6491	101.1923
Real sample 4	11.7193	39.30769	3.9	0.7		
Spike sample4 (100 ppb)	119.2982	130.1538	1.4	2.6	107.5789	90.84615
Real sample 5	8.877193	32.96154	2.9	2.3		
Spike sample 5 (100 ppb)	102.5965	128.9231	0.6	0.1	93.7193	95.96154
Real sample 6	17	17	0.6	0.6		
Spike sample 7 (100 ppb)	116.42857	116.4286	5	5	99.4285714	99.42857
Real sample 8	6.8571429	6.857143	2.4	2.4		
Spike sample 8 (100 ppb)	101.14286	101.1429	5	5	94.2857143	94.28571
Real sample 9	105.6	105.6	0.8	0.8		
Spike sample 9(100 ppb)	220.14286	220.1429	3.8	3.8	114.542857	114.5429
Real sample 10	97.942857	97.94286	4.4	4.4		
Spike sample10(100 ppb)	206.85714	206.8571	1.4	1.4	108.914286	108.9143

binomial, poison, negative binomial, or gamma distribution, many of which better fit non-normal error structures of most ecological data. Thus, GLMs are more flexible and better suited for analyzing ecological relationships, which can be poorly represented by classical Gaussian distributions (Guisan et al. 2002). The predictor data were entered in to the model and the linear models were calculated. Moreover, regression analysis was used to investigate the relationship between metal concentration, fish length, and weight.

Results

The biometric data are shown in Table 3. Total mean weight of fish samples was 363.48 ± 78.57 g with maximum and minimum of 570.00 and 206.00 g, respectively. Total mean length was 27.86 ± 3.10 cm with maximum and minimum of 34.00 and 13.00 cm, respectively.

The mean concentration of nickel was $2.458 \pm 0.910 \text{ mg kg}^{-1}/\text{ww} (3.098 \pm 1.150 \text{ mg kg}^{-1}/\text{dw})$ with maximum and minimum of 4.857 and 0.183 mg kg^{-1}/\text{ww} (6.122 and 0.231 mg kg^{-1}/\text{dw}, respectively). The mean concentration of thallium in muscle samples was 0.781 ± 1.754 mg kg^{-1}/ww (0.984 \pm 2.211 mg kg^{-1}/\text{dw}) with maximum and minimum of 14.860 and 0.241 mg kg^{-1}/\text{dw} (18.730 and 0.304 mg kg^{-1}/\text{dw}). The figures converted to mg kg^{-1}/\text{dw} considering the percentage of dried weight of fish (mg kg^{-1}/\text{ww 0.8178}).

According to the GLM analysis in Table 4 (nickel concentration as a dependent variable, and thallium concentration, length, and weight as independent variables), there was a strong significant reverse relationship observed between the concentrations of nickel and length ($\beta = -27.43$, P = 0.00). Furthermore, thallium concentration and weight showed a less significant direct relationship with the concentration of nickel (with $\beta = 0.231$ and 0.486, respectively, P = 0.00). It was found that the length had the greatest relationship with nickel concentration compared with weight and thallium concentration (Table 4).

Furthermore, considering the concentration of thallium as a dependent variable and nickel concentration, weight and length as independent variables in the GLM analysis, there was a less significant reverse relationship between the concentrations of thallium and weight ($\beta = -1.468$, P = 0.00). In contrast, nickel concentration and the length had significant direct relationship with thallium concentration ($\beta = 1.311$ and 32.051, P = 0.00). It was found that the length had the greatest direct relationship with thallium concentration compared with the other variables.

Furthermore, based on regression analysis, there were significant relationships (equations as below) found between the length and weight (r = 0.65, P = 0.00).

Length = 18.581 + 0.026 weight

Discussion

In this study, the concentration of Ni and Tl was determined in fish samples in Musa estuary. It was found that the maximum concentration of nickel (4.857 mg kg⁻¹/ww) measured in

 Table 3
 The biometric data and concentrations of nickel and thallium in fish

Item	Sampling time	Ν		Mean \pm std. deviation	Minimum	Maximum	
Weight (g)	Day 1	13		375.46 ∓ 52.13	301.00	494.00	
	Day 15	13		391.46 ± 72.39	267	553.00	
	Day 30	13		343.62 ± 57.26	241	486.00	
	Day 45	14		362.93 ± 70.75	246	482.00	
	Day 60	14		345.36 ± 119.55	206	570.00	
	Total	67		363.48 ± 78.57	206	570.00	
Length (cm)	Day 1	13		27.12 ± 5.56	13	31.00	
	Day 15	13		28.77 ± 1.37	26	32.00	
	Day 30	13		27.73 ± 1.60	24.50	30.00	
	Day 45	14		28.25 ± 2.28	25	32.00	
	Day 60	14		27.46 ± 3.03	23	34.00	
	Total	67		27.86 ± 3.10	13	34.00	
Nickel (mg/kg)	Day 1	13	WW	2.481 ± 1.154	0.223	3.680	
			dw	3.127 ± 1.455	0.281	4.638	
	Day 15	13	WW	2.246 ± 0.855	0.229	3.228	
			dw	2.831 ± 1.078	0.289	4.069	
	Day 30	13	WW	2.569 ± 0.190	0.215	2.789	
			dw	3.238 ± 0.239	0.272	3.515	
	Day 45	14	WW	2.374 ± 0.939	0.183	3.913	
			dw	2.992 ± 1.184	0.231	4.932	
	Day 60	14	WW	2.618 ± 1.139	0.423	4.857	
			dw	1.436 ± 1.436	0.533	6.122	
	Total	67	WW	2.458 ± 0.910	0.183	4.857	
			dw	3.098 ± 1.150	0.231	6.122	
Thallium (mg/kg)	Day 1	13	WW	0.538 ± 0.153	0.265	0.819	
			dw	0.678 ± 0.193	0.334	1.032	
	Day 15	13	WW	0.549 ± 0.161	0.246	0.769	
			dw	0.692 ± 0.203	0.310	0.969	
	Day 30	13	WW	0.641 ± 0.168	0.469	11.128	
			dw	0.808 ± 0.212	0.591	14.026	
	Day 45	14	WW	0.586 ± 0.209	0.251	0.981	
			dw	0.739 ± 0.263	0.316	1.237	
	Day 60	14	ww	1.546 ± 3.835	0.241	14.860	
			dw	1.949 ± 4.834	0.304	18.730	
	Total	67	ww	0.781 ± 1.754	0.241	14.860	
			dw	0.984 ± 2.211	0.304	18.730	

ww wet weight, dw dried weight

Cynoglossus arel in this study was about six times higher than the maximum concentration (0.78 mg kg⁻¹/ww) reported for canned fish in Georgia and Alabama study (Ikem and Egiebor 2005) and canned sardines in Nigeria (max $3.26 \text{ mg kg}^{-1}/\text{ww}$) (Iwegbue et al. 2009).

According to studies conducted in this region during the last decade in several fish species, it is shown that the Ni concentration has an increasing trend (Table 5). This may be due to increasing the bioavailability of metals in the estuary and lead to high accumulation of metals in fish muscle tissues consumed by local residents.

Some studies showed that the creeks are more polluted than Musa estuary regarding with the higher nickel content. Parvaneh (Parvaneh et al. 2011) measured nickel concentration in muscle tissue of *Euryglossa orientalis* fish in Khor Ahmadi and Khor Ghanam, which are two creeks in the Musa estuary and located near the industrial plants. It was reported that the mean concentration was 14.47 ± 0.53 mg kg⁻¹/dw, showing that it is about seven times greater than the concentration reported in our study. Moreover, some studies conducted in the Musa estuary showed a high water content of nickel. Abdolahpur Monikh (Abdolahpur Monikh et al. 2013a)

Table 4 The GLM analysis: nickel and thallium as dependent variables

Dependent parameter	Dependent parameter	В	Std. error	95% Wald confidence interval		Hypothesis test		
				Lower	Upper	Wald chi-square	df	Sig.
Nickel	(Intercept)	3850	1.1736	3847.909	3852.509	1.076E7	1	0.000
	Thallium	0.231	7.2760E-5	0.231	0.231	1.008E7	1	0.000
	Weight	0.486	0.0021	0.482	0.490	51,567.679	1	0.000
	Length	-27.4	0.0549	-27.540	-27.325	250,034.901	1	0.000
Thallium	(Intercept)	-3660	1.3837	-3663.025	-3657.601	6,997,738.696	1	0.000
	Nickel	1.311	0.0002	1.311	1.312	5.721E7	1	0.000
	Weight	-1.468	0.0021	-1.472	-1.464	471,631.382	1	0.000
	Length	32.051	0.0551	31.943	32.159	338,540.703	1	0.000

observed the highest concentration of Ni in Khor-Ghazale. On the other hand, the lowest heavy metal concentration was observed in Khor Ghanam with no industrial activity nearby. Furthermore, Imam Port which is one of the biggest ports in Iran is located in the mouth of Khor-Ghazale. Similarly, Khor-Zangi, one of the most polluted creeks in Musa estuary, is situated near petrochemical units close to Mashahr City. The industrial activities in this region may be considered as the main

Table 5Studies on Niconcentration in muscle tissue offish species in Musa estuary

Study	Fish species	Ni concentration	
Hosseini (Hosseini et al. 2015)	Liza abu	42.15 ^a	
	Euryglossa orientalis	42.05 ^a	
	Otolithes ruber	42.01 ^a	
Moghdani (Moghdani et al. 2015)	(Brachirus orientalis)	1.38 ^a	
Farhadiamiri (Farhadiamiri et al. 2013)	(Saurida tumbil)	2.3 ^a	
Abdolahpur Monikh (Abdolahpur Monikh et al. 2013a)	Liza abu	0.47a	
	Johnius belangerii	0.66 ^a	
	Euryglossa orientalis	3.38 ^a	
Khazaei (Hossein Khazaei et al. 2013)	Otolithes rubber	$2.20^{\rm a}$	
	Pampus argenteus	1.08 ^a	
Khorasani (Khorasani et al. 2013)	Otolithes ruber	1.93 ^a	
Abdollahpur Monikh (Abdolahpur Monikh et al. 2012)	Johnius Belangerii	4.32 ^a	
	Euryglossa orientalis	6.27 ^a	
	Cynoglossus arel	3.4 ^a	
Abdolahpur Monikh (Abdolahpur Monikh et al. 2011)	Dasyatis bennettii	1.86 ^a	
Safahieh (Safahieh et al. 2011b)	Liza abu	0.48-2.73	
Safahieh (Safahieh et al. 2011a)	(Johnius belangerii)	2.43	
Sadough Niri (Sadough Niri et al. 2010)	Ilisha Tenualosa	4.004 ^a	
Agah (Agah et al. 2009)	Grunt	0.071 ^b	
	Flathead	0.016 ^b	
	Greasy grouper	0.027 ^b	
	Tiger-tooth	0.024 ^b	
	Silver pomfret	0.039 ^b	
Tatina (Tatina et al. 2009)	Pelates quadrilineatus	0.36 ^b	
Pourang (Pourang and Dennis 2005)	Solea elongata	6.69 ^b	
-	Psettodes erumei	1.09 ^b	
De mora (De Mora et al. 2004)	Epinephelus coioides	0.05 < ^b	
	Lethrinus nebulosus	0.1 ^b	

 a mg kg⁻¹/dw

^b mg kg⁻¹/ww

pollution sources in the estuary and creeks. It may be due to dilution of pollutants in Musa estuary by periodical tidal occurring twice a day in the estuary. Since nickel exist in all crude oils (usually at concentrations far higher than any other metal), it is suggested to be the most important indicator of oil pollution sources (Pourang et al. 2005). It is suggested that the high nickel accumulation observed in fish samples in the present study may be primarily resulted from the pollution originated by the possible anthropogenic sources of the relatively high concentration of Ni such as oil spillage from the oil tankers, refinery activities, and discharge of ballast water by oil tankers, oil drilling platforms in the region. The results of other studies concur with the findings of this study (Jafarian et al. 2012; Love et al. 2013; Mashinchian Moradi et al. 2011; Mohammadi et al. 2011; Osei et al. 2012; Parvaneh et al. 2011). Moreover, the role of sea current in transferring pollutants from other regions and their distribution should also be considered. In addition, Mashinchian Moradi (Mashinchian Moradi et al. 2011) reported that there is a nickel mine under the oceanic crust in this region which is considered as a natural source of nickel for sea water and sediments in the Persian Gulf. Therefore, regarding with contamination of water and sediments in Musa estuary with nickel, its accumulation in fish species would be expectable.

The concentration of Ni decreased significantly with fish size (as shown by the GLM results, Table 4). There are several mechanisms suggested for the negative relationships between size and metal concentration. It is indicated that the tissue growing may be more rapidly than trace metal intake. Therefore, seasonal changes in body condition (weight-length relationship) may show positive trends in a given species collected in different periods, showing relative dilution effect of the lipid content of tissues, as hypothesized by Farkas et al. (Farkas et al. 2003). It is also claimed that young fish have a lower percentage of fat tissue than adults (Merciai et al. 2014). Furthermore, metabolic rate of organisms is dependent on size-specific and is higher in young specimens (Fard et al. 2015) with consequent higher food intake. The net accumulation of heavy metals in an organism may be due to the difference between uptake and depuration (Canli and Atli 2003), the latter being an active process involving energetic expenditure. As young fish consume more energy in growth, less energy may be available for detoxification and this may be more effective in large fish. Moreover, faster short-term uptake by smaller fishes, changes in body surface-volume ratio during growth, size-dependent feeding behavior, and sizedependent concentrations of some biochemical entity involved in accumulation kinetics, such as enzyme concentrations (Liang et al. 1999; Phillips 1980).

Cynoglossus arel is a fish with high growth rate (Rajaguru 1992). Generally, there is a link between decreased muscle metal concentration and a certain length in fishes with higher growth rate (Lavigne et al. 2010) as in a fast growing fish with a

large size metal intake would be diluted (Simoneau et al. 2005). Authman (Authman 2008) indicated that lipid content of tissues may have a relatively dilution effect in reduction of nickel content. Askary sary determined lipid content of *Cynoglosuse arel* fish in 2014 in Persian Gulf (Askary Sary et al. 2014) and showed that its lipid content (% 3/46 ± 0/4) is higher than most of the fish spices in Persian gulf such as *Acanthopagrus latus, Epinephelus coioides, Liza dussmieri, Otolithes ruber, Scomberomorus commerson, and Scomberomorus guttatus.*

The findings of current study are coincide with other studies which showed the negative correlation between fish length and Ni concentration in fish muscle (Agah et al. 2010; Merciai et al. 2014). In contrast to our results, those observed in the literature do not often show significant negative relationship between metal concentrations and fish length (Abdolahpur Monikh et al. 2013b) (Kasimoglu 2014).

In current study, the concentration of nickel in muscle samples of *Cynoglossus arel* exceeded the WHO (0.38 mg kg⁻¹/ ww) and FDA (1 mg kg⁻¹/ww) standards (Commission 2000; Pourang et al. 2004). Therefore, regarding with this issue that the environment in the Persian Gulf is exposed to a high oil contamination, it is suggested that nickel concentration in fish and other marine organisms in this region may also exceed the international standards. Therefore, consuming these sea foods may cause some health problems. Nickel toxicity may lead to some disorders such as allergy, cancer, respiratory disorders (caused by industrial activities), and iatrogenic poisoning (Aundsen et al. 2007). The upper tolerable intake level (UL) of nickel in children (1-3 years old) and males/females (19-70 years old) is 7 and 40 mg/day, respectively (Pazira et al. 2014). The WHO daily recommended dietary allowance for nickel is 35-700 µg/day (WHO 2000).

Thallium is a highly toxic element and its salts are suggested as hazardous substances according to the US Federal Water Pollution Control Act. Thallium is considered as an important environmental pollutant in the Great Lakes Water Quality Agreement between the USA and Canada (Lin et al. 2001) and exists in mainly industrial wastewaters (Peter and Viraraghavan 2005). Thallium usage is increasing due to a growing demand of high-technology industries. The major sources of thallium releases to the environment are not from facilities that produce or use thallium and its compounds, but from processes in which thallium is a trace element of the raw materials (Schoer 1984). The thallium concentration is greater in aquatic environments near industrial areas. Thallium is released to water in the form Tl2O from combustion of sulfide ore smelting, cement production, and combustion of fossil fuels (Lapointe and Couture 2009; Twiss et al. 2003). Fishes available in affected estuaries are exposed to aqueous and dietary metals throughout their life-cycle and hence are prone to metal accumulation and toxicity from the embryo stage. Water and food may be important sources of thallium for aquatic organisms including fish (Lapointe and Couture 2010).

The high concentration of thallium in fish samples in Great estuaries has led to bioaccumulation and bio-magnification of thallium in the aquatic food chain and potential risks of their consumption (Nriagu 1998). Previous studies showed that the concentration of thallium had an increasing trend over a time period in the large estuaries influenced by industrial activities and urban developments with a sever bioaccumulation (Twining et al. 2003). The high concentration of thallium reported in fish tissues from the Great Lakes suggests that the risk of thallium poisoning through fish consumption may be higher than normal (Peter and Viraraghavan 2005). Therefore, it is suggested that contamination of marine systems with thallium should be considered as an emerging issue in industrialized areas such as Musa estuary. In addition, fish consumption in Musa estuary should be concerned due to high levels of pollutants. Generally, most advices of fish consumption in this estuary are based on PCB and mercury levels in fishes and rarely consider contaminants such as thallium. According to our best of knowledge, there are few studies to report information about thallium and its concentration in water, sediments, and fishes in this region. Despite the fact that thallium is a highly toxic element, it has been studied with a lower priority than other toxic elements such as lead, cadmium, or mercury. Agah (Agah et al. 2009) determined the thallium concentration in fish samples ranged between 0.05 and 0.2 ng g^{-1} /ww which was considerably less than those reported in this study $(0.781 \pm 1.754 \text{ mg kg}^{-1}/\text{ww})$. Furthermore, in another study by Agah (Agah et al. 2010), the concentration of some trace metals such as thallium in tissue samples of two commercial fishes collected from the Caspian Sea was 0.3-1.1 and 1.8-3.6 (ng kg⁻¹/ww) which were also lower than our results. Comparing the results in the present study with other studies show an increased levels of thallium in fish in Musa estuary.

Moreover, the thallium concentration in fish samples found in our study was noticeably greater than those reported in other studies carried out in different regions in the world. Ofukany et al. reported thallium concentration in different fish species in Lake Winnipeg ranged between 0.01 and 0.05 mg kg^{-1} /ww (Ofukany et al. 2014a). Agusa showed that the thallium concentration in three fish samples collected from different areas in coastal waters in Malaysia was 0.001 $\mu g g^{-1}$ dw (Agusa et al. 2005). Ikemoto (Ikemoto et al. 2008) also studied nine different species of fishes in the south of Vietnam and reported the ranges between 0.010 and 0.032 $\mu g g^{-1}/dw$ for thallium concentration, which was less than those reported in our study. It is suggested that high concentrations of thallium in this estuary followed by its high bioaccumulation in fishes such as Cynoglossus arel may increase the risk of thallium toxicity. The relationship between fish length and metal concentration in muscle samples was analyzed by GLM. It is indicated that the concentration of thallium related positively and strongly with this fish spices. It means that thallium concentration is greater in tissue of older and larger fish samples compared with younger and smaller samples. Gantner (Gantner et al. 2009) also showed positive relationships between age, length, and trophic position with thallium concentration and suggested that thallium has a possibility of bioaccumulation and bio-magnification. Ofukany also reports that thallium was negatively related to fish length (Ofukany et al. 2014a). Some other studies also found a positive relationship between body size (length, weight, and age) and trace metal concentration (Farkas et al. 2003; Kasimoglu 2014; Linde et al. 1998; Marijic and Raspor 2006; Mastala et al. 1992).

The acceptable daily intake (ADI) for thallium has been estimated at 0.2 μ g/kg or 14 μ g/70 kg. The reference dose of (RFD) for thallium suggested by the U.S. EPA provides a more cautious value (0.07 μ g/kg/day or 5 μ g/70 kg/day) for protecting the public chronic thallium poisoning (Lin et al. 2001). Therefore, it is suggested that the issue of thallium bioaccumulation and bio-magnification in aquatic food webs and its potential risks by fish consumption as members of food chain should be considered.

Thallium is known to destroy fishes slowly at concentrations of 1–60 ppm. It is also lethal to aquatic insects and invertebrates at 2–4 ppm, and it kills tadpoles at concentrations of 0.4 ppm. Algae may be affected at concentrations as low as 0.1 ppm (Peter and Viraraghavan 2005).

Conclusion

It is concluded that the concentration of two heavy metals, nickel and thallium, was higher than the threshold limits for *Cynoglossus arel* fish in the Musa estuary. Therefore, regarding with high consumption of seafood in this region, it is recommended that these fishes should be consumed under a nutritionist counseling specially in vulnerable groups such as children, pregnant, and breastfeeding women.

Moreover, the elimination of heavy metals from huge pollution sources in the region would be necessary. It seems that reducing the pollutant load in the region would be the best approach to decrease the availability of heavy metals and consequently reduce their accumulation in fish tissues.

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