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Evaluation of the Marine Algae *Sargassum latifolium* and *Sargassum crassifolium* for the Biosorption of Heavy Metal Ions from Aqueous Solutions

Bassant S. Farhoud¹, Alaa M. Younis^{1, 2}, Elham M. Ali^{1, 3*}

¹Department of Aquatic Environment, Faculty of Fish Resources, Suez University, Suez, Egypt
²Department of Chemistry, College of Science, Qassim University, Buraidah, 51452, Saudi Arabia
³Department of Environmental Studies, National Authority for Remote Sensing & Space Sciences (NARSS), Cairo, Egypt

*Corresponding Author: elhamali201212@gmail.com, elhamali@narss.sci.eg

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ABSTRACT

A batch experimental study was conducted using biomass of two algal species, Sargassum latifolium and Sargassum crassifolium, collected from the Gulf of Suez to investigate their ability to remove lead (Pb), cadmium (Cd), nickel (Ni) and copper (Cu) ions from aqueous solutions. Those solutions were prepared in the laboratory by dissolving $(CH_3COO)_2Pb.3H_2O, CdCl_2H_2O,$ Ni(NO₃)₂.6H₂O, and CuSO₄.5H₂O (analytical grade) in double distilled water. Biomass of both algal species was grounded and then sieved to three different sizes (0.63, 0.125, and 0.250um). Some experimental conditions (pH. biomass dosage, particle size, agitation speed, contact time, and initial concentrations of metal ions) were optimized to test and define the best conditions for the maximum heavy metals removal attainment. In addition, the tested conditions were also gathered with various factors to determine the most effective combination. Results showed that the algal biomass of both species was significantly efficient in removing the tested metal ions as both Sargassum species performed a percentage of 99, 88, 99, and 99% for the biosorption of Pb, Cd, Ni, and Cu, respectively. Experimental conditions results determined that the maximum metal uptake for both sargassum species was attained at 8 pH during a contact time of 60 minutes, considering these levels as the optimum conditions for the best absorption efficiency. Such results showed the potential use of macroalgae for biosorption functions. Results confirmed that both brown seaweed species were promising, highly competent, and biodegradable biosorbents. These results enabled new methodologies to remove heavy metal pollutants from industrial wastewater while being economically valuable and environmentally safe.

INTRODUCTION

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Industrial wastewater exerts a significant hazardous effect on the environment and human health, even after its treatment (Al Ketife *et al.*, 2020). This is verified since the treated wastewater could contain some harmful residuals (Cruz *et al.*, 2004). Heavy metal ions are one of the most toxic pollutants, even at low concentrations (Ibrahim, 2011; Poo *et al.*, 2018). Several industrial activities, such as mining operations, alloy

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manufacturing, metal plating, fertilizer industry, plastic, textile, battery, and pigments release types of heavy metals via their waste effluents (**Freitas** *et al.*, **2008**) causing adverse health effects for humans, including lung damage, bone lesions, renal disturbances, cancer, hepatic injury, hypertension and also possible DNA modification (**Cechinel** *et al.*, **2016**). Most of those harmful effects couldn't be spotted except when the heavy metal ions are bio-accumulated in the living systems (**Truong** *et al.*, **2022**).

Heavy metals removal or elimination is one of the necessary steps in the industrial wastewater treatment, however their complete removal by conventional methods is difficult. Most conventional methods have a physicochemical approach, such as chemical oxidation or reduction, chemical precipitation, solvent extraction, filtration, ion exchange, electrochemical treatment, and/or evaporation recovery (**Kumar** *et al.*, **2016; Priya** *et al.*, **2022**). Unfortunately, these techniques are extremely expensive and almost ineffective at low concentrations, and in many cases, the combination of two methods or more is essential to increase efficiency and attain today's toxicity-driven limits (**Bordoloi** *et al.*, **2017**). Consequently, with the continuous increase in heavy metal discharge limits, global concerns with restricted regulations have posed the research attention to develop new innovative technologies for the removal of heavy metal ions from wastewater (**Henriques** *et al.*, **2017**).

The biosorption approach is an effective alternative, and recently the most applicable method for removing and recovering heavy metal ions from aqueous solutions via living or nonliving biomasses. The advantages of biosorption have overcome those of conventional methods for its enormous quantities, availability, regeneration of the biosorbent, more efficiency, no need for additives, lower costs, and environmentally friendly (**Barquilha** *et al.*, **2017; Richards** *et al.*, **2019**).

Different biological materials could be used as adsorbents in water bioremediation, including bacteria, fungi, and algae (Rangabhashiyam & **Balasubramanian**, 2019). Using living microorganisms; however, has many limitations especially the possibility of inhibition of microbial growth when the concentration of metal ions is too high in the solution, lowering their adsorption capacity after accumulation of metal ions on their cell wall (Cruz et al., 2004). Using non-living biomass also has similar or greater adsorption efficiencies of metal ions due to the changes that occur in their cell structure after deactivation (Sheng et al., 2004; Moreira et al., 2019). Among most microorganisms, algae have a special interest due to their wide distribution in all ecosystems and their intricate connection with human life (Lee & Chang, 2011). Marine algal biomasses, especially brown seaweeds, have recorded high biosorption capacities for several heavy metal ions (Benaisa et al., 2016). The previous findings revealed that Laminaria hyperborea, Bifurcaria bifurcata, Sargassum muticum, and Fucus spiralis achieved biosorption capacities for lead cadmium, zinc, and ions in the ranges of 32.3-50.4, 23.9-39.5, and 18.6-32.0 mg g⁻¹, respectively (Freitas et al., **2008**). Furthermore, the free *Sargassum* sp. achieved a metal uptake of 1.51mmol of Cu^{2+}

g⁻¹ and 1.23mmol of Ni²⁺ g⁻¹, whereas the immobilized biosorbent reached a removal of 2.06mmol of Cu²⁺ g⁻¹ and 1.69mmol of Ni²⁺ g⁻¹; both were applied in fixed-bed columns (Barquilha et al., 2017).

Therefore, the present study is mainly focused on the application of brown seaweeds, aiming at evaluating their efficiency and adsorptive capacity to remove ions of lead (II), cadmium (II), nickel (II), and copper (II). This research uses 2 species of the brown algal from the genus Sargassum. The investigations included experimental analyses of different parameters to assess the metals uptake at each experimental case and to standardize the conditions under which we can suggest the optimum conditions for maximum removal results of metal removal via adsorption. The study focused on six controlling factors, such as i) Algal dose (0.01, 0.05, and 0.1g), ii) Particle size (0.63, 0.125, and 0.250µm), iii) Initial metal concentration (50, 100, 150, 200, and 250ppm), v) pH (4-8), vi) Agitation speed (50, 100, 150, 200, and 250rpm.), and vii) Contact time (5, 15,30, 60, 90, and 120 minutes). This work aimed to offer a promising avenue for addressing the challenges associated with heavy metal pollution in industrial wastewater by using brown seaweeds as a biosorbent material. The environmentally friendly and economically viable nature of these solutions aligns with the growing emphasis on sustainable and green technologies in water treatment processes.

MATERIALS AND METHODS

Collection and preparation of algae

The brown macroalgae Sargassum latifolium and Sargassum crassifolium were collected from Ra's Ghareb and Ras Shukeir at the Gulf of Suez (Fig. 1).

The collected algae were washed once with an excess tap water and then twice with double distilled water to ensure a complete removal of adhering foreign particles like sand, salts, and debris from their surface. The washed biomasses were first air-dried for two days and then dried in an oven at 40°C until obtaining a constant weight. Dried biomass was cut & ground using an electric mill, and subsequently sieved by a mechanical shaker, and the fraction of $0.063-0.250\mu m$ was selected for use in the sorption tests. The sieved biomasses are stored in a sealed bottle in a desiccator.

Metal ion solutions

Stock solutions (1000mg/ L) of tested heavy metals lead (Pb), cadmium (Cd), nickel (Ni), and copper (Cu) ions were prepared by dissolving (CH₃COO)₂Pb.3H₂O, Ni(NO₃)₂.6H₂O, CdCl₂H₂O, and CuSO₄.5H₂O (analytical grade) in double distilled water. The working solution was prepared by diluting the stock solution with double distilled water to the required volumes.







Fig. 1. Locations of collected Sargassum species (Google Map)

Batch experimental conditions

During the batch experimental process of testing parameters for optimization, the tested parameter was the only changing attribute, while all other factors remained constant, as illustrated in Table (1).

Table 1. The optimized parameters in the batch experiment

Factor name	Biomass dose	Particle size	Metals concentration	Agitation speed	Contact time	рН
Factor	0.01g	0.125µm	150ppm	150rpm	30	5
value					minutes	

Batch adsorption investigation was conducted in 50mL Erlenmeyer flasks containing the dry powder mass of both *Sargassum latifolium* and *Sargassum crassifolium* separately and constant volume (50mL) of the prepared solution of lead (Pb), cadmium (Cd), nickel (Ni) and copper (Cu) ions solution with serial concentrations of metal ions (50, 100, 150, 200, and 250ppm). The samples were mixed with three adsorbent doses (0.01, 0.05, and 0.1g) at different particle sizes (0.063, 0.125, and 0.250µm), and different pH ranges (4– 8) were adjusted by using 0.1N HCL and 0.1N NaOH solutions. The samples were shaken using an orbital shaker with a speed of (50, 100, 150, 200, and 250rpm). The samples were conducted at different time intervals (5, 15,30, 60, 90, and 120min) (25 °C). All experimental samples were conducted in duplicate for error elimination, and the experiment was repeated at room temperature.

Evaluation of heavy metals' concentration

The samples were filtered using membrane filter paper (Whatman No.1, $0.47\mu m$ Millipore), and the solution was filtered to analyze heavy metals left in the solution, then samples were stored in falcon tubes to be analyzed by spectrophotometer. The atomic absorption spectrophotometer (AAS, Thermo Scientific, model S Series) was used to measure the prepared samples and determine the concentration of metal ions. Measurements were made at wavelengths of 341.5nm for Ni, 405.8nm for Pb, 324.8nm for Cu, and 326.1nm for Cd.

Metal removal efficiency

Biosorption capacity (q_e), which represents the amount of metal adsorbed per gram of biosorbent, can be calculated in mg/g as follows:

$$q_e = (c_0 - c_e) v/m \tag{1}$$

Where, C_0 is the initial metal ions concentration (mg/ L); C_e is the equilibrium concentration of metal ions (mg/ L); V is the volume of solution (L), and m is the mass of biosorbent (g). The percentage of metal removal can also be displayed by the percentage metal removal as follows:

$$Metal Removal(\%) = 100(c_0 - c_e)/c_0$$
(2)

Statistical Analysis

Each sample was collected in 2 replicates to ensure the validation of results and error reduction. SPSS version 25.0 for windows was used to analyze the experimental data, and the minimum significant level was set at 0.05. The data shown represent the means \pm standard deviations. The graphs were drawn by Microsoft Excel 365 for Windows.

RESULTS AND DISCUSSION

The experimental results showed variations in the biosorption rate for each heavy metal ion with varying experimental conditions and controlling factors. Results for the six tested parameters are shown below:

1- Particle size

The effect of biomasses particle size has a low significant effect on the adsorption potentiality of all different metal ions. However, the specific conditions and biosorbent characteristics can lead to variations in metal uptake based on particle size. Despite the generally low significance, the results presented in Fig. (2) indicate specific impacts where biomass particle size affects the metal uptake.

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Fig. 2. The influence of different particle sizes of both algae on the uptake of metal ions from solutions

For example: in the case of *Sargassum latifolium*, high metal uptake of 148.3 and 141.3mg.g⁻¹ were obtained with size of 0.063μ m for Pb and Cu, respectively, and of 148.6 and 111.9mg.g⁻¹ for Ni and Cd, respectively, with size of 0.125μ m. While *sargassum crassifolium* showed its highest metal uptake of 148.5 and 143.2mg.g⁻¹ at sizes of 0.063µm for Pb and Cu, respectively, and 115.9mg.g⁻¹ for Cd at the size of 0.250µm, and 148.8mg.g⁻¹ for Ni at size of 0.125µm.

The results are compatible with the findings of other studies. **Putri** *et al.* (2020) reported no significant influence of biomass particle size on the removal of lead (Pb) ions, supporting the indication that the impact of particle size can vary depending on the metal ion and the specific biosorbent used.

In addition, other researchers (**Stirk & Van Staden, 2001**) reported that the particle size of the dried brown algae *Ecklonia maxima* and *Laminaria pallida* (0.5-1.0mm) affected the rate of copper absorbance after 30min. However, after 24h exposure

to copper, there was very little difference in the final ion concentration, regardless of the sorbent size for *L. pallida* and larger than 0.5mm for *E. maxima*; this is due to the smaller particle size of the biosorbent.

2- Biomass dose

The adsorption process is highly dependent on biomass doses since this factor is responsible for the availability of free-binding sites to remove metal ions (**Montazer-Rahmati** *et al.*, **2011**). The effect of biomass dosage on Pb, Cd, Ni, and Cu ions removal is indicated in Fig. (3).





It can be noticed that, *Sargassum latifolium* showed a high removal percent of 98.9, 97, and 82.4% for Pb, Cu, and Cd at 0.1g dose, respectively. Additionally, *sargassum crassifolium* showed the same removal percent result of 99.3, 97.1, and 82.6% for Pb, Cu, and Cd at 0.1g dose, respectively. The increase in the biomass dosage led to a rise in metal removal efficiencies because of the availability of more free-binding sites



for different metal ions (**Ibrahim**, 2011). Kumar *et al.* (2020) also reported that the increase in the adsorbent dosage with the rise in adsorption capacities is attributed to an increase in free surface accessibility, which in sequence increases the number of adsorbate molecules.

In the case of nickel (Ni) ions in the present study, the maximum metal removal was obtained at 0.05g dose, and then decreased gradually at 0.1g dose for both algae species since the removal percent for *Sargassum latifolium* and *sargassum crassifolium* were 99 and 99.2% at a dosage of 0.05g, respectively. This is due to the accessibility formation of partial biomass aggregation at higher doses which reduces the adsorption efficiencies of biomass (**Karthikeyan** *et al.*, **2007**).

This was compatible with Yogeshwaran and Priya's results, in the adsorption of Cr, Pb, and Zn metal ions by Sargassum Wightii; the maximum percentage removal of metal ions was determined at 1g/L dosage and it stayed constant thereafter. Yogeshwaran and Priya also interpreted the constant phase of biosorption after increasing biomass dosage due to a decrease in the aqueous solution's concentration gradient (**Yogeshwaran & Priya, 2022**).

3- Initial metal concentrations

The initial metal concentration has a significant effect on the adsorption process since it provides free-binding molecules which is available to be absorbed. Fig. (4) indicates the fate of metal removal percentages by increasing the initial metal concentrations.

It can be concluded that by increasing the initial metal ions concentrations, the metal sorption capacities of both algal biomasses increased until they reached a constant phase. This behavior, where adsorption efficiency increases at lower concentrations and then plateaus at higher concentrations, is explained by the saturation of active sites on the algal surface (**Ibrahim**, **2016**).

The two algal species had nearly the same adsorption capacity for all four metal ions in all different concentrations, and the highest removal percents were recorded at low concentrations of metal ions. The highest adsorbed metal ions by both species were Pb, Ni, Cu, and Cd, respectively.

Sargassum latifolium (Fig. 4) recorded a high removal percent of 98.3 and 99.1% for Pb and Ni at 100ppm, respectively, and 94.2 and 79.9% for Cu and Cd at 50ppm, respectively. While *sargassum crassifolium* had a high removal percent of 98.2, 99.4, and 79.8% for Pb, Ni, and Cd, respectively, at 50ppm, and 94% for Cu at 100ppm. This is due to the availability of ambient binding sites on the cell wall of biomasses before the saturation state. The higher metal ions adsorption can be clarified by overcoming all mass transfer resistances between the solution and the biomasses by increasing the initial concentrations of metal ions (**Montazer-Rahmati** *et al.*, **2011**).

This was similar to the finding of **Ibrahim** (**2016**) as he found that Sargassum activated carbon (SAC) achieved a high adsorption efficiency of 94% for Cu^{+2} , 91% for Pb⁺², and 90% for Cd⁺² at the initial concentration of metal (80mg/ 1), and beyond this concentration, the adsorption efficiency remained unchanged, even as the initial concentration of metal ions increased. Similar trends were observed for Sargassum algal powder (SAP) but with lower adsorption efficiency compared to SAC (**Ibrahim, W. M., 2016**).



Fig. 4. Different initial metal concentrations influence the metals removal % by algae from solutions

4- Contact time

The two sargassum species observed high adsorption for all four metals. Fig. (5) shows the removal efficiency of metal ions by *Sargassum latifolium* in all time intervals and the maximum biosorption capacities were 148.77 and 143.515mg.g⁻¹ for Pb and Cu,



respectively at 120min., 148.6mg.g⁻¹ for Ni at 30min., and 116.515mg.g⁻¹ for Cd at 5min. Furthermore, the maximum biosorption capacities by *Sargassum crassifolium* was 149.76 and 144.29mg.g⁻¹ for Pb and Cu at 120min., 148.85mg.g⁻¹ for Ni at 30min., and 116.805mg.g⁻¹ for Cd at 5min.



Fig. 5. The influence of different contact time intervals on the capacity of metal uptake by both algae from solutions

While analyzing the data, the adsorption capacities were identical between the time intervals of 60 to 120 minutes. It was unpredictable that at the first-time intervals from 5 to 15 minutes, the two algae species achieved a high performance in the adsorption of heavy metals compared to the equilibrium time of 60 minutes.

The saturation state can be reached when the sorbent cannot adsorb new ions and as the process of adsorption proceeds, the metal ions tend to be desorbed back into the solution (**Montazer-Rahmati** *et al.*, **2011**). In the case of *Sargassum latifolium*, the equilibrium time was achieved at 60 minutes and after that, the metal ions tended to be desorbed, while in the case of *Sargassum crassifolium*, the equilibrium time was achieved

at 60 minutes, and then it is almost constant to 120 minutes. Scientists who studied the adsorption of heavy metals by macroalgae also reported that 90% of the total metal ion sorption is achieved within 60min (Sheng *et al.*, 2004; Ibrahim, 2011).

The removal of chromium by *Sargassum hornschuchii* increased with the increase of contact time to 36hrs, and it decreased with further increase in contact time (**Fakhry** *et al*, **2013**). Over time, the occupation of vacant surface sites of the adsorbent is difficult due to repulsive forces between the solute molecules on the solid and bulk phases (**Kumar** *et al.*, **2010**).

5- Agitation speed

The agitation speed plays a key role in the biosorption process since it controls the efficiency of metal ions uptake. Increasing the agitation speed provides an opportunity for all free sites to be banded with metal ions. In Fig. (6), *Sargassum latifolium* recorded a high metal uptake of 148.6mg.g⁻¹ for Ni at 150rpm, 143.4 and 115.1mg.g⁻¹ at 200rpm for Cu and Cd, respectively, and 149.2mg.g⁻¹ for Pb at 250rpm. In the case of *Sargassum, crassifolium* it recorded 149.89, 115.53, and 143.5mg.g⁻¹ for Pb, Cd, and Cu at 200rpm, respectively, and 148.8mg.g⁻¹ for Ni at 150rpm.

Similar results were observed when the biosorption of lead by *Sargassum ramifolium*, *Ulva fasciata*, *Turbinaria ornata*, and *Sarcodiotheca furcata* increased by increasing the initial agitation speed from 100 to 250rpm (Al-Dhabi & Arasu, 2022).

However, further increasing of agitation speed can cause the desorption of metal ions and hence the biosorption process (**Al-Dhabi & Arasu, 2022**). As in our work, the removal efficiency for Ni (II) decreased after 150ppm by both algae species.

6- pH

The pH of the solution has a significant impact on the biosorption process since it affects the uptake of metals. The metal chemistry in the solution, along with the functional groups present in the biomass's cell walls, influences the dependence of metal uptake. Fig. (7) illustrates the biosorption of lead (Pb), cadmium (Cd), nickel (Ni) and copper (Cu) by *Sargassum latifolium* and *Sargassum crassifolium* at different pH levels.









Fig. 6. The influence of different agitation speeds on the capacity of metal uptake by both algae from solutions

Generally speaking, higher pH values result in higher metal uptake. Both species demonstrate better biosorption performance at pH 8 for all four metal ions. *Sargassum latifolium* exhibits metal uptake of 149.3mg/ g for Pb, 133.1mg/ g for Cd, 149.4mg/ g for Ni, and 149.7mg/ g for Cu. On the other hand, *Sargassum crassifolium* shows a metal uptake of 149.3mg/ g for Pb, 133.3mg/ g for Cd, 149.5mg/ g for Ni, and 149.6mg/ g for Cu. The adsorption of metal ions onto adsorptive centers of biosorbents is influenced by the chemical form of the metal ions in the solution. The solubility of copper and lead ions is decreased in the acidic medium, particularly due to the formation of insoluble metal oxides (**Negm et al., 2018**). That makes their ability to adsorb onto the surfaces of biosorbents diminished.



Fig. 7. The influence of solution pH on the metals uptake of both algae

These results contrast the results obtained by **Ibrahim** (2011), who observed that four red algae species, namely *Corallina mediterranea*, *Galaxaura oblongata*, *Jania rubens*, and *Ptredocladia capillacea*, exhibit high metal uptake at pH 5, with efficiency declining as pH increases. Ibrahim also explained that the decreased metal uptake at higher pH values due to the competition of anionic hydroxide complexes with metal ions on the active sites of the biomass. In addition, other researchers (**Montazer-Rahmati** *et al.*, 2011) reported that biosorption of cadmium (Cd), lead (Pb), and nickel (Ni) by intact and pre-treated brown marine algae, including *Cystoseira indica*, *Sargassum glaucescens*, *Nizimuddinia zanardini* and *Padina australis* were at the highest at pH values of 5, 5.5, and 6.0, respectively. In regards to copper (Cu), it demonstrates high biosorption by *Ulva fasciata* and *Sargassum* sp. when pH values increase from 1 to 4 (**Karthikeyan** *et al.*, 2007).

After obtaining the optimized experimental conditions, the maximum biosorption efficiencies of both algal species for the four metal ions can be shown in (Fig. 8).





Fig. 8. Comparison between *S. latifolium* and *S. crassifolium* in biosorption removal percent for Pb, Cd, Ni and Cu at the the optimum tested conditions



CONCLUSION

This study measured the biosorption of Pb (II), Cd (II), Ni (II) and Cu (II) ions onto brown algal biomass from synthetic aqueous solution. Six parameters, including pH of solution, initial metal concentration, particle size, biomass dosage, agitation speed, and contact time were investigated to study their potential influence on the metal uptake by algae. *Sar. latifolium* showed a high efficiency for removing heavy metal ions from aqueous solution reaching 98.9, 80, 99, and 96.1% for Pb, Cd, Ni, and Cu, respectively. Additionally, *Sar. crassifolium* showed a high efficiency reaching 99.3, 80.5, 99.3, and 96.4% for Pb, Cd, Ni, and Cu, respectively. According to these results, the two brown algal biomasses can be used as an effective, eco-friendly, and alternative biomass for the removal of heavy metal ions from aqueous solutions due to their high metal uptake capacity, cost-effectiveness, availability, and renewable biomass.

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