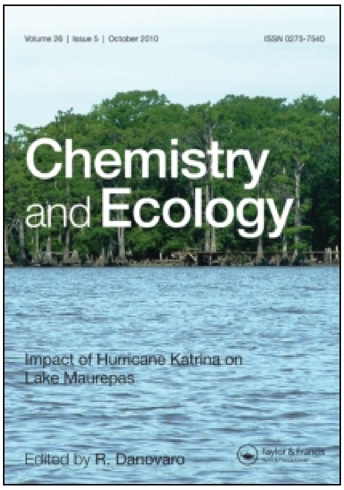

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Comparative study of heavy metal distribution in some coastal seaweeds of Alexandria, Egypt

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Comparative study of heavy metal distribution in some coastal seaweeds of Alexandria, Egypt

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The distribution of Cd, Cu, Fe, Hg, Mn, Ni, Pb, and Zn concentration in five algae species (*Enteromorpha intestinalis, Enteromorpha linza, Ulva lactuca, Corallina mediterranea, Pterocladia capillacea*) collected from three areas having different types of metal contamination along Alexandria coastal waters (Egypt) were determined by Atomic Absorption Spectrometry. The recovery study was carried out using a Certified Reference Material TORT-2. The obtained heavy metal contents indicate that different species demonstrate various degree of metal accumulation. *Enteromorpha intestinalis* (green algae) recorded the highest levels of Fe and Mn while *Corallina mediterranea* (Calcareous red algae) recorded the highest concentration of Pb, Ni, and Cd in the three areas under study. According to the present study, filamentous algae (*Enteromorpha intestinalis*) showed the ability to concentrate greater amount of trace elements than the foliaceous ones (*Enteromorpha linza*). In each area, mercury concentration has nearly the same value for all species under study.

Keywords: Algae; Seaweeds; Heavy metals; Pollution; Egypt

1. Introduction

Toxic and persistent substances are increasing in the environment continuously due to anthropogenic activities. They tend to concentrate in all aquatic matrices in the environment [1] and for this reason, in particular the rapid heavy metal diffusion as an environmental contamination has called attention to their determination at trace and ultratrace level [2–5]. Macrophytic algae, being one of the primary stages in the trophic chain, play a major role in marine ecosystems [6]. Algae interact with the environment through process that include chemical bioconcentration, excretion, organic matter production and decomposition [7]. They have been used as signal for the living status of marine ecosystems and considered as valuable indicators for heavy metals assessment in the major components of the water ecosystems because of their accumulation capacity [8–17]. Their immobility relates them directly to the surrounding environment, and therefore, it must be stressed that seaweeds do not vary their inner metal concentrations when the outer concentrations do so suddenly. However, if the change lasts over a long period of time, then algae will reflect the concentrations of the new environment, especially in the younger parts of the algae as the older ones are more resistant to concentration changes [18].

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Large quantities of seaweed are washed ashore along the Mediterranean Alexandria Coast. Their wide distribution and abundance in many areas could very well be used in assessing their bioaccumulation potential and contamination by heavy metals. Phillips [10] has advocated the use of macroalgae as indicator organisms for depicting the metal levels in a given milieu. Several researchers subsequently have shown that the use of marine algae as an indicator of heavy metals give at least a qualitative picture of heavy metals contamination [19–25].

The purpose of this paper is to give information about the accumulation and the distribution of eight trace metals in two abundant algae species (*Ulva lactuca* and *Corallina mediterranea*, green and red algae, respectively) over the three areas of El-Mex Bay, Eastern Harbour and Abu-Qir Bay, besides a comparative study of heavy metals accumulation for the available algae species within each area.

2. Materials and methods

Algae were collected in Spring 2002 from three locations namely El-Mex Bay, Eastern Harbour and Abu-Qir Bay which represent important fishery areas, play key roles as nursery grounds for commercially relevant fish species and are subjected to different pollutants. El-Mex Bay is affected by industrial wastes, in addition to huge amounts of drainage waters mixed with remains of pesticides and fertilizers. Eastern Harbour, semi closed basin is affected by untreated sewage [26] while Abu-Qir Bay is exposed to industrial and agricultural wastes [27] (figure 1).

Five marine algae species which belongs to green and red algae collected during spring 2002 from the above three areas at depth ranged between half to one meter (Tide 0.25 to 0.50 meter). About 0.5 to 1 kilogram fresh weight were harvested. Two algae species namely *Ulva lactuca* (C) and *Corallina mediterranea* (D) which belong to class *Clorophyta* and *Rodophyta*, respectively, were collected in the three areas under investigation. In addition, *Enteromorpha intestinals* (A) and *Enteromorpha linza* (B) (Clorophyta) were collected only at El-Mex Bay and *Pterocladia capillacea* (E) (Rodophyta) only on Eastern Harbour and Abu-Qir Bay.

The samples were washed in sea water at the sampling site and transferred to the laboratory at the same day in polyethylene boxes under refrigeration (4 °C). After their arrival at the

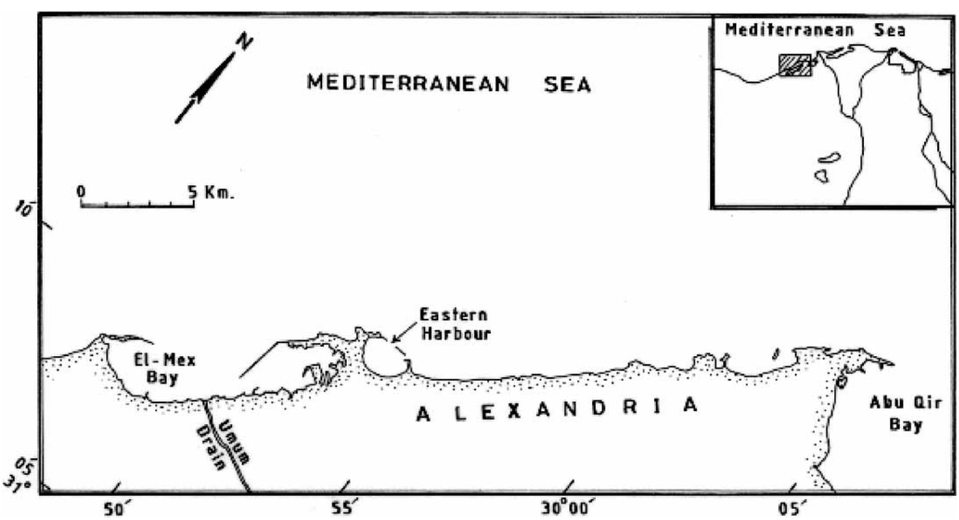


Figure 1. Study area and sampling locations.

Table 1. Recovery study (certified reference material TORT-2).

Element	Certified concentrations (μg/g dw)	Concentration found (μg/g dw)	Recovery %
Cd	26.7 ± 0.6	25.952 ± 0.820	97.199
Cu	106 ± 10	103.241 ± 3.124	97.397
Fe	105 ± 13	105.927 ± 5.317	100.883
Hg	0.27 ± 0.06	0.263 ± 0.026	97.407
Mn	13.6 ± 1.2	14.116 ± 1.164	103.794
Ni	2.5 ± 0.19	2.584 ± 0.261	103.360
Pb	0.35 ± 0.13	0.363 ± 0.146	103.714
Zn	180 ± 6	184.916 ± 5.348	102.731

Note: Number of samples in each measurement is 8.

laboratory, they were washed carefully in sea water to remove sand, particulate matter, and epithytal and epifaunal species, then they were rinsed in distilled water. Finally they were dried at 60 °C (to constant weight), homogenized by crushing each sample in a porcelain pestle and mortar and kept away from metallic materials and dusty conditions to avoid contamination.

Three sub-samples (0.5 to 1.0 g) of each sample were submitted to acid digestion using concentrated 5 ml HNO_3 (Merk, 65%) in Teflon-lined vessel by means of microwave oven (Sharp, Model-800) in pressure-controlled conditions. Digested samples were diluted to 25 ml with double distilled water and analyzed for metal contents [28]. Analysis of cadmium (Cd), copper (Cu), chromium (Cr), iron (Fe), manganese (Mn), nickel (Ni), lead (Pb), and zinc (Zn) were performed with a flame atomic absorption spectrophotometer (Perkin-Elmer, Model 2380). Mercury was analyzed by cold vapor (Spectra AA-10 Plus Varian). Procedural blanks were run within each beach. All glass wares, Plastic devices and Teflon devices were thoroughly acid washed [29]. Reagents of analytical grade were utilized for the blanks and calibration curves. The accuracy of the method was verified using standard reference materials (TORT-2) from National Research Council of Canada. Recoveries were above 95% for the trace metals measured (table 1). The reported results are mean values of triplicate determinations and expressed as μg/g dry weight. Correlations were carried out according to Pearson's correlation analysis.

3. Results and discussion

The results of analysis of eight metals studied in the five species of macro algae from the three sites are summarized in table 2. Iron and manganese were the most abundant metals in the studied algae, followed by zinc, lead, nickel, copper, cadmium, and mercury.

The highest cadmium concentration was recorded in *Corallina mediterranea* (11.08 μg/g dw) from Abu-Qir Bay, the lowest (0.45 μg/g dw) in *Ulva lactuca* from Abu-Qir Bay. Moreover, *Corallina mediterranea* recorded the highest concentration relative to other species in each of the three study areas (figure 2). This may be attributed to the faster rate of calcification in *Corallina mediterranea* in warmer areas which results in less strict regulation of minor and trace element uptake [30–32]. The present data are in agreement with those of Stenner and Nickless [33] who found that cadmium concentration ranged from 0.5 to 2.0 μg/g in *Ulva lactuca* and ranged from 4.4 to 6.2 μg/g in *Corallina mediterranea* from the Atlantic Coast of south west Spain and Portugal. Cadmium level of *Ulva lactuca* in the present study was higher than that reported by El-Sarraf [34] but comparable with that recorded by Abbas [35] along the Alexandria Coast.

Copper is considered to be required by all algae and not replaceable even in part by other elements. It also appears to play a role in the photosynthesis of plastocyanin in some algae [36].

Table 2. Concentration levels of heavy metals in algae species collected from El-Mex Bay, Eastern Harbour and Abu-Qir Bay (Egypt).

		Heavy metal concentrations (μg/g dw)								
Area	Species	Cd	Cu	Fe	Hg	Mn	Ni	Pb	Zn	MPI
El-Mex	A	2.071	9.689	724.433	0.143	54.598	11.767	21.314	38.378	13.432
	B	0.733	14.619	327.308	0.137	36.746	9.958	14.262	29.717	9.639
	C	0.918	10.728	118.656	0.110	32.975	7.234	14.243	18.245	7.289
	D	7.175	8.064	427.297	0.185	49.215	15.383	26.504	53.009	16.252
Eastern	C	1.088	26.143	109.231	1.232	12.629	8.963	9.413	50.429	10.943
Harbour	D	10.038	16.738	236.461	2.387	21.075	27.323	53.313	60.546	25.455
	E	0.541	10.864	303.816	1.246	31.059	8.942	23.897	74.823	13.504
Abu-Qir	C	0.447	5.643	130.073	1.168	28.761	7.429	17.104	7.964	5.093
	D	11.081	4.657	128.777	1.542	36.248	28.196	49.376	13.379	16.982
	E	1.156	5.299	92.837	0.993	32.855	7.301	23.508	49.158	10.576

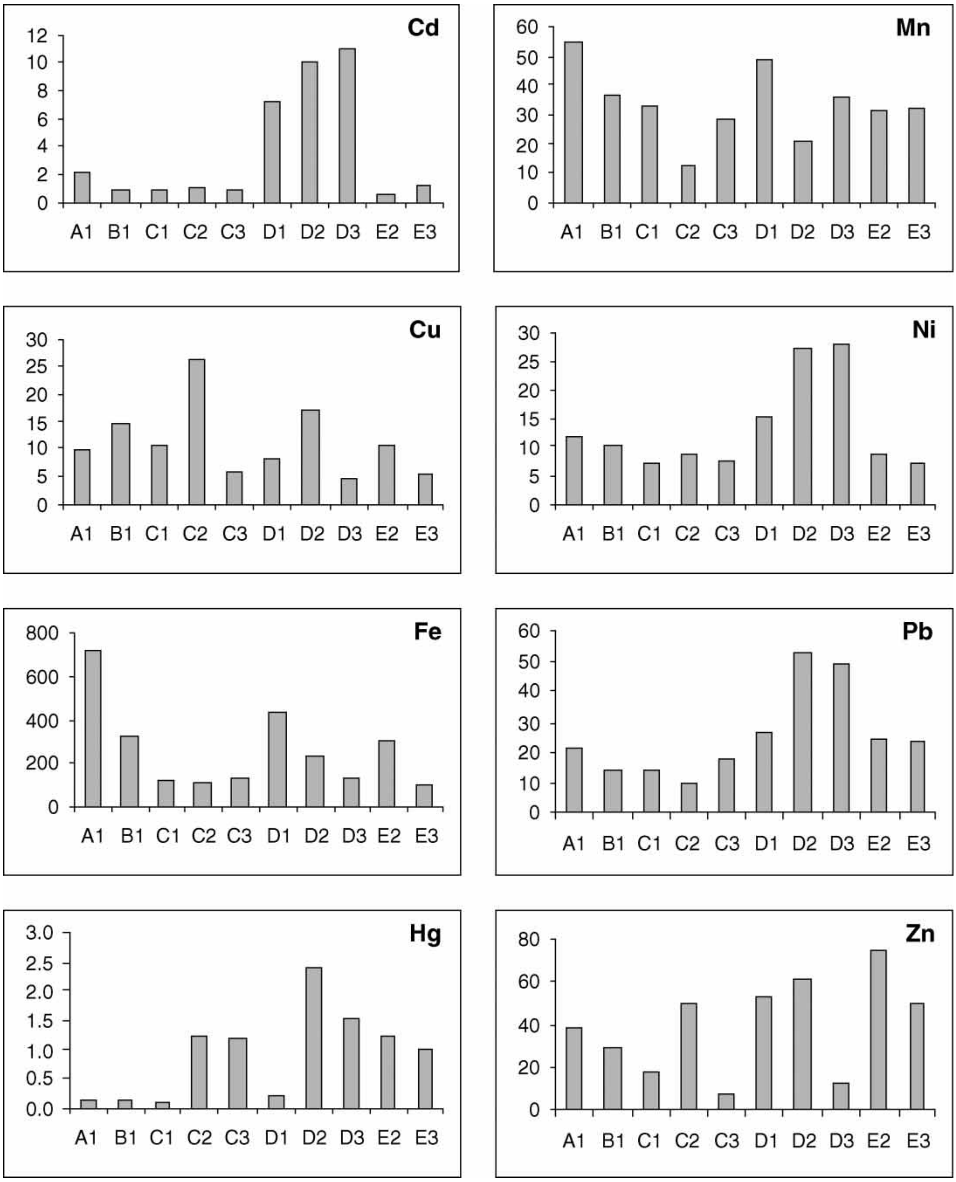
Note: A: *Enteromorpha intestinalis*; B: *Enteromorpha linza*; C: *Ulva lactuca*; D: *Corallina mediterranea*; E: *Pterocladia capillacea*.

The present study reveals that the highest concentration of copper was observed in *Ulva lactuca* collected from Eastern Harbour (26.14 μg/g dw). Copper content depends on the sampling sites as shown by *Ulva lactuca* and *Corallina mediterranea* (table 2). This may be related to the amount of copper concentration in the anthropogenic sources of each area. Furthermore, the concentration of Cu fluctuated between 9.69, 14.62 to 10.728 μg/g dw in *Entermorpha intestinalis, Enteromorpha linza* and *Ulva lactuca* in El-Mex respectively, which belong to the class *Chlorophyceae* (green algae). The Cu concentration for the three species *Ulva lactuca, Corallina mediterranea* and *Pterocladia capillacea* recorded their highest levels in Eastern Harbour followed by El-Mex Bay and Abu-Qir Bay. These high values are attributed to that, Eastern Harbour and El-Mex Bay are said to be the most polluted areas at Alexandria beach [37]. Our results showed that Cu contents are lower than that reported by Buo-Olayan and Subrahmanyam [38] in *Ulva lactuca* and *Enteromorpha intestinalis* along Kuwait Coast but comparable with that observed by Ho [39], Stenner and Nickless [33] in *Ulva lactuca*.

Generally, iron content of the five algae species under study is relatively high because it is essential element for biological activity as reported by many authors [34, 40, 41]. According to the data reported in this study, iron content showed a wide fluctuation with irregular levels in different algal species. The concentration of iron in *Ulva lactuca* follows the pattern Abu-Qir > El-Mex > Eastern Harbour, while its concentration in *Corallina mediterranea* increases in the order El-Mex > Eastern Harbour > Abu-Qir and with the trend Eastern Harbour > Abu-Qir for *Pterocladia capillacea.* This fluctuation may be attributed to that metal is mostly bound to extra cellular sites, so that variation of its accumulation may largely depend on the chemico-physical conditions of the aquatic environment that affect the biosorption of metals on algal surfaces.

The concentration level of Hg in the present study reveals that there is no distinct change in Hg between the algae species except that *Corallina mediterranea* which exhibit higher concentrations in each area under investigation (figure 2).

Manganese concentrations ranged from 54.6 μg/g dw in *Enteromorpha intestinalis* collected from El-Mex Bay to 12.6 μg/g dw in *Ulva lactuca* collected from Eastern Harbour as shown in table 2. Mn concentrations in this study show variable trends, according to the sampling sites: *Enteromorpha intestinalis* > *Corallina mediterranea* > *Enteromorpha linza* > *Ulva lactuca* at El-Mex Bay, *Pterocladia capillacea* > *Corallina mediterranea* > *Ulva lactuca* at Eastern Harbour and *Corallina mediterranea* > *Pterocladia capillacia* > *Ulva lactuca*



Note: A: *Enteromorpha intestinalis*; B: *Enteromorpha linza*; C: *Ulva lactuca*; D: *Corallina mediterranea*; E: *Pterocladia capillacea. 1: El-Mex; 2: Eastern Harbour; 3: Abu-Qir Bay*

Figure 2. Concentration of heavy metals (μg/g dw) in algae species.

for Abu-Qir Bay as showing in figure 2. Correlation analysis of metals in the present study (table 3) reveals that Mn is negatively correlated with Cu ($r = -0.52$, $p < 0.05$) as showing in figure 3, illustrates that the uptake of Mn is strongly dependent on the concentration of Cu since its binds are more stronger than Mn to the cell membrane [42]. Manganese levels of *Ulva lactuca* in the present study was lower compared with those reported in Hong Kong waters [43] and in Pacific Coast of Mexico [44] but similar to that of Kuwait Coast and along Alexandria coast waters [34, 45]. In comparison with other metals, high levels of both Mn and

Table 3. Correlation between heavy metals in algae species.

	Cd	Cu	Fe	Hg	Mn	Ni	Pb	Zn
Cd	**1**							
Cu	−0.091	**1**						
Fe	−0.011	−0.119	**1**					
Hg	0.482	0.148	−0.412	**1**				
Mn	0.211	**−0.520**	**0.699**	**−0.605**	**1**			
Ni	**0.957**	−0.029	0.026	**0.594**	0.120	**1**		
Pb	**0.909**	−0.187	−0.039	**0.651**	0.080	**0.927**	**1**	
Zn	0.009	0.402	0.198	0.168	−0.240	−0.019	0.122	**1**

Note: Significance level at $p < 0.05$.

Fe were accumulated in most algae species under investigation. This reflects, firstly the high bioavailability of these two metals in Alexandria coastal waters [46] and secondly the capacity of the algae to take them up or the same sources for two metals. This is confirmed by the good correlation between the two metals (figure 4) and the previous findings by Ho [38, 47] in *Ulva lactuca* along the Hong Kong coastal waters.

Meanwhile nickel recorded its higher concentrations in the calcareous red *Corallina mediterranea* among all studied species in the three areas. This may be attributed to that the other algae species (*Enteromorpha* species and *Ulva lactuca*) have the ability to regulate the uptake of this metal and hence did not accumulate it to such a great extent. Studies on the levels of Ni in *Enteromorpha flexuosa* and *Gymnogongrus flabelliformis* [43] and *Ulva lactuca* [39] in Hong Kong resulted in similar conclusions. Nickel concentrations in *Ulva lactuca* in the present study are lower than that recorded by Ho [39] but comparable to that observed by Buo-Olayan and Subrahmanyan [38] in Kuwait Coast.

In the three areas under investigation, the highest concentration of Pb is recorded in *Corallina mediterranea*, table 2. This may be attributed to its calcareous structure where lead and calcium are similar in their deposition and mobilization as reported by Forstner and Wittmann [48] and Moore and Ramamoorthy [49]. Moreover, *Enteromorpha intestinalis* recorded relatively high Pb concentration which may be attributed to its filamentous structure. On the other hand, there is a good correlation between Pb, Cd, and Ni in the five species under investigation in the three areas (figures 5 and 6), which illustrate the relation between the three metals and the replacement of Ca in cell algae [50] and or that those metals originated from the same or similar sources. The levels of Pb in *Ulva lacucta* in the present contribution were high compared with those reported by Buo-Olayan and Subrahmanyam [38] along Kuwait Coast but similar to the levels from the Alexandria Coast waters found by El-Sarraf [34]. The maximum level of Pb

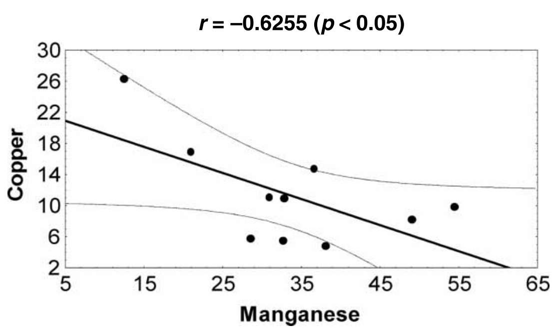


Figure 3. Linear regression analysis between Copper and Manganese.

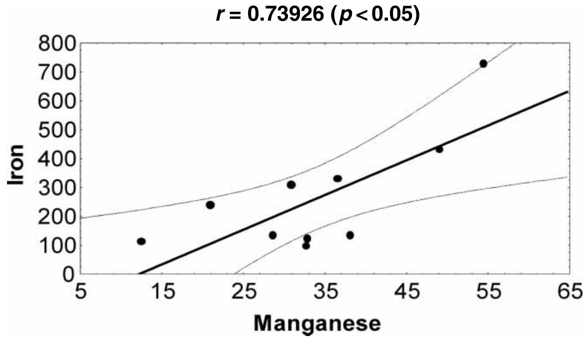


Figure 4. Linear regression analysis between Iron and Manganese.

in *Ulva lactuca* (17.1 μg/g dw in Abu-Qir Bay) was lower than the level reported in that of Hong Kong waters [39, 43].

The present study indicated that Zn accumulation by the same species of algae varied according to the water body where *Ulva lactuca* and *Corallina mediterranea* varies from 7.96 and 13.38 μg/g dw in Abu-Qir Bay to 18.25 and 53.01 μg/g dw in El-Mex Bay while they recorded 50.43 and 60.55 μg/g dw in Eastern Harbour respectively. This finding indicates that the bioavailability of Zn depends on the concentration of Zn in surrounding water, knowing that Eastern Harbour recorded the highest Zn concentration in water as reported by Emara and Shriadah [46]. The concentration of zinc in *Ulva lactuca* and in *Enteromorpha intestinalis* were lower than that observed by Buo-Olayan and Subrahmanyam [38] in Kuwait coast but comparable to that reported by Ho [39] along Hong Kong coastal waters.

To compare the total metal content in algae species in the different sampling sites investigated in this study, the metal pollution index (MPI) was used with the formula [51, 52]:

$$\mathrm{MPI} = (\mathrm{M_1} \times \mathrm{M_2} \times \mathrm{M_3} \times \cdots \times \mathrm{M_n})^{1/\mathrm{n}}$$

where $\mathrm{M_n}$ is the concentration of metal n expressed in μg/g in dw.

The concentration of the total metals in the algae species under investigation notably depending on the location of sampling sites. As shown in table 2, the lowest metal pollution indices for algae species were those of Abu-Qir Bay and El-Mex Bay while the highest indices were found at Eastern Harbour. In El-Mex Bay the MPI in green algae increases in the order *Enteromorpha intestinalis* > *Enteromorpha linza* > *Ulva lactuca* whereas the MPI in red algae increases in the order *Corrallina mediterranea* > *Pterocladia capillacea* in both Abu-Qir Bay and Eastern Harbour.

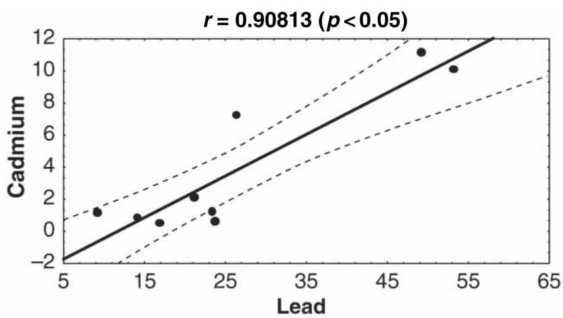


Figure 5. Linear regression analysis between Cadmium and Lead.

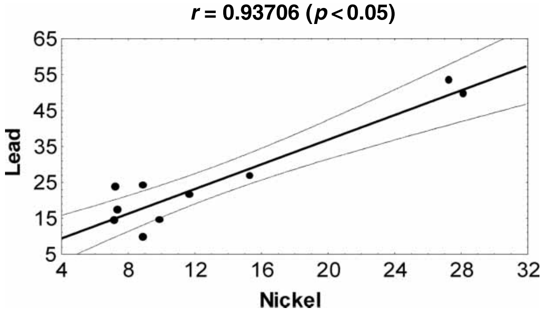


Figure 6. Linear regression analysis between Lead and Nickel.

Conclusively, there are preferential accumulations of metals by different species of algae where *Enteromorpha intestinalis* (filamentous algae) have the ability to concentrate greater amounts of metals than *Enteromorpha linza* (foliaceous algae). On the other hand *Corrallina mediterranea* (calcareous algae) has the ability to accumulate Pb, Cd, and Ni rather than the other species under investigation.

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