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TRACE TOXIC MINERAL LEVELS OF SEA LETTUCE (*Ulva* spp.) FROM COAST OF ISTANBUL

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ABSTRACT

Concentrations of Nickel (Ni), Copper (Cu), Zinc (Zn), Iron (Fe), Arsenic (As), Mercury (Hg), Lead (Pb) and Cadmium (Cd) were determined in the macro algae sea lettuce (*Ulva* spp.), sampled from the coastline of Istanbul old city (Cankurtaran) in summer 2016. The abundance of trace toxic mineral concentrations in sea lettuce were in the following order: Fe > Zn > As > Cu> Ni >Pb> Cd. However, mercury was not detected in any sample. The present study provides a new information to the consumer on the distribution of trace toxic minerals in sea lettuce.

Keywords: Marmara Sea, Istanbul coast, Trace toxic minerals, *Ulva* spp., Sea lettuce

Introduction

Trace toxic minerals are natural substances that have a high atomic weight and a density at least 5 times greater than that of water and these minerals are termed as heavy metals. Some trace minerals taken together with the food have a very important role in human life and are known to be necessary up to a certain concentration for human metabolism, as in the example iron, zinc and copper. Whereas, the majorities of heavy metals are toxic even at low concentrations such as arsenic, cadmium, chromium, lead and mercury. The multiple factors such as industrial, agricultural, medical and technological applications of trace toxic minerals have led their wide distribution in the environment. The trace toxic minerals, in particular, mercury, lead, cadmium and arsenic are frequently detected in aquatic organisms and raised concerns regarding the potential human health impacts. The toxic effects of these trace minerals may depend upon a variety of factors such as dose, exposure route and chemical structure, as well as age, gender, genetics and nutritional status of the exposed individuals (Belitz et al., 2009; Özcan, 2004; Soylak et al., 2005; Tchounwou et al., 2012).

The degree of pollution by trace toxic minerals in marine environments can be estimated by analysis of water, sediment and tissues of organisms (Morillo et al., 2005). Marine algae species are generally used to determine coastal waters trace toxic mineral grades in worldwide. In this process, it takes advantage of the key role of algae in the food chain and its temporal relationship with the pollutants (Topcuoğlu et al., 2010). The analysis of sediments in marine environments always faces limitations and the concentration of a trace toxic mineral in the sediment varies, among other factors, depending on the rate of deposition and the nature of the particles. This does not reflect the bioavailability. Macro algae appear to be the most appropriate indicators of both active and passive minerals (Villares et al., 2010). Sea lettuce (*Ulva lactuca*) is a macro algae which has the potential importance in terms of bio indicators with the tendency to absorb trace toxic minerals and spread in cytoplasmic cells in marine contamination with toxic pollutants (Davis et al., 2003).

Trace toxic mineral-related pollution in coastal regions of Marmara Sea has become a significant problem because of intensive industrial activity, ship wastes, dense population and construction origin pollution and municipality wastewater discharges. Sea lettuce is a type of macro algae and continues its vital activities by fixing itself to rocks and taking nourishment from marine environment. Figure 1 shows the general distributions of this species, which has a large habitat from tropical to polar areas. This species is frequently seen in the rocky coastal lanes of the Black Sea, Marmara and Aegean Sea in Turkey.

Therefore, the aim of the present study is to evaluate the pollution in the Marmara Sea through determination of trace toxic metals in the sea lettuce obtained from old city region Cankurtaran Istanbul coast of the Marmara Sea.

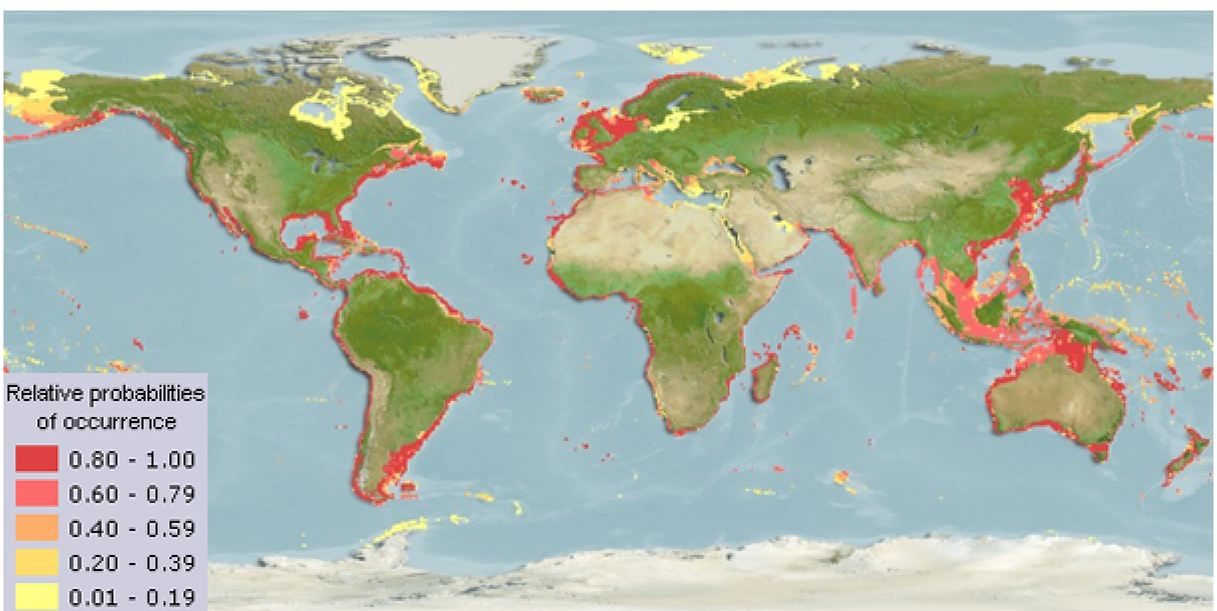

Figure 1. The habitats and distribution (native range) of *Ulva lactuca* in world (AquaMaps, 2016a)

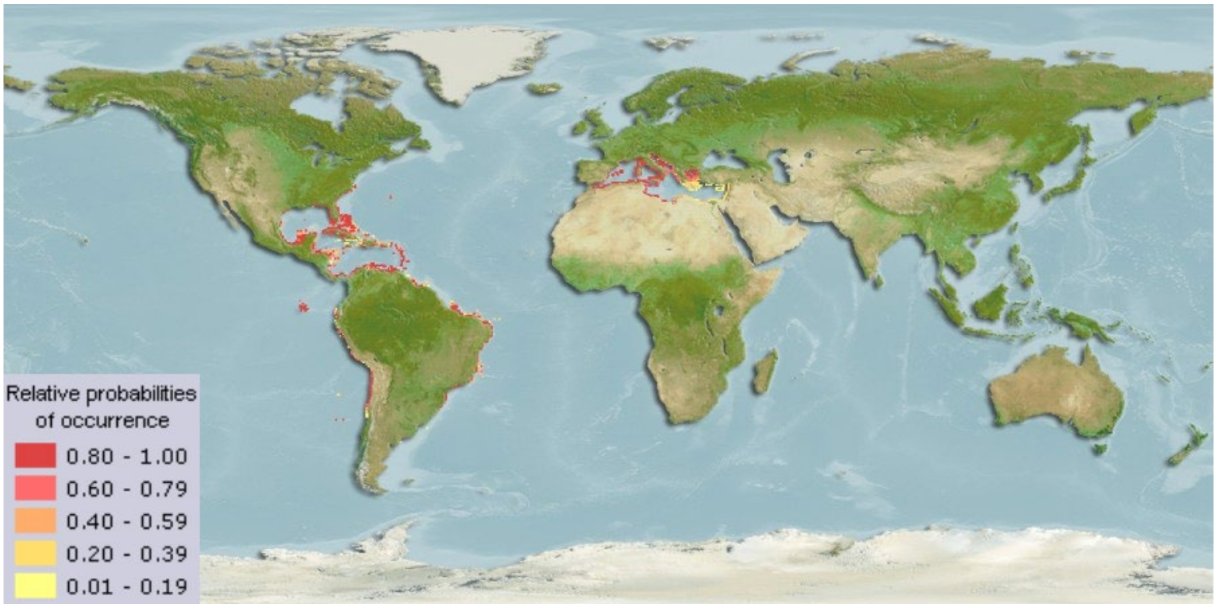

Figure 1. The habitats and distribution (native range) of *Ulva rigida* in world (AquaMaps, 2016b)

Material and Methods

Sea lettuce (*Ulva* spp.) were obtained (5 kg wet sample) from old city region Cankurtaran, Istanbul coast of the Marmara Sea (Figure 1) in year 2016 (end of summer) and dried under shadow (end dry product ~1 kg).

Determination of trace toxic minerals in dry sea lettuce (Ni, Cu, Zn, Fe, As, Hg, Pb, Cd) was carried out using an Inductively Coupled Plasma – Mass Spectrometry (ICP-MS) (Analytic Jena PlasmaQuant® MS, ICP-MS). An aliquot of 250 mg dried sea lettuce sample was weight into a pre-cleaned Teflon tube and 8 mL of concentrated nitric acid (65%) added. The samples digested by microwave assisted digestion system. (Table 1.). After digestion, samples diluted to 50 mL with ultrapure water. The analysis performed with "NMKL No: 186 - Trace Elements - As, Cd, Hg, Pb and Other minerals. Determination by ICP-MS After Pressure Digestion, 2007" method (NMKL "Nordic Committee On Food Analysis", 2007) (Table 2., 3.). The accuracy and precision of the analytical method checked using the certified reference material, powdered muscle tissue (Catalogue No. ERM-BB422). It was found that RSD % did not exceed 5.0%. All trace toxic mineral concentrations were determined on a mg/kg dry weight basis.

Table 1. Microwave Digestion program for sea lettuce sample preparation

Berghof Microwave Unit (Berghof - Speedwave SW4)				
Temperature (°C)	**Pressure (bar)**	**Ramp (min)**	**Time (min)**	**Power (%)**
155	50	8	8	90
205	50	8	30	90
50	0	1	15	0

Figure 1: Istanbul old city coast (Cankurtaran) of Marmara Sea.

Table 2. ICP-MS operating parameters for the quantification of trace toxic minerals in sea lettuce

ICP-MS (Analytic JENA Plasma Quant MS Elite) application parameters	Parameters Used
Nebulizer Gas Flow	1.06 L/ min.
Auxiliary Gas Flow	1.3 L/ min.
Plasma Gas Flow	9.0 L/min.
ICP RF Power	1400 watts
Pump Rate	10 rpm
Stabilization Delay	40 second

Table 2. Trace toxic mineral standards used to establish ICP-MS calibration curve

Specifications of Standards used in ICP-MS					
Element Name	**Trademark**	**Catalogue Number**	**Main Stock Concentration**	**Measured in ICP-MS mass value**	**LOQ (ng/mL)**
Ni	VHG	PNIN-100	999 µg/mL	60	125.36
Cu	VHG	ACUN-100	1001 µg/mL	65	76.77
Zn	VHG	PZNN-100	1000 µg/mL	66	428.05
Fe	VHG	PFEN-100	1000 µg/mL	56	601.42
As	VHG	PASN-100	1008 µg/mL	75	105.04
Hg	VHG	PHGN-100	1002 µg/mL	200	44.57
Pb	VHG	PPBN-100	1005 µg/mL	206	31.50
Cd	VHG	PCDN-100	1000 µg/mL	111	18.53
Y	Inorganic Ventures	GGY1-1	1001 µg/mL	89	-
Y (Ittrium) element is used as internal standard in the device					

The internal standard concentration used was 2.0 ng/mL

Table 3. Concentration levels of the standards for ICP-MS Calibration curve

Element Name	Calibration Points of the Elements						
	Level 1	Level 2	Level 3	Level 4	Level 5	Level 6	
Ni	0.5	2.5	5	10	20	50	
Cu	0.3	2.5	5	10	50	100	
Zn	2	10	50	100	250	500	
Fe	2.5	10	20	50	100	250	**ng/mL**
As	0.5	1	2,5	5	10	50	
Hg	0.2	0.5	1	2.5	5	10	
Pb	0.15	0.5	1	2.5	5	10	
Cd	0.08	0.3	1	2.5	5	10	

Results and Discussion

The present study provides new information on the distribution of trace toxic minerals in sea lettuce (*Ulva* spp.) obtained from Cankurtaran, the old city coast of Istanbul (European seaside). The trace toxic mineral concentrations determined in the algae samples studied are given in Table 1.

The present study reports the trace toxic minerals pollution in the samples collected from the Istanbul old city (Cankurtaran) coastline in the summer of 2016. Similarly, in 2014, Ozyigit et al. studied trace toxic mineral contents of *sea lettuce* samples collected from different parts of Istanbul including Istanbul old city coast (Cankurtaran) station, Büyükada, Fenerbahçe, Maltepe, Bakırköy and Beylikdüzü stations (Ozyigit et. al., 2017). Table 5, shows the trace toxic mineral contents of sea lettuce samples collected from different coastal cities of Marmara Sea. The results of their study is presented in Table 5. As can be seen from the Table, the sea lettuce samples collected from Cankurtaran coastline were found to contain much lower levels of Cu, Zn, Fe, Pb and Cd than other coastal cities of Istanbul including Büyükada, Fenerbahçe, Maltepe, Bakırköy and Beylikdüzü. Consequently, it can be concluded that the Istanbul old city (Cankurtaran) coast is less affected from trace toxic mineral pollution than its other cities with coasts to Marmara Sea.

Trace toxic mineral contents of sea lettuce collected from other cities with coasts to Marmara Sea were also reported in the literature (Table 5 and Table 6).

Culha et al. studied the trace toxic mineral contents of sea lettuce samples collected from city of Yalova located on the coast of Marmara Sea (Culha et al., 2013). They reported lower Cd, Zn, Ni and Pb contents compared to our results for the sea lettuce samples obtained from Cankurtaran, coast of Istanbul old city (Table 6). However, Cu and Fe levels of sea lettuce collected from Cankurtaran were much lower than the samples collected from Yalova. In the same study, no information is reported for As and Hg in the samples of Yalova region.

In another study, Ergul et al. determined the trace toxic mineral contents of sea lettuce collected from **Dilovası** coast shore of Izmit Bay in 2009 (Ergul et al., 2010). The amounts of trace toxic minerals contained in the sea lettuce samples collected during the summer and autumn of 2009 were much higher (Zn:125.95-373.1 mg/kg, Fe: 1518.9-5249.8 mg/kg and Pb: 1.05-1.95 mg/kg) than the samples collected from old city coast of Istanbul (Cankurtaran) during the Summer of 2016 (Table 6).

Table 4. Toxic elements determined in *Ulva* spp. samples

Toxic Lements (mg/kg)	Ni	Cu	Zn	Fe	As	Hg	Pb	Cd
Sea lettuce	1.32 ±0.04	4.92 ±0.10	6.92 ±0.25	88.74 ±3.57	3.65 ±0.11	n.d.	0.26 ±0.01	0.05 ±0.00

n.d. Not Detected <0.02 ng/kg

Table 5. Trace toxic mineral contents (mg/kg) of the sea lettuce samples obtained from present study and those reported by Özyiğit et al. (2017) and Culha et al. (2013)

Sea lettuce	Büyükada	Fenerbahçe	Maltepe	Bakırköy	Beylikdüzü	Istanbul old city coast (Cankurtaran)[a]
Cd	0.45 ±0.01	1.01 ±0.01	1.49 ±0.03	2.22 ±0.04	3.22 ±0.05	**0.05** ±0.00
Cu	6.67 ±0.10	10.92 ±0.13	13.01 ±0.15	15.28 ±0.17	18.31 ±0.20	**4.92** ±0.10
Fe	553.3 ±11.00	686.2 ±13.10	721.23 ±16.12	775.3 ±19.20	989.3 ±29.20	**88.74** ±3.57
Pb	4.93 ±0.07	7.63 ±0.09	8.92 ±0.10	12.35 ±0.20	19.32 ±0.25	**0.26** ±0.01
Zn	15.16 ±0.22	22.23 ±0.52	26.87 ±0.73	31.88 ±0.81	41.23 ±1.02	**6.92** ±0.25
Ni	No Data	No Data	No Data	No Data	No Data	**1.32** ±0.04
As	No Data	No Data	No Data	No Data	No Data	**3.65** ±0.11
Hg	No Data	No Data	No Data	No Data	No Data	**n.d**

n.d. Not Detected <0.02 ng/kg

Table 6. Comparison of the results (mg/kg) obtained from the present study and the data reported in the literature.

Sea lettuce	Istanbul old city coast (Cankurtaran)[a]	Yalova (Culha et al., 2009)	Dilovası/Kocaeli (Ergul et al., 2010) Summer	Autumn
Cd	**0.05** ±0.00	<0.01	No Data	No Data
Cu	**4.92** ±0.10	12.44	No Data	No Data
Fe	**88.74** ±3.57	358.36	1518.9	5249.8
Pb	**0.26** ±0.01	<0,01	1.05	1.95
Zn	**6.92** ±0.25	5.99	125.95	373.1
Ni	**1.32** ±0.04	1.25	No Data	No Data
As	**3.65** ±0.11	No Data	No Data	No Data
Hg	**n.d**	No Data	No Data	No Data

[a] the results obtained from present study n.d. Not Detected <0.02 ng/kg

Conclusion

This study presents new information about the distribution of trace toxic minerals in sea lettuce from the old city coastal area of Istanbul (Cankurtaran). According to the results of the current study, there is no risk for health in the consumption and evaluation of this species. However, if the consumption and evaluation of this product is considered in the future, it is recommended that the water environment of the Black Sea, Marmara and Aegean Sea coastal production / harvest potentials must be monitored periodically.

Compliance with Ethical Standard

Conflict of interests: The authors declare that for this article they have no actual, potential or perceived conflict of interests.

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