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## Biosorption and Recovery of Copper and Zinc from Aqueous Solutions by Nonliving Biomass of Marine Brown Algae of *Sargassum* sp.

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**Abstract:** In this study, the biosorption properties of a pre-treated nonliving biomass of marine brown algae of *Sargassum* species in the removal of Cu<sup>2+</sup> and Zn<sup>2+</sup> ions were investigated. Kinetics, equilibrium isotherms, recovery of metals and regeneration of the *Sargassum* biomass were carried out under different laboratory conditions using batch reactor. Biosorption of Cu<sup>2+</sup> and Zn<sup>2+</sup> was rapidly occurred onto *Sargassum* biomass and most of the sorbed metal was bound in less than 60 min. The removal performance for Zn<sup>2+</sup> by the biomass was found more than Cu<sup>2+</sup>, with maximum uptake values of 1.914 and 1.314 mg g<sup>-1</sup> dry weight biomass for Zn<sup>2+</sup> and Cu<sup>2+</sup>, respectively. Optimum biosorption pH value of Cu<sup>2+</sup> and Zn<sup>2+</sup> was determined as 5 at lab temperature. At the optimal condition, metal ion uptake increased with initial Cu<sup>2+</sup> and Zn<sup>2+</sup> concentration upto 200 and 500 mg L<sup>-1</sup>, respectively. The Cu<sup>2+</sup> and Zn<sup>2+</sup> uptake by *Sargassum* biomass was best described by pseudo-second order rate equation. The results showed that the Freundlich isotherm model was suitable for describing the passive biosorption of Cu<sup>2+</sup> and Zn<sup>2+</sup> by the dead biomass of *Sargassum*. Removal of the biosorbed Cu<sup>2+</sup> and Zn<sup>2+</sup> from *Sargassum* biomass was successfully achieved by eluting with 0.1 M HNO<sub>3</sub> for 15 min and a high degree of metal recovery was observed. For optimum operation in the subsequent metal uptake cycle, regeneration of the *Sargassum* biomass was efficiently performed by 0.1 M CaCl<sub>2</sub> for 15 min. In repeated use of biomass experiment, the Cu<sup>2+</sup> and Zn<sup>2+</sup> uptake capacity of *Sargassum* biomass was approximately retained and no significant biomass change took place after three biosorption-desorption cycles.

**Key words:** Biosorption, desorption, heavy metal, marine algae, *Sargassum* sp., aqueous solution

### INTRODUCTION

The removal and recovery of heavy metals from wastewater is important in the protection of the environment and human health (Kaewsarn, 2002). Some industrial processes result in the release of heavy metals in the natural water systems (Jalali *et al.*, 2002). Since copper and zinc are widely used materials, there are many actual or potential sources of copper and zinc pollution. Generally, any processing or container using copper and zinc material may contaminate the product, such as food, water or drink. Copper and zinc are essential to human life and health but, like all heavy metals, are potentially toxic as well (Vijayaraghavan *et al.*, 2004; Antunes *et al.*, 2003). Conventional methods for removing heavy metals from industrial effluents (e.g., chemical precipitation, chemical oxidation and reduction, ion exchange, reverse osmosis, membrane separation, electrochemical treatment and

evaporation) are often ineffective and costly when applied to dilute and very dilute effluents with heavy metal concentration of less than 100 mg L<sup>-1</sup> (Jalali *et al.*, 2002; Valdman and Leite, 2000; Gupta *et al.*, 2001; Matheickal and Qiming, 1999; Volesky, 1990). Biosorption is an alternative technology in which an increased amount of study is being focused (Antunes *et al.*, 2003; Hamdy, 2000a). Biosorption is a term that describes the removal of heavy metals by the passive binding to nonliving biomass from aqueous solution (Davis *et al.*, 2003). The biological materials that have been investigated for heavy metal uptake include fungi, bacteria, yeast, micro-algae and macro-algae (Hamdy, 2000b; Dönmez *et al.*, 1999; Matheickal and Qiming, 1996; Heravi, 1993; Volesky, 1990). Marine algae are biological resources, which are available in large quantities in many parts of the world (Kaewsarn, 2002). Some seaweeds collected from the ocean have indicated impressive biosorption of materials.

Brown marine algae tend particularly to sequester heavy metals (Davis *et al.*, 2003; Volesky, 1990). Brown seaweeds (phaeophyceae) constitute an algal group containing the characteristic pigment fucoxanthine, responsible for their brown color (Antunes *et al.*, 2003). In brown algae *Sargassum* biomass, alginate in the cell wall is the main component responsible for the metal sorption. It is present in a gel form in the cell wall, which appears very porous and easily permeable to small ionic species (Davis *et al.*, 2003; Vieira and Volesky, 2000). Alginic acid is present in the seaweeds usually as calcium, magnesium, sodium and potassium salts, mainly in the cell wall. It is a structural polysaccharide with strong ion exchange properties (Antunes *et al.*, 2003). Ion exchange has been confirmed to be highly involved to a large degree in the metal sequestering by algal biomass (Vieira and Volesky, 2000). The main objective of this study was to investigate the ability of nonliving biomass of marine brown algae of *Sargassum* sp. as a biosorbent for copper and zinc ions from aqueous solutions. The influences of different parameters on copper and zinc uptake, such as sorption time, pH, initial copper and zinc concentration and algae dose was investigated. The Freundlich and Langmuir isotherm models were used to analyze the biosorption equilibrium. Copper and zinc desorption and regeneration of the *Sargassum* biomass was also examined.

## MATERIALS AND METHODS

**Preparation of biosorbent:** Fresh samples of brown algae of *Sargassum* sp. used in this study were harvested from the Oman Sea on the coast of Chabahar, Iran. The biomass of *Sargassum* sp. was extensively washed with distilled water to remove dirt and particulate material from their surface and oven-dried at 60°C for 24 h. Dried biomass was ground in a laboratory blender. The biomass of *Sargassum* sp. (20 g) was treated with 0.1 M CaCl<sub>2</sub> solution (400 mL) for 15 min under slow stirring (Protonation). The calcium treated biomass was washed several times with deionized water to remove excess calcium from the biomass and kept on a filter paper to reduce the water content. After that, the nonliving biomass was heated in an oven at 60°C for 24 h and then sieved for particle size 400-600 µm (500 µm). Finally, the nonliving biomass of *Sargassum* sp. was stored in desiccators until they were used.

**Copper and zinc solutions:** Stock copper and zinc solutions (1000 mg L<sup>-1</sup>) were prepared by dissolving 3.93 g of CuSO<sub>4</sub>·5H<sub>2</sub>O and 4.4 g of ZnSO<sub>4</sub>·7H<sub>2</sub>O (Merck, Germany) in 1000 mL of Deionized Distilled Water (DDW). Copper and zinc solutions of different concentrations

were prepared by adequate dilution of the stock solutions with DDW (APHA, 1998). The range in initial concentrations of copper and zinc prepared from stock solutions varied between 20-500 mg L<sup>-1</sup>.

**Determination of the copper and zinc contents in the solutions:** The concentration of copper and zinc in the solutions before and after the equilibrium was determined by flame atomic absorption spectrophotometry (FAAS), using a Perkin Elmer 2380 atomic absorption spectrophotometer at the wavelength of 324.8 nm and 214 nm for copper and zinc, respectively (APHA, 1998).

**Kinetic experiments:** Biosorption studies were conducted in a routine manner by the batch technique. Preliminary experiments were performed to determine equilibrium time for biosorption of copper and zinc by *Sargassum* biomass. For this purpose, 500 mg of dried biomass (size of particles  $d = 400-600 \mu\text{m}$ ) was added to 50 mL metal solution with a known concentration (20 mg L<sup>-1</sup>) and initial pH of 5 in 100 mL Erlenmeyer flasks. The flasks placed on a rotating shaker (SINA 2000, Iran) with constant shaking at 200 rpm, at 21±2°C. The pH of solutions during the contact period (5-420 min) was adjusted at 5±0.2 using small amount of 0.1 M H<sub>2</sub>SO<sub>4</sub> or 0.1 M NaOH as required. All pH measurements were carried out with a pH meter model CG-710. Samples were periodically withdrawn from the shaker and the solutions were separated from the biomass by filtration through filter papers (Whatman No. 40 Ashless). After appropriate dilution, the concentrations of copper and zinc in the filtrate were determined by Flame Atomic Absorption Spectrophotometry (FAAS). The effect of pH values (1-11), initial copper and zinc concentration (20-500 mg L<sup>-1</sup>) and *Sargassum* biomass dose (100-500 mg 50 mL<sup>-1</sup>) on the biosorption of Cu<sup>2+</sup> and Zn<sup>2+</sup> by nonliving biomass of *Sargassum* was studied.

**Equilibrium experiments:** The equilibrium isotherms were determined at 21±2°C under optimized conditions, changing the biomass dose into the range of 100-500 mg 50 mL<sup>-1</sup> and using an equilibrium time equal to 300 min. Metal free and biosorbent free blanks were used as control. All biosorption experiments were carried out in duplicates and the average value were used for further calculations.

**Recovery of metals and repeated use of biomass:** Following the copper and zinc biosorption batch experiment, metal-laden biomass (*Sargassum* sp.) was separated by filtration and suspended into 50 mL of the eluent solution (0.1 M HNO<sub>3</sub>). Desorption of Cu<sup>2+</sup> and

Zn<sup>2+</sup> from biomass was carried out on a rotary shaker (200 rpm) for 15 min. The biomass was separated by filtration and thoroughly washed with distilled water. The concentration of the Cu<sup>2+</sup> and Zn<sup>2+</sup> released into the eluent solution was determined by FAAS. After desorption, the unloaded biomass was regenerated with 50 mL of 0.1 M CaCl<sub>2</sub> for 15 min, twice washed with distilled water, then filtered and finally oven-dried overnight at 60°C. The regenerated biomass suspended in the new solution for 5 h. The biosorption-desorption experiment was performed in three successive cycles.

**Data evaluation:** The amount of metal bound by the biosorbent was calculated as follows:

$$q = \frac{v(C_i - C_f)}{m}$$

Where q is the metal uptake (mg metal g<sup>-1</sup> of the biosorbent), v the liquid sample volume (mL), C<sub>i</sub> the initial concentration of the metal in the solution (mg L<sup>-1</sup>), C<sub>f</sub> the final (equilibrium) concentration of the metal in the solution (mg L<sup>-1</sup>) and the amount of the added biosorbent on dry basis (mg).

The Langmuir isotherm model,  $q = \frac{QbC_f}{1 + bC_f}$

The linear form of the Langmuir isotherm model,

$$\frac{1}{q} = \frac{1}{Q} + \frac{1}{bQ} \frac{1}{C_f}$$

Where, Q is the maximum metal uptake under given conditions, b a constant related to the affinity between the biosorbent and sorbate.

The Freundlich isotherm model,  $q = kC_f^{\frac{1}{n}}$

The linear form of the Freundlich isotherm model,

$$\log(q) = \log(K) + \frac{1}{n} \log(C_f)$$

Where, k and n are Freundlich constants.

## RESULTS

The biosorption of copper and zinc in aqueous solution on pre-treated nonliving biomass of *Sargassum* sp. were examined by optimizing various physicochemical parameters such as contact time, pH, initial copper and zinc concentration and *Sargassum* biomass dose. Figure 1 shows the effect of contact time on the removal of Cu<sup>2+</sup> and Zn<sup>2+</sup> by *Sargassum* biomass. According to Fig. 1, this species of brown algae removed zinc most efficiently than copper from aqueous solution. Figure 2 shows the biosorption kinetics plot of Cu<sup>2+</sup> and Zn<sup>2+</sup> onto *Sargassum* biomass. According to Fig. 2, kinetics of biosorption obeyed a pseudo-second order equation. The biosorption data for the metal uptake versus contact time

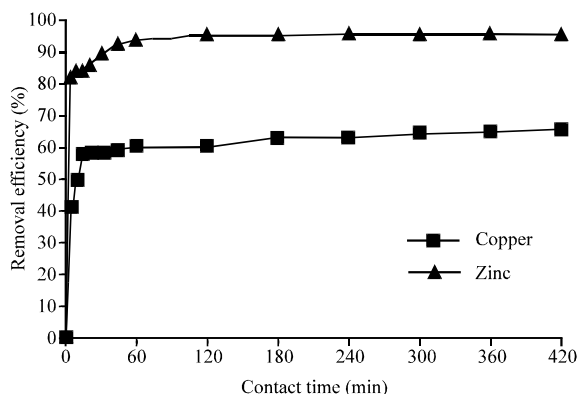


Fig. 1: Effect of contact time on the removal of Cu<sup>2+</sup> and Zn<sup>2+</sup> by nonliving biomass of *Sargassum* sp.

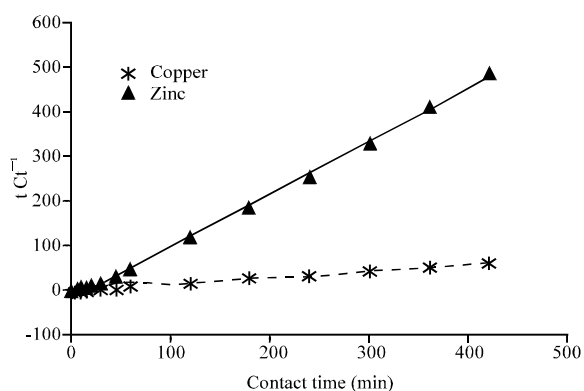


Fig. 2: Pseudo-second order kinetics plot of Cu<sup>2+</sup> and Zn<sup>2+</sup> onto nonliving biomass of *Sargassum* sp.

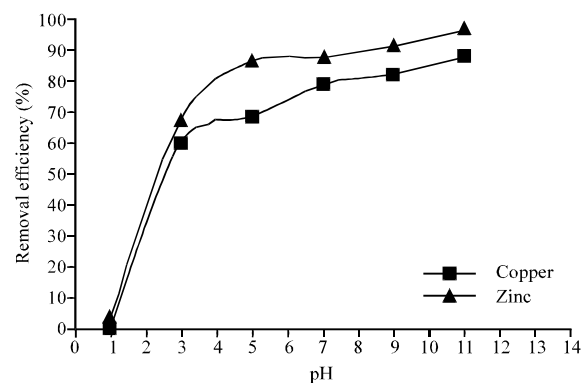


Fig. 3: Effect of pH on the removal of Cu<sup>2+</sup> and Zn<sup>2+</sup> by nonliving biomass of *Sargassum* sp.

at 20 mg L<sup>-1</sup> initial copper and zinc concentration with 500 mg 50 mL<sup>-1</sup> of biomass were carried out in pH value of 5±0.2. The results show that Cu<sup>2+</sup> and Zn<sup>2+</sup> removal increases with time and attains equilibrium in 5 h. An increasing uptake of the metals by biosorbent with increasing the pH was demonstrated in Fig. 3. This figure

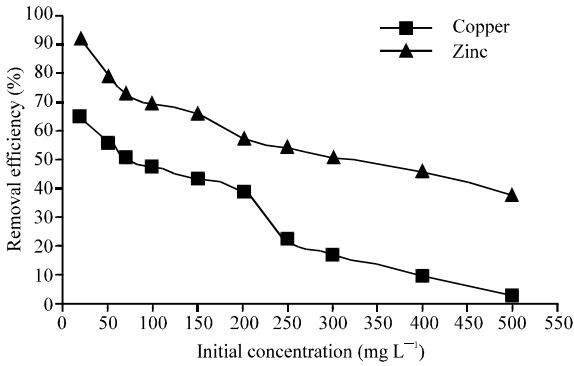


Fig. 4: Effect of initial Cu<sup>2+</sup> and Zn<sup>2+</sup> concentration on their removal by nonliving biomass of *Sargassum* sp.

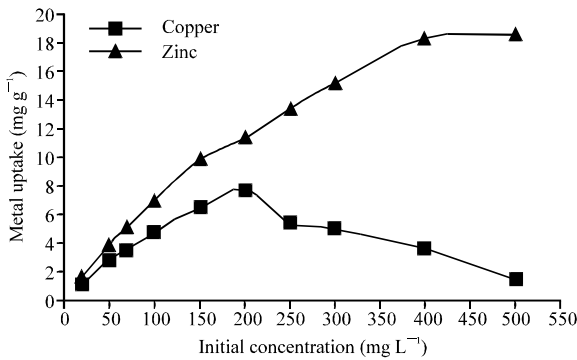


Fig. 5: Effect of initial Cu<sup>2+</sup> and Zn<sup>2+</sup> concentration on the capacity of their biosorption by nonliving biomass of *Sargassum* sp.

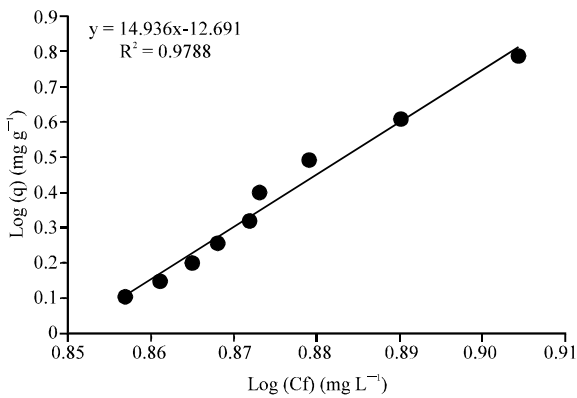


Fig. 6: Freundlich biosorption isotherm for copper with nonliving biomass of *Sargassum* sp.

represents the effect of initial pH on the removal of Cu<sup>2+</sup> and Zn<sup>2+</sup> by nonliving biomass of *Sargassum*. Biomass dose, initial Cu<sup>2+</sup> and Zn<sup>2+</sup> concentration and equilibrium time were 500 mg 50 mL<sup>-1</sup>, 20 mg L<sup>-1</sup> and 5 h, respectively.

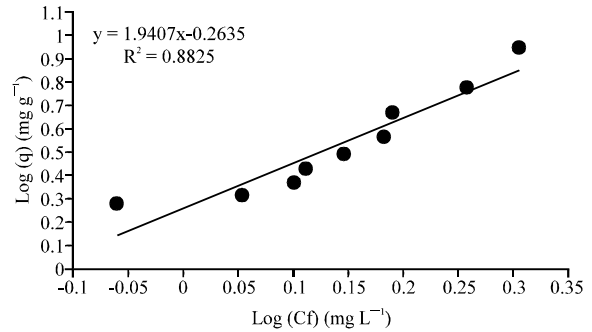


Fig. 7: Freundlich biosorption isotherm for zinc with nonliving biomass of *Sargassum* sp.

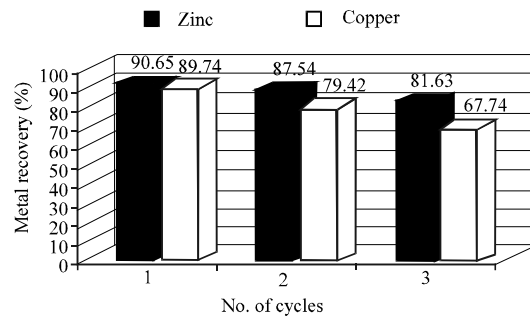


Fig. 8: Percent recovery of biosorbed Cu<sup>2+</sup> and Zn<sup>2+</sup> onto nonliving biomass of *Sargassum* sp. in three successive cycles

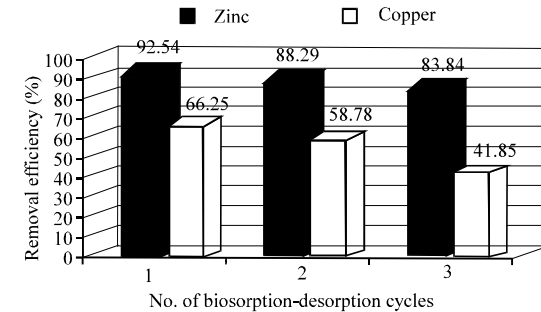


Fig. 9: Effect of regeneration of nonliving biomass of *Sargassum* sp. on the removal of Cu<sup>2+</sup> and Zn<sup>2+</sup> in three successive biosorption-desorption cycles

*Sargassum* biomass was added separately in the pH range 1-11 and the results are depicted in Fig. 3. Figure 4 shows the removal of Cu<sup>2+</sup> and Zn<sup>2+</sup> as a function of their initial concentration by *Sargassum* biomass. The capacities of Cu<sup>2+</sup> and Zn<sup>2+</sup> biosorption at equilibrium time by biomass obtained from experimental data at different initial concentrations are presented in Fig. 5. Algae dose, equilibrium time and pH were 500 mg 50 mL<sup>-1</sup>, 5 h and 5, respectively. The linearized Freundlich biosorption

isotherms of *Sargassum* sp. for  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  are shown in Fig. 6 and 7, respectively. Attempt to desorption of the loaded  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  from the biomass was investigated by 0.1 M  $\text{HNO}_3$  that is shown in Fig. 8. Regeneration without damaging the capacity of the biosorbent is very important factor for the success of the biosorbent technology development. Figure 9 shows the  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  removal efficiency by biomass after three biosorption-desorption cycles. Algae dose, initial  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  concentration, equilibrium time and pH were  $500 \text{ mg } 50 \text{ mL}^{-1}$ ,  $20 \text{ mg L}^{-1}$  and 5 h, respectively.

## DISCUSSION

**Effect of contact time:** In the biosorption of  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  by *Sargassum* biomass, most of the metal ions were sequestered from solution within the first 60 min and almost no increase in the level of bound metal occurred after 5 h. So, 5 h was used as the equilibrium time for *Sargassum* biomass. As seen from the results, the kinetics of  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  binding to the biosorbent follow the pseudo-second order rate equation. The very fast sorption kinetics observed with *Sargassum* biomass represents an advantageous aspect when effluent treatment systems are designed. These results indicated that nonliving biomass of *Sargassum* sp. removed zinc most efficiently than copper from aqueous solution. Differences between algal species in the magnitude of change in metal ion binding capacity may be due to the properties of the metal sorbate (e.g., ionic size, atomic weight, or reduction potential of the metal) and the algae (e.g., structure, functional groups and surface area, depending on the algal division, genera and species) (Dönmez *et al.*, 1999). Marine brown algae in particular are suited for binding metallic ions due to their polysaccharide material content (alginates, xylofocoglycuronans, xylofocoglucans and homofucans). These polysaccharides contain carboxyl and sulfate groups that have identified as the main metal-sequestering sites (Vijayaraghavan *et al.*, 2004; Davis *et al.*, 2003; Jalali *et al.*, 2002; Vieira and Volesky, 2000).

**Effect of pH:** The initial pH of the solution is a very important factor in  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  sorption uptake by *Sargassum* biomass. Earlier studies have indicated that solution pH is an important parameter affecting biosorption of heavy metal ions and the concentration of the counter ions on the functional groups of the biomass cell wall. So, pH is an important parameter on biosorption of metal ions from aqueous solutions (Vijayaraghavan *et al.*, 2004; Antunes *et al.*, 2003; Jalali *et al.*, 2002; Kaewsarn, 2002; Matheickal and Qiming,

1999). *Sargassum* sp. presents a high content of ionizable groups (carboxyl groups from mannuronic and guluronic acids) on the cell wall polysaccharides, which makes it, at least in theory, very liable to the influence of the pH. According to the results, the uptake of free ionic copper and zinc depend on pH, increasing with the increase in pH from 2 to 3 and then reaching a plateau in the range 4-5. Similar results were reported on literature (Antunes *et al.*, 2003; Cossich *et al.*, 2002; Jalali *et al.*, 2002; Kaewsarn, 2002; Matheickal and Qiming, 1999). At the pH values lower than 3,  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  removal was strongly decreased, possibly as a results of the competition between hydrogen and  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  ions on the sorption sites, with an apparent preponderance of hydrogen ions, which restricts the approach of metal cations as in consequence of the repulsive force. As the pH increased (pH = 3.5-5), the ligands such as carboxylate groups in *Sargassum* sp. would be exposed, increasing the negative charge density on the biomass surface, increasing the attraction of metallic ions with positive charge and allowing the biosorption onto the cell surface. As seen from the results, in pH values higher than 5, insoluble copper and zinc start precipitating from the solution. So, these pH values making true sorption studies impossible and we chose pH = 5 for further experiments.

**Effect of initial copper and zinc concentration:** As seen from results, at the optimal conditions, metal ion uptake increased with initial  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  ion concentration upto 200 and  $500 \text{ mg L}^{-1}$ , respectively. Increasing the initial  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  concentration would increase the mass transfer driving force and therefore the rate at copper and zinc molecules pass from the bulk solution to the biomass surface. This would results in higher  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  biosorption capacity (Dönmez *et al.*, 1999). On a relative basis, however, the percentage biosorption of copper and zinc decreases as their initial concentration increases. The equilibrium uptake and biosorption yield were highest for the zinc, which was expected.

**Biosorption isotherms:** Several models have been published in the literature to describe experimental data of biosorption isotherms. The Freundlich and Langmuir models are the most frequently employed models. In this study, both models were used describe the relationship between the amount of copper and zinc biosorbed and their equilibrium concentration for *Sargassum* biomass. The results showed that the Freundlich isotherm model was suitable for describing the passive biosorption of  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  by the nonliving biomass of *Sargassum*.

**Recovery of metals and repeated use of biomass:** Recovery of the biosorbed  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  on *Sargassum* biomass was carried out by 0.1 M  $\text{HNO}_3$  for 15 min.

According to the results, a high degree of metal recovery was observed and  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  released to this dilute mineral acid with 90.65 and 89.74% elution efficiency, respectively after the first cycle. On the other hand, mineral acids ( $\text{HNO}_3$ ,  $\text{HCl}$  and  $\text{H}_2\text{SO}_4$ ) are able to elute and concentrate the biosorbed metals such as copper and zinc. This was also achieved previously (Vijayaraghavan *et al.*, 2004; Jalali *et al.*, 2002; Hamdy, 2000b). For optimum operation in the subsequent metal uptake cycle, regeneration of the *Sargassum* biomass was efficiently performed by 0.1 M  $\text{CaCl}_2$  for 15 min. Regeneration without damaging the capacity of the biosorbent is a very important factor for the success of the biosorbent technology development. Heavy metal biosorption by inactive biomass had been improved by  $\text{Ca}^{2+}$  and/or  $\text{Mg}^{2+}$  saturation of biomass (Jalali *et al.*, 2002; Volesky, 1990). Regeneration of the biomass, after desorption of the bound  $\text{Cu}^{2+}$  and  $\text{Zn}^{2+}$  and subsequent water washing with 0.1 M  $\text{CaCl}_2$  was efficient. As seen from the results, in repeated use of biomass experiment, the copper and zinc uptake capacity of *Sargassum* biomass was approximately retained and no significant biomass damage took place after three biosorption-desorption cycles. Similar results were reported on other works (Vijayaraghavan *et al.*, 2004; Jalali *et al.*, 2002; Hamdy, 2000b). According to the results, removal capacity of  $\text{Ca}^{2+}$  regenerated *Sargassum* biomass after three uptake/elution cycles and decrease in the pH of solution (in batch experiment performed without pH adjustment) revealed that an ion exchange between the metal and  $\text{H}^+$  or  $\text{Ca}^{2+}$  occurred.

#### ACKNOWLEDGMENTS

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