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PROOF RESEARCH PAPER

Assessment of Potential Health Risks of Heavy Metals to the General Public in Turkey via Consumption of Red Mullet, Whiting, Turbot from the Southwest Black Sea

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Abstract

Trace metals in red mullet (*Mullus barbatus*), whiting (*Merlangius merlangus*) and turbot (*Psetta maxima)*, from the Southwest Black Sea were determined and compared with the permitted limits. The maximum and minimum concentrations (mg.kg-1) in red mullet, whiting and turbot samples were found as 1.36-11.85, 1.57-3.35 and 0.39-26.36 for Cu; 20.80-34.94, 13.70-36.49 and 4.89-72.13 for Zn; 3.37-5.87, 1.98-7.53 and 0.70-2.90 for As; 0.02-0.05, 0.01-0.04 and 0.00-0.05 for Cd; 0.01-0.03, 0.01-0.02 and 0.00-0.02 for Hg; 0.03-1.70, 0.05-1.22 and 0.00-1.44 for Pb, respectively. The 2% of turbots exceeded the permitted limit for Cu, while 4% of them contained Zn above the limit. Lead concentration exceeded the permitted limit in the 25% of red mullets, 33% of whitings and 15% of turbots. The estimated weekly intakes (EWI) of metals were lower than established provisional tolerable weekly intakes (PTWI).

Target hazard quotients (THQs) due to metal uptake from consuming each of these fish species were below 1, indicating no health risks. Besides, consumption of these demersal species together would not cause potential health risks since hazard index (HI) was lower than 1.

Keywords: Heavy metals, fish, target hazard quotients (THQs), Hazard Index (HI), PTWI.

Introduction

Fish has been acknowledged as an important food source for humans by nutritionists. Omega-3 fatty acids have been advised for health, and it is possible to obtain these fatty acids by eating fish twice a week (Kris-Etherton, Harris, & Appel, 2002). Moreover, fish is a good source of high-quality proteins, phospholipids, vitamins and minerals and it has been contributed to the normal neurodevelopment in children (Luten, 2009).

On the other hand, toxicologists regard fish as the main source of heavy metals. Because, it may accumulate toxic elements in edible tissues. Heavy metals are emitted to the marine environment as a result of human activities such as transportation, agriculture, industry, urbanization, mining and approaches to optimise water use (Gorur, Keser, Akcay, & Dizman, 2012; Robu *et al*., 2015). The Black Sea is exposed to these threats because of its closed basin, inputs from large rivers, discharge wastewater from factories and numerous coastal activities (Jitar *et al*., 2013; Plavan *et al*., 2017). The main source of pollution in the Black Sea is the pollutants carried by the rivers. This is because, in addition to the six countries that have openings at the Black Sea, the toxic wastes from wastewater industry of 18 countries are discharged through the rivers to the Black Sea. The Danube, Yeşilırmak, Kızılırmak (Jitar ve diğ., 2013) and Sakarya rivers are important factors of pollution in the Black Sea. The Sakarya River is spilling from the southwest region to the Black Sea and is known to carry heavy metal in high quantities (Ozata *et al*., 2011).

Copper is an essential element for health but elevated concentrations of this metal may cause liver and kidney damages (Ikem & Egiebor, 2005). Similarly, zinc is naturally found in a wide variety of foods, but it can be toxic at high concentrations (Celik & Oehlenschlager, 2004). Fish has been known to contain higher concentrations of arsenic than other foods. But it is mostly organic, and organic arsenic in seafood appears to be much less toxic than the inorganic forms. Fish may also contain highly toxic metals in edible tissues, such as cadmium, mercury and lead. Cadmium has negative effects on the kidney, the skeletal system and the respiratory system, and fish consumption is the main source of exposure to cadmium in human (Keskin *et al*., 2007; Castro-Gonzalez & Mendez-Armenta, 2008). Methylmercury, the organic form of mercury, is highly toxic to humans and may increase the risk of coronary heart disease. Lead is a cumulative toxicant that negatively affects the nervous system, hematological system, cardiovascular system and kidney (Jarup, 2003).

Since contaminated fish provides the largest dose of heavy metals to humans, determining metal concentrations in commercial fish species is a global public health concern (Mathews & Fisher, 2009). Red mullet, whiting and turbot are popular species (Rad, 2002; Ozden, Erkan, & Ulusoy, 2010) and the potential for the accumulation of trace metals may be greater in the muscles of such demersal fish (Cronin *et al*., 1998). Red mullets (*Mullus barbatus*) and whitings (*Merlangius merlangus*) tend to concentrate high amounts of contaminants, since they are carnivore bottom dwellers (Uluozlu, Tuzen, Mendil, & Soylak, 2007; Zorita *et al*., 2008). Turbot, *Psetta maxima*, is a carnivore demersal fish of a high commercial value, and Turkey is one of the main fishery nations of this species (Imsland, 2010). All these demersal species are mainly caught from the Black Sea, which is an important fishery resource for Bulgaria, Georgia, Romania, Russia Federation, Ukraine and Turkey (Duzgunes & Erdogan, 2008). Analyzes of toxic elements in fish from Black Sea are needed to assess possible public health risks, considering increased pollution (Tuzen, 2009).

There are some studies on heavy metal concentrations in the edible tissues of fish from the Black Sea (Harmelin-Vivien *et al*., 2009; Jitar, Teodosiu, Oros, Plavan, & Nicoara, 2015; Mendil, Demirci, Tuzen, & Soylak, 2010; Tepe, Türkmen, & Türkmen, 2008; Topcuoğlu *et al*, 2002; Turan, Dural, Oksuz, & Ozturk, 2009; Tuzen, 2009; Uluozlu *et al*., 2007). But, the amount of periodical fish intake by the consumer is also very important to determine possible health risks for human. There is a lack of information on public health risk associated with toxic metals in fish from the Black Sea. Target hazard quotient (THQ) based risk assessment method indicates the public health risk associated with the heavy metals in fish and provides an indication of the risk level associated with contaminant exposure.

The aim of this study was to determine the concentrations of Cu, Zn, As, Cd, Hg and Pb in popular demersal fish (red mullet, whiting, and turbot) from the Southwest Black Sea. The dietary intakes of metals derived from the consumption of aforementioned species were also estimated. Health risk associated with heavy metals in fish was evaluated by using THQ. Since exposure to more than one chemical may increase the health risk, total THQ (TTHQ) was also calculated.

Materials and Methods

Materials

In this study, 12 individuals of red mullet (*Mullus barbatus*), 12 individuals of whiting (*Merlangius merlangus*), and 42 individuals of turbot (*Psetta maxima*) were caught by trawl from the Southwest Black Sea, during spring season, 2013. The sampling area was shown in Figure 1. Samples were transferred to the laboratory in foamed polystyrene boxes with ice and biometric measurements of red

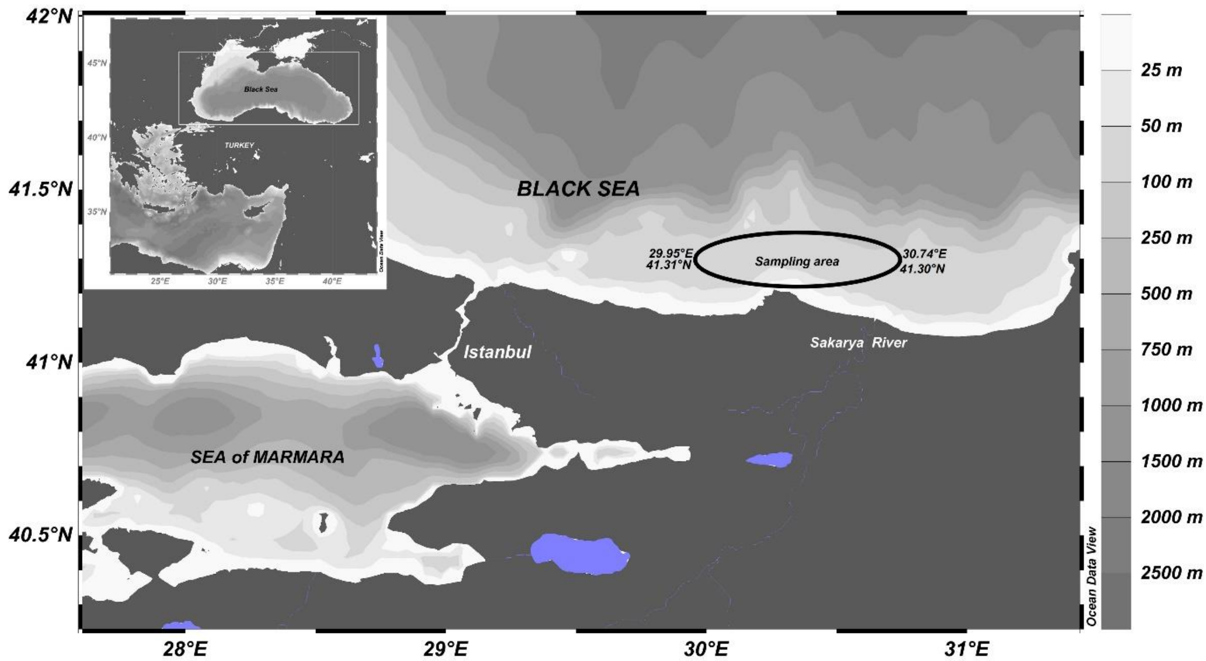

Figure 1. Sampling area (Southwest Black Sea, Turkey).

mullets (FL: 14.19 ±2.59 cm; weight: 13.14 ± 4.12 g), whitings (FL: 17.65 ± 2.10 cm; weight: 36.95± 5.01 g) and turbots (FL: 32.99 ±5.87 cm; weight: 363.77 ± 24.22 g) were performed. Approximately 2 g of muscle sample was dissected from each fish and kept frozen until analysis. To avoid metal contamination and adhesion, any metal or glass equipment were not used during sampling and analysis.

Methods

Samples were weighed (0.3 and 0.5 g) and placed in a Teflon digestion vessel with 7 ml of a high-quality nitric acid (65%) and 1 ml of hydrogen peroxide (30%). Then Ethos D (Plus 1) microwave lab station from Milestone Inc. (Monroe, Ct, USA) was used for digestion. The microwave program were as follows: step 1: 25–200°C for 10 min at 1,000 W; step 2: 200°C for 10 min at 1,000 W. Then the digests were made up to 25 ml using deionized water. Thermo electron X2 ICP-MS, (model X series, UK) was used to analyze the digested samples for Cu, Zn, As, Cd, Hg and Pb. The instrument detection limits were 20 ppt for Cu, 100 ppt for Zn, 25 ppt for As, 2 ppt for Cd, 50 ppt for Hg, and 10 ppt for Pb. Calibration standards were obtained from High-Purity Standards (Charleston, SC, USA). Catalogue numbers were (1000±3µg/ml in 2% HNO_3) Cu: 100014-1, Zn: 100068-1, As: 10003-1, Cd: 10008-1, Hg: 100033-1 and Pb: 100028-1. The standard solutions for calibration were prepared just before the analysis with high purity deionized water, and ICP-MS was calibrated (US EPA, 1994). ICP-MS operating conditions were: Nebulizer gas (Ar) flow 0.91 L/min, Radio frequency (RF) 1,200 W, Lens voltage 1.6 V, Cool Gas 13.0 L/min, and Auxiliary Gas 0.70 L/min. Certified reference material (catalogue no. SRM 2976, Gaithersburg, MD) was analyzed (n=3) to validate the method for accuracy (Table 1). The analyses were triplicated, the significance level was chosen as 0.05, and results were analyzed by means of analysis of variance (ANOVA). The statistical package SPSS 17.0 (SPSS, Inc., Chicago, IL, USA) was used for statistical analyses.

EWI (estimated weekly intakes) values were calculated by multiplying the mean concentrations of each element and the amount of weekly consumed fish. The weekly consumption of fish in Turkey is 148 g (Speedy, 2003; TUIK, 2009). The average weight of a person was considered as 70 kg (Turkmen, Turkmen, Tepe, Tore, & Ates, 2009) and multiplied by PTWI (provisional tolerable weekly intakes) values, proposed for each metal. Then the percent PTWI was calculated.

The health risks for Turkish consumer caused by consuming red mullet, whiting and turbot from the Southwest Black Sea were assessed based on THQ. This method offers an indication of the risk level due to the heavy metal exposure. The following equation was used to determine THQ (Han *et al*.,1998; Chien *et al*., 2002; Storelli, 2008):

$$THQ = \frac{(EF \times ED \times FIR \times C)}{(RFD \times WAB \times TA)} x10^{-3}$$

Where E_F is the exposure frequency (365 days/year), E_D is the exposure duration (70 years, the average lifetime according to Bannett, Kastenberg, & McKone, 1999. F_{IR} is the food ingestion rate (21.09 g/day for Turkish consumers, according to Speedy (2003), C is the determined metal concentration (mg . kg-1), R_{FD} is the oral reference dose (mg . kg-1 /day), W_{AB} is the average body weight (70 kg), according to Kumar, Verma, Naskar, Chakraborty, & Shah, 2013, T_A is the average exposure time for noncarcinogens (365 days/year x E_D, assuming 70 years in this study).

The oral reference doses (R_{FD}) for Cu, Zn, Cd, Hg and Pb (mg . kg-1 , day) have been suggested as 4 x 10^{-2}, 3x10^{-1}, 1 x 10^{-3}, 1.6 x 10^{-4} and 4 x 10^{-3}, respectively (US EPA, 2009).

Results

The mean concentrations of Cu in red mullet, whiting and turbot were 3.30 mg. kg-1; 2.44 mg.kg-1 and 4.29 mg.kg-1 (P>0.05), respectively (Table 2). The mean concentrations of Zn were 26.65 mg.kg-1 in red mullet, 23.54 mg.kg-1 in whiting and 17.57 mg. kg-1 in turbot. The mean concentrations of As in all species were below 5 mg.kg-1 and the highest As concentration was determined as 7.53 mg.kg-1 in whiting. The mean concentrations of Cd were not above 0.05 mg. kg-1 in all species. The highest concentration of Hg was 0.03 mg.kg-1, which was found in the red mullet. The mean concentrations of Pb were 0.29 mg. kg-1, 0.36 mg.kg-1 and 0.17 mg. kg-1 in red mullet, whiting and turbot, respectively. However, Pb concentration exceeded 0.3 mg.kg-1 in

Table 1. Certified, observed values (mg.kg-1) and recoveries (%) of trace metal concentrations in standard reference material

	Certified value	Observed value	Recovery (%)
Cu	4.02	4.16	103.48
Zn	137.0	136.60	99.71
As	13.30	14.70	110.53
Cd	0.82	0.77	93.90
Hg	61.00	67.81	111.16
Pb	1.19	1.08	90.76

the 25% of red mullets, 33% of whitings and 15% of turbots.

EWI and PTWI values of red mullet, whiting and turbot were presented in Table 3. The maximum EWI (µg/ week b.w.) values were 3944.20 for Zn, 4.44 for Cd and 2.96 for Hg in red mullets, while it was 634.92 for Cu in turbot and 53.28 for Pb in whiting. THQ were shown in Table 4, and this value remained below 1 in all samples.

Discussion

Concentrations of Heavy Metals

The permissible limit for Cu is 30 mg . kg-1 , according to FAO (1983), WHO (1996) and Maff (1995), but this limit is suggested as 20 mg . kg-1 in Turkish Food Codex (2002). All individuals of red mullets and whitings contained Cu below these limits in our study. The concentration of Cu was reported as 3.48 mg . kg-1 in red mullets from the Black Sea (Jitar *et al*., 2015), similar to that of our study. Likewise, Cu concentrations in red mullet and whiting from Black Sea (Mendil *et al*., 2010; Tepe *et al*, 2008; Tuzen, 2009; Uluozlu *et al*., 2007) were well below the recommended limits. The mean concentration of Cu in turbot was also well below the recommended limits, but some individuals (2%) contained Cu above 20 mg . kg-1 (Table 2).

The concentration of Zn remained below the limit value of 50 mg . kg-1 (Maff, 1995; Turkish Food Codex, 2002) in red mullets and whitings, similar to the former studies (Mendil *et al*., 2010; Ozden *et al*., 2010; Tepe *et al*., 2008). The reported concentration of Zn in turbot is generally below 50 mg . kg-1 (Guerin *et al*., 2011; Tuzen, 2009). Likewise, the mean concentration of this metal in turbot was determined as 17.57 mg . kg-1 in this study. But, some individuals (4%) contained this metal above the permitted limit.

The highest As concentrations in red mullet, whiting and turbot samples were 5.87 mg. kg-1 , 7.53 mg . kg-1 and 2.90 mg . kg-1 , respectively. Ozden *et al*. (2010) and Olmedo *et al*. (2013) reported lower As concentrations in red mullets and whitings than that of the present study. Permitted limit for As is 1 mg . kg-1 in fish, according to Turkish Food Codex (2002). However, suggested limits vary from 0.1 mg . kg-1 to 10 mg . kg-1 (De Gieter *et al*., 2002), and typical concentrations of this metal in fish are generally between 1-20 mg . kg-1 (Francesconi, 2007). It is clear that fish contains higher amounts of As than the other foods. But an important part of As is organic, which is not toxic as the inorganic form

Table 2. Ranges and mean concentrations (mg. kg-1 wet weight) of heavy metals for red mullet, whiting and turbot

Metal		Red mullet	Whiting	Turbot
Cu	min.-max.	1.36 – 11.85	1.57 – 3.35	0.39 – 26.36
	mean ± SD	3.30 ± 2.92[a]	2.44 ± 0.54[a]	4.29 ± 5.85[a]
Zn	min.-max.	20.80 – 34.94	13.70 – 36.49	4.89 – 72.13
	mean ± SD	26.65 ± 4.80[a]	23.54 ± 6.77[ab]	17.57 ± 14.46[b]
As	min.-max.	3.37 – 5.87	1.98 – 7.53	0.70 – 2.90
	mean ± SD	4.76 ± 0.94[a]	4.09 ± 2.30[a]	1.60 ± 0.52[b]
Cd	min.-max.	0.02 – 0.05	0.01 – 0.04	0.00 – 0.05
	mean ± SD	0.03 ± 0.01[a]	0.02 ± 0.01[a]	0.01 ± 0.01[b]
Hg	min.-max.	0.01 – 0.03	0.01 – 0.02	0.00 – 0.02
	mean ± SD	0.02 ± 0.01[a]	0.01 ± 0.01[b]	0.01 ± 0.01[c]
Pb	min.-max.	0.03 – 1.70	0.05 – 1.22	0.00 – 1.44
	mean ± SD	0.29 ± 0.47[a]	0.36 ± 0.42[a]	0.17 ± 0.29[a]

[a, b,c]: Different letters in the same row show statistical differences (P<0.05).

Table 3. The estimated weekly intakes (EWI) and percent PTWI's for red mullet, whiting and turbot from Black Sea, consumed by adult people in Turkey

Metal	PTWI*	PTWI**	Red mullet		Whiting		Turbot	
			EWI***	Percent PTWI	EWI	Percent PTWI	EWI	Percent PTWI
Cu	3 500 [a]	245 000	488.40	0.20	361.12	0.15	634.92	0.26
Zn	7 000 [a]	490 000	3944.20	0.80	3483.92	0.71	2600.36	0.53
Cd	7 [a]	490	4.44	0.91	2.96	0.60	1.48	0.30
Hg	1.6 [b]	112	2.96	2.64	1.48	1.32	1.48	1.32
Pb	25 [a]	1 750	42.92	2.45	53.28	3.04	25.16	1.44

* The established Provisional Tolerable Weekly Intake (µg/week/kg body weight)
** PTWI for a 70 kg adult (µg/week body weight)
*** Estimated Weekly Intake (µg/week body weight)
[a] FAO/WHO, 2004
[b] EC, 2006

Table 4. Estimated Target Hazard Quotients (THQs) for metals caused by consuming red mullet, whiting and turbot from the Black Sea

Metal	Red mullet	Whiting	Turbot	TDHQ
Cu	0.025	0.018	0.032	0.075
Zn	0.027	0.024	0.018	0.069
Cd	0.009	0.006	0.003	0.018
Hg	0.038	0.019	0.019	0.076
Pb	0.022	0.027	0.013	0.062
TTHQ	0.121	0.094	0.085	0.300 (HI)

TDHQ=The Total Diet THQ of each metal
TTHQ= Total THQ of heavy metals due to the consumption of each species
HI= Hazard Index, the sum of TTHQs

(Castro-Gonzaleza & Mendez-Armenta, 2008). So, it is difficult to judge potential health risk related to arsenic concentration in fish.

In the present study, none of the samples contained Cd above the permitted limit of 0.05 mg.kg-1 (EC 2014; Turkish Food Codex, 2011). Alkan *et al*. (2016) reported Cd levels in anchovy, sprat and horse mackerel from the Black Sea above the permissible safety levels for human consumption. Likewise, higher concentrations of Cd have been reported for red mullet, whiting (Mendil *et al*., 2010; Tuzen, 2009; Uluozlu *et al*., 2007) and turbot (Tuzen, 2009) from the Black Sea. Heavy metals in fish tissues have been regarded as a threat for human consumption (Turan *et al*., 2009). Therefore, Cd concentrations in fish from the Black Sea should be controlled periodically considering human health.

The maximum limit for Hg has been suggested as 0.5 mg . kg-1 for fish (EC, 2006) and 1 mg . kg-1 for halibut, a turbot-like species, and for *Mullus* sp. (EC, 2008; Turkish Food Codex, 2011). Even the mercury accumulation in red mullet has been regarded as an important health risk (Harmelin-Vivien *et al*., 2009); all fish samples, including red mullets contained Hg below the maximum permitted levels in this study. Likewise, Tuzen (2009) reported mercury concentrations in red mullet, whiting and turbot from the Black Sea lower than the legal limits.

The maximum permitted concentration of Pb is 0.3 mg. kg-1 (EC, 2008; Turkish Food Codex, 2011) for fish. Lead concentration exceeded this limit in the 25% of red mullets, 33% of whitings and 15% of turbots. The Black Sea is considerably affected from the sources of pollution (Turan *et al*., 2009), and the formerly reported Pb concentrations in fish from Black Sea were higher than that of our study. Jitar *et al* (2015) reported the mean concentration of Pb as 0.32 mg . kg-1 in red mullets from the Black Sea. Likewise, the mean concentrations of Pb in red mullet and whiting from Black Sea were reported as 0.84 mg . kg-and1, 0.93 mg . kg-1, respectively (Uluozlu *et al*., 2007). It was concluded that demersal species from the Black Sea should be checked periodically, considering the concentrations of Pb in their muscles.

Dietary Intake of Heavy Metals

For many years, the determination of heavy metal concentrations in fish has been used to assess human health risks. But additional information is needed for a better risk assessment, since consumption amounts of various communities may be very different (Olmedo *et al*., 2013). Even if the toxicological limits are not exceeded for an average consumer, heavy consumers may be under the risk of heavy metal exposure (Leblanc *et al*., 2005). For this reason, the EWI and percent PTWI values for red mullet, whiting and turbot consumed by adults in Turkey were investigated, in this study.

The established PTWI and PTWI for a 70 kg adult were presented in Table 3, and compared with the EWI values. In our case, all heavy metal intakes through the consumption of demersal fish from the Southwest Black Sea were lesser than tolerable weekly intake limits**.** Turkmen *et al*. (2009) reported EWI of Cu, Zn, Cd, and Pb for individual metals by consumption of a demersal fish, *Trigla lyra*, from Turkish waters as 347 µg per person/week, 1172 µg per person/week, 18.2 µg per person/week, and 58.8 µg per person/week, similar to our results. Gorur *et al*. (2012) studied the Zn, Cu, Pb concentrations of various fish from the Black Sea and reported lower daily intakes than the recommended intake values. Turkmen and Dura (2016) studied heavy metal concentrations in *Mullus barbatus* from South Western Black Sea, and reported EWI values below the recommended values. Low EWI values for Hg, Cd and Pb have also been reported for *Merluccis merluccis* and *Mullus barbatus* from the Adriatic Sea (Storelli, 2008). Bat and Arici (2016) assessed health risk of heavy metals in Atlantic bonito from Southern Black Sea and reported average weekly intakes of heavy metals (Cu, Zn, Cd, Hg and Pb) below the established PTWI values.

The EWI and percent PWTI values of arsenic were not calculated and shown in the tables. Because, the former PTWI value of As (15 µg/kg) is no longer appropriate according to JECFA (2011) and withdrawn by the Committee, since an important part (about 98%) of the As in fish is the organic and non-toxic form. Olmedo *et al*. (2013) did not calculate the PTWI percentages for As for the same reason.

Potential Health Risk of Metals

The THQ-based risk assessment has been regarded as a useful method to indicate possible health risk associated with heavy metal exposure (Yi, Yang, & Zhang, 2011). The THQ values below 1 show that the estimated exposure is not considered as a potential concern (Basim & Khoshnood, 2013). Since THQ values remained below 1 in this study (Table 4), no significant health risk was concluded due to the consumption of red mullet, whiting or turbot from Southwest Black Sea. Similar results were reported by Zheng *et al*. (2007). They studied the health risk due to dietary intake of heavy metals in China and reported THQ values lower than 1 through consumption of sea products. Likewise, Çulha *et al*. (2016) reported THQ values for heavy metals (Al, Cu, Ni, As, Cd, Hg, Pb, U) below 1 in scorpionfish, a demersal species caught from Black Sea, and reported no risk for human consumption. Storelli (2008) studied the potential human health risks via seafood consumption from the Adriatic Sea and reported the THQs for individual metals by consumption of *M. merluccis* as 0.22 for Hg, 0.02 for Cd and 0.01 for Pb; and by consumption of *M. barbatus* as 0.84 for Hg, 0.01 for Cd and 0.01 for Pb. Fish from Yangtze River, China were also regarded as safe, considering the THQ values below 1 (Yi *et al*., 2011). Han *et al*. (1998) studied possible health hazards caused by seafood consumption in Taiwan and reported THQ values below 1 for various seafood containing Cd and Zn. Likewise, Wang, Sato, Xing, & Tao (2005) studied health risks of heavy metals to the population in Tianjin, China through the consumption of fish and vegetables. They reported THQ values lesser than 1, but underlined that potential health risks may arise due to the consumption of both vegetables and fish, when THQ values close to 1.

It is inaccurate to calculate THQ for arsenic since RfDo given by US EPA is for inorganic As only. It is known that an important part of the As (about 98%) in fish is non-toxic, organic form (EFSA, 2009). Therefore, THQ values were not calculated for As in this study. Likewise, Bandowe *et al*. (2014) did not calculate THQ values for As due to the same reason.

Since exposure to more than one contaminant may result in combined effects (Hallenbeck, 1993), following formula was used to determine TTHQ of heavy metals for each species (Chien *et al*., 2002; Storelli, 2008; Zeng *et al*., 2007):

$$\text{TTHQ (individual fish)} = THQ\ (toxicant\ 1) + THQ\ (toxicant\ 2) + THQ\ (toxicant\ n)$$

In the present study, the TTHQ values were determined below 1 (Table 4), indicating no significant health risk for the Turkish consumer by consuming red mullet, whiting and turbot. A similar result was reported for *M. merluccis* and *M. barbatus* from the Adriatic Sea (Storelli, 2008). Total THQ of heavy metals due to sea product consumption in China was also reported as 0.490 by Zheng *et al*. (2007). In our study, the total diet THQs (TDHQ) of each metal were also below 1, indicating no potential risk.

HI to human population due to Cu, Zn, Cd, Hg and Pb exposure, through red mullet, whiting and turbot consumption was calculated as follows (Zheng *et al*., 2007):

$$\text{HI} = TTHQ\ (red\ mullet) + TTHQ\ (whiting) + TTHQ\ (turbot)$$

Since the HI values were below 1 (Table 4), consumption of these demersal species from the Southwest Black Sea might not be hazardous to the consumer with respect to observed levels of Cu, Zn, Cd, Hg and Pb alone or in combination with each other.

Conclusion

The mean concentrations of heavy metals were below permitted limits for demersal species, except Pb concentrations of whiting. Besides, some individuals of all species contained Pb higher than 1 mg. kg-1. Some individuals of turbot exceeded the limits, suggested for Cu and Zn. However, EWI values remained far below PTWI, showing there is no health risk to the consumer from red mullet, whiting and turbot consumption. THQ values also remained below 1, showing no health risk due to the intake of individual heavy metal by consuming one of red mullet, whiting of turbot. Likewise, HI for combined heavy metals due to the consumption of all fish species were lower than one, showing no potential health risk to human health.

As well as its numerous health benefits, fish has also been considered as an important route of exposure to heavy metals. This study, of course, is a selective fish investigation from the Southwest Black Sea. Heavy metal concentrations might be different due to the fish species, catching area, and habitat. Health risks may also be different for various populations depending on their consumption amounts. So, contaminant levels of more seafood species from commercial catching areas should be monitored and possible health risks must be assessed periodically. Consumers should be informed both with the benefits and risks of fish. Therefore, minimizing risks and increasing benefits of fish consumption will be possible.

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