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Risk assessment for exposure to potential toxic metals via dietary intake of black scorpionfish from the Black Sea

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ABSTRACT

The concentrations of lead (Pb), cadmium (Cd), mercury (Hg), copper (Cu) and zinc (Zn) were measured in black scorpionfish (*Scorpaena porcus* Linnaeus, 1758) from Sinop coasts of the Black Sea, Türkiye. The study found that the levels of these metals were below Türkiye and international permissible values. For each metal, it was discovered that the estimated daily intake (EDI), hazard quotient (HQ) and risk index (RI) values were significantly lower than the corresponding reference doses, indicating that the fish were not found to pose a significant health risk to children or adult consumers based on the available data.". This is important because *S. porcus* has become a popular food choice due to the decrease in naturally caught fish and its abundance in the area. The findings suggest that black scorpionfish can be a healthy and sustainable food option for consumers.

Introduction

For many years, the Black Sea has been a globally productive region, as noted by Mee [1] and Ivanov & Beverton [2]. Even though there are 189 species of fish in the Black Sea [3], only a small number of these species are economically significant [4]. Moreover, the supply of fish is restricted due to intensive fishing, as well as the fact that urbanization and industrialization have led to a decline in fisheries for the most popular species. The dumping of wastes in the Black Sea provides a significant source of metals input [5–8]. Heavy metals are one of the most harmful pollutants in the marine environment because of their toxicity, persistence, and bioaccumulation issues. One of the major issues associated with heavy metal persistence is the possibility of bioaccumulation and biomagnification, resulting in higher exposure for aquatic organisms than is present in the marine environment. Most heavy metals are found in seawater in trace amounts, but when concentrations are higher than what is necessary for proper nutrition, they can harm marine biota by way of the food chain and endanger consumers of marine food [9,10]. Certain nutrients, like zinc (Zn) and copper (Cu), are necessary for <0.01 % of the mass of the organism in the diet, and they can cause severe illness if they are not consumed [11]. Others, like mercury (Hg), cadmium (Cd), and lead (Pb), are referred to as non-essential because they have no biological function and can be poisonous even in very small amounts [11,12]. However, all heavy metals pose a risk to living organisms, even at low levels of exposure [11]. Global environmental concern is raised by the contamination of marine biota by heavy metals. For example, in Japan, people have died from cadmium poisoning (known as Itai-Itai disease, which means "it hurts-it hurts") and mercury poisoning (known as Minamata disease) after consuming seafood from heavily polluted water. According to Phillips [12], there needs to be a clear correlation between the levels of heavy metals in the environment and the organism for it to serve as a useful indicator of heavy metal pollution. To Phillips and Rainbow [13], marine organisms can be used to estimate the levels of toxic metal pollution in the aquatic environment. Because commercial fish are an important source of protein, their pollution levels must be carefully investigated and controlled.

The study focused on determining the levels of heavy metals (Pb, Cd, Hg, Zn, Cu) in black scorpionfish (*Scorpaena porcus* Linnaeus, 1758) caught by local fishermen and sold by fish market in Sinop, particularly between May and July of 2022 and 2023. In 2022, a total of 101.4 tonnes of scorpionfish were caught from the seas of Türkiye and the amount off the coast of Sinop was 27,580 kg [4]. *S. porcus* live in all Turkish seas. The black scorpionfish is solitary and sedentary and feeds on small fish (gobies, blennies), crustaceans and other invertebrates [14]. They prefer shallow and stony littoral areas covered with algae and are active in semi-darkness. The spines on the dorsal fin and gill cover are poisonous. When stimulated, they can erect their fin and gill cover spines. They show very good colour harmony. As soon as they come out of the water, their stomach contents come out and this vomit distorts

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their colour [15]. For this reason, they are either consumed in restaurants or only the meat, cleaned from the bones and skin, is purchased. It is a preferred fish because its meat is very tasty. Soup steamed and skewers are consumed with pleasure [15]. Its consumption is especially high in Sinop.

To date, numerous studies have reported heavy metal contamination in marine species from the Black Sea; however, research specifically focusing on *S, porcus* remains limited despite its increasing importance as a commercial and subsistence fish species in the region. This study provides novel insights by examining heavy metal accumulation in *S. porcus* collected from, Sinop coast of the southern Black Sea, using sensitive analytical techniques. In contrast to previous studies, our work incorporates a detailed human health risk assessment for both adult and child consumer employing established indices such as Estimated Daily Intake (EDI), Target Hazard Quotient (THQ), and Hazard Index (HI). The findings contribute important data for regional environmental monitoring and offer public health insights relevant to seafood safety and dietary exposure guidelines.

Material and methods

Fish sampling

The samples of the fish species *S. porcus* were purchased from the fishermen, at landing on Sinop coasts of the Black Sea. The fishes were caught between May and July 2022 and 2023. Every month, five fish were sampled, resulting in a total of thirty fish subjected to metal analyses. Mean lengths and weights black scorpionfish specimens were measured as 18.3 ± 3.2 cm and 78±7 g, respectively (Fig. 1). They were instantly stored in an icebox kept in polystyrene bag and shifted to the laboratory where they are weighed, and the total length measured. They were kept in deep freezer at −21 °C for metal analysis.

Chemicals and reagents

All reagents and solutions were prepared in water obtained from a Milli-Q water purification system (Millipore). All chemicals used for samples preparation were of analytical reagent grade or higher purity (Merck, Darmstadt, Germany). A standard reagent (Merck, Germany), prepared using the same acid matrix as the samples, was employed to analyse the concentration of metals in the black scorpionfish.

All glassware and equipment were acid washed to prevent any potential contamination. To begin sample analysis, the system was flushed with the method blank solution both before and after each sample to prevent instrument contamination.

Fig. 1. *Scorpaena porcus* Linnaeus, 1758.

Metal analysis

Measurements were conducted using EPA Method 200.3 [16], which is the Sample Preparation Procedure for Spectrochemical Determination of Total Recoverable Elements in Biological Tissues. Using a microwave digestion system (Milestone SK10), fish tissues up to 1.5 g were digested in Teflon vessels with a combination of concentrated grade HNO_3 (Suprapur® %65) and H_2O_2 (Suprapur® %30) (7:1) in accordance with (HPR-FO-67) temperature and pressure profile. Teflon vessels were filled with acid, sealed, and heated for 30 min at 200 °C. After the solution had been broken down, it was poured into a 50 mL polypropylene falcon tube and filled to the brim with Milli-Q water. An Inductively Coupled Plasma Mass Spectrophotometer (ICP-MS, Agilent Technologies 7700X) was used to measure the elemental concentrations. Lobster TORT-2, a CRM (National Research Council of Canada), and triplicate measurement were used for quality assurance and control. Calibration curves were created using multi-element standard solutions from Agilent Technologies with27-element mix consisting of 8500–6940 2A (contains 10 mg/L of Ag, Al, As, Ba, Be, Ca, Cd, Co, Cr, Cs, Cu, Fe, Ga, K, Li, Mg, Mn, Na, Ni, Pb, Rb, Se, Sr, Tl, U, V, Zn in a matrix of 5 % HNO_3) and 8500–6940 Hg (contains 10 mg/L Hg in a matrix of 5 % HNO_3).. Within ±10 %, the analytical precision was achieved. In this investigation, samples were continuously analysed using 1 mg/L internal standard (Agilent 5188–6525). Important parameters including the limit of quantification (LOQ) and the limit of detection (LOD) were covered in the validation [17,18]. LOD were <0.001 μg/L for Cd and Pb, 0.01 μg/L for Hg and Zn, and 0.0001 μg/L for Cu. The LOQ values were LOD × 3. The data was analysed using the mean values, and each analysis was carried out in triplicate. The concentrations of measured metals were expressed as mg/kg of wet weight (wt.). Metal analyses were conducted at Sinop University's Scientific and Technological Research Application and Research Center (SUBİTAM) via service procurement.

Human health risk assessment

In the present study, health risk indices including the estimated daily intake (EDI) of five heavy metals by humans, hazard quotient (HQ), total hazard quotient (THQ), and risk index (RI) were assessed.

Estimated daily intake (EDI)

The EDI (mg/kg/day) is given by the equation below [19–22].

$$EDI = \frac{C_f \ x \ FIR}{Bwt}$$

where C_f is the concentration of heavy metal in black scorpionfish (mg/kg, wet wt.), FIR is the daily fish ingestion rate (27.4 and 41 g/day for children, and adults, respectively) [23] and B_{wt} is the mean body weight (30 and 70 kg for children and adults, respectively) [20]. The estimated weekly intake (EWI) is calculated by multiplying the EDI by 7 [19–24].

Hazard quotient (HQ) and total hazard quotient (THQ)

The noncarcinogenic risk related to the consumption of fish and its associated heavy metals was evaluated using the hazard quotient (HQ).

$$HQ = \frac{EDI}{Rf.D.}$$

where Rf. D. is the oral reference dose which is the estimated maximum permissible health risk associated with daily consumption of metals in black scorpionfish. The Rf. D. values for Cd, Hg, Cu and Zn are 0.0001, 0.0003, 0.04 and 0.3 mg/kg/day, respectively [25,26]. However, the Rf. D. for Pb is not given.

The THQ estimates the cumulative risk associated with exposure to multiple heavy metals.

$$THQ = \sum HQ$$

It is accepted that the likelihood of a health risk is inversely correlated with the combined impacts of metals on the same target organ. THQ >1 discloses potential chronic danger while THQ <1 implies no potential health risk.

Risk index (RI)

The RI is one metric to measure the carcinogenic risk. The equation below represents RI in terms of:

$$RI = EDI \ x \ SF$$

The SF (slope factor) establishes the risk associated with a lifetime average contaminant dose. SF value for Pb is 0.0085 per mg/kg-day [25, 26].

Statistical analysis

The non-parametric tests were implemented during the analysis. Kruskal Wallis test was implemented to investigate the differences in the concentrations of heavy metals among the months (May, June and July) and the Mann Whitney U test was implemented to show differences between the years (2022 and 2023) for edible tissues of *S. porcus*. The statistical significance level was accepted as $p < 0.05$.

Results and discussion

Heavy metals in S. porcus

The results of heavy metal concentrations in *S. porcus* obtained in 2022 and 2023and statistical differences were shown in Fig. 2. It is well known that essential and non-essential metals accumulate in the food web in marine ecosystems [27,28]. In both years, Zn and Cu were the highest in black scorpionfish, whereas the accumulation concentration of Cd, Hg and Pb were at the lowest levels. The order of metal accumulation in the edible tissues of *S. porcus* were Zn > Cu > Pb > Hg > Cd.

While it competes with calcium ions at the sites where the organism's enzymes are located, Cd is not a necessary metal. The maximum allowable limit of Cd concentration is 0.05 mg/kg wet wt., according to the Turkish Food Codex [29–32] and European Commission Regulations [33,34], Commission Regulation [35]. Due to its strong nephrotoxic potentials, Cd can have toxic effects on the kidneys and liver when consumed in excess [36]. *S. porcus* had Cdconcentrations that were below the legal limit.

The Hg value in 2023 was higher than in 2022 and this was statistically significant ($p < 0.05$). Eating fish that has Hg levels above the recommended level can have detrimental effects on the endocrine and central nervous systems [37]. The Hg concentration in *S. porcus* was found to be below the allowable limits set by the European Commission Regulations [33], Commission Regulation [38] and the Turkish Food Codex [29–32]. The permissible limits are set at 0.5 mg/kg wet wt.

Pb concentration in all *S. porcus* samples were lower than the level 0.3 mg/kg wet wt. set by the European Commission Regulations [33], Commission Regulation [39], and the Turkish Food Codex [29–32].

Cu and Zn have been recognized as the essential elements; they are required for many different types of enzymes and other cellular components that play significant roles in the biota. On the other han d, excessive Cu and Zn consumption is bad for human health since it can cause toxicity and other related conditions. Cu and Zn levels found in the edible tissues of *S. porcus* are less than the Ministry of Agriculture, Forestry and Fisheries (MAFF) and Turkish Food Codex permitted limits of 20 and 50 mg/kg, respectively [40,41].

Comparison of metal levels in S. porcus with previous studies

Comparisons of literature results for metal levels in *S. porcus* studied from different locations have been summarized in Table 1. The average metal concentrations reported in the present study were lower than the previous studies in different locations. The highest Hg levels were found in the Sevastopol coast of the Black Sea [42], followed by the Sinop coast [43]. The Hg levels found in the present study were the lowest. The highest amount of Cd in the bumper fish was found in the Sea of Marmara and the Aegean Sea. Cd values in the present study are at the lowest levels. Likewise, the highest amount of Pb was found in the Sea of Marmara [44]. The Pb values in the present study are like the previous study conducted on the coasts of Sinop [45]. Cu and Zn, which are essential metals, were found mostly in the Sea of Marmara [44]. The mean concentrations of Cu and Zn in this study were like previous studies in Sinop coasts [45].

Health risk analysis evaluation

Table 2 presents the EDIs, the EWIs, the HQ, the THQ and the RI of

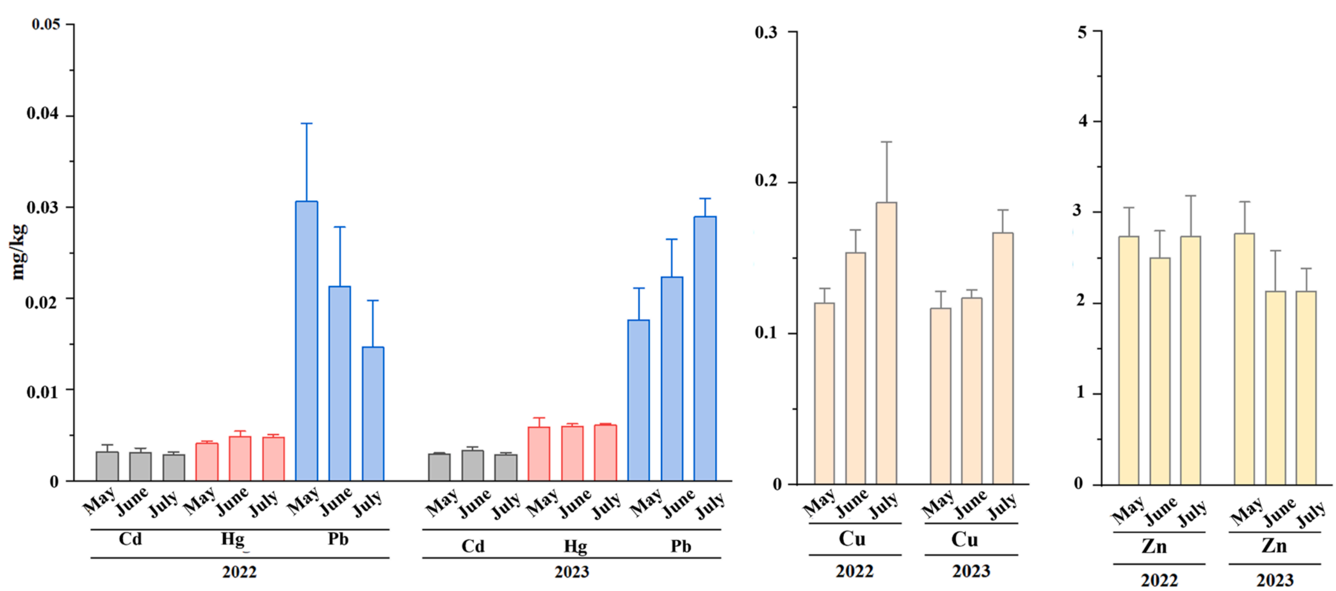

Fig. 2. The trace elements in the edible tissues of *S. porcus* collected from Sinop coasts of the southern Black Sea.

Table 1
Comparison of heavy metal concentrations (mg/kg wet wt.) in *S. porcus* with the other studies (ND: not detected; *: dry wt.).

Location	Metals					References
	Hg	Cd	Pb	Cu	Zn	
Aegean Sea	–	0.005–0.04	0.01- 0.08	0.03–0.09	0.92–2.23	[46]
Aegean Sea*	–	0.80 ± 0.06	0.66 ± 0.06	0.73 ± 0.06	95.3 ± 8.7	[47]
Sea of Marmara	–	0.20 ± 0.07	0.83 ± 0.19	5.29 ± 1.88	26.2 ± 4.31	[44]
Black Sea- Sevastopol	0.06 ± 0.01	0.04 ± 0.01	0.16 ± 0.04	0.60 ± 0.10	5.14 ± 1.25	[42]
Black Sea- Sinop	–	0.02–0.023	0.03- 0.07	0.88–1.7	7.44–12.3	[48]
Black Sea- Sinop	0.01 ± 0.00	ND	0.04 ± 0.03	0.07 ± 0.009	–	[43]
Black Sea- Sinop	ND-0.012	ND-0.01	ND-0.02	0.05–0.14	0.9–3.22	[45]
Black Sea- Sinop	0.004 ± 0.0005 (0.0039–0.0055)	0.0031 ± 0.0005 (0.0023–0.0039)	0.022 ± 0.009 (0.009–0.039)	0.153 ± 0.036 (0.11–0.21)	2.65 ± 0.33 (2.2–3.2)	This study 2022
Black Sea- Sinop	0.006 ± 0.0005 (0.0048–0.0068)	0.0031 ± 0.0003 (0.0027–0.0037)	0.023 ± 0.006 (0.014–0.031)	0.135 ± 0.025 (0.11–0.18)	2.34 ± 0.44 (1.7–3.1)	This study 2023

Table 2
The EDIs and EWIs for all heavy metals, HQs and THQ for Cd, Hg, Cu, and Zn and risk index (RI) for Pb in age groups consuming *S. porcus*.

Metal	Year	EDI (mg/kg/day)		EWI (mg/kg/week)		Rf. D. (mg/kg/day)	SF (mg/kg-day)	HQ		RI	
		(children)	(adults)	(children)	(adults)			(children)	(adults)	(children)	(adults)
Cd	2022	2.81×10^{-6}	1.80×10^{-6}	1.97×10^{-5}	1.26×10^{-5}	1.0×10^{-3}		2.81×10^{-3}	1.80×10^{-3}		
	2023	2.79×10^{-6}	1.79×10^{-6}	1.95×10^{-5}	1.25×10^{-5}	1.0×10^{-3}		2.79×10^{-3}	1.79×10^{-3}		
Hg	2022	4.20×10^{-6}	2.69×10^{-6}	2.94×10^{-5}	1.89×10^{-5}	3.0×10^{-4}		1.40×10^{-2}	8.98×10^{-3}		
	2023	5.49×10^{-6}	3.52×10^{-6}	3.84×10^{-5}	2.46×10^{-5}	3.0×10^{-4}		1.83×10^{-2}	1.17×10^{-2}		
Pb	2022	2.03×10^{-5}	1.30×10^{-5}	1.42×10^{-4}	9.11×10^{-5}		8.5×10^{-3}			1.73×10^{-7}	1.11×10^{-7}
	2023	2.10×10^{-5}	1.35×10^{-5}	1.47×10^{-4}	9.43×10^{-5}		8.5×10^{-3}			1.79×10^{-7}	1.15×10^{-7}
Cu	2022	1.40×10^{-4}	8.98×10^{-5}	9.80×10^{-4}	6.29×10^{-4}	4.0×10^{-2}		3.50×10^{-3}	2.25×10^{-3}		
	2023	1.24×10^{-4}	7.94×10^{-5}	8.67×10^{-4}	5.56×10^{-4}	4.0×10^{-2}		3.10×10^{-3}	1.98×10^{-3}		
Zn	2022	2.43×10^{-3}	1.56×10^{-3}	1.70×10^{-2}	1.09×10^{-2}	3.0×10^{-1}		8.08×10^{-3}	5.18×10^{-3}		
	2023	2.14×10^{-3}	1.37×10^{-3}	1.50×10^{-2}	9.61×10^{-3}	3.0×10^{-1}		7.14×10^{-3}	4.58×10^{-3}		
THQ	2022							2.84×10^{-2}	1.82×10^{-2}		
	2023							3.13×10^{-2}	2.01×10^{-2}		

heavy metals for children and adults consuming *S. porcus* from Sinop coasts of the Black Sea.

The EDI values of Cd for *S. porcus* consumers (children, adults) are quite below the respective Rf. D. (0.0001), indicating that the Cd levels consumed through this fish is within safe levels. No difference was found between Cd EDI values in 2022 and 2023. The EDI values of Hg for both children and adults' consumers are also below the Rf. D. (0.0003), suggesting that the consumption of *S. porcus* does not pose significant health risks associated with Hg exposure. Among the Hg EDI values, 2023 is slightly higher than 2022. The EDI values of Cu for children and adults are highly lower than the Rf. D. (0.04), suggesting that the amount of Cu consumed through *S. porcus* is safe. The EDI values of Zn for *S. porcus* consumers are well below the Rf. D. (0.3), indicating that the consumption of this fish does not pose any significant health risks. Considering the EDI values for both Cu and Zn, 2022 is slightly >2023. Like other metals, the EDI values of Pb are below the SF (0.0085), indicating that the consumption of *S. porcus* does not lead to harmful Pb exposure [25,26].

The THQ values for Cd, Hg, Cu, and Zn are below 1, indicating that the noncarcinogenic health risk associated with these metals' intake through *S. porcus* consumption is negligible. Similarly, the RI values for Pb are exceptionally low, implying a minimal carcinogenic risk associated with Pb intake for consumers. As described in Table 2, it can be pointed out that the concentration of heavy metals in *S. porcus* from Sinop coasts of the Black Sea in 2022 and 2023 is within safe and acceptable limits for human consumption. The black scorpionfish is considered safe for consumption, and the levels of heavy metals in edible tissues do not pose significant health risks to consumers, including children and adults.

Despite initial concerns regarding cleaning difficulties associated with dorsal fin toxins, the study underscores the safety of consuming this species, supporting its suitability as a sustainable food source amidst declining wild fish stocks.

Conclusion

Fish tissues are bioaccumulated with heavy metals like Cd, Hg, Pb, Cu, and Zn instead of decomposing naturally. Consuming these fish can have a negative impact on one's health, even at low concentrations, and can interfere with the function of essential components. Even so, metals like Cd, Hg, and Pb have harmful effects, and concentrations that exceed safety thresholds can be lethal. But, if present within the allowed range, metals like Cu and Zn are essential for the regular operation of the metabolic process. According to the current study, the levels of metals found in edible tissues of the black scorpionfish are lower than those allowed by EU and Turkish regulations, which deems them safe for ingestion by humans. The study concludes that black scorpionfish sourced from Sinop coasts of the Black Sea are safe to eat as they do not contain excessive levels of heavy metals. Given the species' popularity amid declining wild fish stocks, *S. porcus* supports its viability as a sustainable and healthy food choice. Future studies should expand to include broader geographical areas and consider long-term environmental impacts to ensure comprehensive assessment of health risks associated with consuming this species.

Ethics approval

Not applicable.

Consent to participate

Not applicable.

Consent to publish

Not applicable.

CRediT authorship contribution statement

Levent Bat: Writing – review & editing, Writing – original draft, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Ayşah Öztekin:** Writing – review & editing, Visualization, Software, Methodology.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Data availability

No data was used for the research described in the article.

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