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Primary study on metal amounts in *Lophius piscatorius* Linnaeus, 1758 obtained from fish markets in Sinop, Turkey

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Abstract

The levels of five heavy metals (mercury, cadmium, lead, copper and zinc) were analyzed in edible tissues of *Lophius piscatorius* Linnaeus, 1758 marketed in Sinop coasts of the Black Sea by using inductively coupled plasma mass spectrometry. With the present study, heavy metal values of this fish in the Black Sea were examined for the first time. The mean concentrations in mg kg^{-1} wet wt. of Hg, Cd, Pb, Cu and Zn were 0.022, 0.009, 0.035, 6.3 and 16 mg kg^{-1} wet wt., respectively. The results of the present study indicate that the consumption of muscle from anglerfish can be considered safe in terms of permissible legal limits. It is clear that, Zn showed the high accumulation in muscle tissue followed by Cu, while non-essential metals Hg, Cd and Pb showed the low accumulation. This could be explained by the fact that Zn and Cu are essential elements in the bodies of living organisms and has an important role in different physiological processes. In the present study, heavy metal levels in angler fish were low. Likewise, the calculated HI values were lower than one. In conclusion, the results of the present study indicate that the consumption of muscle from anglerfish can be considered safe in terms of permissible legal limits and hazard index values

Keywords: Black Sea, Sinop, Heavy Metals, Lophius piscatorius, Hazard Index

Major classification: Health Science (Environmental Safety and Engineering)

1. Introduction

The pollution of the Black Sea has been studied for many decades (Bat et al., 2018). The Black Sea is not only contaminated by the its coastal countries but also by contaminants from several European countries via River Danube. The maximal serious of these contaminants are heavy metals. In a review, Bat et al. (2018) showed that the heavy metal contaminations in the Black Sea with industrial, agricultural and domestic discharges from rivers, fisheries and touristic activities are main sources. It is particularly important for benthic organisms, as heavy metals remain in the seawater and in the final destination sediment without degradation and degradation for a long time (Bat & Ö zkan, 2019). Therefore, studies on the determination of the amount of heavy metals in marine organisms are very important. The European Parliament stated in the area of Marine Environment Policy **M**arine **S**trategy

Framework **D**irective (Official Journal of the European Union, 2008), ecological characteristic and unity in estuarial, the inshore and offshore systems, has been improved to preserve and restore. MSFD is set on the tenet of ecological system-based management makes allowances for all forces of the seas and approaches to the sea regionally. The goal of the directive in the EU by 2020 is to make sure Good Environmental Status of the seas (GES). The objective of the MSFD with concern to Descriptors 8 and 9 is to supply that amounts of pollutants are at quantities not displaying increase to pollution effects. The appraisal of accomplishment of GES should be based on basis of monitoring programmes covering the MSFD indicators. MSFD Descriptor 8 "Concentrations of contaminants are at levels not giving rise to pollution effects" and Descriptor 9 "Contaminants in fish and other seafood for human consumption do not exceed levels established by Community legislation or other relevant standards", are aiming the subject of marine contamination. Necessary metals, like Cu and Zn, play a fundamental role in biological systems by getting involved in myriad enzymatic reactions, while other heavy metals like Hg, Cd and Pb are non-essential and are depicted by their big toxicity even at very nominal levels.

Aims of the current study were to determine the amounts of Hg, Cd, Pb, Cu and Zn in muscles of *Lophius piscatorius* Linnaeus, 1758 marketed in Sinop coasts of the Black Sea and to evaluate the intake of these metals resulting from the consumption of anglerfish.

2. Material and Methods

2.1. Collection of animals

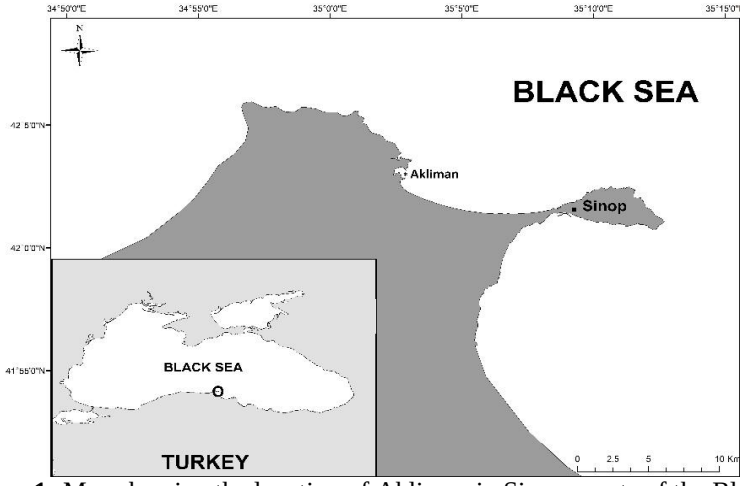

Figure 1: Map showing the location of Akliman in Sinop coasts of the Black Sea.

Figure 2: *Lophius piscatorius* Linnaeus, 1758 (photo L. Bat)

2.2. Analytic procedure

The edible tissues of anglerfish samples were made for metal analysis pursuant to the method depicted by Bernhard (1976). For heavy metal analysis, the edible tissues of the anglerfish digested with Suprapur® HNO_3 (nitric acid) using a Milestone Systems, Start D 260 microwave digestion system. In parallel, blanks and certified reference material samples were made similarly to verify the accuracy and precision of the method using the certified reference material NRCC-TORT-2 lobster hepatopancreas. The retrieval percentages got with these reference materials ranged from 96 to 103%. Determination and quantification limits were considered the instrumental reaction of the equipment. Tissues were analyzed in triplicate by an ICP-MS, Agilent Technologies, 7700x and the results were expressed as mg kg^{-1} wet weight (wt.).

2.3. Calculation of Intake Levels

The mean heavy metals daily intake was calculated according to the following formula:

Intake level = mean metal content X daily consumption of fish per person/ body weight

Estimated weekly intake (EWI) values were calculated from estimated daily intakes (EDI) values. According to health risk, the tolerable daily/weekly intakes were estimated by means of references for edible soft tissues of fish consumed by people. Considering an average diurnal eating of fish muscle for the adult person of 16.7 g/day for 2015 issued in the paper on seafood consumption by the Turkish Ministry of Agriculture and Forestry Fishery Statistics (BSGM, 2019).

2.4. Appraisal hazard index (HI) of studied metals in anglerfish

Risk from heavy metals taken into the body through ingestion may be defined employing a hazard index (HI) as the ratio of the calculated metal dose (EDI mg/kg of body wt. per day) and the reference dose (Rf. D. mg/ kg day^{-1}). The HI was found by using the calculation below:

HI = EDI/ Rf. D.

If HI > 1.0, then the EDI of a particular metal exceeds the Rf. D., showing that there is a potential risk associated with that metal. It depends on both the metal levels and the amount of consumption of fish.

3. Results

The results between the analytical and the certified values were in good agreement, together with the Relative Standard Deviation (RSD) percentage which was about ±7% for all studied metals.

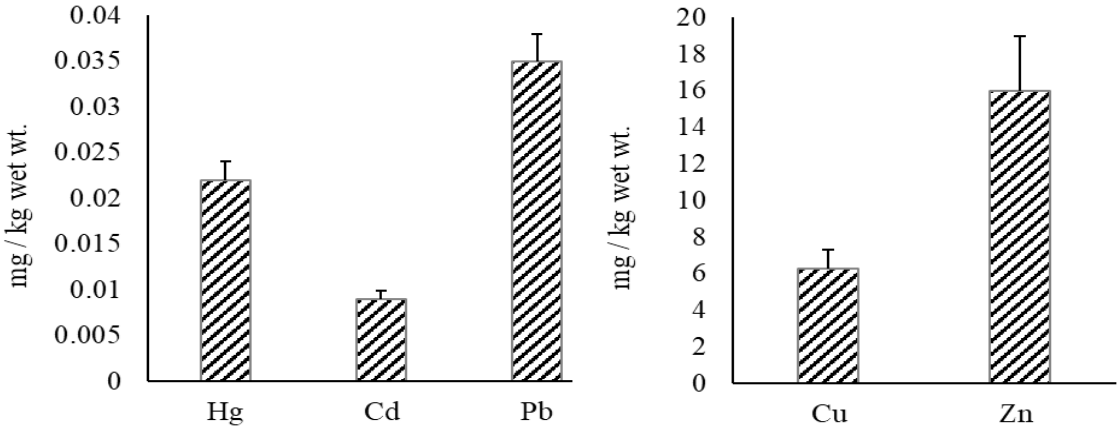

Figure 3: The means with standard deviations (vertical line) of Hg, Cd, Pb, Cu and Zn concentrations (mg/kg wet wt.) in edible tissues of *Lophius piscatorius* marketed in Sinop coasts of the Black Sea.

Figure 3 shows the means amounts of Hg, Cd, Pb, Cu and Zn as mg kg^{-1} wet wt. in edible tissues of *L. piscatorius* marketed in Sinop coasts of the Black Sea. Of the five heavy metals analyzed, Zn was the metal with the highest concentrations in edible tissues followed by Cu, Pb, Hg and Cd.

The EDI and EWI values given in Table 1 were estimated by assuming that a 70 kg person will consume 16.7 g fish /day which are 117.9 g seafood/week (BSGM, 2019). The tolerable daily and weekly intake of heavy metals, are set by the Food and Agriculture Organization/World Health Organization (FAO/WHO) Joint Expert Committee on Food Additives (JECFA) (Council of Europe, 2001; FAO/WHO, 2010 and 2011; WHO, 1989, 1996, 2000 and 2004). HI was calculated from EDI and Rf. D. values for each metal and given in Table 1.

Table 1: Mean levels (mg/kg wet wt.) in edible tissues of anglerfish, Rf. D. (mg/kg body wt. /day), EDIs (EWIs) and HIs values for consumers.

Metal	Mean Levels	Rf. D.	EDI (EWI) mg/kg body wt. /day	Hazard index (HI)
Hg	0.022	0.0003	$5.25x10^{-6}$ ($3.67x10^{-5}$)	0.017495238
Cd	0.009	0.001	$2.15x10^{-6}$ ($1.5x10^{-5}$)	0.002147143
Pb	0.035	0.0035	$8.35x10^{-6}$ ($5.85x10^{-5}$)	0.002385714
Cu	6.3	0.04	$1.50x10^{-3}$ ($1.05x10^{-2}$)	0.037575
Zn	16	0.3	$3.82x10^{-3}$ ($2.67x10^{-2}$)	0.01272381
		Total		0.072326905

4. Discussion

The mean levels of Hg, Cd and Pb (0.022, 0.009 and 0.035 mg kg^{-1} wet wt.) in the edible tissues of anglerfish did not passed the maximal levels set by a European Commission regulation (No 1881/2006) (1 mg of Hg kg^{-1} wet wt., 0.05 mg of Cd kg^{-1} wet wt. and 0.3 mg of Pb kg^{-1} wet wt., respectively for *Lophius* spp.). The European Commission regulation does not give acceptable values for essential heavy metals. However, the amounts of Cu and Zn allowed by the Turkish authorities in fish is 20 and 50 mg kg^{-1} wet wt. (Official Gazette of Republic of Turkey, 1995). The Cu and Zn levels in muscles of anglerfish were 6.3 and 16 mg kg^{-1} wet wt., respectively. All these results showed that the studied heavy metals were below acceptable values in the anglerfish from Sinop shores of the southern Black Sea.

Till now heavy metal accumulation studies related to angler fishes in the Black Sea have not been found. There is no study to compare the results obtained in the Black Sea. For this purpose, the results of the available studies carried out different seas and current study are compared and are given in Table 2.

Comparing the heavy metals levels in edible tissues of *Lophius* spp. with different regions of the world, it is obtained that similar amounts of Hg (0.02-1.02 mg kg^{-1} wet wt.), Pb (0.001-0.051 mg kg^{-1} wet wt.) and Cu (6.24 mg kg^{-1} wet wt.) were reported in *L. americanus* of N Atlantic (Hall et al., 1978), *L. americanus* of NW Atlantic (Johnson et al. 2011) and *L. budegassa* of Iskendurun Bay (Yilmaz et al. 2010), respectively.

It is also noted that the amounts of the studied metals in the current study were appreciably lower than those normally obtained in the scientific study on *Lophius* spp. (Table, 2) except Cu in *L. piscatorious* of Rockall Through (Mormede & Davies, 2001) and in *L. budegassa* of NE Mediterranean Sea (Perçin Olgunoglu et al. 2015), and Zn in *L. americanus* of N Atlantic (Hall et al. 1978) and in *L. budegassa* of NE Mediterranean Sea (Perçin Olgunoglu et al. 2015).

Heavy metal concentrations in benthic fish species depend on many factors such as length and weight and age of fish, seasons, physicochemical properties of the water bodies and many properties of sediment such as organic matter, particle size affect the bioaccumulation of metals.

The adverse effects of toxic contaminants such as Hg, Cd and Pb on seafood including fish have been discussed in detail and recommendations have been made (EFSA, 2010, 2012a,b,c). *L. piscatorius* is very tasty and economic value is very high fish. Their bodies are without scales; their skin is mucus and soft. They can live for a long time after getting out of the water. Their teeth are hooked. It's a very glutton fish. They mostly feed on fish. They prefer muddy-sandy floors where they can be easily buried (Bat et al., 2011). They are marketed fresh and frozen. Since the meat is tasty, it is used in all processed forms such as steamed, sautéed, broiled, boiled, fried, microwaved and baked (Froese and Pauly, 2019). The daily intakes (EDIs) of Hg, Cd, Pb, Cu and Zn were calculated considering the means of these heavy metals in the muscles of *L. piscatorius* and the average eaten of that anglerfish daily for adults as indicated by United States Environment Protection Agency (US-EPA, 2000). Table 1 indicates that EDIs of

hazard metals (Hg, Cd and Pb) and necessary metals (Cu and Zn) for adults are notably lower than the permitted amounts of national and international regulations. HIs of each heavy metal suggest that metals levels in edible tissues do not show any apparent hazard to the people, where the total HI of all these studied metals was under the value of 1 as given by Table 1.

Table 2: Comparison of the amounts of metals in *Lophius* spp. from the different locations in the world seas (nd=below limit of detection).

Species	Region	Unit	Tissue	Metals					References
				Hg	Cd	Pb	Cu	Zn	
Lophius americanus	N Atlantic	mg/kg wet wt.	muscle	0.02-1.02	0.02-0.16	0.13-0.78		2.5-9.5	Hall et al. 1978
			liver	0.01-0.05	0.04-1.07	0.22-1.0		8.94-31.25	
Lophius piscatorious	Italy	mg/kg wet wt.	muscle	0.61-2.22					Storelli et al. 2000
Lophius budegassa				0.22-1.62					
Lophius piscatorious	Rockall Through	µg/g dry wt.	muscle	nd	<0.002	<0.002	0.15	nd	Mormede & Davies, 2001
			gonad		<0.005				
			gill		0.021				
			liver		0.19				
Lophius budegassa	Iskenderun Bay	µg/g wet wt.	muscle		0.02	0.17	6.24	20.8	Yilmaz et al. 2010
			liver		0.26	1.77	31.0	38.2	
			skin		0.09	1.69	9.49	40.8	
Lophius americanus	NW Atlantic	mg/kg wet wt.	muscle	0.103-0.59	0.002-0.0023	0.001-0.051		3.4-14.6	Johnson et al. 2011
			liver	0.037-0.445	0.032-1.436	0.004-0.111		8.0-24.0	
			gonad	0.029-0.103	0.001-0.039	0.001-0.124		8.1-37.6	
Lophius budegassa	Iskenderun Bay	µg/g dry wt.	liver	nd	0.26	1.77			Renieri et al. 2014
			skin	nd	0.09	1.69			
			muscle	nd	0.02	0.17			
Lophius budegassa	NE Mediterranean Sea	µg/g dry wt.	muscle		nd	nd	0.19	5.88	Perçin Olgunoglu et al. 2015
			gill		nd	nd	0.98	14.5	

5. Conclusion

The Black Sea receive large quantities of many pollutants including heavy metals. The release of untreated sewage, agricultural and industrial wastes is the source for enriching these pollutants at the water column and sediments of coastal area remarkably in the locality of the hot points. In particular, the bottom organisms living in these regions, including fish may be exposed to heavy metals. This can cause serious health problems in humans via consumption of contaminated fish.

It is clear that, Zn showed the high accumulation in muscle tissue followed by Cu, while non-essential metals Hg, Cd and Pb showed the low accumulation. This could be explained by the fact that Zn and Cu are essential elements in the bodies of living organisms and has an important role in different physiological processes. In the present study, heavy metal levels in angler fish were low. Likewise, the calculated HI values were lower than one.

In conclusion, the results of the present study indicate that the consumption of muscle from anglerfish can be considered safe in terms of permissible legal limits and hazard index values.

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7. Conflict of interest statement

The authors declare that there are no conflicts of interest.

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