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Full Length Research Paper

Brown macroalgae as bio-indicators for heavy metals pollution of Al-Jubail coastal area of Saudi Arabia

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Wastes from both industrial and domestic sources, as well as habitat destruction have a substantial impact on the coastal environments. It causes serious problems in many countries and for several seas and oceans which leads to the extinction of several plant and animal species. Many water resources are no longer suitable for drinking or for agriculture as a result of pollution. The main aim of this study was to investigate the efficiency of four brown macroalage as bioindicators for toxic heavy metals (manganese (Mn), copper (Cu), zinc (Zn), arsenite (As), cadmium (Cd), and lead (Pb)) along Al-jubail industrial city coast at Persian Gulf (Saudi Arabia). Brown macroalage samples were collected from three different sites in three time points, January, March and May, 2010. The four collected brown macroalgae were identified as *Sargassum angustifolium, Sargassum boveanum*, *Sargassum latifolium*, and *Padina gymnospora*. The algal samples were cleaned using sea water and distilled water, dried, and the concentrations of various toxic metals were determined. The average concentrations of Mn, Co, Ni and Cd were within the expected limits of un-contaminated areas. However, the results indicate the high level of Zn ion accumulation in all tested brown algae, showing highest concentration in *S. angustifolium* > *P. gymnospora* > *S. latifolium* > *S. boveanum* with highest Zn concentration of 991 ± 49.1, 988 ± 47.5, 980 ± 44.2, and 911 ± 39.7 μg g^{-1} dry weights, respectively. In addition, Cu was detected at high concentration of 92.1 ± 3.7 μg g^{-1} dry weight in *S. boveanum*. These results clearly indicate the high pollution levels of Al-jubail industrial city coast with Zn and Cu toxic heavy metals, which is mostly due to uncontrolled disposal of industrial waste into coastal area. Furthermore, the consistency of Zn concentrations in all tested brown algae indicated the efficiency of the tested algae, including *P. gymnospora, S. angustifolium*, *S. latifolium*, and *S. boveanum*, for bioaccumulation and bio-monitoring studies of Zn.

Key words: Brown algae, heavy metals, bio-indicators, *Sargassum* sp., *Padina* sp.

INTRODUCTION

Wastes from both industrial and domestic activities usually introduce huge amounts of pollutants into the marine environment, causing significant and permanent disturbances in the marine ecosystem systems and consequently, environmental and ecological deterioration (Buffle et al., 2009; Akcali and Kucuksezgin, 2011). This phenomenon is especially significant in coastal area, owing to the fact that these are the main drainages of most anthropogenic pollutants. It has long been reported that heavy metals in the marine environment have a particular importance in the ecotoxicology, as they are highly persistent and can be very toxic even in very low concentration (Simon et al., 2011).

Many toxic pollutants are found in only trace amounts in water, and often at elevated levels in sediments. Therefore, risk assessments based only on data derived from water analyses are usually misleading (Gosavi et al., 2004; Melville and Pulkownik, 2007). In water, pollutants concentrations are usually below the detection limits, and vary greatly both spatially and temporally (Villares et al., 2002). On the other hand, data from

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sediments may not be representative of contaminant concentrations in the overlying water column and cannot give information on patterns of contamination, especially at the higher levels of the food chain (Binelli and Provini, 2003; Torres et al., 2008). In addition, the concentrations available to biota are affected by several factors, including pH, salinity, particle size and organic content (Karez et al., 1994; Jothinayagi and Anbazhagan, 2009). Consequently, some aquatic organisms have become increasingly used in the assessment of contamination, as bioindicators. Algae and molluscs are among the organisms most used for this purpose (Villares et al., 2002; Simon et al., 2008; Lavoie-Michel et al., 2009; Topcuoğlu et al., 2010; Rajfur et al., 2010).

The main advantage of bio-monitoring approach using marine organisms compared to direct measurement in water or sediment is to provide a direct and time-integrated assessment of the metal fraction that is actually available to the organisms (Coteur et al., 2003; Danis et al., 2004; Metian et al., 2008). However, several criteria have been established to ensure appropriate organisms to be used as biomonitors and bioindicators. The organisms should ideally be sedentary, and thus reflect only pollutant specific to a particular site, easy to identify, and cosmopolitan, ensuring wide geographic relevance. Furthermore, bioindicators should be sensitive to contaminants and tolerate high contaminant concentrations and also provide sufficient tissue for analysis (Conti and Cecchetti, 2003; Melville and Pulkownik, 2008).

Of the many types of biosorbents including fungi, bacteria and yeasts, recently investigated for their ability to sequester heavy metals, brown algal biomass has proven to be highly effective as well as reliable and predictable in the removal of several heavy metals (such as Pb^{2+}, Cu^{2+}, Cd^{2+}, and Zn^{2+}) from aqueous solutions. The main mechanisms of heavy metals biosorption by brown algae include some key functional groups such as carboxylic groups, which are generally the most abundant acidic functional group in the brown algae. They constitute the highest percentage of titratable sites (typically greater than 70%) in dried brown algal biomass. The adsorption capacity of the algae is directly related to the presence of these sites on the alginate polymer, which itself comprises a significant component (up to 40% of the dry weight). Ion-exchange is another important concept in biosorption by brown algae (Davis, 2003; Moacir et al., 2008; Buffle et al., 2009; Akcali and Kucuksezgin, 2011).

The main objective of this work was to investigate the suitability of some brown macroalgae to act as bio-indicator for heavy metals pollution of Al-Jubail industrial city coastal area at the Persian Gulf (Saudi Arabia).

MATERIALS AND METHODS

Study area

Al-Jubail (27° 0′ N, 49° 40′ E) (Figure 1) is a city located in the Eastern province on the Persian Gulf coast of Saudi Arabia. It consists of the Old Town of Al-Jubail, which was originally a small fishing village. In 1975, Al-Jubail was designated as a site for a new industrial city by the Saudi government, and has seen rapid expansion and industrialization since. The seventh census report for Al-Jubail Industrial City, prepared in 2009, gives a resident population of 150,367. Al-Jubail city is considered as the largest industrial complex in the Middle East. It holds the Middle East's largest, and the world's 4th largest petrochemical company, fertilizer plants, steel works, industrial port and a huge number of support industries. There is also the Royal Saudi Naval Base, plus a separate Commercial Port and Military Air Base. In addition, Al-Jubail industrial city is home to the world's largest seawater desalination plant. It provides 50% of the country's drinking water through desalination of seawater from the Persian Gulf. Other industries include electroplating, leather tanning, dye and pigment manufacturing, wood processing, textile dyeing, steel and alloy industries, photographic sensitizer manufacturing and others.

Sampling procedures

Algal samples were collected in triplicates from three sites along Al-jubail industrial city coast using five liter polyethylene acid-washed bottles. Algal samples were collected during three surveys, in January, March and May, 2010. The samples were transferred to the laboratory in refrigerated box where they were cleaned with distilled water and identified according to previously reported protocols (Basson, 1979; Basson et al., 1988). All algal samples were air dried at 90°C and then kept for further analysis.

Analytical methods

The concentrations of manganese, copper, zinc, arsenic, cadmium and lead were determined in the collected algal samples according to previously reported methods (Levkov and Krstic, 2002; Murphy et al., 2008; Al-Homaidan, 2008; Al-Homaidan et al., 2011). 500 mg (dry weight) of each algal dried sample were placed in acid washed digestion tubes. 25 ml of concentrated analaR grade nitric acid was added to each tube and the contents were evaporated to about dryness. After cooling, 20 ml of double deionized water was added to each tube and the contents were filtered through 0.45 μM Millipore filters. The solutions were then transferred to 25 ml acid washed volumetric flasks and the volumes were completed to 25 ml with double distilled deionized water. The concentrations of various metals including Mn, Cu, Zn, As, Cd, and Pb were measured in aliquots of algae, using Inductively Coupled Plasma-Optical Emission Spectrometer (Optima 4300 DV). All samples were analyzed in triplicates and the concentrations were expressed in μg (metal) per gram dry weight (alga) and the mean values were recorded.

Statistical analysis

All analysis was made in three replicates. Statistical analysis was based on SPSS (Version 11.0) program. One-way analysis of variance (ANOVA) was used to evaluate the inter-specific significance between algal metal accumulation and between metal levels in different sites with $p = 0.05$ (Jothinayagi and Anbazhagan, 2009; Al-Homaidan et al., 2011).

RESULTS

In this study, four brown macroalagae were collected in triplicates from three different sites in three survey trips in

Figure 1. Location of Al-Jubail industrial city at the Persian Gulf coast of Saudi Arabia.

January, March and May, 2010. These algae were chosen to conduct the investigation as bioindicators for heavy metals pollution of Aljubail industrial city coastal area at the Persian Gulf (Saudi Arabia), due to the fact that they were available in all sites during all field expeditions. Based on Basson et al. (1979, 1988) protocols, the collected brown algae were identified as *P. gymnospora*, *S. angustifolium*, *latifolium*, and *boveanum* (Figure 2). Their proliferation in this area is probably attributed to the elevated levels of nutrients which prevailed in the stream and to the high concentrations of total dissolved solids. The concentrations of various toxic heavy metals were determined in all of the collected algal samples, including Mn, Cu, Zn, As, Cd and Pb.

The mean concentrations of the tested metals (Mn, Cu, Zn, As, Cd and Pb) in *P. gymnospora* are presented in Figure 3. The results indicate that the average concentrations of various metals were in the normal range except the concentrations of Zn, which was high and ranged from 668 ± 33.6 to 988 ± 47.5 µg g^{-1} dry weights, and highest concentration of 988 µg g^{-1} dry weights was recorded in the *P. gymnospora* samples collected in March collection. Figure 4 demonstrates the average concentrations of various metals in *S. angustifolium* which indicated high content of Zn with concentration range from 537 ± 27.2 to 991 ± 49.1 µg g^{-1} dry weights, with highest concentration in samples collected in January. However, the other tested metals were in normal range.

In contrast to other algae, the average concentrations

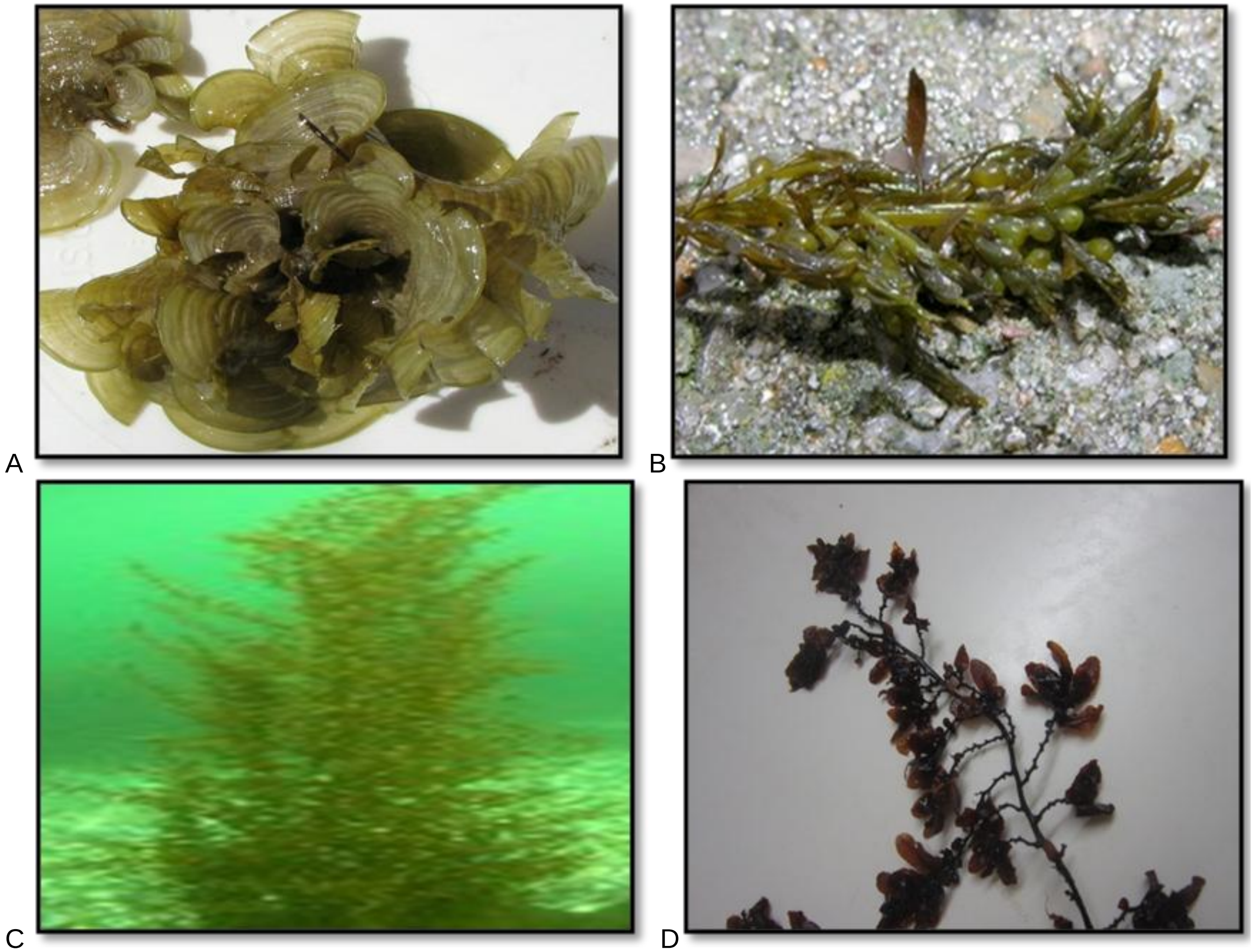

Figure 2. (A) *Padina gymnospora*; (B) *Sargassum angustifolium*; (C) *Sargassum latifolium* and; (D) *Sargassum boveanum*.

of Cu was high in *S. boveanum*, ranged from 6.1 ± 0.4 to 92.1 ± 3.7 μg g^{-1} dry weights, with highest concentration in samples collected in January. In addition to copper, average concentration of Zn was also high in this *S. boveanum* as well with highest concentration (911 ± 39.7 μg g^{-1} dry weight) in samples collected in March collection trip, while the other heavy metals (Mn, As, Cd and Pb) were in normal levels (Figure 5). The concentrations of various metals in *S. latifolium* are shown in Figure 6. Once again, Zn concentration was high and ranged from 1.0 to 980.0 ± 44.2 μg g^{-1} dry, with highest concentration in *S. latifolium* samples collected in March collection trip.

DISCUSSION

The passive removal, termed biosorption, of toxic heavy metals by inexpensive biomaterials requires that the substrate exhibits high metal accumulation ability and selectivity, suitable mechanical properties, as well as low cost for application in remediation technology (Davis et al., 2003; Torres et al., 2008).

In the last decade, many low-cost sorbents have been investigated, however the brown algae have proven to be the most effective and promising substrates for bioremediation of the toxic heavy metals from the aqueous solutions (Torres et al., 2008). This is mainly due to their basic biochemical constitution that is responsible for this enhanced performance among other biomaterials. It is the properties of cell wall constituents, such as alginate and fucoidan, which are chiefly responsible for heavy metal chelation (Davis et al., 2003). The brown algae are an important large assemblage of algae that are classified in about 265 genera, with more than 1500 species (Conti et al., 2007). They derive their characteristic brown color from the large amounts of the carotenoid fucoxanthin contained in their chloroplasts and the presence of various pheophycean tannins (Conti et

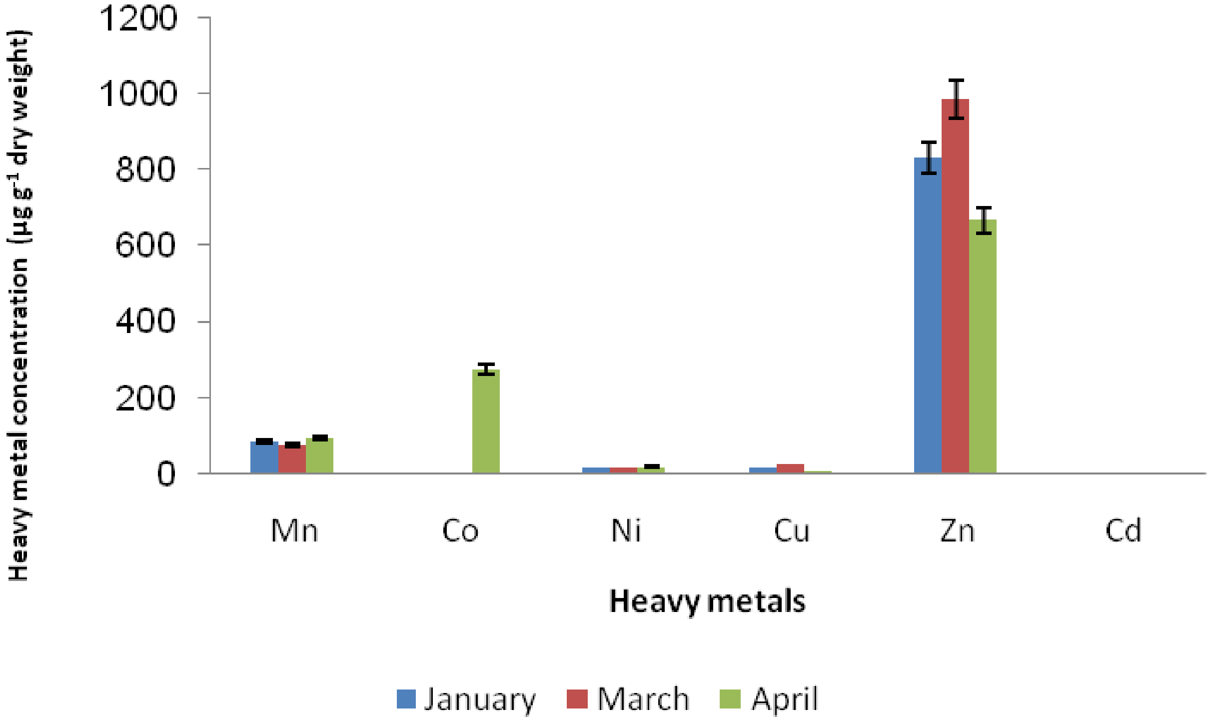

Figure 3. Concentrations of heavy metals in *P. gymnospora*. Various heavy metals were measured in the aliquots of algae using Inductively Coupled Plasma-Optical Emission Spectrometer (Optima 4300 DV). All samples were analyzed in triplicates and the concentrations are expressed in µg (metal) per gram dry weight (alga) and the recorded values are means and the error bars are indicated.

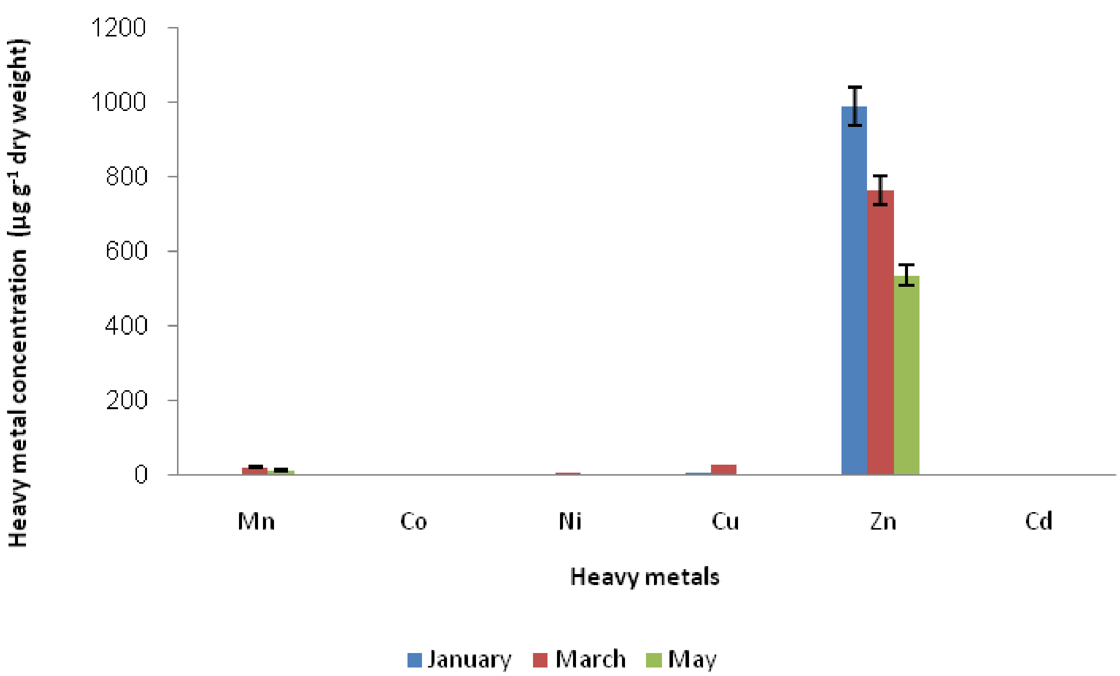

Figure 4. Concentrations of heavy metals in *S. angustifolium*. Various heavy metals were measured in the aliquots of algae using Inductively Coupled Plasma-Optical Emission Spectrometer (Optima 4300 DV). All samples were analyzed in triplicates and the concentrations are expressed in µg (metal) per gram dry weight (alga) and the recorded values are means and the error bars are indicated.

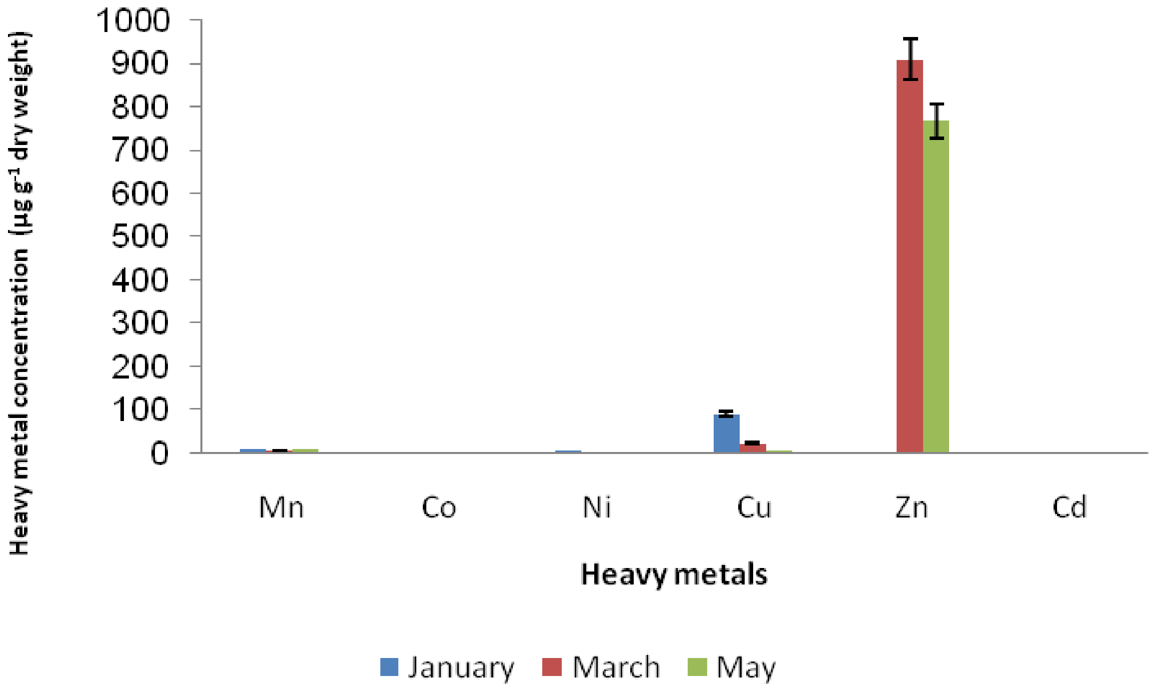

Figure 5. Concentrations of heavy metals in *S. boveanum*. Various heavy metals were measured in the aliquots of algae using Inductively Coupled Plasma-Optical Emission Spectrometer (Optima 4300 DV). All samples were analyzed in triplicates and the concentrations are expressed in µg (metal) per gram dry weight (alga) and the recorded values are means, and the error bars are indicated.

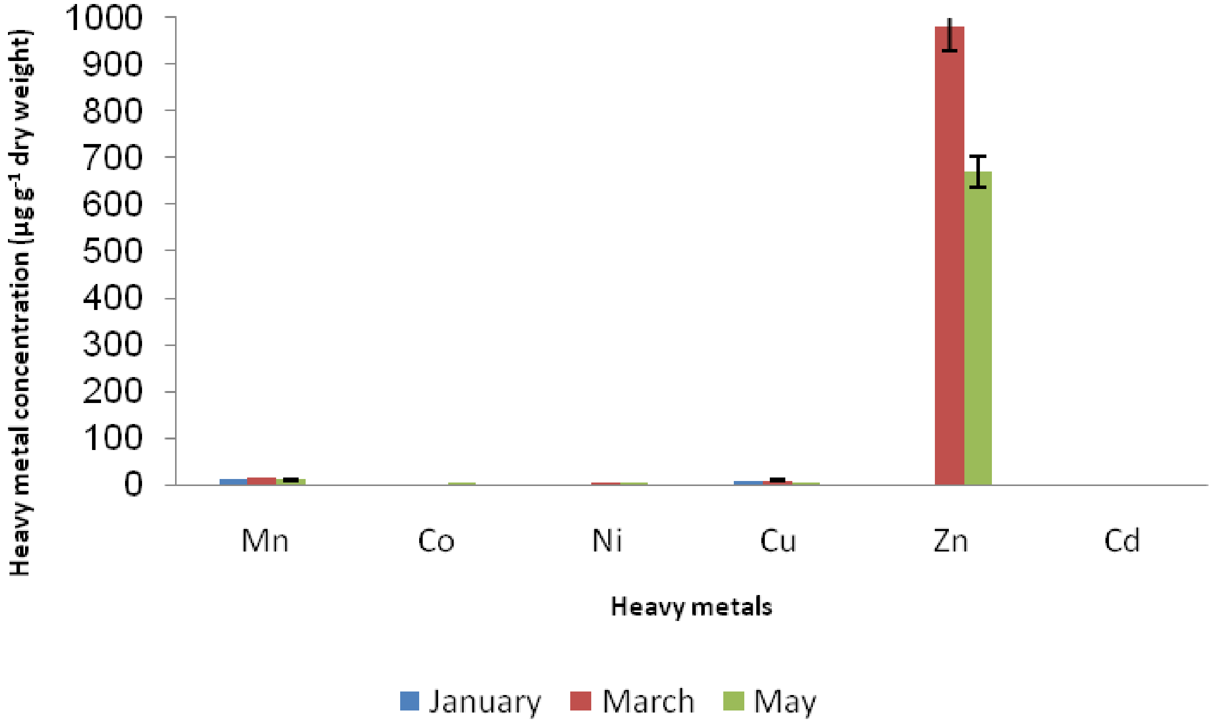

Figure 6. Concentrations of heavy metals in *S. latifolium*. Various heavy metals were measured in the aliquots of algae using Inductively Coupled Plasma-Optical Emission Spectrometer (Optima 4300 DV). All samples were analyzed in triplicates and the concentrations are expressed in µg (metal) per gram dry weight (alga) and the recorded values are means and the error bars are indicated.

al., 2007). They occur mainly in the marine environment, where they appear as an intertidal component. Some marine forms penetrate into brackish environments, and can be an important part of the salt marsh fauna. Brown algae flourish in temperate to subpolar regions where they exhibit the greatest diversity in species and morphological expression (Lee, 1989).

In this study, four brown macroalgae, including *P. gymnospora*, *S. angustifolium*, *latifolium*, and *boveanum* were selected for evaluation of their efficiency as bioindicators of heavy metals pollution at Al-Jubail industrial city coastal area of Saudi Arabia at Persian Gulf. These algae were selected to conduct the investigation due to the fact that they were distributed in sites during all field expeditions. Their proliferation is probably attributed to the elevated levels of nutrients which prevailed in the stream, and to the high concentrations of total dissolved solids. Furthermore, brown algae in general and in particular species of *Padina* and *Sargassum* were reported to be the best species for biomonitoring and phytoremediation of toxic heavy metals from coastal tropical environment (Davis et al., 203; Murphy et al., 2008; Jothinayagi and Anbazhagan, 2009). The concentrations of several toxic heavy metals were measured in aliquots of the four algal species collected in three surveys (January, March, and May, 2010). The results indicate the high level of Zn ion accumulation in all tested brown algae, showing highest concentration in *S. angustifolium* > *P. gymnospora* > *S. latifolium* > *S. boveanum*, with highest Zn concentrations of 991 ± 49.1, 988 ± 47.5, 980 ± 44.2, and 911 ± 39.7 µg g^{-1} dry weights, respectively. Such high concentrations are not usually encountered in these algae in unpolluted waters and much lower values have been reported by other workers from different countries (Storelli et al., 2001; Topçuoĝlu et al., 2003; Akcali and Kucuksezgin, 2011).

It has been reported that the average zinc residues in algae collected from polluted waters are within the range of 100 to 500 µg g^{-1} dry weight (Melville and Pulkownik, 2007; Al-Homaidan et al., 2011). By comparing the findings of this study with other investigations, it can be said that there is high degree of zinc pollution in the study area, Al-jubail industrial city coast. Furthermore, the consistency of Zn concentrations in all tested brown algae indicated the efficiency of the tested algae, including *P. gymnospora S. angustifolium*, *latifolium* and *boveanum*, for bioaccumulation and bio-monitoring studies of Zn.

In addition to Zn, Cu ion was detected at high concentration of 92.1 ± 3.7 µg g^{-1} dry weight in only *S. boveanum*. These high levels of copper are much higher than what is expected for uncontaminated coasts. It has been reported that a range of 10 to 100 µg g^{-1} dry weight as typical of attached algae species inhabited polluted waters (Moore and Ramamoorthy, 1984; Donat and Dryden, 2001; Sawidis et al., 2001).

These results clearly indicated the high pollution levels of Al-jubail industrial city coast with Zn and Cu toxic heavy metals, which is mostly due to uncontrolled disposal of industrial waste into coastal area. Moreover, the fact that the average concentrations of Mn, Co, Ni and Cd in all tested algal samples were within the expected limits of un-contaminated areas, and could be due other factors. It has been reported that the high levels of some heavy metals in the algae reflect firstly the high bioavailability of the metals in the study area and secondly the capacity of the alga to accumulate them. Many factors may influence the bioavailability of metals in algae including pH, salinity, temperature, light, particulate matters and organic matters (Jothinayagi and Anbazhagan, 2009). In addition to variations in the available metals concentrations in the ambient water, other factors such as water conditions, and the stage of development and variation in growth and chemical composition of the algae may influence the pattern of accumulation (Wai-Yin and Wen-Xiong, 2001; Jothinayagi and Anbazhagan, 2009).

Conclusion

The results of the current study clearly indicate the high pollution levels of Al-jubail industrial city coast with Zn and Cu toxic heavy metals, which is mostly due to uncontrolled disposal of industrial waste into coastal area. Furthermore, the consistency of Zn concentrations in all tested brown algae indicated the efficiency of the tested algae, including *P. gymnospora*, *S. angustifolium*, *latifolium*, and *boveanum* to act as bioindicators of Zn pollution. Also, the results indicate the high capability of *S. boveanum* for biosorption of Cu ion than other heavy metals, and its suitability as bioindicator of copper pollution. Finally, the results indicate that a special attention is required for treatment of industrial waste of Aljubail industrial city before disposal into the coast to meet the internationally accepted procedures for environmental/ecological impact and risk assessment to manage human impact on coastal environments.

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