High content of heavy metals in seaweed species: A case study in the Persian Gulf and the Gulf of Oman in the southern coast of Iran

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Abstract

The contamination of heavy metals is a serious environmental challenge which threatens human health through food chain. This study focused on pollution level of six heavy metals (Cu, Zn, Pb, Ni, Fe^{+2} , and Cd) by investigating 12 seaweed species collected from different intertidal areas in the Persian Gulf and the Gulf of Oman. In addition, the uptakes of heavy metals in seaweed species were examined. The results confirmed the heavy metals contamination in the studied seaweed species. In addition, the uptake level was affected by the type of heavy metal ($P \le 0.0001$). Additionally, the type of seaweed species and collection site affected the heavy metal uptake. Accordingly, the highest content of Fe^{+2} (2844) ppm) was found in *Dictyota* sp. and Nickel (Ni) was observed in *Padina gymnospora* (105.97 ppm) and *Hypnea* sp. (100.41 ppm). Furthermore, the highest concentrations of Zinc (58.46 ppm) and Copper (32.44 ppm) were found in *Sargassum angustifolium,* and *S. boveanum*, respectively. Additionally, Cadmium had the lowest concentration ranging from 4.8 ppm in *S. angustifolium* to 10.7 ppm in *Dictyota* sp. The lowest content of all tested heavy metals was observed in *Gracilariopsis persica*. Further, the results revealed that brown macroalgae (Phaeophyta) contaminated more than green (Chlorophyta) and red (Rhodophyta) macroalgae.

Keywords: Heavy metals, Persian Gulf, Seaweeds, Environmental pollution

Introduction

Contamination by oil fractions may persist in the marine environment for many years after an oil spill, depending on characteristics of oil such as type, spill size, and location (Tansel, 2014). However, the environment may recover quickly (within 2-10 years) in areas such as salt marshes and mangrove swamps. A spill can remain for more than 25 years if it is not related to the physical removal of oil (Kingston, 2007).

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Chronic oil inputs cause a range of biological effects such as repeated small spillages in coastal waters, as well as local and longterm impacts (Carpenter, 2019).

Heavy metals are considered as the most common environmental pollutants which prevent the presence of natural or anthropogenic sources (Khoshnam et al., 2017). Previous research showed that seaweeds can rapidly accumulate high concentrations of metals such as Cd and Cu (Jarvis and Bielmyer-Fraser, 2015). Further, the toxic metals may deposit in human body where enter seaweeds grown in aquatic systems. In addition, Cd, Hg, and Pb can be toxic even in very small quantities and biologically essential elements might cause toxic effects in high concentrations. Additionally, heavy metals can accumulate in fatty tissues and internal organs of the human body, which may affect the central nervous system. In another instance, arsenic as a metalloid element in organic and inorganic forms can produce different toxicities. This element causes genotoxic damage and is classified as a human carcinogen such as liver, bladder, lung, and skin cancers (Rose et al., 2007). There are some concerns about ecological and global public health issues regarding environmental contaminations caused by heavy metals (He et al., 2005). Metals can cause oxidative stress by increasing the formation of free radicals, and high levels of heavy metals in soils could make the environment unsuitable for plant growth and decrease biodiversity (Ghosh and Singh, 2005). Furthermore, in biological systems,

heavy metals damage cell membrane, mitochondria, lysosome, endoplasmic reticulum system, nuclei, and some enzymes with functional roles in metabolism, detoxification, and damage reparation (Wang and Shi, 2001). Further, metal ions interact with other cell components (e.g., DNA and nuclear proteins) and cause conformational disorders and changes in DNA structure (Chang, 1996; Beyermann and Hartwig, 2008).

Despite the widespread negative effects of metal pollution on marine ecosystems and humans, this type of pollution has received less attention. Previous studies showed that it can threaten all types of life in aquatic and moist terrestrial environments (Tawfiq and Olsen, 1993; Moreno et al., 2011). Anthropogenic sources of metals are derived from mining, petrochemical industries, printing, electronic industry, and municipal waste, which are ultimately discharged into the marine environment (Wang et al., 2013). Further, heavy metals polluting water inhibit growth, body size, and reproduction of fishes (Sarnowski and Jezierska, 2007). Although many coastal countries depend on desalinated seawater as a source of potable water for domestic and industrial use, heavy metals are considered as serious pollutants of the aquatic environments due to their accumulative behavior (Abdolahpur Monikh et al., 2015; Forouhar Vajargah et al., 2018). The chemical and biological analysis methods for tracing environment pollution efficiently have increasing the importance of environmental waste management (Allah et al., 1997). Different animals and plants (e.g., seaweeds) are often used as bio-indicators for checking the quality of effluent and surface water (Trifanet al., 2015).

Persian Gulf, bordered by several wealthy countries, is one of the most important traditional marine regions with 800 oil and gas platforms (Srinivasan and Swain, 2007). Over the past five decades, the gulf has been the main pathway for oil transportation and was damaged by oil leakages (Kazemi et al., 2012), which may potentially have destructive effects on its marine ecosystem. Furthermore, the increasing trend of urbanization and industrialization polluted the coastline by heavy metals (Kazemi et al., 2012). Although little is known about the toxicity of metals in seaweeds and their health risks, several studies have been conducted on different aspects of seaweeds in Persian Gulf and the Gulf of Oman (Sohrabipour et al., 2004; Jasbi et al., 2013; Moein et al., 2015; Pirian et al., 2018). The present study investigated the heavy metal absorption level

in 12 seaweed species collected from geographically distinct sampling sites in the Persian Gulf and the Gulf of Oman.

Material and Methods

Sampling

First, 12 seaweed species including green, brown, and red algae were collected from different intertidal regions of the Persian Gulf and the Gulf of Oman (Table 1 and Fig. 1) from May to June 2018. Species identification was carried out using standard keys (Bellorin et al., 2008; Sohrabipour and Rabiei, 2008; Kokabi and Yousefzadi, 2015).

The seaweed specimens were thoroughly washed with tap water. Then, all epiphytes and organic and inorganic debris were manually removed. Next, all samples were rinsed in distilled water and air-dried (24 ºC) for 72 h. Finally, the dried samples were stored at -20 ºC for further analyses.

Fig. 1. Map of the five regions in the south of Iran where seaweed species were collected

Heavy metal analysis

Samples were dried in the oven at 155 ºC for 30 minutes (Rattanasomboon et al., 2018). Dried and finely ground seaweed materials (0.5 g) were transferred into poly tetra fluoroethylene digestion vessels, and 5.0 ml of concentrated HNO3 (ultrapure 65%) was added and incubated at ambient temperature for 2 hours. Then, the samples were heated for 5 hours at 100 ºC until 1 ml of the acid remained. Next, the digested cool solutions were filtered by Whatman filter paper (No 41) and transferred to polypropylene volumetric tubes. Ultra-pure deionized water was added to make up 50 ml volume for instrumental analysis in three replicates. The blank was prepared based on Trifan et al.'s method (2015). The concentration of six heavy metals including Zn, Pb, Cu, Ni, Cd, and Fe^{+2} was measured using the atomic absorption device (Thermo Electron Corporation, S series, UK). The multiparameter analyzer (HACH, HQ 40 d, USA) and refractometer (ATAGO, S/Mill-E, Japan) were used for seawater analysis, pH, salinity, and EC parameters.

Statistics analysis

One-way ANOVA was used to compare significant differences using SAS software (Version 9.4) (SAS, 1998), and mean grouping was performed by the Least Significant Difference (LSD) test ($P < 0.05$).

Results

Average uptake concentration of Zn, Pb, Cu, Ni, Cd and Fe were significantly different among 12 studied species. The F value, df, and mean square of error for each element were as follows: $F_{11,0.005} = 92708.9$, $F_{11,0.000}$ = 54626.9, $F_{11,0.001}$ = 91288.3, $F_{11,0.000}$ = 4496553, $F_{11,0.002} = 4626.06$, and $F_{11,0.09} = 3.66$ for Zn, Pb, Cu, Ni, Cd, and Fe, respectively $(P \le 0.0001)$ (Table 2).

S. angustifolium collected from Bandare-Lengeh and *G. persica* collected from Qeshm Island had the highest (58.46 ppm) classified in group A, and the lowest Zn content (19.67 ppm) was placed in class L, respectively (Table 3 and Fig. 2).

The *Dictyota* sp. collected from Qeshm Island had the highest Pb content (19.17 ppm) and bunched in class A. The species *G. folliifera* collected from Bushehr with 7.41 ppm Pb concentration had the lowest content of Pb and grouped in class K (Table 3

and Fig. 3).

S. boveanum and *S. angustifolium* collected from Bandare-Lengeh had the highest $(32.44$ ppm) and lowest $(11.17$ ppm) Cu content and were grouped in class A and L, respectively (Table 3 and Fig. 4).

The highest and lowest content of Ni was

Fig. 2. Variation in Zn concentration in studied species

Fig. 3. Pb concentration level in studied species

Fig. 4. Cu concentration in studied seaweeds

Fig. 5. Ni concentration in different seaweed species

Fig. 6. Cd concentration in different seaweed species.

Fig. 7. Iron (Ferro) concentration in studied species

observed in *P. gymnospora* (105.97 ppm) and *S. angustifolium* (12.89 ppm) collected from Qeshm Island and Bandar-e-Lengeh, respectively (Table 3 and Fig. 5).

However, *Dictyota* sp. and *S. boveanum* var. *aterrimum* collected from Bushehr had the highest content of Cd (10.7 and 10.66 ppm, respectively) and were placed in class A. *S. boveanum* collected from Bandar Abbas (4.8 ppm) had the lowest content and was placed in class H (Table 3 and Fig. 6).

Accordingly, the descending order of Fe concentration was observed in *Dictyota* sp. (2843.96 ppm), *P. gymnospora* collected from Bushehr (2606.78 ppm), *Hypnea* sp. (2556 ppm), *C. glubifera* (2447.15 ppm), *S. boveanum* var. *aterrimum* (1906.16 ppm), *S. boveanum* collected from Bandar-e-Lengeh (926.1 ppm), *G. folliifera* (772.6 ppm), *P. gymnospora* collected from Qeshm Island (644.74 ppm), *Ulva* sp. (440.57 ppm), *S. angustifolium* (231.3 ppm) and *G. persica* (196.08 ppm), respectively (Table 3 and Fig. 7).

Among the studied heavy metals, Fe had the highest concentration and variance in concentration (196.08-2843.96 ppm) followed by Ni (12.89-105.97 ppm), Zn (19.67-58.46 ppm), Cu (11.17-32.44 ppm), Pb (7.41- 19.17 ppm), and Cd (4.8-10.7 ppm) (Table 3 and Figs. 2-7). While the lowest levels of EC (52.8 ms/cm) and salinity (37/6 PPT) were measured in Chabahar, the highest levels of EC (62.7 ms/cm), salinity (42 PPT), and pH (8.27) were observed in Bushehr (Table 4). Table 5 presents the comparison of the results of the current study with previous studies.

Discussion

The present study provided valuable information related to heavy metal contamination in some marine seaweed species collected across Iranian coastlines of the Persian Gulf and the Gulf of Oman. The results showed that Fe, Ni, Zn, Pb, Cu, and Cd were found in all of the studied seaweed species. The content of metals in seaweed tissues varied from 4.8 to 10.7 ppm for Cd, 11.17 to 32.44 ppm for Cu, 8.37 to 19.17 ppm for Pb, 19.67 to 58.46 ppm for Zn, 12.89 to 105.97 ppm for Ni, and 196 to 2843.96 ppm for Fe. Juma and Al-Madany (2008) examined seawater contamination in the territorial water of the Kingdom of Bahrain (Persian Gulf) and observed 5 ppm of Cd, 5 ppm of Cu, 25 ppm of Pb, 40 ppm of Zn, 30 ppm of Ni, and 1000 ppm of Fe content in seawater. The results are consistent with the published data of the United States Environmental Protection Agency (USE-PA), reporting 8.8 ppm of Cd, 3.1 ppm of Cu, 8.1 ppm of Pb, 81 ppm of Zn , 610 ppm of Ni, and 300 ppm of Fe. Al-Abdali et al. (1996) studied the bottom sediments of the Persian Gulf and found that the amounts of all metals were within the permissible range $(Zn = 0.60, Pb = 15-30, Cd = 1.2-2.0, Ni =$ 70-80, Fe = $10,000$ -20,000 and Cu = 15-30 ppm). In addition, the content of Cd in shallow water was higher than those at the bottom of the sea. In another study, Janadeleh and Jahangiri (2016) investigated the heavy metal concentrations in sediments and fish

Region	EC (ms/cm)	pH	Salinity (PPT)
Bandar-e-Lengeh	56.6	7.97	38.4
Chabahar	52.8	8.1	37.6
Bushehr	62.7	8.27	$42/0*$
Qeshm Island	56.6	7.94	38.9
Bandar Abbas	56.9	7.49	39.3

Table 4. Seawater analysis in different collection sites

*Samples with ≥40 PPT salinity, were detected by refractometer device.

body of the Persian Gulf (Hormuz strait) and represented that the contents of Cd, Zn, Ni, Pb, and Fe were 0.12-0.43, 25.83-112.31, 24.63-42.38, 5.32-10.12, and 10800-22400 ppm, respectively. The consistency of the recent results with other findings (Table 5) determining pollution level in seaweeds can provide more information about contamination of seaweeds, especially those used for food and cosmetic purposes.

The comparison of the findings of this study with those of the previous studies indicated that the values of Ni and Cu increased significantly by increasing moisture-proof paints (Srinivasan and Swain, 2007). Dadolahi et al. (2011) studied the heavy metal absorption in seaweed and associated sediment harvested from the Strait of Hormuz and showed that the amounts of Pb, Cd, Cu, Ni, Zn, and Fe were 13.3-30.5, 0.7-7, 6.35-16.87, 21.46-71.6, 39.65-54.93, and 7441-14867 in different seaweed species, respectively. Indeed, the results confirmed the differences in metal contamination in different classes of dry seaweeds. For instance, Pb content was 23.95, 20.53, and 22.82 in Chlorophyta, Phaeophyta, and Rhodophyta, respectively. Cd content was 3.27, 5.09, and 5.05, while it was 11.95, 11.68, and 10.92 for Cu. Regarding Ni, it was 47.04, 40.72, and 52.98, while it was 45.99, 46.53, and 51.03 for Zn. Finally, it was 11550, 6371, and 11085 for Fe. In addition, the results of this study could confirm these achievements. Qari (2015) studied heavy metal contamination in *Padina pavonia* and *P. tetrachromatic* species and found that the level of Fe, Zn,

Pb, Cu, Ni, and Cd was 0.25-1.64, 0.091- 0.76, 0.25-0.63, 0.047-1.22, 0.037-0.42, and 0.017-0.38 ppm, respectively. Accordingly, the Persian Gulf was more polluted than the open seas.

The results also indicated a relationship between contamination level in seaweeds and the collection site. For instance, *S. boveanum* collected from Bandar-e-Lengeh and Bandar Abbas had different levels of contamination, which were also true for *P. gymnospora* collected from Qeshm Island and Bushehr. Additionally, the results revealed that brown macroalgae (Phaeophyta) had a higher level of contamination compared with green (Chlorophyta) and red (Rdodophyta) macroalgae (Dadolahi et al., 2011; Qari, 2015). Additionally, contamination level was different based on the seaweed species. For instance, *S. angustifolium, S. boveanum* var. *aterrimum*, *Dictyota* sp., *S. boveanum*, and *P. gymnospora* showed a higher level of concentration for all six heavy metals while *G. persica, G. folliiferda,* and *Ulva* sp. had the lowest level of Fe, Ni, Pb, and Zn.

Furthermore, the findings indicated contamination variability in the same seaweed species collected from different location, while Fe and Ni had the highest rate in *Dictyota* sp., *P. gymnospora*, *Hypnea* sp., and *C. glublifera* had only high content in *Hypnea* sp. and *P. gymnospora* collected from Qeshm Island. Further, *Sargassum boveanum, S. angustifolium, Hypnea* sp., *C. glublifera, P. gymnospora*, and *Dictyota* sp. could greatly uptake Zn. Furthermore, Cd was more accumulated in *Dictyota* sp. (10.7 ppm), and high concentration of Pb was observed in *Dictyota* sp*. Sargassum boveanum* var. *aterrimum, P. gymnospora*, and *S. boveanum* exhibited higher levels of Cu. Additionally, the brown seaweed species were more efficient biosensors for studying Cu and Pb (Figs. 3 and 4). The findings are consistent with the results of a study by Hiroyuki (2015) who found *P. gymnospora* was an effective biosensor for Cu and the concentration of heavy metals (Cu, Fe, and Pb) was different in either different seaweed species or collection sites. For instance, the concentration of Cu ranged from 1 to 20 ppm in brown seaweeds and 2 to 40 ppm in red seaweeds. Regarding the collection site, the Cu content ranged from 3 to 20 in green seaweed collected from the north and south of Kii Peninsula and 2 to 80 ppm in green seaweeds in Kanayama. Additionally, the high concentration of Cu in marine ecosystems made the environment unsuitable for fishes, seaweeds, planktons, and other marine organisms. This contamination may be caused by oil-producing and shipping companies, antifouling coating paints, and industrial and agricultural centers (Giusti, 2001; Caliceti et al., 2002).

The concentration value of Pb varied from 0.2 to 20, 0.2 to 10, and 2 to 40 ppm in the same seaweed species collected from different collection sites (Hiroyuki, 2015). This difference was observed in the present study based on seaweed species and collection site. For example, *S. boveanum* collected from Bandar-e-Lengeh absorbed more concentration of Pb, Cu, Ni, and Fe than *S. boveanum* collected from Bandar Abbas. Additionally, Zn content in *S. boveanum* from Bandar Abbas was higher than the species from Bandare-Lengeh. However, the concentration of Cd in the same species collected from these two sampling sites remained unchanged.

In addition, some environmental factors such as salinity, pH, light intensity, and metabolic factors can affect the concentration of heavy metals in seaweeds (Zbikowski et al., 2006). For instance, Fe+2 contents in *P. gymnospora*, collected from Qeshm Island (644 ppm), were much lower than those collected from Bushehr (2606 ppm). In fact, the values of salinity in these two sampling sites were different, which could affect the results (Qeshm Island =36.8 PPT and Bushehr =42.0 PPT) (Table 4). Furthermore, metal pollution may affect other heavy metal contamination, which is considered as an important issue confirmed based on the results of the present study (Andrade et al., 2004). Foster (1993) reported that increased Zn contamination inhibited Cd uptake by seaweeds due to competition for binding sites. In other words, a positive or negative relationship may be observed between the uptakes of heavy metals in different seaweed species (synergistic or antagonistic interactions of ions in binding with the anionic sites). Furthermore, variation in concentration of different heavy metals may be due to the electronegativity values of metals. Fe, Ni, Zn, and Cu had higher electronegativity values and concentration than Cd. However, the high contents of Fe and Zn in the studied species could be related to the necessity of these elements for seaweeds, which is in line with those of Dadolahi et al. (2011). In addition, a relationship was reported between seaweed species and the concentration of heavy metals, which may be related to the morphological attributes of seaweed species. For instance, the filamentous seaweeds (*G. persica* and *G. folliifera*) represented lower metal uptake than membranous and thick-wall species such as *S. boveanum* and *P. gymnospora* (Trifan et al., 2015). Trifan et al. (2015) confirmed that filamentous seaweed species, *Enteromorpha intestinalis*, *Cladophora vagabunda*, *Ceramium rubrum*, and *Phyllophora pseudoceranoides* were able to uptake more Ni, Hg, Zn, Cd, Cu, Cr, Mn, and Pb than other morphological types of seaweed species. They suggested that these species were widely distributed and could be used as bio-indicators for assessing heavy metal contamination along sea coastlines.

During the recent decades, water sampling has been a common approach for tracing heavy metals contamination in seas and oceans (Hiroyuki, 2015). Based on this approach, a large number of sampling should be considered, which is a labor-intensive procedure. Furthermore, the results in the water sampling approach may change based on the variation in tidal range, temperature, salinity regimes, dissolved nutrients, geological structure of the study area (Dadolahi et al., 2011), sampling method, and sampling time or sampling site, which may lead to an inaccurate estimation about pollution level (Hiroyuki, 2015). Consequently, seaweed-based tracing of heavy metals is an easier and more cost-benefit approach which can be replaced by other sampling approaches. Based on the results, seaweed species can be used as worth biosensors for the assessment of heavy metal contamination. Thus, this information can be utilized as a benchmark for further studies on heavy metal contamination of the seaweed species in the Persian Gulf and the Gulf of Oman. It is evident that the endophytic bacteria, which are related to marine plants, can induce resistance to heavy metals in hosts. For example, *Shewanella* sp. and *Idiomarina* sp., which were isolated from red seaweeds, can reduce the uptakes of several metal ions (Konishi et al., 2007; Seshari et al., 2012; YokeshBabu et al., 2014). Additionally, the association of endophytes with different seaweed species reported in previous studies confirmed the important role of these microorganisms in seaweeds (Suryanarayanan, 1992; Devarajan and Suryanarayanan, 2002; Flewelling et al., 2015; venkatachalam et al., 2015; Kaaria et al., 2015). It seems that these microorganisms could act as a deterrent and decrease metals uptake by seaweeds (YokeshBabu et al., 2014). This study provided insights into the Iranian seaweed species, which can be properly used as a metal biosensor. Based on the findings, it is strongly recommended that seaweed species be used as proper biosensors to trace other heavy metals not included in this study.

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