Bioremediation of heavy metals by chemically-modified biomass of algae and *Eichhornia* sp.

Walaa S. Abd El Monsef, Awad A. Ragab and Emad A. Shalaby*

Biochemistry Department, Faculty of Agriculture, Cairo University, Giza, 12613, Egypt.

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The current work aimed to determine the bioremoval capacity of different aquatic organisms and its modified forms by oxidizing by potassium permanganate (PC), or treated with sodium hydroxide to remove soluble lead and cadmium from water solution. The aquatic organisms (micro-macro algae and *Eichhornia* sp) were obtained from the Cairo University, Abu Qir (Alexandria) and El-Zamar canal (Giza), respectively. The viable aquatic organisms were exposed to lead (from Pb(NO₃)₂) or cadmium (from Cd(NO₃)₂) at a concentration of 10 mg/l for a time period of 240 min. All measurements were done in triplicate and performed in accordance with standard methods. These data were used to calculate the removal efficiency with respect to time, and to provide the necessary empirical constants to model the removal behavior. The obtained results showed that *Eichhornia* sp had high capacity for bioremoval of lead and cadmium (97.15 and 97.48%) during 15 min. while *Gellidium* sp had highest efficiency for removal of cadmium (96.80%) but during 30 min.

Key words: Bioremediation, bioremoval capacity, aquatic organisms, modified biomass.

INTRODUCTION

Environmental pollution and contamination with heavy metals have become a key-focus of concern. Changes in technology and manufacturing practice are providing relief to these problems. However, some of the present methods for environmental cleaning up result in the production of harmful by-products (Gardea-Torresdeya et al., 2000). Environmentally friendly processes need to be developed to clean up the environment without creating harmful waste products. An advantage of using viable organisms (microorganisms and plants) over non-viable biomass is that they experience rapid growth and regenerate a supply of unsaturated metal-removing biomass. Thus, the removal of the metal becomes a problem of removing the plant biomass. Biosorption has provided an alternative treatment of industrial effluents from that of the traditional physico-chemical methods. A few species of marine macro algae, commonly known as brown algae, play an important role in the research and development of new biosorption materials due to their high uptake capacities, similar to commercial ion-exchange resins and their availability in nearly unlimited amounts from the ocean. *Laminaria japonica*, an abundant waste product in China can be economically used as a potential biosorbent for heavy metals removal from aqueous solutions. The algae were pretreated in order to reinforce it for sorption process applications and also to enhance the sorption performance.

Metal uptake in plants is believed to occur through a wide variety of functional groups present on cell walls. Some of these groups include sulfhydryl, carboxyl, carbonyl, and hydroxyl groups. Previous experiments conducted on functional groups showed that by hydrolyzing *Datura innoxia* cell wall ester groups, the carboxylate content was increased and this in turn caused an increase in metal uptake (Drake et al., 1996). These carboxyl groups have been shown to be very important for metal binding, algal cell carboxyl groups were transformed into ester groups and a decrease in copper and aluminum uptake was observed by determining the amounts that these constituents make up on the cell wall surface and their affinity to bind metal ions, chemical modification of existing groups can lead to enhanced metal-binding abilities.

Pretreatment may be in terms of hardening the cell wall structure through a cross-linking reaction using epichlorohydrin (Kim et al., 1999), or increasing the
negative charge on the cell surface by NaOH treatment (Gurisik et al., 2004), or opening of the available sites for the adsorption by acid treatment (Yang and Volesky, 1999), and enhancing ion exchange by Ca²⁺ solution treatment (Kratochvil et al., 1998). However, only a limited number of studies have so far been focused on the use of pretreated algae for Lead (II) removal from wastewater (Gong et al., 2005).

_E. crassipes_ is water hyacinth, found in large amounts around the fields of irrigations and in the fresh water bodies through the year in tropical and subtropical countries including Egypt (Schneider et al., 1995). The potential of using _E. crassipes_ as alive or a dead biomass to remove metal ions from solutions was recently investigated. The results showed that it is a promising cheap biosorbent source for metal ions (Schneider et al., 1995; Soltan and Rashed, 2003). Little is known about the types and amounts of functional groups located on _E. crassipes_ as well as proton and Cu²⁺ binding constants with _E. crassipes_ (Schneider et al., 1995; Soltan and Rashed, 2003). Moreover, extensive researches have been done on the roots parts while little is known about leaves and stems. Therefore, the present work aims to investigate the physicochemical characteristics of the leaves and stems of _E. crassipes_ biomass in their dead state that help studying their reactivity towards copper adsorption at different pH's and hence suggesting a mechanism for the biosorption process.

The present work aimed to evaluate the bio-removal capacity of different aquatic organisms and its modified biomass against lead and cadmium heavy metals.

**MATERIALS AND METHODS**

**Samples collection**

**Microalgae**

One green microalga, _Dictyochloropsis splendida_ Geitler, was isolated from Ain Helwan, Cairo, Egypt, during spring season (March 2013), cultured on Bold’s basal medium (Bischoff and Bold, 1963) at 20°C under 16/8 light/dark cycles and light intensity of 40 µE / m²/s. The cyanobacterium, _Spirulina platensis_ Geitler cultured on Zarrouk medium (Zarrouk, 1966) was obtained from the culture collection of Botany Department, Faculty of Science, Cairo University, Egypt.

Each microalgal species was harvested at the stationary phase (22 day for _Dictyochloropsis splendida_ and 25 day for _Spirulina platensis_) by centrifugation (3000 rpm) pellets were finally dried at 80°C for 20 min.

**Macroalgae**

Two macroalgae were collected from Abu Quir beaches at Alexandria city. The algae belong to different families: Ulvaceae (_Enteromorpha_ sp, super littoral and intertidal zones, 7 - 15.5 cm), Gelidiaceae (_Gelidium_ sp, sub littoral and intertidal zones, 7.5 - 8.0 cm). These algae were collected during Spring 2012. Thallus of different algae was cleaned from sand and foreign materials by washing with sea water followed by fresh water.

**Eichhornia sp**

_E. crassipes_ (Mart) Solms. was collected from the river Nile at the El-Zomor canal (Giza) during April 2013, cleaned from any debris, washed several times with tap, distilled and then sterilized water, air dried (or lyophilized), ground then stored at -20°C until use.

**Identification of collected species**

Algal species and _Eichhornia_ sp, were identified by Dr. Sanaa Shanab, professor of phycology, Botany Department, Faculty of Science, Cairo University according to Aleem 1993.

**Biosorption experiments**

All chemicals used in this study were analytical grade and distilled water was used to prepare all the solutions. A batch laboratory method was used for the time dependence studies of metals binding to cells of different aquatic organisms including micro, macroalgae and _Eichhornia_ sp. The thallus of different algae was washed for several times with deionized water, air dried and weighted for 1 g (d.wt)/100 ml dist. water. However, the metal ion concentration were 10 mg/l of lead (II), 10 mg/l Cd (II) and each of the metal ion solution was adjusted at pH = 5.4, and the samples (algae) in water contain ions were shacked in a shaker water bath at ≥ 100 rpm for 240 min. At intervals time (Zero, 15, 30, 60, 120 and 240 min.) the concentration of cadmium and lead samples were determined by ICP Spectrometer (iCAP 6000 series, Thermo scientific) and their levels were calculated from stander curve. The performance of the seaweed was expressed as removal capacity (RC) and removal efficiency (RE) (Chan et al., 2004). The terms were calculated by the following equations:

\[
RC = V (C_i - C_f) / m
\]

\[
RE (%) = \frac{C_i - C_f}{C_f} \times 100
\]

Where V is the volume of solution, C_i the initial concentration of metals, C_f the equilibrium concentration of metals and m the mass of biosorbent added.

**Cell chemically modification**

**Sodium hydroxide treatment**

A 6 g sample of dried creosote leaves was weighed and
washed twice with 0.1M HCl and centrifuged at 2500 rpm. The biomass was then reacted with 50 ml of 0.1 M sodium hydroxide for 24 h. The sample was centrifuged and supernatant was removed.

Potassium permanganate oxidized

A modified procedure of Jeon et al. (2002) was used. Powered biomass (10 g) was oxidized in 10 mM solution of potassium permanganate at 30°C for 30 min. Reacted mixture was separated by centrifugation and washed thoroughly with distilled water and dried in an oven at 60°C, hereafter abbreviated as PC. (c) Raw alga. The powered biomass was washed with distilled water and dried in an oven at 60°C until a constant weight was reached, hereafter, abbreviated as DW.

Chemical composition

Determination of phosphorus

The total phosphorus in different algae was extracted as reported by Soltanpour (1985) and spectrophotometrically determined according to procedures of Olse and Watanab, (1965).

Determination of sulfate contents

Sulfate contents in aquatic organisms were determined by turbidimetric method using gelatin-barium reagent according to APHA method (APHA 1998).

Determination of chloride content

Chloride contents in aquatic organisms were determined by turbidimetric method according to Kraemer and Stamm (1924).

Determination of total hydrolysable carbohydrate

Total hydrolysable carbohydrates were spectrophotometrically determined using 5% phenol / sulfuric acid reagent (Dubios et al., 1956).

Determination of total nitrogen and total protein

The determination of total nitrogen was carried out according to Micro-Kjeldahel method. The crude protein was calculated by multiplying total nitrogen percent by the factor of 6.25 (AOAC, 1990).

Determination of titratable acidity

Titratable acidity of different samples was determined according to Harborne (1973).

RESULTS

Bioremoval of heavy metals by aquatic organisms

Biosorption has always been reported as a promising method to treat various kinds of pollutants. Aquatic organisms are the most important biosorbers (Shanab et al., 2012). Table 1 showed the biosorption of heavy metals (lead and cadmium) using dead cells by two macroalgae (Gelidium and Enteromorpha sp.), two microalgae (Spirulina and Dicyochloropsis sp. and Eichhornia sp.). The obtained results showed that Eichhornia sp. had high capacity for bioremoval of lead and cadmium (97.15 and 97.48%) during 15 min. while Gelidium sp. had highest efficiency for removal of cadmium (96.80%) but during 30 min. on the other hand, Dicyochloropsis sp. had the high efficiency but for removal of cadmium (96.05%) during 15 min. However, Spirulina sp. had the lowest bioremoval efficiency against both lead and cadmium (23.21 and 39.64%, respectively) as maximum biosorption during 30 min contact time.

Modified biomass

Raw biosorbents (especially the promising ones from Table 1) have generally been modified with chemical treatments such as, Sodium hydroxide and potassium permanganate to increase their sorption capacity. Table 2 presents the results from experiments on adsorption of lead and cadmium onto modified biomass (with Sodium hydroxide) of Gelidium and Eichhornia sp. The equilibrium could be achieved after having been shaking more than 3 h. these changes in lead and cadmium uptake may be due to the fact that, initially, all adsorbent sites were vacant and the solute concentration was high. After that period, only a very few increase in the lead and cadmium uptake was observed because there are few surface active sites on the cell wall of organism (algae or water hyacinth). The quick equilibrium time is due to the particle size. The effective surface area is high for small particles.

Physicochemical characteristics of biomass

Table 4 shows the results of elemental analysis, total protein, carbohydrates for promising aquatic organisms as bio-adsorbent (E. crassipes and Gelidium biomass). The biomass shows a significant content of total protein (41.25 and 17.68% respectively) and carbohydrates (27.3 and 15.6% respectively) as well as the elemental analysis (as phosphorus, sulfur, nitrogen and chloride) is
Table 1. Removal capacity (mg g\(^{-1}\)) and removal efficiency (%) of different aquatic organisms.

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>Lead</th>
<th>Cadmium</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>RC(^a) (mg g(^{-1}))</td>
<td>RE(^b) (%)</td>
</tr>
<tr>
<td>Zero time</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>15</td>
<td>4.84</td>
<td>72.77</td>
</tr>
<tr>
<td>30</td>
<td>3.24</td>
<td>48.47</td>
</tr>
<tr>
<td>60</td>
<td>2.36</td>
<td>35.27</td>
</tr>
<tr>
<td>120</td>
<td>3.46</td>
<td>51.70</td>
</tr>
<tr>
<td>240</td>
<td>1.93</td>
<td>28.84</td>
</tr>
</tbody>
</table>

**Gelidium pectinatuns (red algae)**

| Zero time  | 0.0 | 0.0 | 0.0 | 0.0 |
| 15         | 3.85 | 57.63 | 7.46 | 83.92 |
| 30         | 4.96 | 74.32 | 7.75 | 87.14 |
| 60         | 5.73 | 85.72 | 7.6 | 85.50 |
| 120        | 5.22 | 78.15 | 7.61 | 85.52 |
| 240        | 5.60 | 83.75 | 7.05 | 79.32 |

**Enteromorpha compressa (green algae)**

| Zero time  | 0.0 | 0.0 | 0.0 | 0.0 |
| 15         | 1.54 | 23.21 | 6.52 | 39.64 |
| 30         | 1.50 | 22.41 | 0.61 | 6.90 |
| 60         | 0.0 | 0.0 | 0.0 | 0.0 |
| 120        | 0.0 | 0.0 | 0.0 | 0.0 |
| 240        | 0.0 | 0.0 | 0.0 | 0.0 |

**Spirulina platensis (cyanobacteria)**

| Zero time  | 0.0 | 0.0 | 0.0 | 0.0 |
| 15         | 6.41 | 96.05 | 6.94 | 84.78 |
| 30         | 6.22 | 93.11 | 7.54 | 84.78 |
| 60         | 6.24 | 93.35 | 6.57 | 73.85 |
| 120        | 6.44 | 94.61 | 6.74 | 75.85 |
| 240        | 6.40 | 95.90 | 7.18 | 80.78 |

**Dictyochloropsis splendidida**

| Zero time  | 0.0 | 0.0 | 0.0 | 0.0 |
| 15         | 6.19 | 96.05 | 6.94 | 84.78 |
| 30         | 6.22 | 93.11 | 7.54 | 84.78 |
| 60         | 6.24 | 93.35 | 6.57 | 73.85 |
| 120        | 6.44 | 94.61 | 6.74 | 75.85 |
| 240        | 6.40 | 95.90 | 7.18 | 80.78 |

**Eichhornia crassipes leaves**

| Zero time  | 0.0 | 0.0 | 0.0 | 0.0 |
| 15         | 6.54 | 95.43 | 6.59 | 74.10 |
| 30         | 6.22 | 93.11 | 7.54 | 39.78 |
| 60         | 6.50 | 97.33 | 7.89 | 88.73 |
| 120        | 6.19 | 77.61 | 8.10 | 91.03 |
| 240        | 6.16 | 12.22 | 8.0 | 89.95 |

**Eichhornia crassipes root**

| Zero time  | 0.0 | 0.0 | 0.0 | 0.0 |
| 15         | 6.32 | 94.58 | 4.69 | 52.79 |
| 30         | 6.37 | 95.36 | 7.79 | 87.84 |
| 60         | 6.0 | 89.82 | 7.68 | 86.33 |
| 120        | 5.86 | 87.85 | 8.16 | 91.88 |
| 240        | 6.33 | 94.82 | 8.18 | 92.06 |

**Eichhornia crassipes Whole plant**

| Zero time  | 0.0 | 0.0 | 0.0 | 0.0 |
| 15         | 6.49 | 97.15 | 8.66 | 97.48 |
| 30         | 6.52 | 97.60 | 7.72 | 86.80 |
| 60         | 6.30 | 94.31 | 8.32 | 93.61 |
| 120        | 6.22 | 44.31 | 8.53 | 95.97 |
| 240        | 6.0 | 93.47 | 8.88 | 99.97 |

\(^a\)RC: removal capacity; \(^b\)RE: Removal efficiency.

Illustrated in Table 4. This reflected that the biomass tissue has abundant function groups (ACOOH, ANH\(_2\), ANHA, AOH, C, O, and PO\(_4\)\(^3-\)) that giving a primary anticipation for the biomass capability to react with the
Table 2. Removal capacity (mg g⁻¹) and removal efficiency (%) of different aquatic organisms treated by sodium hydroxide.

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>Lead</th>
<th>Cadmium</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>RC²</td>
<td>RE² (%)</td>
</tr>
<tr>
<td>Zero time</td>
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<td>0.0</td>
</tr>
<tr>
<td>15</td>
<td>2.10</td>
<td>23.60</td>
</tr>
<tr>
<td>30</td>
<td>3.22</td>
<td>38.64</td>
</tr>
<tr>
<td>60</td>
<td>3.75</td>
<td>45.00</td>
</tr>
<tr>
<td>120</td>
<td>3.98</td>
<td>48.13</td>
</tr>
<tr>
<td>240</td>
<td>6.05</td>
<td>72.98</td>
</tr>
</tbody>
</table>

Gellidium Pectinatuns (red algae)

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>Lead</th>
<th>Cadmium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Zero time</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>15</td>
<td>2.34</td>
<td>25.45</td>
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<tr>
<td>30</td>
<td>3.02</td>
<td>36.37</td>
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<tr>
<td>60</td>
<td>3.66</td>
<td>43.29</td>
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<tr>
<td>120</td>
<td>4.50</td>
<td>53.46</td>
</tr>
<tr>
<td>240</td>
<td>5.82</td>
<td>70.09</td>
</tr>
</tbody>
</table>

Eichhornia crassipes (leaves)

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>Lead</th>
<th>Cadmium</th>
</tr>
</thead>
<tbody>
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<td>Zero time</td>
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<td>0.0</td>
</tr>
<tr>
<td>15</td>
<td>1.81</td>
<td>21.60</td>
</tr>
<tr>
<td>30</td>
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<td>48.13</td>
</tr>
<tr>
<td>240</td>
<td>3.65</td>
<td>72.98</td>
</tr>
</tbody>
</table>

Figure 1. Total acidity (%) of different aquatic biomass and its modified form (using KMnO₄).

cadmium and lead through chelation with those sites (Komy et al., 2006).
The titratable acidity of treated and treated biomass was shown in Figure 1. The results indicated that, the total acidity of modified biomass with permanganate increase from 0.32 to 0.61% in Eichhornia sp. and from 1.81 to 3.23% in Gellidium sp. These results may be due to the oxidation of alkyl groups on plant cell wall and increase the carboxyl groups and led to increase the acidity and increase the biomass capability to react with cadmium and lead through chelation and formation the ionic bond between negative and positive charge (Shanab et al., 2012).

Discussion

Bioremoval of heavy metals by aquatic organisms

The results of biosorptive activities (Table 1) may be due to the aquatic organisms contents especially water hyacinth and algae from phycocolloide, sulfate,
Table 3. Removal capacity (mg g\-1) of and removal efficiency (%) different aquatic organisms treated by potassium permanganate.

<table>
<thead>
<tr>
<th>Time (min)</th>
<th>Lead</th>
<th>Cadmium</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>RC\a (mg g-1)</td>
<td>RE\b (%)</td>
</tr>
<tr>
<td>Gellidium sp (Red algae)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Zero time</td>
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<td>0.0</td>
</tr>
<tr>
<td>15</td>
<td>1.88</td>
<td>20.27</td>
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<tr>
<td>30</td>
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</tr>
<tr>
<td>240</td>
<td>5.39</td>
<td>64.54</td>
</tr>
<tr>
<td>Eichhornia crassipes (leaves)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Zero time</td>
<td>0.0</td>
<td>0.0</td>
</tr>
<tr>
<td>15</td>
<td>2.58</td>
<td>29.37</td>
</tr>
<tr>
<td>30</td>
<td>3.35</td>
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<td>3.80</td>
<td>45.33</td>
</tr>
<tr>
<td>120</td>
<td>4.50</td>
<td>53.29</td>
</tr>
<tr>
<td>240</td>
<td>5.70</td>
<td>68.19</td>
</tr>
</tbody>
</table>

phosphorus and nitrogen (Shalaby, 2008). However, the obtained results indicated that the maximum bioremoval capacity 97% for tested organisms occurred after 15 min of the experimental duration which was decreased progressively during 240 min of contact with the heavy metals. These may be due to break the bonds between the algal cell and metals by some microorganism e.g. fungi and bacteria. The present results are in agreement with those obtained by Chan et al. (2004) they showed that from different species of seaweed have good removal capacity for different metals, Sargassum sp. showed the best removal ability. Sandau et al. (1996) who reported that sorption capacities are generally similar between live and dead biomass of a specific type. Dead biomass (heat, acid, and/or otherwise chemically treated) had great biosorption capacity, probably due to the uncovering of mask binding sites. The two principle mechanism involved in biosorption appear to be the ion exchange where ions such as Na\+, Mg\sup{2+} and Ca\sup{2+} became displaced by heavy metals ions, and the complexation between metal ions and various functional groups such as carboxyl, amines, thiol, hydroxyl, phosphate and hydroxyl-carboxyl, that can interact in a coordinated way with heavy metal ions (Awadalla and Pesic, 1992).

Also, Kumar et al. (2007) found that, biosorbent materials primarily weak acidic and basic functional groups. It follows from the theory of acid-base equilibrium that, in the pH range 2.5 - 5, the binding of heavy metal cation is determined primarily by the state of dissociation of the weak acidic groups. Carboxyl groups (-COOH) are the important groups for metal uptake by biological matter. At pH 5, there are lower numbers of competing hydrogen ions and more ligands are exposed with negative charges, resulting in greater metal sorption. But for pH values from 6 to 10, lower adsorption capacity was observed due to the precipitation and lower polarity of metal at high pH. Biosorption of lead by eight brown, green and red marine algae was investigated by Jalali et al. (2002). They found that biosorption of lead was rapidly occurred onto algal biosorbents and most of the sorbed metal was bound in < 30 min of contact.

**Modified biomass**

Pretreatment of raw cell with sodium hydroxide may be in terms of hardening the cell wall structure through increasing the negative charges on the cell surface as reported by Gurisik et al. (2004). According to experimental data, the maximum uptakes of lead and cadmium were done during 240 min contact time. The lead has increased in both species (Gellidium and Eichhornia sp) from 28.84 and 12.22 % (in raw cells) to 72.98 and 70.09% (in modified cells) during 240 min respectively. However, the uptake of cadmium using modified cells not significantly increased and lower than metal uptake by raw cells and these results may be due to the particle size of cadmium metal or its needed to long time more than 240 min for complete absorption (Luo et al. 2006).

Chemical modifications in aquatic organism using potassium permanganate have been applied to enhance the uptake of lead and cadmium through oxidation reaction with KMnO\textsubscript{4}. This modification method is favor to increasing carboxylic groups and the lead uptake capacity is increased in the process.

According to experimental data (Table 3), the maximum uptakes of lead and cadmium were done during 240 min
contact time. The lead has increased in both species (Gelidi um and Eichhornia sp.) from 28.84 and 12.22 % (in raw cells) to 64.54 and 68.19% (in modified cells by KMnO4) during 240 min respectively. However, the uptake of cadmium using modified cells not significantly increased and lowers than metal uptake by raw cells and these results may be due to the particle size of cadmium metal or it's needed to long time more than 240 min for complete absorption (Luo et al., 2006).

**Physicochemical characteristics of biomass**

The results in Table 4 confirmed the ability of aquatic organisms to binding and uptake of heavy metal and were in agreement with the results obtained by Awadalla and Pesic (1992) who found that algal biomass has great biosorption capacity, probably due to the uncovering of masked binding sites. The two principle mechanisms involved in biosorption appear to be: (1) ion exchange wherein ions such as Na, Mg, and Ca become displaced by heavy metal ions, and (2) complexation between metal ions and various functional groups such as carboxyl, amino, thiol, hydroxyl, phosphate, and hydroxyl-carboxyl, that can interact in a coordinated way with heavy metal ions.

**Conclusions**

From the present study it can be concluded that Eichhornia and Gelidi um sp. especially its modified forms can be effectively used for lead (II) and Cd (II) bioremoval from aqueous solutions, and can be used more than eight times to remove more than 50% from heavy metals concentration.

**Acknowledgements**

The authors are thankful to the supports of Faculty of Agriculture, Cairo University, and Giza, Egypt.

**REFERENCES**


<table>
<thead>
<tr>
<th>Compound/elements</th>
<th><em>Eichhornia crassipes</em> (leaves)</th>
<th><em>Gelidium pectinatums</em></th>
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<tbody>
<tr>
<td>Phosphorus</td>
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<td>0.21</td>
</tr>
<tr>
<td>Sulfur</td>
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<tr>
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</tr>
<tr>
<td>Nitrogen</td>
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<td>2.83</td>
</tr>
<tr>
<td>Chloride</td>
<td>0.0355</td>
<td>0.046</td>
</tr>
<tr>
<td>T. Protein</td>
<td>41.25</td>
<td>17.68</td>
</tr>
<tr>
<td>T. Carbohydrate</td>
<td>27.3</td>
<td>15.6</td>
</tr>
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heavy metals by some microalgae species (Egyptian Isolates). Plant
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