Biosorption of Copper by the Immobilized Biomass of Marine Brown Algae(Phaeophyta) *Hizikia fusiformis*

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Abstract

It was investigated the biosorption performances of copper by the immobilized biomass of nonliving marine brown algae *H. fusiformis* by each of the Ca-alginate method(Ca-ALG), Ba-alginate method(Ba-ALG), polyethylene glycol method(PEG), and carrageenan method (CARR). The copper removal performance increased but the copper uptake decreased as the biomass amount was increased. However, the copper uptake by the immobilized biomass increased with increasing initial copper concentration. The copper uptake by the immobilized biomass of the immobilization method decreased in the following sequence : Ca-ALG〉Ba-ALG〉PEG〉CARR among the immobilization methods. The copper uptake by the immobilized biomass followed the Langmuir isotherm better than the Freundlich isotherm.

Key words : marine brown algae, immobilization, H. fusiformis, Ca-alginate method, Ba-alginate method, polyethylene glycol method, carrageenan method

Introduction

Different and unusual metal-binding properties of various algal species and cell preparations have been known[1,2]. The reason for the variations in metal binding among algae depends on the fact that the various algal divisions and genera have different cell-wall compositions[3]. The different cell-wall compositions result in different metal-adsorptive moieties being present on or in the cell walls[4].

Free algal cells are not suited for use as a column packing since the cells tend to clump together and excessive hydrostatic pressures are required in order to generate suitable flow rates. Furthermore, since the algal cells inherently are fragile, high pressures may cause a disintegration of the free biomass. The fragility problem has been alleviated by the use of algae immobilized in a suitable porous matrix. Polyacrylamide, calcium alginate, and silica are among the porous matrices that have been used to immobilize nonviable algae and to permit their use in packed columns for metal ion recovery[5,6]

A number of methods have been employed in order to immobilize nonviable algal cells, including the adsorption or covalent binding to an insoluble matrix[2,7]. One of the matrices that has been used for metal recovery by both viable and nonviable algae has been the entrapment of algal cells in matrix of insoluble calcium alginate. Kennehy et al.[8] and Nakajima et al.[6] observed that the alginate beads have been shown excellent resistance to hydrostatic pressures. and that the polyacryla-

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mide matrix improved the stability of the algal cells to mechanical degradation. Darnall et al.[9] used polyacrylamide-immobilized C. *vulgaris* both to biosorb and to selectively recover a number of heavy metal ions. Even though algae immobilized in a polyacrylamide gels can accomplish to broad a distribution of pore sizes, the use of polyacrylamide gels for commercial applications probably will be minimal[8]. One immobilized algae preparation that is currently being used commercially for metal recovery is AlgaSORB®. This proprietary material contains algal cells immobilized in a silica matrix[10]. The silica-immobilized algae are suitable for use in either batch or column procedures for industrial applications.

In this study, the biomass of nonliving *H. fusiformis* which are known as predominant heavy metal biosorbent materials among easily available and preferably abundant marine brown algae in the earlier studies[11,12], were immobilized by calcium alginate method(Ca-ALG), barium alginate method(Ba-ALG), polyethylene glycol method(PEG), and carrageenan method(CARR) and the biosorption performances of copper were investigated in the batch-stirred reactor for each of the immobilized biomass by the immobilization method.

MATERIALS AND METHODS

Materals

The marine brown algae(Phaeophyta) *Hizikia fusiformis* harvested in the sea near Cheju Island were employed as biomass. These materials were washed fully with water, stored in refrigerator for one day, and removed water fully by the freezing dryer, and was ground and sieved(70/100mesh), and stored in desiccator.

Analytical grades of $Cu(NO_3)_2 \cdot 3H_2O$, sodium alginate, H_3BO_3, NaOH(Yakuri Pure Chem. Co., Japan), κ-carrageenan, polyethylene glycol(PEG) 4000, $K_2S_2O_8$ (Junsei Chem. Co. Ltd., Japan), $CaCl_2 \cdot 2H_2O$, N,N,N', N'-tetramethyleneethylenediamine(Sigma Chem. Co., USA), Na_2CO_3(Hayashi Pure Chem. Co., Japan), and

HNO_3(Hori Pharm. Co., Japan) were used and the solutions were prepared with deionized distilled water. The stock solution of copper ion were prepared as 1000 mg/L and diluted as desired concentrations with water.

Immobilization Method

Ca-ALG and Ba-ALG : Fixed amount of biomass was added to the 250mL Erlenmeyer flask containing 100 mL of 1% sodium alginate solution, stirred throughly with magnetic stirrer, and the mixture was dropped to the solution containing 1% $CaCl_2$ or 1% $BaCl_2$ using a syringe. The bead of spherical immobilized biomass formed left for one day to increase its stability, and then washed with 2% KCl and then water.

PEG : Fixed amount of biomass was added to the 250mL Erlenmeyer flask containing 100mL of 40% PEG solution, stirred throughly with magnetic stirrer. To this solution, 1mL of 3% $K_2S_2O_8$ and N,N,N',N'-tetramethyleneethylenediamine were added, stirred, and then the mixture was dropped to the solution containing 1% $CaCl_2$ using a syringe.

CARR : Fixed amount of biomass was added to the 250mL Erlenmeyer flask containing 100mL of 4% carrageenan solution in a constant water bath controlled at 60℃, stirred throughly with magnetic stirrer, and the mixture was dropped to the solution containing 1% $CaCl_2$ using a syringe.

Methods

The immobilized biomass with fixed amount and copper concentration with fixed concentration were placed in a 1-liter flask container. The solution was stirred with a magnetic stirrer. During the experiment, 2mL of sample in the solution was taken at a given time interval and used for the measuring metal ion concentrations in the solution. It was investigated the effects of initial copper concentration, the biomass amount, pH, and biosorption isotherm.

The metal uptake for batch reactor experiments was

determined as follows :

$$q = 1000V(C_o - C_e)/M$$

where q is the metal uptake(the amount of metal adsorbed per unit weight of biomass, mg/g), V is the volume of the solution in the contact batch flask(mL), C_o is the initial concentration of metal in solution(mg/L), C_e is and equilibrium concentration of metal in solution (mg/L), and M is the mass of biomass(g). M is calculated by the sum of the amount of biomass and sodium alginate or carrageenan contained in immobilizing carrier, because the latter is responsible for metal biosorption as a component of the algae[13].

Water bath was used to maintain the constant temperature in the system. All experimental vessels were pyrex glass and all glasswares were leached in a 14% nitric acid and washed distilled water prior to use. The concentrations of copper in solution were determined by an atomic absorption spectrophometer(GBC 904AA) at 324.7nm.

RESULTS AND DISCUSSION

Effects of initial copper concentration on copper removal were investigated. Each of the immobilized biomass containing 0.6g of biomass for Ca-ALG, Ba-ALG, and PEG or the immobilized biomass containing 1.5g of biomass for CARR was added to the solution containing 25, 50, 100, 200, 300, and 500mg/L. As shown in Fig. 1~2 represented as the solution concentration with time, all the immobilized biomass attained the final equilibrium copper uptake after 2hr. This result shows much more time is required, compared to the native biomass[11,12]. This result indicates that the immobilization agent can give higher mass transfer resistance than the native biomass.

Fig. 3 shows the effect of initial copper concentration on the biosorption performance. As shown in figure 3, the experimental result shows the better copper removal as
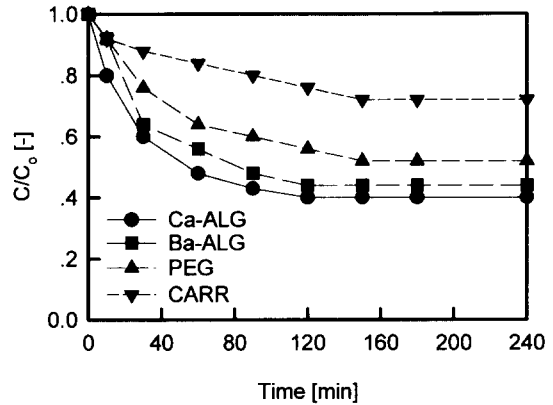

Fig. 1. Concentration profile for copper biosorption by the immobilized biomass of H. fusiformis (initial copper concentration : 25mg/L, biomass amount : 0.6g for Ca-ALG, Ba-ALG, and PEG, 1.5g for CARR).
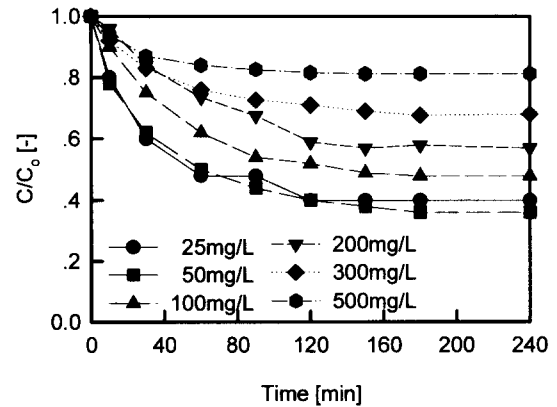

Fig. 2. Concentration profile for copper biosorption by the immobilized biomass of H. fusiformis(immobilization method : Ca-ALG, biomass : 0.6g).

initial copper concentration is increased. This feature causes the increase of driving force of mass transfer with increasing the initial copper concentration. Fig. 4 shows the comparative copper uptake among each of the immobilization methods. As shown in Fig. 4, the maximum copper uptake is about 50mg/g, 45mg/g,

and 17mg/g for Ca-ALG, Ba-ALG, PEG, and CARR, res-pectively, when 500mgCu/L of initial concentration was used. For the case of Ca-ALG, the highest capacity
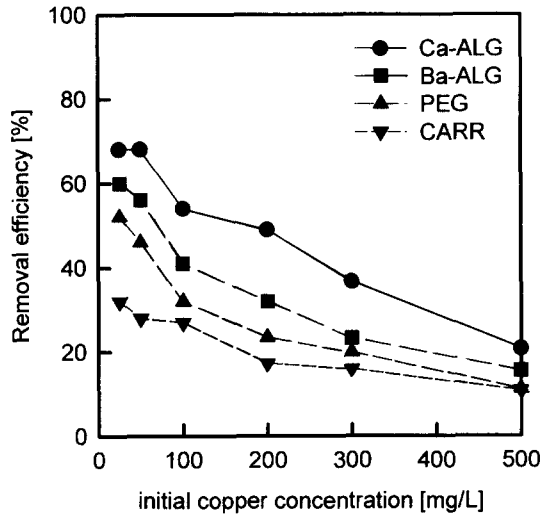

Fig. 3. Effect of initial solution concentration on remo-val efficiency for copper biosorption by the im-mobilized biomass of *H. fusiformis*(biomass amount : 0.6g for Ca-ALG, Ba-ALG, and PEG, 1. 5g for CARR).
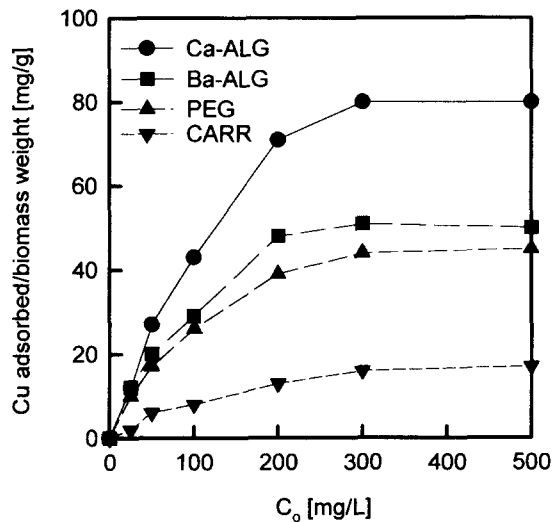

Fig. 4. Effect of copper uptake on initial copper concen-tration for biosorption by the immobilized bio-mass of *H. fusiformis*.

about 50mg/g is shown. It is considered that Ca-ALG is the most efficient among the immobilization methods used in this study for removing copper to the practical water and wastewater treatment.

The effect of biomass amount on the metal removal for each of the immobilization methods used was examined. Each of the immobilized biomass containing 0.15, 0.3, and 0.6g of biomass for Ca-ALG, Ba-ALG, and PEG, 0.375, 0.75, and 1.5g of biomass for CARR was added to 500 mL solution containing 25mg Cu/L at 20℃.

Fig. 5 shows the comparison of copper removal perfo-rmance with the biomass amount for each of the immo-bilization methods. The copper removal performance in-creased as the biomass amount added was increased. This result means that the increase of biomass amount contributes to the increase of surface available for bio-sorption. This figure also shows that different copper re-moval efficiency was obtained according to the immobi-lization method used. The copper removal efficiency was in the following sequence : Ca-ALG⟩Ba-ALG⟩PEG⟩CARR among the immobilization methods.
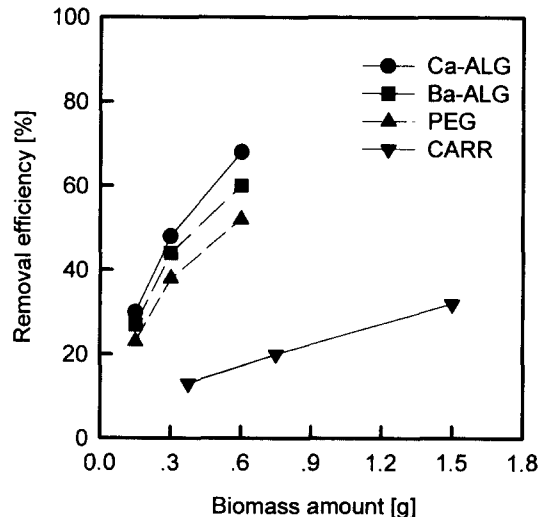

Fig. 5. Comparison of copper removal efficiency with the biomass amount of *H. fusiformis* for each of the immobilized methods(initial copper concent-ration : 25mg/L).

Fig. 6 illustrates the copper uptake(q), i.e., the amount of copper adsorbed per unit weight of dry biomass, at constant loading on the variation of biomass amount. These figures show that the equilibrium copper uptake decreases as the amount of biomass added is increased.
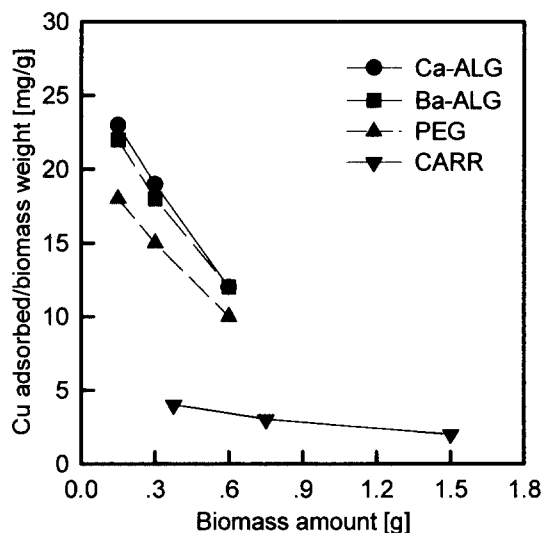

Fig. 6. Effect of biomass on the copper uptake by the immobilized biomass of *H. fusiformis* for each immobilization method(initial copper concentration : 25mg/L).

Fig. 7 shows the effect of pH on the copper uptake by the immobilized biomass in a solution containing 25 mg/L. A constant pH for batch equilibrium biosorption experiment was maintained at pH 2, 3, 4, 5, and 6, respectively. Each pH was controlled with $Na_2CO_3(10^{-4}$ N) and HNO_3 or NaOH. As shown in figure, a low pH of 2 resulted in a markedly lower copper uptake and a higher uptake was obtained in the range of pH 4.0-6.0. The reason for this is to be expected due to a higher hydrogen ion concentration, which in turn prevents the immobilized biomass-copper binding the more. The experimental result demonstrates that pH of the solution is a very important factor in copper uptake by the im-

mobilized biomass. It is considered that copper ions and protons compete for the same binding sites, because the pH dependence of the copper-cation binding generally occurs when the active metal-binding sites can also bind protons.
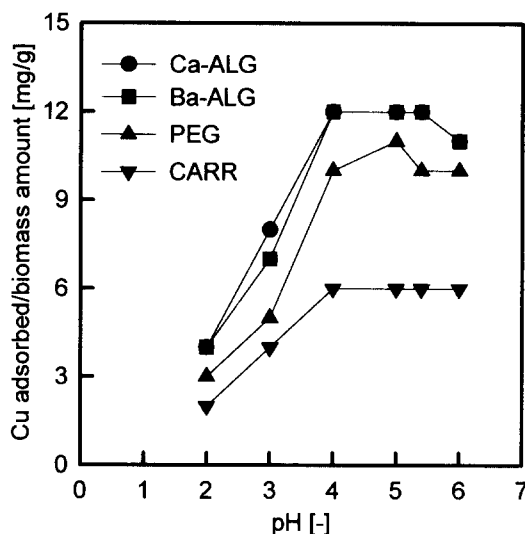

Fig. 7. Effect of pH on the copper uptake by the immobilized biomass of *H. fusiformis*(initial copper concentration : 25mg/L, biomass amount : 0.6g for Ca-ALG, Ba-ALG, and PEG, 1.5g for CARR).

Adsorption from aqueous solution at equilibrium is usually correlated by one of the two following adsorption isotherm relationships. Eq. (1) is the Langmuir isotherm and Eq. (2) is the Freundlich is otherm.

$$q = q_{max} \ b \ C_e(1 + b \ Ce) \qquad (1)$$

$$q = K \ C_e^{1/n} \qquad (2)$$

Fig. 8 shows copper isotherms for the immobilized biomass by each of the Ca-ALG, Ba-ALG, PEG, and CARR, in the range of initial metal concentration, 25~500mg/L. The data demonstrated that immobilized biomass by Ca-ALG had relatively high biosorption capacity for copper. However, the copper biosorption capacity by CARR were very low, compared to those by the

other immobilization methods. The Langmuir and Freundlich constant values obtained by the non-linear regression method for the immobilized biomass sorption sys-
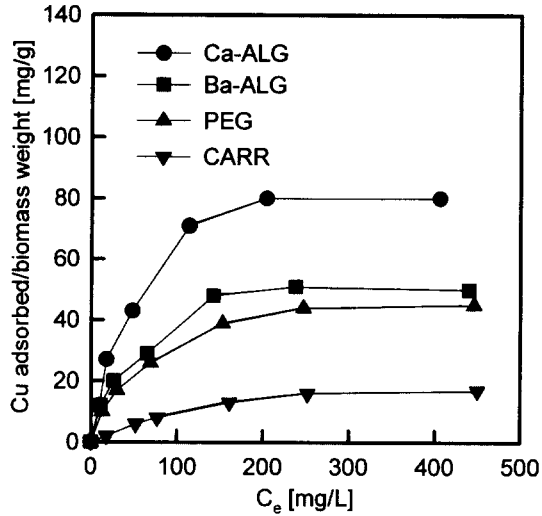

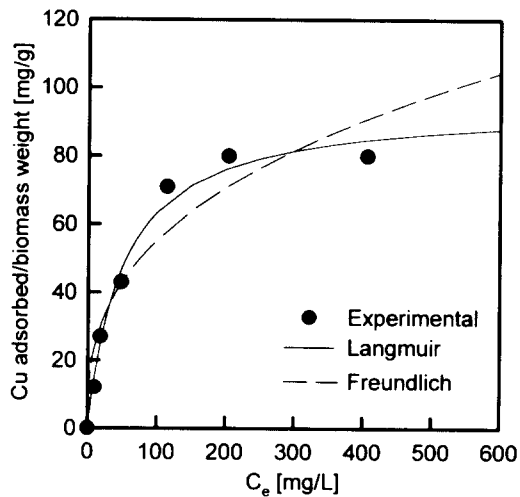Fig. 8. Copper biosorption isotherms for the immobilized biomass of *H. fusiformis*.

Fig. 9. Comparison of calculated values by the Langmuir and Freundlich model with experimental data of copper biosorption isotherms for the immobilized biomass of *H. fusiformis* by Ca-ALG.

Table 1. Langmuir and Freundlich isotherm parameters for copper uptake by the immobilized biomass.

Immobilization methods	Langmuir			Freundlich		
	q_{max}	b	r^2	K	n	r^2
H. fusiformis						
Ca-ALG	104	0.020	0.964	5.37	2.000	0.886
Ba-ALG	53	0.026	0.985	5.01	2.418	0.937
PEG	49	0.019	0.994	3.63	2.252	0.955
CARR	20	0.010	0.998	3.11	1.442	0.951

tems are summarized in Table 1. As shown in Table 1, the maximum adsorption capacity was about 104, 53, 49, and 20mg/g for the case of Ca-ALG, Ba-ALG, PEG, and CARR, respectively. Ca-ALG method showed an excellent adsorption capacity compared to the other methods.

Fig. 9 illustrates the comparison of calculated values by the Freundlich and Langmuir correlation with the experimental data of copper biosorption for the immobilized biomass of *H. fusiformis* by Ca-ALG. The experimental results fits well with Langmuir isotherm.

CONCLUSIONS

The biomass of nonliving *H. fusiformis* which are known as predominant heavy metal biosorbent materials among easily available and preferably abundant marine brown algae, were immobilized by calcium alginate method(Ca-ALG), barium alginate method(Ba-ALG), polyethylene glycol method(PEG), and carrageenan method(CARR) and the biosorption performances of copper were investigated in the batch-stirred reactor for each of the immobilized biomass by the immobilization method.

The experimental result showed that each of the immobilized biomass-Cu^{2+} system attained the final equilibrium plateau after 2hr. The copper removal performance increased but the copper uptake decreased as the

biomass amount was increased. However, the copper uptake by the immobilized biomass increased with increasing initial copper concentration. The copper uptake by the immobilized biomass of the immobilization method decreased in the following sequence : Ca-ALG>Ba-ALG>PEG>CARR among the immobilization methods.

Equilibrium parameters based on the Langmuir and the Freundlich isotherm were determined. The experimental data of the copper biosorption by the immobilized biomass are fitted to the Langmuir isotherm better than the Freundlich isotherm.

NOMENCLATURES

b   Langmuir constant(related to energy of adsorption)

C   concentration of copper in the solution [mg/L]

C_e equilibrium concentration of copper in the solution [mg/L]

C_o initial concentration of copper in the solution [mg/L]

K   Freundlich constant(adsorbent capacity)

n   Freundlich constant(adsorption intensity) [−]

q   copper uptake(amount of copper adsorbed per unit weight of adsorbent) [mg/g]

q_{max} maximum copper uptake

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초록 : 해양 갈조류인 톳의 고정화된 생물질에 의한 구리의 생흡착

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제주도 근해에서 다량 채취되고 있는 해양갈조류인 톳의 살아있지 않은 건조한 생물질을 Ca-alginate법 (Ca-ALG), Ba-alginate법(Ba-ALG), polyethylene glycol법(PEG), κ-carrageenan법(CARR) 등으로 고정화시켜 고정화방법에 따른 Cu의 흡착능을 검토하였다.

생물질의 첨가량이 증가함에 따라서 Cu의 제거율은 증가하였으나 흡착량은 감소하였다. 그러나 초기 농도가 증가함에 따라서는 Cu의 흡착량이 증가하였다. 고정화법에 따른 Cu의 흡착량은 Ca-ALG〉Ba-ALG〉 PEG〉CARR의 순으로 감소하였다. 고정화된 생물질에 의한 Cu의 흡착은 Freundlich 흡착 등온식 보다는 Langmuir 흡착 등온식을 잘 따름을 알 수 있었다.