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Research Article

An evaluation of target hazard quotation of mercury and Arsenic in four commercially fish species of the Oman Sea, Iran

Narjes Okati *, Mohsen Shahriari Moghadam, Fatemeh Einollahipeer

[1] Department of Environment, Faculty of Natural Resources, University of Zabol, Zabol, Sistan and Baluchestan, Iran

*Email: narjesokati@uoz.ac.ir

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Abstract

Increasing the presence of mercury (Hg) and Arsenic (As) in aquatic ecosystems as two unnecessary and dangerous elements in the environment has raised many concerns worldwide. This research aimed to evaluate associated risks of Hg and As in four fish species (*Lethrinus crocineus*, *Otolithes ruber*, *Rhabdosargus haffara*, and *Epinephelus coioides*) that are generally consumed by the people residents on the Coast of the Oman Sea in Iran. The maximum of the mean Hg and As concentrations were 0.38 ± 0.014 and 0.94 ± 0.124 µg g-1 wet weight (ww) for Orange-spotted grouper (*Epinephelus coioides*), respectively. Target hazard quotient (THQ) values of Hg for *Epinephelus coioides* and Tiger tooth croaker (*Otolithes ruber*) were higher than 1 for children and adult groups, but all THQ values of As were below one. The lifetime cancer risk (CR) for inorganic As was above 10-5. Estimation of health risks of Hg and As showed that there are no consumption limits for children and adults due to the amount of As in fish tissue, but the consumption of *Epinephelus coioides* and *Otolithes ruber* for both children and adult groups indicated the potential risk for them.

Keywords: Bioaccumulation, Fish, Oman Sea, risk assessment, toxic elements

Introduction

Marine ecosystems are being polluted by the adverse effects related to the development of anthropogenic and industrial activities. Untreated depletion of wastewaters is among the significant origin of pollution in aquatic environments (Attaran-Fariman, 2010). High concentrations of toxic metals affect living organisms and create substantial environmental risks (Kortei et al., 2020).

Increasing mercury (Hg) and Arsenic (As) levels in aquatic ecosystems as two unnecessary and dangerous elements in the environment, through natural and anthropogenic sources, has raised many concerns around the world (Wang et al., 2014; Okati & Esmaili-sari, 2018). The toxic elements exert their effects through various mechanisms, being chronic exposures at low doses of complex metal mixtures the responsible for the effects observed in wild animal populations and communities, with implications at the ecosystem level. Hg and As can be transported from plants to higher strata of the food chain, representing the threat to biodiversity and ecosystem integrity (Tovar-Sánchez et al., 2018). The toxicity of these elements in the environment is due to their non-biodegradability and accumulation in various plant and animal tissues (Rahman et al., 2013). These pollutants are stored in aquatic animals' tissues, such as fish, and bioaccumulate in the marine ecosystem along the food chain (Giri & Singh, 2014). People can be exposed to Hg and As in the environment or through their food. Most of the Hg and As in the food referred to the fish consumption (Okati & Esmaili-sari, 2018; Kortei et al., 2020). Higher bioavailability of Hg and As and higher biological half-life, as well as their potential to be accumulated in different parts of the body, make them serious contaminants, whose risks of toxicities in humans are one of the significant global public health concerns (Raissy & Ansari 2014, Kumari et al., 2016, Schneider et al., 2018). Mercury is a toxic, dangerous element that can severely impact the central nervous system (CNS) and kidney. It can also create deleterious effects on humans' respiratory and psychological issues (Gyimah et al., 2018). The harmful health effects of As poisoning in humans are multiple. The impact of acute As toxicities include skin rash, toxic cardiomyopathy, abdominal pain, vomiting, and diarrhea (Ratnaike, 2003).

The positive effect of fish meat on human health and its characteristics as a rich source of essential elements, amino acids, vitamins, and omega-3 (Silva et al., 2019) made them a healthier dietary food (Farrugia et al., 2015). Therefore, rigid national and regional standards should be applied to prevent the risk of such toxic elements in human diets through fish consumption (Shahbazi et al., 2016). Many countries in the world evaluate the amounts of harmful ingredients in fish that are locally consumed to conserve their health. The European and Food Safety Authority (EFSA) established a range of benchmark doses between 0.3 and 8 μg per kg body weight (BW) per day for As (ESFA, 2009). Also, a potential total weekly intake (PTWI) of 5 μg per kg BW per week for total Hg has been set by the Joint Food and Agriculture Organization/World Health Organization Expert Committee on Food Additives (JECFA) (JECFA, 2014). The EFSA established the PTWI value for Hg of 1.6 μg per kg BW per week, which is sufficient to preserve the embryo and fetus from neurodevelopment hazard risk (ESFA, 2009).

Various methods have been proposed to estimate the effects of carcinogenic and non-carcinogenic effects of different pollutants on human health by consuming polluted fish (Usese et al., 2017). The evaluation of health risk through Target Hazard Quotients (THQ) estimation methodology is an accurate and acceptable method initially proposed by the United States Environmental Protection Agency (USEPA), which has been cited by many researchers (Wang et al., 2005; Gu et al., 2017). Because there is limited information on the health risk assessment of fish consumption for fish species of the Oman Sea (Ziyaadini et al., 2017), and fish consumers commonly do not have sufficient information to select the healthy fish for their diets. Since fish is served as the primary food item for the people in the Oman Sea coastal area (including Iran and the other littoral countries of the Oman Sea), we compared the concentrations of Hg and As in fish species with the permissible levels. We also aimed to evaluate associated risks of Hg and As in four fish species on the shorelines of the Oman Sea for adults and children in Iran.

Material and methods

Study area

The Oman Sea (Fig.1) is a triangular strait situated between Iran, Oman, and Pakistan. It is surrounded by land on three sides and connected to the Indian Ocean high Sea on the other hand. The Strait of Hormuz joined it to the Persian Gulf. Between November 2018 and February 2019, a total of 51 samples from four commercially essential and highly consumed fish were analyzed. The selected species were Orange-spotted grouper, *Epinephelus coioides*; Tiger tooth croaker, *Otolithes ruber*; Stump nose, *Rhabdosargus haffara*, and Yellow tail emperor, *Lethrinus crocineus*, (Table 1). Specimens were purchased from fishers and fish markets in Chabahar city located on the north coasts of the Oman Sea in Iran (Fig.1) and processed according to the USEPA guidelines (USEPA, 2000). The total length and weight of the fish samples were recorded before cutting. The specimens were separately packaged in polyethylene bags with a numbered label and kept at four °C in portable refrigerators while they were transported to the laboratory.

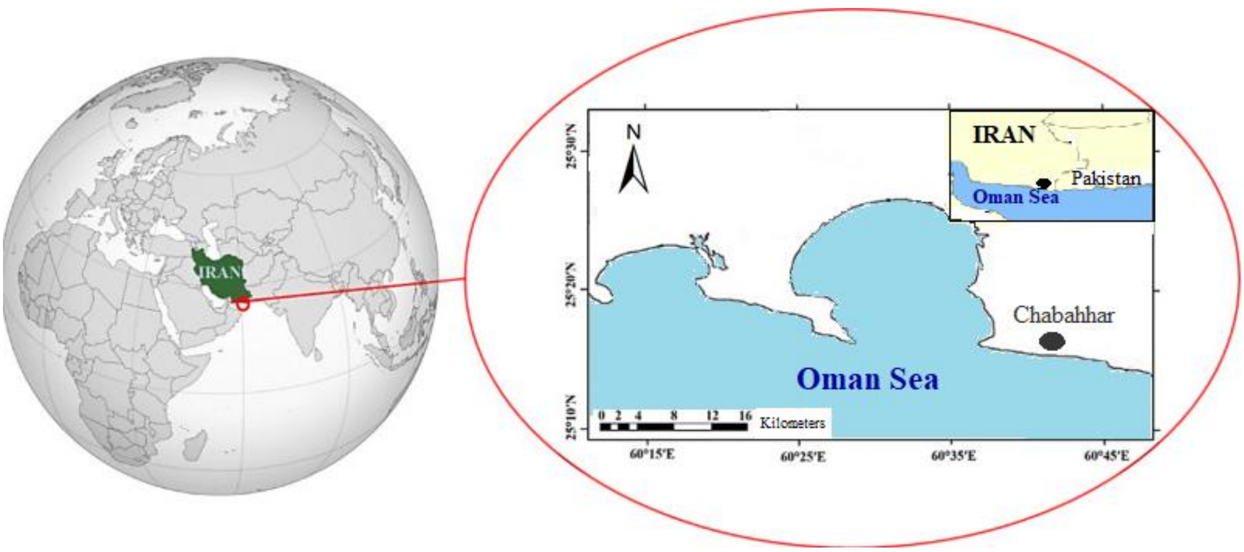

Figure 1. The map of the study area

Sample preparation and analysis

At first, fish samples were cleaned several times with deionized water, put in the polyethylene bags, and kept in a freezer at -20°C until experiment time (USEPA, 2000; Malakootian et al., 2016). Approximately 100 g of each sample's lateral-dorsal muscle was dissected, and then freeze-dried (Freeze dryer OPERON, Model; FDCF-12012) at -54°C for 24 h to make them thoroughly dried, and then were converted to powder using pounder (Okati & Esmaili-sari, 2018). The sample was weighed before and after freeze-drying. Then we calculated the moisture percentage of the muscle tissues. All containers used were washed with dilute nitric acid, distilled water, and dried, respectively.

Table 1. Sampled fish species and biometric characteristics of fish

Species name		n	The total weight(g) Mean±SE	Total Length(cm) Mean±SE
Common name	**Scientific name**			
Orange-spotted grouper	*Epinephelus coioides*	10	1750.0±54.26	69.90±0.98
Yellowtail emperor	*Lethrinus crocineus*	15	1020.8±50.1	30.75±0.92
Tiger tooth croaker	*Otolithes ruber*	12	1250.0±33.7	36.91±1.25
Stump nose	*Rhabdosargus haffara*	14	1078.6±77.1	32.64±1.42

For Arsenic measurement, about one gram of the dry tissue sample (muscles) was precisely weighed and digested with six ml of concentrated HNO_3 65% (Merck, Darmstadt, Germany) and left at room temperature for 12 hours. Then, we added four ml of H_2O_2 30% (Merck) to it. The polyethylene tubes were placed on a digestive apparatus at 80 °C for 3 hours and a temperature of 150 °C for digestion (Liu et al., 2018; Salgado-Ramírez et al., 2017). After completing the digestion procedure, the solution was filtered using Whatman filter paper 42 and polyethylene funnel in a 25-ml balloon. Finally, using deionized water, the volume of the solution was diluted to 25 ml. Using a HP-4500 (made in the USA) equipped with the Asus-520 Autosampler, the amount of Arsenic was read at the wavelengths of 193.7 nm using ICP-MS. Then, the concentration of As calculated using the following Eq. (1) (Okati et al., 2020):

$$M = \frac{(C \times V)}{w} \times A \quad (1)$$

M: concentration of As (μg g^{-1}), C: concentration of the device (μg l^{-1}), V: final volume of the sample in l (0.125), W: prototype weight for acid digestion (g), and A: dilution factor

In fish muscle tissue's inspection regarding the Arsenic remainings, the concentration of total mercury (THg) in the sample is measured rather than methyl mercury (MeHg) because of the more costly MeHg analyses in comparison of THg. It is admitted that in the health risk assessment experiments, 100% of THg is supposed to be MeHg In this way, caution is taken (Health Canada, 2007; Doke & Gohlke, 2014; Garrigues, 2015). So, in this study, we have measured THg concentration in muscle tissue of fish samples. To detect the Hg amount, about 0.03 to 0.05 g dry weight of each sample was placed in the mercury analyzer's nickel boat. LECO AMA 254 Advanced Mercury Analyzer (USA), as stated in ASTM, standard no. D 6722, at a wavelength of 253.7 nm, was applied to analyze Hg in the studied samples. Detection of Hg in samples using LECO AMA 254 is without sample pretreatment or sample pre-concentration (Okati & Esmaili-sari 2018; Khoshnood et al., 2012).

Quality control

The standard solutions were provided from stock solutions (Merck, multi-element standard). We replicated measurements three times for Hg and As. The accuracy and precision of the obtained concentrations were compared by inspecting the certified reference material (CRM, Dorm 2) prepared from the dogfish muscle, National Research Council, Canada. Analytical quality control (AQC) for the determination of As and Hg in fish muscles are summarized in table 2. Our results demonstrated that there was good communication among certified and analytical data. The used Standard Reference materials (SRM) for checking the accuracy of the Hg analysis were three samples of the National Institute of Standard and Technology (NIST) (NIST 1633b, NIST 2709, & NIST 2711a) (Okati & Esmaili- sari, 2018). The percent of Recovery for Hg was in the range of 98.7 to 103.6%. The detection limits (LOD) for Hg and As were obtained 0.001 and 0.01 μg g^{-1} of dry weight, respectively. The reproducibility of the methods was examined by the three times detection of 10% of the samples. The coefficient of variation of Hg and As ranged between 0.05-2.5% and 1-4.2%, respectively.

Table 2. Results of analytical quality control for the determination of As and Hg (data as means in μg g^{-1} dry weight)

Element	Reference Material	Certified	Observed[1]	SD[2]	Recovery (%)
Arsenic	CRM-DORM[3]-2	0.065	0.069	0.009	94.2
Mercury	NIST[4]-1633b	0.141	0.138	0.015	103.6
	NIST-2709	1.400	1.412	0.146	99.1
	NIST-2711a	7.420	7.511	0.419	98.7

[1] Each value is the mean of 10 analysis.; [2] Standard Deviation; [3] Certified Reference Material -Dogfish muscle; [4] National Institute of Standard and Technology

Human health risk through fish consumption

The health risk assessment, as stated by the levels of As and Hg through human consumption of fish, was estimated using the target hazard quotient (THQ) and Cancer Risk (CR) according to the USEPA guideline (USEPA, 2011). The THQ, which displays the risk of non-carcinogenic effects, implicates non-obvious risk. If the THQ is lower than one (THQ<1), it is unlikely that person will experience evident adverse effects during a person's lifetime, and if THQ>1, there is a potential health hazard (Taweel & Ahmad, 2013; Traina et al., 2019). The equation for estimating THQ was as follows (Eq. 2):

$$\mathrm{THQ} = \frac{\mathrm{MC} \times \mathrm{IR} \times 10^{-3} \times E\mathrm{F} \times \mathrm{ED}}{\mathrm{RfD} \times \mathrm{BW} \times \mathrm{AT}} \quad (2)$$

Where, MC: The mean As and Hg concentration in fish species (μg g^{-1}, ww)

IR: Ingestion rate (g day^{-1}) = 29.23 g day^{-1} (AFS, 2010). In this study, an IR for children was assumed to equal 60% of IR for adults = 17.53; BW is body weight (70 kg for adults and 32 kg for children) (USEPA, 2009).

EF: exposure frequency (365 days $year^{-1}$)

ED: Exposure duration (adults: 70 years; children: 10 years) (USEPA, 2011)

RfD: reference dose (μg g^{-1} day^{-1}); (1×10^{-4} μg $g^{-1}day^{-1}$ for Hg and 3×10^{-4} μg $g^{-1}day^{-1}$ for As) (USEPA, 2011)

BW: body weight (kg); (adults: 70 kg; children: 32 kg)

AT: Averaging time, non-carcinogens (day $year^{-1}$) = (365×ED) (USEPA, 2011)

Because people are usually exposed to more than one pollutant with combined or synergistic effects (Li et al., 2013; Traina et al., 2019), the total target hazard quotient (TTHQ) was estimated as the sum of the THQ_i amounts (Eq. 3):

$$TTHQ = \sum THQ_i \quad (3)$$

International Agency for Research into Cancer (IARC) classified Arsenic as a carcinogenic element. To assess the possibility for an individual to develop cancer over a lifetime, as a result of exposure to this potential carcinogen, the lifetime cancer risk (CR) was calculated. It was estimated with Eq. (4) (USEPA, 2010):

$$CR = \frac{MC \times IR \times EF \times ED \times CSF}{BW \times AT} \quad (4)$$

CSF: Cancer Slop Factor

Since CSF (µg g^{-1} day^{-1}) is the oral carcinogenic slope factor from the Integrated Risk Information System (IRIS) (USEPA, 2010), CSF values are presented only for Arsenic (1.5 µg g^{-1} day^{-1}). As so long, the amount is higher than 10^{-5}, and it shows the chance greater than one over 100,000 of an individual of expanding cancer (USEPA, 2014). AT for carcinogenic effects is 70 years × 365 days $year^{-1}$ (Vieira et al., 2011; Traina et al., 2019). It is incorrect to estimate THQ for total Arsenic since the oral RfD set by the USEPA is for inorganic As only. Hence, we determined the risk factors (THQ, CR, and CR_{lim}) for the inorganic Arsenic, assuming that 3% of the total arsenic concentration is the inorganic form (Vieira et al., 2011; ESFA, 2012; Martinez-Gomez et al., 2012; Copat et al., 2015; Traina et al., 2019).

The maximum allowable concentration rate per day CR_{lim} (Kg day^{-1}) is calculated according to the concentration of contaminants stored in the muscle of fish (Eq. 5) set by the USEPA. It was estimated for adult, and children with an age of 10 years old (32 kg BW) (USEPA, 2000):

$$CR_{lim} = \frac{RfD \times BW}{C_m} \quad (5)$$

Also, CR_{lim} (Kg day^{-1}) can be applied to estimate the maximum allowable fish consumption rate per month (CR_{mm}) with Eq. (6) (USEPA, 2000):

$$CR_{mm} = \frac{CR_{lim} \times T_{ap}}{MS} \quad (6)$$

T_{ap}: time averaging period (365 days per year and 30.44 days per month)

MS: meal size (0.227 kg for adult and 0.136 kg for children)

Statistics

All statistical analysis was carried out using the SPSS software packages, version 17.0. Hg Mercury and As concentrations were tested for normality by the Kolmogorov-Smirnov test. Data were normality distribution. Levene's test showed the homogeneity of variances. So, the analysis of variance one way ANOVA with Tukey posthoc test was performed to survey differences between group means of fish species for significance ($P<0.05$). Linear regression with Pearson's correlation analysis was conducted to assess the relationship between metals concentrations and each fish species' length.

Results

The mean Hg and As concentrations in the fish muscles are summarized in Table 3. The lowest and highest of the mean ± SE for Hg and As concentrations in sampled fish were obtained for *Lethrinus crocineus* (Hg: 0.05 ± 0.010 µg g^{-1} ww; As: 0.15 ± 0.017 µg g^{-1} ww), *Otolithes ruber* (Hg: 0.26 ± 0.021 µg g^{-1} ww; As: 0.80 ± 0.062 µg g^{-1} ww), *Rhabdosargus haffara* (Hg: 0.10 ± 0.014 µg g^{-1} ww;

As: 0.19 ± 0.029 µg g^{-1} ww), and *Epinephelus coioides* (Hg: 0.38 ± 0.014 µg g^{-1} ww; As: 0.94 ± 0.124 µg g^{-1} ww), respectively. The results of statistical analysis showed that there is a significant difference between the concentration of Hg between all fish species ($p<0.001$) (Fig. 2a). Also, the mean As concentrations of As in *Epinephelus coioides* and *Otolithes ruber* were significantly ($p< 0.001$) higher than *Lethrinus crocineus* and *Rhabdosargus haffara* (Fig. 2b). The comparison of Hg and As concentrations in fish species to the maximum standard level of JECFA and USEPA safety level showed in Fig. 2 (a, b). In this study, the mean Hg concentrations in all fish species did not increase the recommended maximum standard level of JECFA for Hg in fish (0.5 µg g^{-1} ww). According to the USEPA safety level of 0.3 µg g^{-1} ww (USEPA, 2009; Vahabzadeh et al., 2013), only the mean Hg concentration of *Epinephelus coioides* (0.38 ± 0.014 µg g^{-1} ww) was exceeded from this level (Fig. 2 (a)). Although the mean concentration of As in *Epinephelus coioides* (0.94 ± 0.124 µg g^{-1} ww) was not exceeded than the JECFA standard level for As in fish (1 µg g^{-1} ww) (JECFA, 2014), but As levels were higher than this level in some of the samples of *Epinephelus coioides* (n = 4) (Fig. 2 (b)). Linear regression showed that the relationship between Hg concentrations with fish length was significant for all fish species (Fig. 3). The significant positive correlations were seen between Hg concentrations with fish length for *Epinephelus coioides* ($r = 0.61$; $p = 0.03$), *Otolithes ruber* ($r = 0.61$; $p = 0.03$), *Rhabdosargus haffara*: ($r = 0.61$; $p= 0.03$), and *Lethrinus crocineus*: ($r = 0.61$; $p = 0.03$), respectively (Fig. 3). In contrast, there was no significant correlation between As levels in muscle tissue and the length of fish species (Fig. 3).

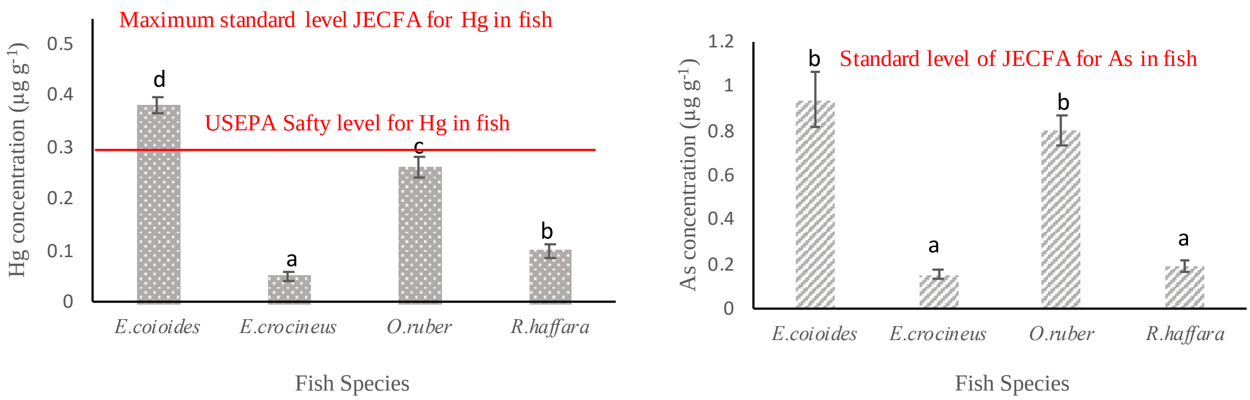

Figure 2. The comparison of Hg and As concentrations in intra fish species and international standards in fish (µg g^{-1}, ww) (JECFA, 2014; USEPA, 2011)

THQ values of Hg for *Epinephelus coioides* and *Otolithes ruber* were higher than 1 for children and adult groups, but all THQ values of As were below than 1 (Table 4). TTHQ amounts were higher than 1 in *Epinephelus coioides* and *Otolithes ruber* for children and adult groups (Table 4). The CR_{As} values were found to range between 5.2×10^{-9} for *Lethrinus crocineus* (for children) to 1.7×10^{-7} for *Epinephelus coioides* (for adults) (Table 4). The human health risks assessments due to the usage of Hg and As contents polluted fish are given according to CR_{lim} and CR_{mm} for the two groups of population (children and adults) in Table 5. According to Hg concentration in fish species, the minimum and maximum allowable consumption rates were obtained for *Epinephelus coioides* for children (0.008 kg day^{-1} ~ 1.8 meals $month^{-1}$), and *Lethrinus crocineus* for adults (0.140 kg day^{-1} ~ 18.7 meals $month^{-1}$), respectively. According to As levels in fish, the minimum and maximum allowable consumption rates were obtained for *Epinephelus coioides* for children (0.34 kg day^{-1} ~

76 meals month^{-1}) and *Lethrinus crocineus* for adult (4.66 kg day^{-1} ~ 625 meals month^{-1}), respectively.

Table 3. The results of measuring the mercury and arsenic concentrations in (μg/g, ww) in fishes of Oman Sea

Taxon	Mercury concentration					Arsenic concentration				
	Mean	Minimum	Maximum	[a] SE	[b] SD	Mean	Min.	Max.	SE	SD
Epinephelus coioides	0.38	0.32	0.44	0.014	0.044	0.94	0.24	1.65	0.124	0.392
Lethrinus crocineus	0.05	0.01	0.12	0.010	0.033	0.15	0.03	0.25	0.017	0.068
Otolithes ruber	0.26	0.10	0.38	0.021	0.072	0.80	0.56	1.23	0.062	0.215
Rhabdosargus haffara	0.10	0.04	0.21	0.014	0.053	0.19	0.03	0.38	0.029	0.111

[a] SE: Standard Error of Mean; [b] SD: Standard Deviation; [c] Min.: Minimum; [d] Max: Maximum

Table 4. THQ, TTHQ and CR for each analyzed metals in different fish species for children (C) and adult (A) groups

Scientific name	THQ_{Hg}		THQ_{As}		TTHQ		CR_{As}	
	C	A	C	A	C	A	C	A
Epinephelus coioides	2.08	1.58	0.051	0.039	2.13	1.62	3.3E-08	1.7E-07
Lethrinus crocineus	0.27	0.20	0.008	0.006	0.28	0.21	5.2E-09	2.8E-08
Otolithes ruber	1.42	1.08	0.043	0.033	1.46	1.11	2.8E-08	1.5E-07
Rhabdosargus haffara	0.54	0.41	0.010	0.007	0.55	0.42	6.7E-09	3.5E-08

Discussion

Hg and As levels in fish species

In this study, 51 individual fish caught from the Oman Sea were studied for Hg and As contents in muscle tissues. The mean concentrations of Hg in fish species were summarized in table 3. The mean concentrations of Hg and As in fish species followed *E. coioides*> *O. ruber*> *R. haffara*> *L. crocineus*.

Table 5. CR_{Lim} (kg day^{-1}) and CR_{mm} (meals month^{-1}) for mercury and Arsenic in different fish species for children and adult groups

Scientific name	CR_{Lim}				CR_{mm}			
	Mercury		Arsenic		Mercury		Arsenic	
	C[a]	A[b]	C	A	C	A	C	A
Epinephelus coioides	0.008	0.018	0.34	0.74	1.8	2.4	76	99
Lethrinus crocineus	0.064	0.140	2.13	4.66	14.3	18.7	477	625
Otolithes ruber	0.012	0.027	0.4	0.87	2.7	3.6	89	117
Rhabdosargus haffara	0.032	0.07	1.68	3.6	7.1	9.3	376	494

a: Children; **b**: Adult

The comparison of Hg and As concentrations in fish species to the maximum standard level of JECFA and USEPA safety level showed in Fig. 2 (a, b). In this study, the mean Hg concentrations in all fish species did not increase the recommended maximum standard level of JECFA for Hg in fish (0.5 µg g^{-1} ww). According to the USEPA safety level of 0.3 µg g $^{-1}$ ww (USEPA, 2009;

Vahabzadeh et al., 2013), only the mean Hg concentration of *Epinephelus coioides* (0.38 ± 0.014 μg g^{-1} ww) was exceeded from this level (Fig. 2 (a)). Although the mean concentration of As in *Epinephelus coioides* (0.94 ± 0.124 μg g^{-1} ww) was not exceeded than the JECFA standard level for As in fish (1 μg g^{-1} ww) (JECFA, 2014), but As levels were higher than this level in some of the samples of *Epinephelus coioides* (n = 4) (Fig. 2 (b)). According to standard values of Hg and As in fish, it might be a health hazard about the concentration of Hg, and As in *Epinephelus coioides* caught off the Oman Sea. The great concern about the Hg and As toxicity in humans is related to low levels of Hg and As in their diet linked to the potential neurotoxicity of MeHg and acutely toxic of As in both children and adults (Authman et al., 2015).

The comparison of the Hg and As concentrations (μg g^{-1} ww) in fish of Oman Sea with other researchers showed in Table 6. However, in our study, the Hg levels of 4 four studied fish species were lower than other studies (Storelli et al., 2005; Al-Reasi et al., 2007; Saei-Dehkordi et al., 2010; Martinez-Gomez et al., 2012; Alina et al., 2012; Kortei et al., 2020) or even higher than others (Fu et al., 2010; Thiyagarajan et al., 2012; Traina et al., 2019). Also, in this study, the As levels were below than other researches (Shah et al., 2009; Julshamn et al., 2012; Leung et al., 2014; Traina et al., 2019; Ranjbar Vakil Abadi et al., 2015), and they were higher than other studies (Fu et al., 2010; Raissy & Ansari, 2014; Leung et al., 2014; Alina et al., 2012; Martinez-Gomez et al.,, 2012; Storelli et al., 2005; Ranjbar Vakil Abadi et al., 2015; Kortei et al., 2020). Mercury and As levels are different in various studies. Its reason might be related to other factors, such as modality nature and intensity of pollution, alkalinity, water pH, temperature, the physiology of the studied fishes, body weight and age, habitat, trophic level, and also the period of the study (Raissy & Ansari 2014; Kortei et al., 2020).

There was a significant difference of Hg ($p < 0.001$) between all fish species using one way ANOVA test (Fig. 2a). Hg concentration in *E. coioides* significantly higher than *O. ruber*, *R. haffara*, and *L. crocineus*. As contents in *E. coioides* and *O. ruber* were significantly ($p < 0.001$) higher in comparison with *R. haffara*, and *L. crocineus* (Fig. 2b). The differences of toxic metals concentrations such as Hg and As in fish species could be the result of the different habitats and ecological factors such as environmental requirements, metabolic regulations, growth, breeding, food supply, and environmental situations, feeding patterns, behavior, and the bio-concentration capacity of fish species, and period of study (Rozon-Ramilo et al., 2011; Monikh et al., 2013; Traina et al. 2019). Thus, the variations in levels of total As between the fish species and between individuals within a specific fish species could result from the different factors, such as the variability of As in the prey existing for the fish (Julshamn et al., 2012). Atobatele and Olutona (2015), showed the significant differences in As concentrations of fish species from Aiba Reservoir, Iwo, Nigeria. Today, it has become clear that feeding is the leading way to intake Hg, especially MeHg, in aquatic animals (Borgå et al., 2012). MeHg can easily store within animals' tissues at significantly higher concentrations than its concentration in the water and the sediments in the marine ecosystem (Beltrame et al., 2010). Fish species at high trophic levels generally have more Hg levels in their tissues than fish that locate in the low food levels (Vieira et al., 2011, Lavoie et al., 2013). In this study, all sampled fish were carnivores, and *E. coioides* had the highest Hg and As concentrations than other studied fish species. An increase of Hg and As concentrations in *E. coioides* have usually been ascribed to its habitat and feeding pattern. These fish species tend to be near the sediment region (Bahnasawy et al., 2009; Elnabris et al., 2013). Mirzaei et al. (2016)

demonstrated the heavy metals present in water and sediments of the Oman Sea along the Chabahar coast might be due to discharging the sewage directly into a nearby body of water, discharged wastes from factories and industrial zone, chemicals of industrial effluent and products of ship and boats.

Relationships between Hg and As concentrations with fish length

The results of linear regression showed that the relationship between Hg concentrations with fish length was significant for all fish species (Fig. 3). The significant positive correlations were seen between Hg concentrations with fish length for *Epinephelus coioides* (r = 0.61; p = 0.03), *Otolithes ruber* (r = 0.61; p = 0.03), *Rhabdosargus haffara*: (r = 0.61; p= 0.03), and *Lethrinus crocineus*: (r = 0.61; p = 0.03), respectively (Fig. 3). The highest correlation was recorded for *E. coioides*. In contrast, there was no significant correlation between As levels in muscle tissue and the length of fish species (Fig. 3). While Besada et al. (2006), could not find any correlation between the size of fish and the As concentration, but Miloškovic and Simić (2015), reported the higher significant correlation among total As the level in muscle tissue and fish size (correlation coefficient= 1.00; p < 0.05) of pike. Also, Milačič et al. (2017) found no As bioaccumulation in some fish species. They reported As the level in fish muscle to a large extent reflects As the level in water-soluble. According to De Gieter et al. (2002), and Copat et al. (2015), interspecies differences in As concentrations of fish are related to their available food and the reality that As is metabolized and does not show to biomagnify along the food chain. The size of aquatic animals plays an important role in metal amounts of fish tissues (Dang & Wang, 2012). The correlation between metal levels and the fish length was widely explored and generally reported as positive (Monsefrad et al., 2012; Yi & Zhang, 2012; Noël et al., 2013), and sometimes, negative (McKinley et al., 2012; Tekin-Özan et al., 2012; Merciai et al., 2014; Traina et al., 2019). The length has been introduced as a variable to which Hg levels are well correlated with some fish species because of the bioaccumulation of Hg in muscular tissues all over their life (Burger et al., 2007; Vieira et al., 2011).

Assessment of human health risks

According to Table 4, the maximum and minimum THQ for Hg were obtained for *Epinephelus coioides* (THQ=2.08) for children and *Lethrinus crocineus* (THQ=0.20) for adults. Observation of Hg THQ greater than 1 for *Epinephelus coioides* and *Otolithes ruber* for both children and adult groups indicated a potential risk for consumers. In contrast, As THQ values were lower than 1. Also, THQ of As were within the 0.006 (*Lethrinus crocineus*) for adults to 0.051 (*Epinephelus coioides*) for children. Thus, the obtained THQ of As does not show any possible non-carcinogenic risk for consumers. Because exposure to further than one pollutant may result in a combined effect (Storelli, 2008), TTHQ was calculated for each species. This study's TTHQ values were higher than 1 for *Epinephelus coioides* and *Otolithes ruber* for both children and adult groups. It also indicates the significant health risks for Iranian consumers by using these two fish species. Usese et al. (2017) reported that except for populations with high fish consumption, the THQ was lower than 1 (guideline value of USEPA) for all fish species in a tropical open lagoon, Southwest-Nigeria. The annual per capita fish consumption in Iran is reported about 10-11 kg per person (AFS, 2010). This value increases significantly in residents in the coastal communities (Okati & Esmaili-sari, 2018). Therefore, there is an additional risk of non-carcinogenic health consequences due to increased fish consumption rates.

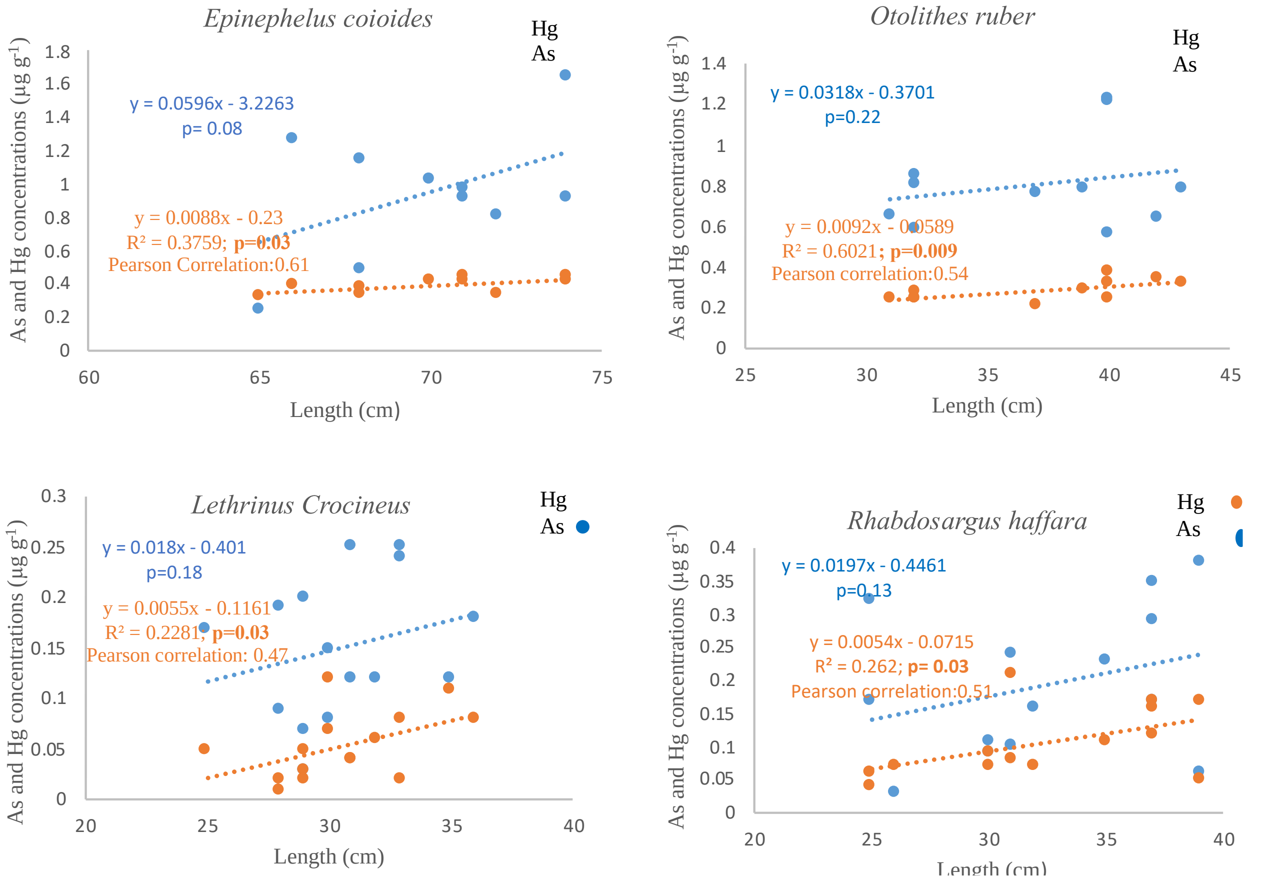

Figure 3. The relationship between metals concentrations (As and Hg) and length for each fish species. Statistically, significant differences are indicated in bold.

The carcinogenic risk obtained from the intake of As was also estimated because this element may develop both non-carcinogenic and carcinogenic effects related to the exposure dose. Inorganic Arsenic is categorizing in USEPA Group A, a known carcinogen (USEPA, 2010). The lifetime cancer risk (CR) for inorganic Arsenic in all four fish species is more than 10^{-5} (Table 4). Thus, there is no health-threatening concern for inorganic As due to the consumption of studied fish species of Oman Sea, according to 29.23 g day^{-1} fish consumption for adults in Iran.

Alamdar et al. (2017) also reported the people who frequently consume fish from the river Chenab were exposed to As pollution with carcinogenic (10^{-4} to 10^{-6}), and non-carcinogenic (THQ>1) risks, especially from the intake of *Cirrhinus reba*. Also, Yi et al. (2017) demonstrated HQ values < 1, which showed no toxic health effects of heavy metals for people were discovered through everyday fish consumption from the Yangtze River in China. Again, the TTHQ of 1.659 obtained by them exceeded one because people might be exposed to non-carcinogenic health risks from the aggregate effect of heavy metals they studied. The findings of Zhong et al. (2018) displayed the levels of heavy metals in freshwater fish from both central and eastern North China were low and did not cause major people health risks.

Table 6. Comparisons of the Hg and As concentrations (µg/g, ww) in fish of Oman Sea with other researches

Location	Fish species	Hg levels	As levels	Reference
Mediterranean Sea	*Thunnus thynnus*	(Mean: 1.53)	(Mean: 0.10)	Storelli et al (2005)

Gulf of Oman (between Oman and Iran)	Different Species	0.003–0.76	-	Al-Reasi et al. (2007)
Manchar Lake and Indus River (Pakistan)	Different species	-	2.11-14.1	Shah et al. (2009)
Hunan (China)	Different Species	0.0027-0.243	0.009-0.152	Fu et al. (2010)
Persian Gulf (Iran)	Different species	0.12-0.52	0.15-0.83	Saei-Dehkordi et al. (2010)
The Straits of Malacca (Malaysia)	*Psettoddes erumei*	1.7 and 3.7	0.59 and 1.06	Alina et al. (2012)
Norwegian waters	Different species	-	0.3-110	Julshamn et al. (2012)
Mediterranean Sea, Spain	*Mullus barbatus*	Mean: 17.70	Mean: 0.09	Martinez-Gomez et al. (2012)
Southwest Coast, India	*Lutjanus russelli*	Mean:0.09	-	Thiyagarajan et al (2012)
Pearl River delta, China	*Lutjanus griseus* *Lutjanus stellatus*		(Mean: 1.53) (Mean: 0.36)	Leung et al (2014)
Persian Gulf (Iran)	Different Species	0.049-0.402 (Mean: 0.133)	0.168-0.479 (Mean: 0.312)	Raissy & Ansari (2014)
Persian Gulf (Iran)	*Otolithes ruber*	-	0.27-0.46 (Mean: 0.36)	Ranjbar Vakil Abadi et al. (2015)
	Scomberomorus guttatus		0.83-1.63 (Mean: 2.93)	
Mediterranean Sea (Sicily Channel), Sciacca, Italy	*Sardina pilchardus*	Mean: 0.11	Mean: 7.86	Traina et al. (2019)
Ankobrah and Pra Rivers, Ghana Pra River, Ghana	*Clarias anguillaris*	(Mean: 0.40)	(Mean: 0.04)	Kortei et al. (2020)
	Oreochromis niloticus	(Mean: 0.40)	(Mean:0.04)	
	Clarias anguillaris	Mean:0.48	(Mean: 0.04)	
	Oreochromis niloticus	(Mean: 0.60)	(Mean: 0.04)	
Oman Sea (Iran)	*Epinephelus coioides*	0.32-0.44 (Mean: 0.38)	0.24-1.65 (Mean: 0.94)	This study
	Lethrinus crocineus	0.01-0.12 (Mean: 0.05)	0.03-0.25 (Mean: 0.15)	
	Otolithes ruber	0.10-0.38 (Mean: 0.26)	0.56-1.23 (Mean: 0.80)	
	Rhabdosargus haffara	0.04-0.21 (Mean: 0.10)	0.03-0.38 (Mean: 0.19)	

Consumption of fish is always recommended due to its high protein values, vitamins, beneficial mineral compounds, and unsaturated fatty acids (PUFA) (Storelli, 2008). But the presence of pollutants such as Hg and As in the edible tissue of fish can reduce the benefits of consuming this useful food (Chan & Egeland, 2004). To estimate the non-cancerous effects, the maximum allowable fish consumption rate per day (CR_{Lim}) has been assessed for Hg and As in all four fish species (Table 5). The maximum permissible fish consumption rate per month (CR_{mm}) is also estimated to assess how many meals of these four fish species of the Oman Sea can be consumed securely without undesirable non-cancerous effects per month (Table 5). According to Hg concentration in fish species, the minimum and maximum allowable consumption rates were obtained for *Epinephelus coioides* for children (0.008 kg day^{-1} = 1.8 meals month^{-1}) and *Lethrinus crocineus* for adults (0.140 kg day^{-1} ~ 18.7 meals month^{-1}), respectively. According to As levels in fish, the minimum and maximum allowable consumption rate were obtained for *Epinephelus coioides* for children (0.34 kg day^{-1} ~ 76 meals month^{-1}) and *Lethrinus crocineus* for adults (4.66 kg day^{-1} ~ 625 meals month^{-1}), respectively. In other studies, the restrictions on fish consumption for some population groups have also been reported by other researchers. For example, Mozaffarian and Rimm (2006), reported the advantages of the modest fish consumption (1–2 meals $week^{-1}$) for women of childbearing age to reduce the possible hazards due to Hg exposure in fish. Also, the maximum allowable concentration rate per day for an adults with mean 71.5 kg BW, was 55 and 93 g day^{-1} based on Hg and As concentrations, fish caught off the Persian Gulf, respectively (Raissy & Ansari, 2014). According to USEPA guideline for pollutants in fish, there is no consumption limit for CR_{mm} > 16 meals $month^{-1}$ (USEPA, 2009). In this study, only the obtained CR_{mm} for Hg in *Lethrinus crocineus* for adults was more than 16 meals per month but for other fish species (*Epinephelus coioides*, *Otolithes ruber* and *Rhabdosargus haffara*) were less than 16 meals for adults and children groups. The obtained CR_{mm} for As in all four fish species was more than 16 meals per month. It was indicated there is no consumption limit for all fish species, according to As levels in fish muscles.

Conclusion

The main facet assessing Hg and As levels in fish species is their toxicity to humans. Fish consumption has various health advantages, and fish has been known as an essential pathway of exposure to Hg and As. This work was a risk assessment research about the selective fish from the Oman Sea, Iran. Observation of THQ for Hg, and TTHQ greater than one for *Epinephelus coioides* and *Otolithes ruber* caught off the Oman Sea for both children and adult's groups indicated the potential risk. In this study, there is no consumption limit for Hg in *Lethrinus crocineus* for adults, but for other fish species (*Epinephelus coioides, Otolithes ruber,* and *Rhabdosargus haffara*) must be considered consumption limits for children and adult's groups. The obtained CR_{mm} for As in all four fish species showed that it is safe for adults and children. The correlation between Hg concentrations and length of fish species showed the bioaccumulation of Hg in muscle tissue throughout their life. Health risks of pollutants can also vary for different populations depending on their frequency and fish consumption rate. So, other pollutant levels of more fish species and other seafood in this region should be monitored, and potential health risks must be assessed periodically. Consumers must be knowledgeable both about the benefit and hazard of fish consumption. Hence, minimizing health risks and raising the advantage of seafood consumption will be feasible.

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