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The measurement of nickel concentration in muscle tissues of *Otolithes ruber* in Persian Gulf waters, Iran

Abdol-Rahim Pazira[1*], Saeid Moghdani[2], Farshad Ghanbari[2], Narges Javad Zadeh[3]

[1]*Department of Engineering, College of Natural Resources, Islamic Azad University, Bushehr Branch, Iran*

[2]*Young Researchers and Elite Club, Bushehr Branch, Islamic Azad University, Bushehr, Iran*

[3]*Department of Fisheries, Khouzestan Science and Research Branch, Islamic Azad University, Ahvaz, Iran*

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Abstract

In order to examine and compare nickel accumulation levels in muscle tissues of *Otolithes ruber* in Persian Gulf waters (Bushehr province region), sampling the fish was done in both Bushehr and Asalouyeh stations during the summer 2013. After biometry, muscle tissues of the samples were separated and chemical digestion was done. Nickel accumulation levels in tissues were measured by using graphite furnace atomic absorption instrument. Based on the obtained results, mean concentrations of Nickel in muscle tissues were 0.262 ± 0.014 $mgkg^{-1}$ dw in Bushehr station and 0.293 ± 0.011 $mgkg^{-1}$ dw in Asalouyeh station, and it indicated no statistically significant differences between the two stations ($P>0.05$). The obtained concentrations and analysis done indicate that based on the WHO standard, the amount of nickel was higher than the standard levels and its use poses risks to human.

* **Corresponding Author:** Abdol-Rahim Pazira ✉ abpazira@gmail.com

Introduction

An increase in the amount of the released pollutants in marine environment have been considered in a lot of studied during the last decade. One of the main issues attracting a large number of researchers' attention is heavy metal contaminations and their influences on the environment (Henry *et al.,* 2004, Yilmaz *et al.,* 2007). Heavy metals are accumulated in tissues and organs of the fish after they entered the aquatic ecosystems and finally they enter the food chains and are considered as a potentially toxic factor for microorganisms (Chen & Chen., 1999). Heavy metals are entered the marine environments through various anthropogenic resources such as the petrochemical wastewater, mineral and agricultural runoffs, the oil transport and domestic wastewater (Karadede *et al.,* 2004). Age, length, weight, sex, ecological needs, feeding habits, heavy metal concentrations in water and sediments, exposure period of the fish to the aquatic environment, fishing season, and physical and chemical properties of the water (salinity, pH, hardness and temperature) are the effective factors in heavy metal concentrations in different organs of the fish (Canli & Atli., 2003).

Among animal species, fishes are apt to absorb these metals and their harmful influences and have high absorption levels of these metals because of long exposure to the contaminants (Olaifa *et al.*, 2004). Metals are constantly entering the aquatic environment through nature and anthropogenic resources and due to their toxicity, extensive permanency in the environment, bioaccumulation and bio-magnification in food chains, these metals are considered as a serious threat for human health (Papagiannis *et al.*, 2004). Heavy metal contamination chain nearly always follows the following cycle: industry, atmosphere, soil, water, phytoplankton, zooplankton, fish and human (Mendil *et al.*, 2010; Askary Sary and Mohammadi., 2012). Fishes which are usually located in the last levels of aquatic food chains are considered as a main passageway of metals for the transference to human bodies (Svensson *et al.*, 1992; Abdolahpour Monikh *et al.*, 2004).

The Persian Gulf is the progress of the Indian Ocean waters in the south area of Iran on the edge of the Indian Ocean located in west-northern Oman Sea. The Persian Gulf is a shallow water basin with the average depth 35-40 meters and the area about 240 km^2. This region is connected to the international waters via Hormoz strait (Anon, 1995; Banat *et al.*, 1998; Moghdani *et al.,* 2014). This sea is considered as an important resources for accessing to the great food resources that is invaded by the various pollutants in recent years. According to the studies, the water rotation and exchange time of this sea is estimated about 3 to 5 years probably indicating the pollutants remain in the Persian Gulf for a significant period. It's clear that the effect of pollutants on aquatic environment should be more significant due to semi-closed, shallowness, water limited rotation, salinity and high temperature which are the characteristics of the north part of the Persian Gulf (Pourang *et al.*, 2005; Ghanbari *et al.,* 2014). Heavy metals are not only a threat to the aquatic animals but also are considered a great risk to the consumers of marine foods contaminated with these metals (Abel, 1989). Therefore, studies done in the field of heavy metal contamination in aquatic ecosystems are very important from the human health and public sanitation viewpoints. Thus, the objective of this study is to measure the nickel levels in muscle tissues of *Otolithes ruber* in the Persian Gulf waters (Bushehr and Asalouyeh Seaports) and to compare them with the international standards.

Materials and methods

Study area

Bushehr is located in 28°55´19.84" N and 50°50´4.76" E of southwestern Iran and on the edge of the Persian Gulf. Asalouyeh is located in 28°28´24.48" N and 52°36´ 49.79" E on the edge of the Persian Gulf, 300 kms east of Bushehr and 570 kms west of Bandar Abbas and has a distance of 100 kms to the South Pars gas area located along the Persian Gulf.

Sampling

20 samples were caught by trawl net in both regions,

Bushehr and Asalouyeh seaport during summer season 2013 to do this research. Then, the samples were placed in a plastic bag and coded and were placed in an ice bucket full of ice in order to be transferred in the laboratory. The samples were transferred to Islamic Azad University Bushehr branch laboratory after fishing. The fish samples were kept at a temperature of -30° C by the analysis time in the laboratory.

Fig. 1. Location of the sampling areas.

Sample preparation

First all lab dishes which were going to be used were placed in nitric acid for 24 hours and then they were washed by using distilled water and finally they were placed in an oven at a temperature of 80° C to prevent contamination. The samples were removed from the fridge. When they reached the environment temperature, biometry operation (total length, standard length, total weight) was done. Then the muscle tissues were separated and some sample muscles were transferred into the complete clean dishes (washed using nitric acid) and were placed in an oven at a temperature of 80° C for 18 hours to be dried completely . Dried samples were transferred into a mortar to be grinded completely. After grinding the samples they were placed in a desiccator to prevent them absorbing air moisture. Acid digestion is performed to release all metal connections with tissues. In this regard, 1 g dried and constant tissues were transferred into a beaker and 10 ml concentrated nitric acid were added to digest the dish contents and the samples were placed in the room temperature for 30 minutes until the primary digestion was done. Then the samples were heated in a heater located under the hood having a steam system in a temperature of 90° C to be dried. When the samples were cooled and reached the environment temperature, they were sieved from a 45 mm Whatman filter paper and were transferred to 25 ml dishes and were reached the necessary volume. Finally, the samples were transferred into lidded polyethylene dishes to be injected into the instrument (MOOPAM., 1999). A graphite furnace atomic absorption instrument was used to measure the nickel metal levels.

Statistical analysis

One sample Kologorov-Smirnov test in SPSS®18 was used to check the validity of the data normalization. Then, one way sample T-test was used to check interactions between heavy metals and stations. Data have been presented in diagrams as Mean±SDs with 95% of the confidence interval. Excel software was used to draw diagrams (Zar., 1999).

Results

Biometric results

Biometric results indicated that there was no significant different between the mean weight in both stations. Mean weight in Bushehr was 376.82 g and mean weight in Asalouyeh was 369.40 g. Biometric results are presented in table 1.

Table 1. Biometric results of *Otolithes ruber* in Bushehr and Asalouyeh stations (N=10).

	Bushehr Station		Asalouyeh Station	
	Mean	SD	Mean	SD
Total weight	376.82	14.9	369.4	4.96
Total length	34.25	1.46	33.7	0.33
Standard length	30.14	1.55	29	0.67

Nickel concentration

The obtained results show that the lowest and the highest nickel concentration levels in muscle tissues in Bushehr station was equal to 0.175 and 0.350 $mgkg^{-1}$ dw and in Asalouyeh was 0.125 and 0.625 $mgkg^{-1}$ dw, respectively. According to the obtained statistical results mean and standard deviation (SD) with the confidence interval in 95% level of nickel in Bushehr station was 0.262±0.014 $mgkg^{-1}$ dw and in Asalouyeh station was 0.293±0.011 $mgkg^{-1}$ dw. Based on T-test analysis, no significant differences were observed between nickel levels in muscle tissues in both stations (P=0.626). Figure 2 indicates nickel levels in muscle tissues in both Bushehr and Asalouyeh stations.

Table 2. Comparison of Nickel concentrations in muscle tissues of *Otolithes ruber* with WHO standards ($mgkg^{1}$).

Standard	Nickel
WHO (FAO, 1976)	0.2
Otolithes ruber, Bushehr	0.262
Otolithes ruber, Asalouyeh	0.293

Table 3. Comparison of nickel concentrations in the present study with the other researches ($mgkg^{-1}$).

Refrence	Ni	Region	Species
Pourang *et al.*, 2005	1.56	Northern Persian Gulf	*Epinephelus coioides*
Pourang *et al.*, 2005	1.2-14.5	Northern Persian Gulf	*Solea elongates*
Pourang *et al.*, 2005	0/1-5.8	Northern Persian Gulf	*Psettodes erumei*
Bu-Olayan., 1996	3.6	Persian Gulf, Kuwait shores	*Solea bleekeri*
Bu-Olayan., 1996	2.3	Persian Gulf, Kuwait shores	*Gastrophysus lunaris*
Bu-Olayan., 1996	4.7	Persian Gulf, Kuwait shores	*Acanthopagarus latus*
Bu-Olayan., 1996	0.9	Persian Gulf, Kuwait shores	*Mugil macrolepis*
Bu-Olayan., 1996	20.4	Persian Gulf, Kuwait shores	*Sillego sihana*
Ashraf., 2005	1.62	Eastern shores of Saudi Arabia	*Epinephelus microdon*
Abdolahpour Monikh *et al.*, 2012	4.32	Musa estuary, Persian Gulf	*Johnius belangerii*
Abdolahpour Monikh *et al.*, 2012	6.27	Musa estuary, Persian Gulf	*Euryglossa orientalis*
Abdolahpour Monikh *et al.*, 2012	3.41	Musa estuary, Persian Gulf	*Cynoglossus arel*
Fatih fidan *et al*, 2008	0.05-0.12	Eber lake, Turkey	*Carassius carassius*
Alkan *et al.*, 2012	0.02-0.67	Black sea	*Bullus barbatus ponticus*
Alkan *et al.*, 2012	0.01-0.71	Black sea	*Merlangius merlangus euxinus*
Moghdani *et al.*, 2014	1.378	Persian Gulf, Bushehr	*Brachirus orientalis*
Moghdani *et al.*, 2014	2.208	Persian Gulf, Asalouyeh	*Brachirus orientalis*
Current study	0.262	Persian Gulf, Bushehr	*Otolithes ruber*
Current study	0.293	Persian Gulf, Asalouyeh	*Otolithes ruber*

Based on the obtained concentrations and comparison done it as specified that based on WHO standard, the amount of nickel in *Otolithes ruber* tissues was higher than the standard permissible levels.

Discussion

Nowadays all animals' habitats are in humans' hands more than any other time. Human activities are the main factor of pollution in nature and different types of the environmental polluting resources are also created with the industry development and day by day they are increasing. Mining oil resources, oil tanker traffic, discharging industrial and domestic waste and drainage and agricultural drainage and production of petrochemical materials are among the destruction factors and consequently the destruction of aquatic environment. The presence of these pollutants cause different chemical combinations especially heavy metals to enter the aquatic ecosystem (Wicker & Gantt, 1994; Plaskett & Potter, 1979). *Otolithes ruber* is one of the migratory and coastal fish. This species is found in coastal waters highly in regions with muddy bed. The species is benthopleagic to the effect that they live both in bed and water surface. *Otolithes ruber* feeds on smaller fish, crustaceans like shrimps and the other invertebrates (Moghdani *et al.*, 2014). Heavy metal accumulation in benthopelagic species in comparison with benthic species probably has a relation with the fish diet (Boustamant *et al.*, 2003). However, these findings can prove that mental concentrations are highly under the influence of habitat, feeding habits, mental accumulation capacity and kind of species (Bustamante *et al.,* 2003; Agah *et al.,* 2009). In this study, the muscle was chosen as the purpose organ since it has an important role in human feeding and the necessity of the assurance to its health for consumption has a particular importance.

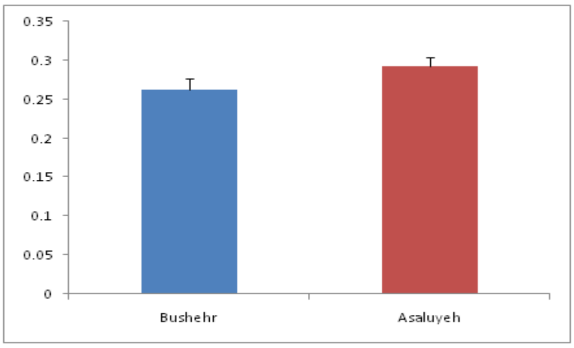

Fig. 2. Comparison of nickel levels in muscle tissues of *Otolithes ruber* in Bushehr and Asalouyeh stations.

The results of this study generally showed that the measured nickel levels in both Bushehr and Asalouyeh stations didn't have significant differences. Bushehr which is a fishing region is also a place for mooring the fishing boats. Beside this fishing pier, the Bushehr nuclear power station, the customs, and the National Shrimp Research Institute was located. The National Shrimp Research Institute has some shrimp farming pools and its discharge drainage is directly discharged into the sea. In Asalouyeh station, in addition to the fishing pier which is located near Asalouyeh city, the largest world gas and oil installations, South Pars oil particular region are located there influencing the environment directly and indirectly. All installations of the South Pars region located near the Asalouyeh city and pier are located along the shoreline due to the location of oil and gas fields in the sea and also several oil rigs are located on the sea near it. Examination of the obtained results shows that the nickel levels in tissues of *Otolithes ruber* in Asalouyeh station (0.293±0.011) and Bushehr station (0.262±0.014) are higher than the international permissible levels. Nickel is widely spread in the environment and its concentration is a function of fossil fuel and its meaning from the oil fields and refineries. In other words we can say that the highest concentrations of nickel in sediments are mainly caused by human sources such as ship traffic, oil tankers, petroleum and industrial and domestic wastewaters (De Astudillo *et al.*, 2005; Pourang *et al.*, 2005; Beg *et al.*, 2001).

Nickel toxicity is divided into 4 groups: (1) allergy (2) cancer (3) respiratory disorders (which all these three cases are caused by industrial activities) and (4) iatrogenic poisoning (Amundsen *et al.*, 2007).

World Health Organization (WHO) hasn't specified the bearable absorption amount of nickel. The environment Protection Agency of the United States (EPA) suggested the maximum permissible concentrations, 20 $mgkg^{-1}$ per day and the maximum bearable daily mean 1/2 mg for a 60 kg person. However, this amount cause skin inflammation in people having greater sensitivity to nickel (Esmaeili Sari., 2002). Based on the studies which are done by Mashinchian Moradi (Mashinchian Moradi., 1993) in Hormuz strait and areas around the Persian Gulf, elements like nickel in these regions have mines under the Oceanic crust. Thus, another important and significant reason for the high amounts of mean nickel concentrations is its crust origin. Moreover, the Persian Gulf has caused various contaminations by petroleum spillage in the Persian Gulf aquatic ecosystem due to its huge oil and gas resources, ship traffic and abundant oil tankers and refinery activities. De Mora *et al.* (De Mora *et al.*, 2004) by examining the nickel concentrations in south sediments of the Oman sea found that the segments of the coasts have high concentrations of nickel. These researchers found that nickel in sediments of this regions has mainly natural origin which have introduced Ophiolites stone in bed as its origin. Nickel combinations have rather high toxicity and this toxicity increases in the presence of zinc. Nickel is accumulated in fish livers, gills, kidneys and muscles (Van-Doijn., 2000).

Eslami *et al* (Eslami *et al.*, 2011) determined the concentrations of some heavy metals in muscle tissues of *Rutilus frisii Kutum* in Tajan river. The results indicated that the nickel metal levels in muscle tissues were equal to 2.650±0.094 µgg-1 which this metal along with lead which are among non-essential metals had higher levels in muscles in comparison to essential metals. Moreover, the existing nickel levels in muscle tissues were higher than the international standards.

According to the results of the study of Fatih Fidan *et al.* (Fatih Fidan *et al.*, 2008) which was done on the heavy metal concentration levels in muscle tissues, gills and livers of *Carassius Carassius* in Eber lake in Turkey, the highest concentration levels of nickel in muscle were related to the winter season with the amount of 0.12±0.03 µgg-1 fish weight. In addition, Alkan *et al.* (Alkan *et al.*, 2012) in the study on heavy metal levels in spices including *Mullus barbatus ponticus* and *Merlangius merlangus euxinusin* in southwestern region of the Black sea, calculated the nickel levels in spices *Mullus barbatus ponticus* 0.02-0.67 µgg-1 and in species *Merlangius merlangus euxinus* 0.01-0.71 µgg-1 respectively.

Based on the obtained concentrations and the performed comparisons it was specified that according to the WHO standards, the amount of nickel in muscles of *Otolithes ruber* were higher than the standard permissible levels. Therefore, based on this research finding, we can conclude that the Persian Gulf environment has high oil contamination so that nickel which is one of the oil metals is accumulated higher than the international standard rates in the studied fish muscles, and if it is used by human beings, it will certainly cause some harm for them.

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