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Investigation of the Column Performance of Cadmium(II) Biosorption by *Cladophora crispata* Flocs in a Packed Bed

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ABSTRACT

In this study the biosorption of cadmium(II) ions to dried flocs of *Cladophora crispata*, a kind of green algae, was investigated in a packed bed column. The cadmium(II) removal performance of the column was investigated as a function of the cadmium(II)-bearing solution flow rate and the inlet cadmium(II) concentration. Removal and total removal percentages of cadmium(II) related to flow volume were determined by evaluating the breakthrough curves obtained at three different flow rates for two different constant inlet concentrations. At the lowest flow rate the effect of inlet cadmium(II) concentration on the column capacity was also investigated. Data confirmed that early saturation and lower cadmium(II) removals were observed at higher flow rates and at higher cadmium(II) concentrations. Column experiments

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also showed that maximum specific cadmium(II) uptake values of *C. crispata* flocs were as higher as those of other biomass sorbents.

Key Words. Packed bed column; Flocs of *Cladophora crispata*; Cadmium(II) biosorption

INTRODUCTION

Cadmium, well recognized for its negative effect on the environment where it accumulates in the food chain, is used in a wide variety of industrial processes, e.g., mining and metallurgy of cadmium, cadmium electroplating, battery and accumulator manufacturing, pigments, and ceramic industries. Heavy metal ion concentrations are higher than values allowed by water standards, and they must be reduced to the desired values (1–4).

Using microorganisms as biosorbents for heavy metals offers a potential alternative to existing methods for detoxification and for the recovery of toxic or valuable metals from industrial discharge water. Many aquatic microorganisms such as algae can adsorb dissolved heavy metals from their surroundings. Metal ion binding to nonliving cells occurs rapidly by cell surface (passive) adsorption and is called "biosorption." The use of dead microbial cells is more advantageous for water treatment in that dead organisms are not affected by toxic wastes (1–17).

The biosorption of metal ions by the algal biomass arises from the coordination of the ions to different functional groups on the algal cell. These coordinating groups (provided by proteins, lipids, and carbohydrates) include amino, carboxyl, sulfhydryl, phosphate, and hydroxyl groups as well as the sulfate. The amino groups in the proteins on the cell wall and the nitrogen and oxygen of the peptide bond could also be available for bonding metallic ions. Such bond formation could be accompanied by the displacement of protons dependent in part on the extent of protonation as determined by the pH. Metallic ions could also be electrostatically bonded to unprotonated carboxyl oxygen and sulfate. The kinetics of metal uptake, thought to be physical adsorption at the cell surface, is very rapid and occurs in a short time after the microorganism comes into contact with the metal (1, 2, 5, 6, 8, 9, 12, 14, 15, 17).

A major consideration with any biosorption scheme is the separation of liquid and solids after batch or continuous contacting. Centrifugation or filtration, as routinely used in the laboratory, are not generally practical in industrial processes; thus, immobilized or free microbes packed into columns are generally used. Packed-bed adsorption has a number of advantages related to its process engineering. It is a simple, high yield operation and relatively easily scaled up from a laboratory-scale procedure. The stages in the separation protocol can be automated and high degrees of purification can often be achieved in a single-step process (1, 3, 7, 11, 16, 17).

Immobilizing biomass in a granular or polymeric matrix may improve the biomass performance, biosorption capacity, and facilitate separation of biomass from metal-bearing solution. But diffusion limitations caused a decrease in the biosorption rate, and the lack of mechanical strength of immobilized cells for large-scale systems with higher costs are the major disadvantages of immobilized systems. As an alternative, the direct use of colonies or flocs of microbial cells without immobilizing can be proposed for the biosorption of heavy metal ions in a packed-bed column. Algal colonies and flocs are capable of achieving high cell densities while minimizing diffusion limitations (1–3, 6, 7, 11, 16, 17).

Continuous biomass packed into a column system is simply a piece of pipe, standing on its end and filled with biomass flocs. Fluid containing the metal ion of interest flows into one end of the pipe and out the other end. Initially, most of the metal ion is adsorbed, so that the solute concentration in the effluent is low. As biosorption continues, the effluent concentration rises, slowly at first, but then abruptly. When this abrupt rise or "breakthrough" occurs, the flow is stopped. The general position of the breakthrough curve along the volume axis depends on the capacity of the column with respect to the feed concentration and flow rate. The breakthrough curve would be a step function for favorable separations, i.e., there would be an instantaneous jump in the effluent concentration from zero to the feed concentration at the moment the column capacity is reached.

Much of the information needed to evaluate column performance is contained in plots of adsorbed metal ion concentration (C_{ad} = inlet metal ion concentration − outlet metal ion concentration) or normalized concentration defined as the ratio of effluent metal ion concentration to inlet metal ion concentration (C/C_0) as a function of flow time (t) or effluent volume (V_{eff}) calculated from Eq. (1).

$$V_{eff} = Qt \tag{1}$$

where V_{eff} = effluent volume (mL)
t = flow time (min)
Q = volumetric flow rate (mL/min)

Removal or total removal percentages of metal ion with respect to flow volume can also be found from the ratio of adsorbed or total adsorbed quantities to the amount or total amount of metal ion sent to the column. The total adsorbed quantity in the column for a given feed concentration is equal to the area under the breakthrough curve obtained from the adsorbed concentration vs effluent volume plot. Total amount of metal ion sent to the column is calculated from Eq. (2).

$$\text{Total amount of metal ion sent to column} = C_0 Q t_{\text{total}}/1000 \qquad (2)$$

where C_0 = inlet metal ion concentration (mg/L)
t_{total} = total flow time (min)

Maximum specific metal uptake (q_{total}) is defined by Eq.(3) as the total amount of metal ion sorbed per gram of dried flocs of the alga at the end of total flow time (11, 18, 19).

$$q_{\text{total}} = \frac{\text{total adsorbed quantity in the column}}{X} \qquad (3)$$

where q_{total} = maximum specific metal uptake of biosorbent in the column (mg/g)
X = amount of biosorbent (dried algal flocs) in the column (g)

The aim of this study was to investigate the column performance of the cadmium(II) biosorption by the dried flocs of *Cladophora crispata* in a packed bed column as a function of flow rate and inlet cadmium(II) concentration. This is the first detailed study on the direct use of *C. crispata* flocs for cadmium(II) adsorption in a packed bed.

EXPERIMENTAL

Microorganism and Cadmium(II) Solutions for Biosorption

C. crispata, a species of green alga collected from irrigation water channels near Firat University, Elazığ, Turkey, was used in this study. For the biosorption studies the harvested fresh cell flocs were rinsed with tap water, washed several times with distilled water, and then inactivated in an oven at 90°C for 24 hours. For the biosorption studies in the batch system, 1.0 g of dried biomass was suspended in 100 mL of double-distilled water and homogenized in a lidded, stainless steel Waring mixer for 35–40 minutes and then stored in a refrigerator. For the biosorption studies in the column, 3.7 g of dried algae was prepared as described before. After waiting for 24 hours, the dried algae solution was filtered using Whatman 1 filter paper. The wet algae were filled in the column with small silicone rings, 0.4 cm in diameter and 0.4 cm in height together, to reduce the pressure drop and to prevent flow canalization. Fresh biomass was always used in the trials.

Cadmium(II) solutions were prepared by diluting 1.0 g/L of stock cadmium(II) solution which was obtained by dissolving exact quantities of anhydrous $CdCl_2$ in double distilled and deionized water. Before biosorption in the batch or continuous column system, the pH of each test solution was adjusted to the required value with 1 M H_2SO_4.

Biosorption Studies in the Batch and Column Systems

The effects of pH and temperature on cadmium(II) biosorption by dried algal flocs were examined in the batch system. For the batch system studies, 10 mL of dried algae solution was contacted with a known concentration of 90 mL metal-bearing solution in Erlenmayer flasks at the desired temperature and pH. The flasks were agitated on a shaker for 120 minutes, which is more than ample time for sorption equilibrium. Three milliliter samples of solution were taken at definite intervals and centrifuged at 6000 rpm for 2 minutes. Then the liquid was separated and analyzed for cadmium(II) ions.

Continuous fixed-bed column studies was performed in a column with an i.d. of 3.0 cm and 3.7 g of dried algae. The bed depth was kept at 20.0 cm. The temperature of the column during the experiment was kept constant at 25°C. The column was preconditioned to pH 5.0 (by eluting the column with 0.01 M H_2SO_4). The metal ion solution at a known concentration was passed continuously through the stationary bed of biosorbent (algae). The flow rate was regulated with a variable speed pump by a Masterflex L/S digital drive and easy-load pump head. The effluent samples were taken out from the effluent at precise time intervals and analyzed for cadmium(II) ions as described below. The experiment was continued until the effluent concentration of cadmium(II) ion reached a constant concentration.

Cadmium(II) Analysis

The concentration of unadsorbed cadmium(II) ions was determined by an atomic absorption spectrophotometer (GBC Model 932) with a detection limit of 3.0 ppm at a wavelength of 228.8 nm. Reagent blanks were run for every sample.

RESULTS

Biosorption of heavy metal ions by algal cells is affected by several factors. These factors include the specific surface properties of the organism and such physicochemical parameters of the solution as pH, temperature, initial metal ion concentration, and biomass concentration. The batch system studies showed that the adsorption rate of cadmium(II) ions to the dried green alga was also affected by the temperature and the initial pH of the cadmium(II) ion solution. It was observed that equilibrium cadmium(II) uptake increases as the initial pH of the adsorption medium increases to a value of 5.0 (Fig.

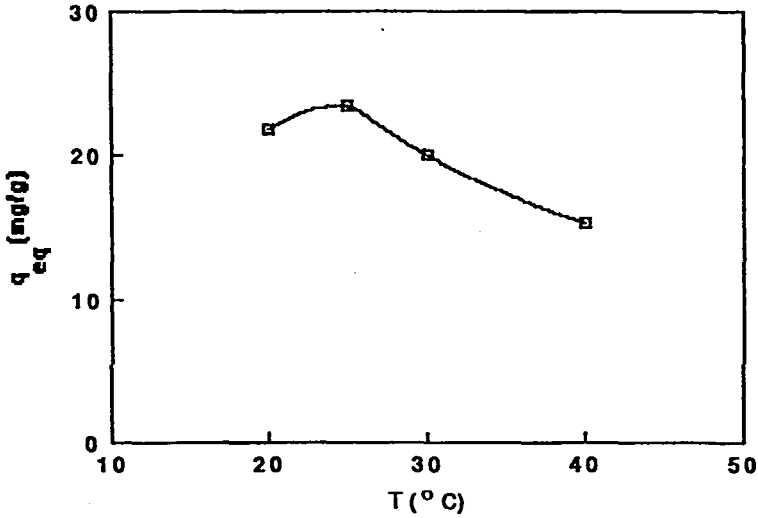

FIG. 1 The effect of temperature on the equilibrium cadmium(II) uptake. Initial pH: 5.0. Initial cadmium(II) concentration: 75 mg/L, Biomass concentration: 1.0 g/L. Agitation rate: 150 rpm.

1). When the initial pH of the adsorption medium is adjusted to values higher than 5.0, precipitation is observed. The effect of temperature on cadmium(II) uptake is less significant than that of pH. Figure 2 shows that the cadmium(II) uptake is only slightly affected by variations in temperature. In general, biosorption of cadmium(II) ions by green algae is normally exothermic, thus the extent of biosorption increases with decreasing temperature. The optimum adsorption temperature of cadmium(II) ions to the dried green algae was determined as 25°C. Packed-bed column studies were performed at these optimum conditions.

The effects of flow rate and inlet cadmium(II) concentration on the column performance of cadmium(II) biosorption on *Cladophora crispata* flocs were investigated in a packed-bed column. In the first stage of the removal studies in the packed bed, the flow rate was changed from 3.2 to 9.0 mL/min while the inlet cadmium(II) concentration in each experiment was held constant at 25 or 175 mg/L. The adsorption breakthrough curves obtained at different flow rates at 25 and 175 mg/L of inlet cadmium(II) concentrations are given in Figs. 3 and 4. At 25 mg/L of inlet cadmium(II) concentration and at the lowest flow rate of 3.2 mL/min, the biosorption, although continuous with time, is very efficient in the initial steps of the process. This fact is probably associated to the availability of reactional sites able to capture metal ions

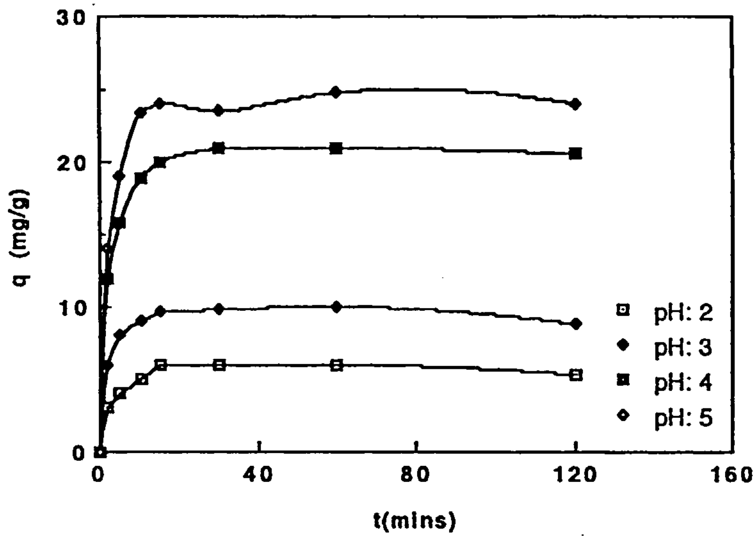

FIG. 2 The effect of initial pH on the cadmium(II) uptake. Temperature: 25°C. Initial cadmium(II) concentration: 75 mg/L. Biomass concentration: 1.0 g/L. Agitation rate: 150 rpm.

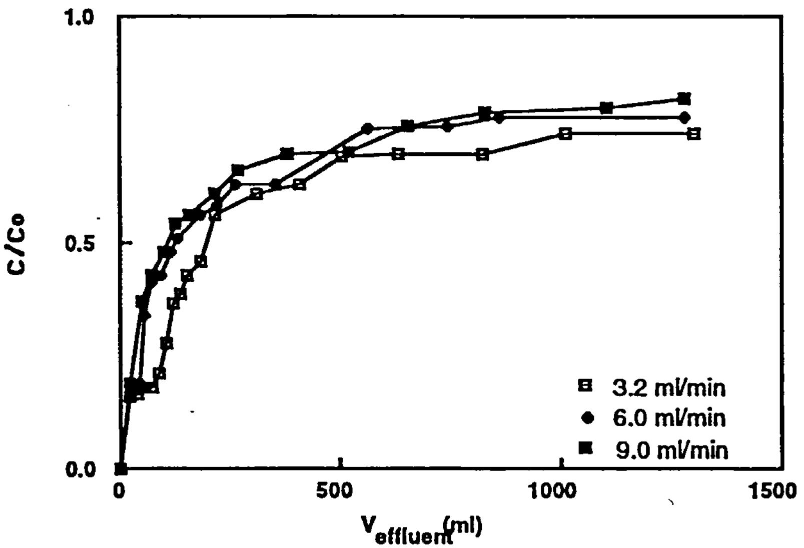

FIG. 3 The adsorption breakthrough curves obtained at different flow rates at 25 mg/L of inlet cadmium(II) ion concentration. Temperature: 25°C. pH of feed cadmium(II) solution: 5.0.

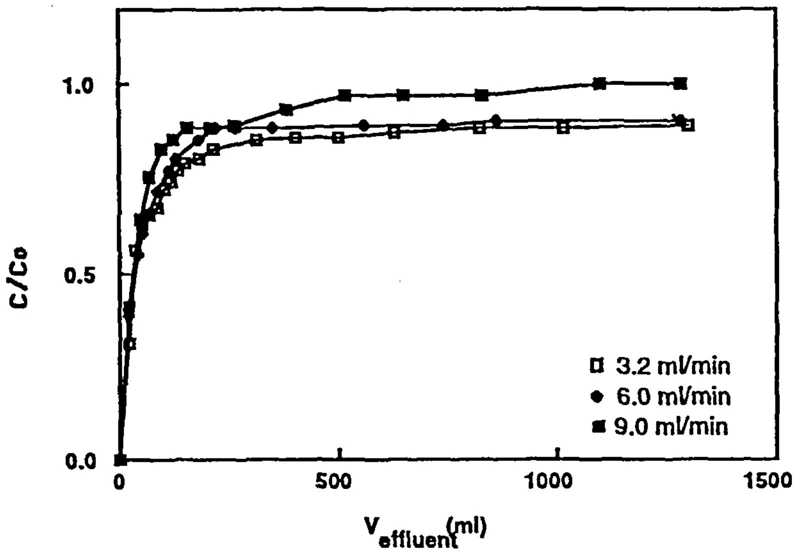

FIG. 4 The adsorption breakthrough curves obtained at different flow rates at 175 mg/L of inlet cadmium(II) ion concentration. Temperature: 25°C. pH of feed cadmium(II) solution: 5.0.

around or inside the cells. Rapid cadmium(II) uptake by the algal flocs also shows that biosorption is not affected by solute diffusion. In the second stage, with the gradual occupancy of these sites, the uptake become less effective. Even after breakthrough occurs, the column is still capable of accumulating cadmium(II), although at a progressively lower efficiency. As indicated in Fig. 3, as the flow rate increases the breakthrough curve becomes steeper and the breakpoint time and adsorbed metal ion concentration decrease with increasing flow rate. The reason for this behavior can be explained in the following way: Adsorption equilibrium between cadmium(II) and biosorbent occurs within 8–10 minutes, at the end of a rapid physical adsorption (Fig. 2). If the residence time of the solute in the column is not long enough for adsorption equilibrium to be reached at that flow rate, the metal ion solution leaves the column before equilibrium occurs. So at higher flow rates, the contact time of cadmium(II) ions with the algal flocs is very short. As indicated in Fig. 4, much sharper breakthrough curves are obtained with dried green alga at 175 mg/L of inlet cadmium(II) concentration and at all studied flow rates. At the lowest flow rate of 3.2 mL/min, relatively higher uptake values observed for cadmium(II) adsorption at 175 mg/L of inlet cadmium(II) concentration at the beginning of column operation could be due to the magnitude of the concentration driving force. But, as solution continues to flow, the concentration of cadmium(II) in the effluent rapidly increases, the bed becomes saturated with metal ion, and the concentration of solute in the effluent suddenly rises to the inlet cadmium(II) concentration. This behavior can also explain why cadmium(II) biosorption by algal flocs is not affected by solute diffusion and the number of active sites and ionic groups of algal flocs for biosorption is limited. It was also observed that the adsorbed metal ion concentration decreases with increasing flow rate at 175 mg/L of inlet cadmium(II) concentration.

Plots of the comparative adsorbed cadmium(II) concentration vs effluent volume at different flow rates are given in Figs. 5 and 6 for 25 and 175 mg/L of inlet cadmium(II) concentrations. As may be seen from Figs. 5 and 6, the breakthroughs for 25 and 175 mg/L inlet cadmium(II) concentrations occur at the end of 22.2 and 7.2 minutes, respectively, at a 3.2 mL/min flow rate. The lowest flow rate and lowest inlet cadmium(II) concentration gave a more gentle breakthrough curve. Metal adsorption by the dried algal flocs in the packed column depends on the volume of flow passed. The effluent volume shows whether metal uptake by the biosorbent is subject to saturation limits or not. By evaluating the adsorption curves given in Figs. 5 and 6, adsorbed

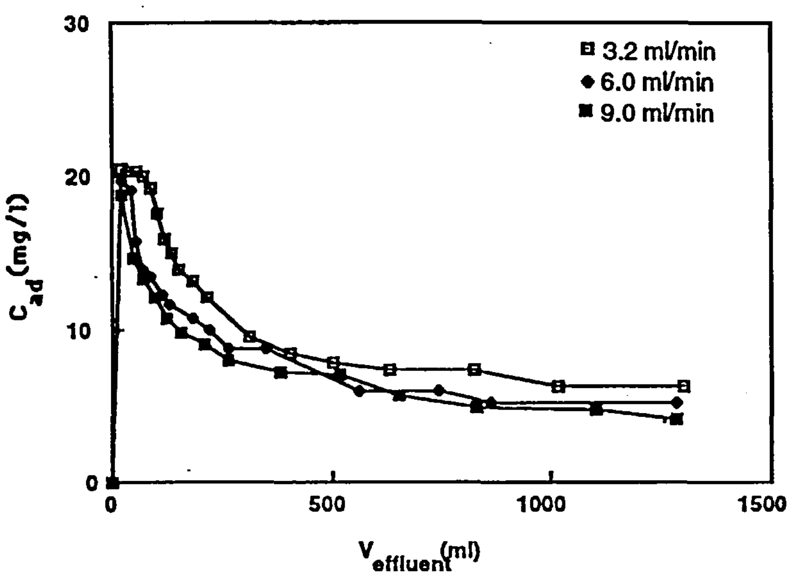

FIG. 5 The change of adsorbed cadmium(II) concentration with effluent volume at different flow rates at 25 mg/L of inlet cadmium(II) ion concentration. Temperature: 25°C. pH of feed cadmium(II) solution: 5.0.

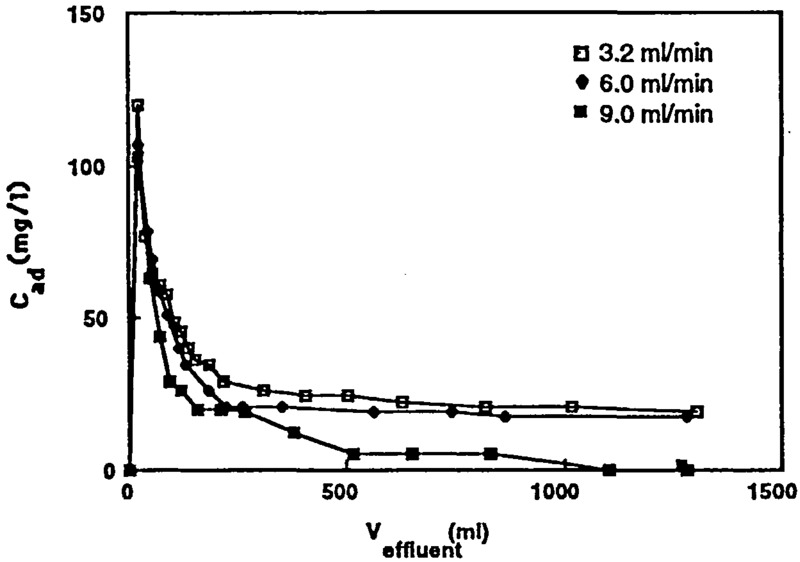

FIG. 6 The change of adsorbed cadmium(II) concentration with effluent volume at different flow rates at 175 mg/L of inlet cadmium(II) ion concentration. Temperature: 25°C. pH of feed cadmium(II) solution: 5.0.

and total adsorbed cadmium(II) quantities are determined by the computer. Removal percentages of cadmium(II) with respect to the volume of flow passed at different flow rates for 25 and 175 mg/L of inlet cadmium(II) concentrations are presented in Table 1. In general, increasing the flow volume decreases the total cadmium(II) removal percentages for all the flow rates and inlet cadmium(II) concentrations. Total cadmium(II) removal percentages at different flow rates obtained with a 25 mg/L inlet cadmium(II) concentration are much higher than those of a 175 mg/L inlet cadmium(II) concentration. The results found at the lowest flow rate also show that at the end of 1282 mL of flow volume, total removal percentages of cadmium(II) are obtained as 38.0 and 15.8% for 25 and 175 mg/L of inlet cadmium(II) concentrations, respectively. It is obvious that early saturation and lower cadmium(II) removal percentages are observed at higher flow rates and at higher cadmium(II) concentrations.

As generally expected, a change in the inlet metal ion concentration of the feed affects the operating characteristics of the packed column Figure 7 shows the effect of inlet metal ion concentration on cadmium(II) biosorption at the 3.2 mL/min flow rate. The adsorption breakthrough curves at 25, 50, 75, 125,

TABLE 1
Effect of Flow Volume on the Removal and Total Removal of Cadmium(II) at Various Flow Rates for 25 and 175 mg/L of inlet metal ion concentrations (T = 25°C; pH of inlet cadmium(II) solution = 5.0)

3.2 mL/min		6.0 mL/min		9.0 mL/min	
Flow volume (mL)	Cd(II) removal (%)	Flow volume (mL)	Cd(II) removal (%)	Flow volume (mL)	Cd(II) removal (%)
		C_0 = 25 mg/L			
23	83.8	23	82.7	23	80.7
71	83.0	71	73.0	68	66.6
103	81.0	113	64.8	122	58.5
135	76.2	185	56.8	212	49.8
183	70.8	263	51.5	266	46.4
311	56.8	352	44.8	382	41.9
503	47.6	563	37.5	518	38.7
823	41.8	743	34.4	833	32.5
1015	39.6	863	32.6	1103	28.9
1282	38.0	1282	29.1	1282	27.3
		C_0 = 175 mg/L			
23	68.9	23	61.3	23	58.5
71	48.5	71	46.6	68	40.6
103	43.3	113	40.8	122	29.7
135	38.5	185	35.8	212	21.0
183	34.2	263	27.9	266	17.3
311	27.2	352	20.0	382	14.9
503	22.3	563	16.7	518	12.0
823	18.4	743	12.0	833	8.6
1015	17.2	863	10.5	1103	7.5
1282	15.8	1282	7.7	1282	6.6

and 150 mg/L inlet metal ion concentrations at this flow rate are also given in Fig. 8. As seen from Figs. 7 and 8 the breakpoint time decreases with increasing inlet metal ion concentration. At lower inlet metal ion concentrations the breakthrough curves are dispersed, breakthrough occurs very late, and the surface of the adsorbent is saturated with metal after a longer time.

Based on an evaluation of Fig. 8, adsorbed and total adsorbed cadmium(II) quantities are determined and removal percentages of cadmium(II) by dried *C. crispata* flocs with respect to flow volume at different inlet metal ion concentrations are shown in Table 2. As seen from Table 2, higher removal percentages of cadmium(II) ions are obtained at the beginning of the biosorption. Maximum cadmium removal by dried alga at all inlet concentrations is

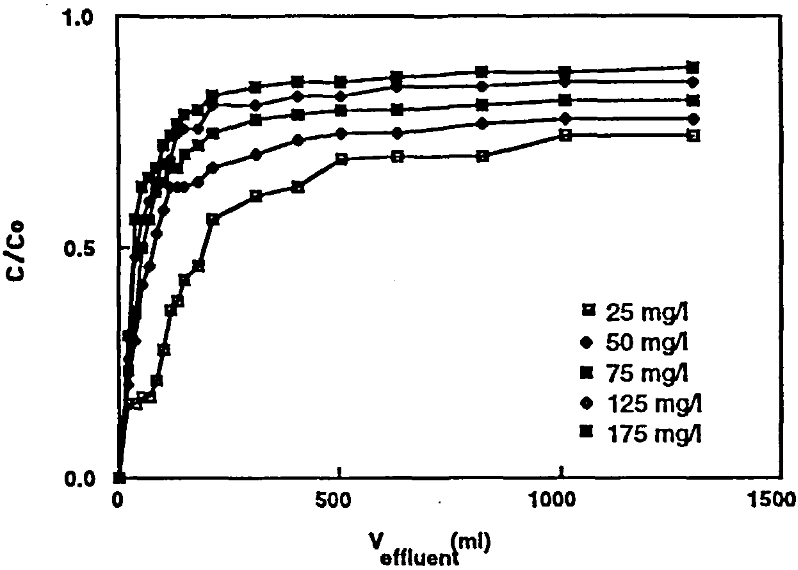

FIG. 7 The adsorption breakthrough curves obtained at different inlet cadmium(II) ion concentrations at 3.2 mL/min of flow rate. Temperature: 25°C. pH of inlet cadmium(II) solution: 5.0.

obtained after approximately 23 mL of flow. This is because diffusion limitations are not important in sorption with respect to initial cadmium(II) concentration. Metal uptake by dried alga is subject to saturation limits, especially at higher inlet metal ion concentrations and higher flow rates. For example, when the column is loaded with a flow containing 25 or 175.0 mg/L of cadmium(II) at a flow rate of 3.2 mL/min, column activity is invariant with the volume of flow passing from 71 to 23 mL, representing loading times of 22.2 and 7.2 minutes, respectively, at which point the experiments had to be terminated. After this point, packed-bed columns need to have continuous adsorption–desorption circuits or marginal metal removal yields would have been obtained. If processing time is important, a recycle or linked-column series may be also employed in order to obtain higher metal removal yields at higher flow rates and higher inlet metal ion concentrations. If the quantity of algae per unit volume of the column is increased (decrease of the void fraction of the column), metal uptake will also be higher for concentrated metal solutions because of the increase in the active adsorption sites.

Maximum specific metal uptakes of the algal flocs obtained at different inlet cadmium(II) concentrations at a 3.2 mL/min flow rate are compared in Table 3. Although the total cadmium(II) removal percentages obtained at

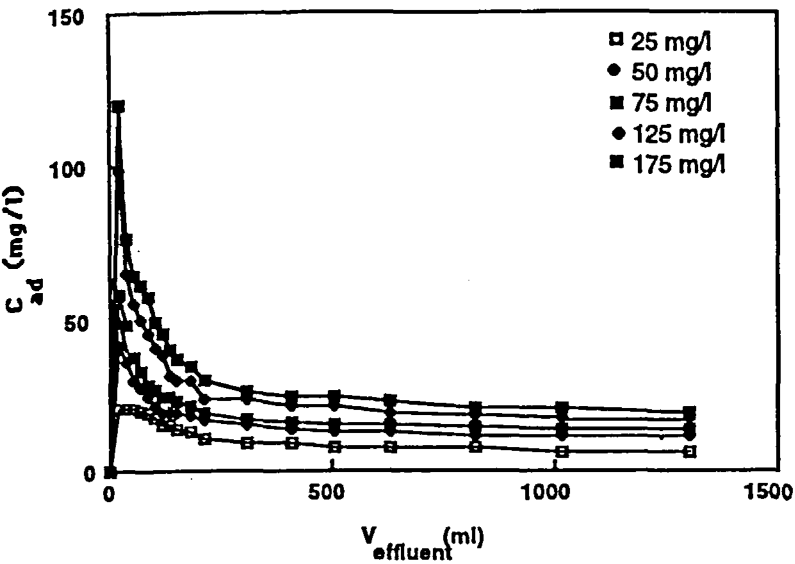

FIG. 8 The change of adsorbed cadmium(II) concentration with effluent volume at different inlet cadmium(II) concentrations at 3.2 mL/min of the flow rate. Temperature: 25°C. pH of inlet cadmium(II) solution: 5.0.

TABLE 2
Effect of Flow Volume on Cadmium Removal at Various Inlet Metal Ion Concentrations at a Flow Rate of 3.2 mL/min (*T* = 25°C; pH of inlet cadmium(II) solution = 5.0)

Flow volume (mL)	Cd(II) removal %				
	At 25 mg/L	At 50 mg/L	At 75 mg/L	At 125 mg/L	At 175 mg/L
23	83.8	79.3	77.0	77.6	68.9
71	83.0	66.7	59.9	56.3	48.5
103	81.0	59.8	52.9	49.5	43.3
183	70.8	49.8	43.5	39.8	34.2
311	56.8	42.9	35.9	31.2	27.2
503	47.6	36.6	30.0	25.7	22.3
823	41.8	31.5	25.9	21.6	18.4
1015	39.6	29.7	24.4	20.1	17.2
1282	38.0	28.0	23.1	18.9	15.8

TABLE 3
Effect of Inlet Cadmium(II) Concentration on the Maximum Specific Cadmium(II) Uptake of the Alga at a Flow Rate of 3.2 mL/min (T = 25°C; pH of inlet cadmium(II) solution = 5.0)

Inlet cadmium(II) concentration (mg/L)	q_{total} (mg/g)
25	3.4
50	4.9
75	6.1
125	8.3
175	9.7

lower inlet cadmium(II) concentrations are higher than those of higher inlet cadmium(II) concentrations, it is seen that the maximum specific cadmium(II) uptake of the alga increases from 3.4 to 9.7 mg/g with an increase of inlet cadmium(II) concentration from 25 to 175 ppm owing to the large difference in concentration between the sorbent surface and the metal ion solution. It is suggested that competition among metal ions for the binding sites of algal flocs results in a higher uptake of cadmium(II) ions.

The accumulation of cadmium(II) by flocs of dried alga were compared to the results of Duncan et al. (10) where granular yeast was used as a biosorbent in a packed-bed column. As noted by Duncan et al., their biomass had a 10 mg/g capacity at a 100 mg/L inlet cadmium(II) concentration and a 0.4 mL/min flow rate. Half a gram of freeze-dried *Scenedosmus quadricauda*, a marine algae, was immobilized in a crosslinked copolymer of ethyl acrylide–ethylene glycol dimethacrylate and used for cadmium(II) removal by Harris and Ramelow (6) in a packed bed. They found that at a flow rate of 0.7 mL/min and a feed cadmium(II) concentration of 4 mg/L, the maximum specific cadmium(II) uptake of the green alga was 3.9 mg/g. Wehrheim and Wettern (8) reported that the uptake value of the whole cells of *Chlorella fusca*, a green alga, was 3.4 mg/g dry biomass. Comparison of the accumulation capacities of *C. crispata* flocs and the other free or immobilized biomasses showed that *C. crispata* flocs reached similar or even higher uptakes than the other biosorbents.

CONCLUSIONS

Although higher adsorption rates and capacities for the selective or simultaneous removal of metal ions can be carried out in a batch reactor by adjusting the pH of the wastewater and diluting the wastewater to lower levels of metal ion concentrations, the direct use of algal flocs, without immobilization, in continuous columns for the removal of toxic metal ions from waste streams appears much more promising. Biosorption of heavy metal ions by dried algal flocs in a packed-bed column, where the biomass behaves as an ion exchanger, is a technically efficient and economically feasible technology for removing metal ions from solution. This process only slightly decreases the metal biosorption properties of the biomass.

The breakthrough curves for column biosorption of cadmium(II) from dilute solutions using algal flocs of *C. crispata* have been measured at 25°C. The dependence of the shapes of the breakthrough curves on experimental parameters can be explained by the mutual effects of the biosorption capacity and biosorption rate. Column studies showed that the adsorption of cadmium(II) ions is dependent on the flow rate, inlet metal ion concentration, and time, and that these parameters directly affect the saturated capacity. The saturated capacity of the algal bed is greater under conditions of a lower concentration of metal ions and a lower flow rate. Adjusting such operating characteristics of the packed column as flow rate, metal ion concentration, algal biomass, and particle size can also be employed to obtain higher cadmium(II) removal yields.

This study has attempted, at least in part, to more clearly define the cadmium(II) bioaccumulation potential of the dired algal flocs. We believe that application of biosorption by green alga for the purification of wastewater is suitable for large-scale column exploitation.

NOTATION

C_0	Inlet (feed) cadmium(II) concentration (mg/L)
C	cadmium(II) concentration in the effluent (mg/L)
C_{ad}	$(= C_0 - C)$. Adsorbed cadmium(II) concentration by the alga in the column (mg/L)
q	adsorbed cadmium(II) quantity by the alga in any time in the batch system (mg/g)
q_{eq}	adsorbed cadmium(II) quantity by the alga in the batch-stirred reactor at equilibrium (mg/g)
q_{total}	maximum specific cadmium(II) uptake of dried *Cladophora crispata* flocs in the column (mg/g)
Q	flow rate (mL/min)
t	flow time (min)
t_{total}	total flow time (min)
V_{eff}	effluent volume (mL)
X	amount of dried alga in the column (g)

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