Biosorption of Metal Ions by Seaweed Alginate, Polyguluronate, and Polymannuronate

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Based on $P_{1/2}$ values, relative affinities of alginate, polyguluronate, and polymannuronate for metal ions are, in order, as follows; 1) seaweed alginate: $Cu^{2^+} > Cd^{2^+} > Pb^{2^+} > Fe^{3^+} >> Zn^{2^+} > Sr^{2^+} > Ca^{2^+} > Co^{2^+} >> Cr^{6^+} > Mn^{2^+} >> Hg^{2^+}, Mg^{2^+}, Rb^+, 2)$ polyguluronate: $Cd^{2^+} > Cu^{2^+} > Pb^{2^+} > Fe^{3^+} >> Ca^{2^+} > Ca^{2^+} > Sr^{2^+}, Zn^{2^+}, Co^{2^+} >> Mn^{2^+} > Cr^{6^+} >> Hg^{2^+}, Mg^{2^+}, Rb^+, and 3)$ polymannuronate: Cd^{2^+} , $Cu^{2^+} > Fe^{3^+} > Pb^{2^+} > Ca^{2^+} > Zn^{2^+} > Sr^{2^+} > Co^{2^+} > Cr^{6^+} >> Mn^{2^+} >> Hg^{2^+}, Mg^{2^+}, Rb^+.$ Amounts of the metal ions, Cd^{2^+} , Cu^{2^+} , Fe^{3^+} , Pb^{2^+} , and Zn^{2^+} , bound to 1 g of seaweed alginate, were measured as 363.5 ± 45.0 , 226.3 ± 9.2 , $1,299.4\pm81.3$, 500.7 ± 27.7 , and 165.9 ± 11.4 mg, respectively. Amounts of the metal ions, Cd^{2^+} , Cu^{2^+} , Fe^{3^+} , Pb^{2^+} , and Zn^{2^+} , bound to 1g of polyguluronate, were 354.5 ± 26.5 , 177.6 ± 8.7 , $1,288.6\pm60.1$, 424.0 ± 7.4 , and 140.2 ± 28.5 mg, respectively, whereas those bound to 1 g of polymannuronate were 329.0 ± 10.3 , 206.9 ± 1.9 , $1,635.6\pm11.1$, 419.8 ± 12.6 , and 251.0 ± 49.1 mg, respectively. Due to its higher solubility than alginate and higher affinity for metal ions than polyguluronate, polymannuronate can be used for bioremediation or biosorption of toxic and/or noble metal ions.

Key words: Alginate, polyguluronate, polymannuronate, biosorption, metal ions

Introduction

Seaweed alginate is a linear copolymer with homopolymeric blocks of $(1\rightarrow 4)$ -linked β -D-mannuronate and its C-5 epimer, α-L-guluronate [12,17,21]. The molecular weight (MW) and molar ratio of D-mannuronate to L-guluronate (M/G ratio) in alginate are dependent on algal species and harvest time, as well as the location of the polymer in plants [7]. The affinity of seaweed alginate for divalent ions has been studied since the discovery that the addition of calcium ions to a solution of seaweed alginate caused gel formation and precipitation. The minimum amount of divalent metal ions that initiate precipitation of alginate isolated from L. Hyperborea stipe and A. nodosum increased in the following order: Pb²⁺, Cu²⁺, Ca²⁺, Ni²⁺, Co²⁺, Zn²⁺, and Mn²⁺ [8]. But smaller amounts of Pb2+ and Ca2+ were required to precipitate alginate from L. hyoerborea stripe (rich in guluronate residues) than that which was required to precipitate alginate from A. modosum (rich in mannuronate residues).

Alginate is used in foods and generally applied in many fields of industries because of its unique colloidal ability to

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thicken, stabilize, emulsify, suspend, and produce gel with high safety [2]. The potential of biopolymers as safe and cost-effective materials to remove heavy metals from dilute aqueous solutions has long been recognized [10,26]. And also, seaweed alginate has been one of the most extensively investigated biopolymers for binding heavy metals [11,23,24]. Recently, biosorption of heavy metals has received a lot of concerns about application of hazardous waste treatment. We are interested in the role of polyguluronate and polymannuronate, which have a lower molecular weight than alginate, to remove of heavy metals as another biosorption material. The metal biosorption characteristics of polymannuronate and polymannuronate were investigated in this study and compared with that of alginate.

Materials and Methods

Seaweed alginate, polyguluronate and polymannuronate

Alginate from brown seaweed *Macrocystis pyrifera* was purchased from Sigma-Aldrich Chemicals (St. Louis MO). The molecular weight (MW) of alginate was about 1,300 kDa. Polyguluronate and polymannuronate were kindly donated by Korea Bio-solution Co., Ltd (Busan, Korea). The molec-

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ular weights of polyguluronate and polymannuronate, isolated from partially hydrolyzed alginate, ranged from 30 to 50 kDa. The molar percentages of guluronate in polyguluronate and mannuronate in polymannuronate were more than 98% by HPLC analysis, respectively [13].

Precipitation of alginate, polyguluronate, and polymannuronate by metal ions

Metal salts were dissolved in deionized water at concentrations of 0 to 100 or 500 mM with 4 intermediate concentrations. Metal salts used in this study were CaCl₂, CdCl₂, CoCl₂, CrCl₆, CuCl₂, FeCl₃, HgCl₂, MgCl₂, MnCl₂, PbCl₂, RbCl₂, SrCl₂, and ZnCl₂. All metal salts were purchased from Sigma-Aldrich Chemical Co. (St. Louis, MO, USA). The concentrations of alginate, polyguluronate, and polymannuronate were adjusted to 500 µg/ml. Four volumes of alginate, polyguluronate, and polymannuronate were mixed with one volume of each metal salt solution. The mixture was incubated for 2 hr at room temperature and centrifuged at 4,500× g for 20 min to precipitate the mixture of metal ion and alginate, polyguluronate, or polymannuronate. Unbound metal ions in the supernatant were removed by NAPTM 25 columns (Amersham Pharmacia Biotech AB, Sweden). The concentrations of polymers in each supernatant were measured by the phenol-sulfuric acid method [3]. The concentration of precipitated polymers was calculated and its value was used to determine the relative affinity of alginate, polyguluronate, and polymannuronate for metal ions.

Biosorption of metal ions by alginate, polyguluronate, and polymannuronate

Metal salts of CdCl₂, CuCl₂, FeCl₃, PbCl₂, and ZnCl₂ were dissolved in deionized water at concentrations ranging from 0 to 500 or 4,000 μ g/ml with 6 intermediate concentrations. The concentrations of alginate, polyguluronate, and polymannuronate were fixed to 500 μ g/ml. Four volumes of alginate, polyguluronate, and polymannuronate were also mixed with one volume of metal salt solution. The mixture was incubated for 2 hr at room temperature and centrifuged at 4,500× g for 20 min. The concentration of metal ions in each supernatant was measured by atomic absorption spectrophotometer (Perkin Elmer Analyst 300).

Analytic methods

The molecular weights of polyguluronate and poly-

mannuronate were measured by gel chromatography equipped with Sepharose CL-6B and Sepharose CL-4B [27]. The size of the column was 97.6×1.2 i. d. cm. The calibration curve for determining total sugar was plotted by the pullulans (Showa Denko, Japan). Deionized water was used as a mobile phase at a flow rate of 1.0 ml/min. The sample concentration and injection volume were 5.0 mg/ml and 100 μ l, respectively. All of the sample solutions were filtered through 0.45 μ m-pore-size filters (Adbentec MFS, Inc., Japan) before injection.

The molar compositions of polyguluronate and polymannuronate were measured with high pressure liquid chromatography (HPLC) using a Spectra Physics P2000 system controller equipped with a Whatman partisil 10-SAX anion exchange column (250×4.6 mm i. d.) [4]. The L-gulucuronic acid lactone and D-mannuronic acid lactone used for standard materials were purchased from Sigma-Aldrich Chemical Co. (St. Louis, MO, USA). The elution buffer was 0.02 M KH₂PO₄ (pH 4.6) solution containing 5% methanol.

Results and Discussion

Precipitation of alginate, polyguluronate, and polymannuronate by metal ions

The relative abilities of metal ions to precipitate alginate, polyguluronate and polymannuronate were examined. Metals used in this study were CaCl₂, CdCl₂, CoCl₂, CrCl₆, CuCl₂, FeCl₃, HgCl₂, MgCl₂, MnCl₂, PbCl₂, RbCl, SrCl₂, and ZnCl₂. About 80% of alginate, polyguluronate, and polymannuronate in the mixture were precipitated by 25 mM CaCl₂ and more than 60% of these biopolymers were precipitated by 10 mM CaCl₂, as shown in Fig. 1. The relative affinity of calcium ions for polymannuronate was higher than those for alginate and polyguluronate. More than 80% of alginate, polyguluronate, and polymannuronate in the mixture were precipitated by 25 mM CdCl₂. Cadmium ions showed relatively high affinity for alginate, polyguluronate, and polymannuronate as shown in Fig. 2. About 90% of alginate and polymannuronate in the mixture were precipitated by 20 mM CoCl₂, whereas less than 80% polyguluronate was precipitated by the same concentration of CoCl₂ as shown in Fig. 3. The relative affinity of cobalt ions for polyguluronate was lower than those for alginate and polymannuronate.

More than 70% of polymannuronate in the mixture was precipitated by 40 mM CrCl₂, whereas about 60% of alginate

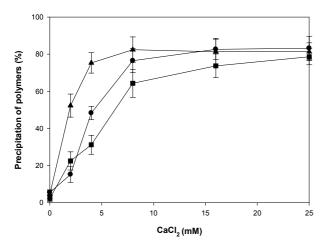


Fig. 1. Precipitation of alginate, polyguluronate, and polymannuronate by CaCl₂ (●; alginate, polyguluronate, and ▲; polymannuronate).

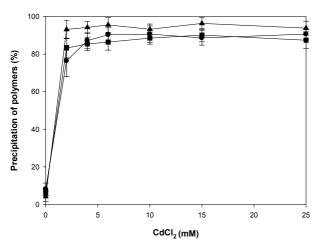


Fig. 2. Precipitation of alginate, polyguluronate, and polymannuronate by CdCl₂ (●; alginate, polyguluronate, and ♠; polymannuronate).

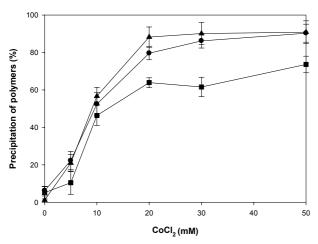


Fig. 3. Precipitation of alginate, polyguluronate, and polymannuronate by CoCl₂ (●; alginate, □; polyguluronate, and ▲; polymannuronate).

and 40 % of polyguluronate in the mixture were precipitated by the same concentration of CrCl₂, as shown in Fig. 4. The relative affinity of chromium ions for polyguluronate was also lower than those for alginate and polymannuronate. About 90% of alginate and polymannuronate and 85% of polyguluronate in the mixture were precipitated by less than 5 mM CuCl₂, as shown Fig. 5. Copper ions as well as cadmium ions showed a relatively high affinity for alginate, polyguluronate, and polymannuronate. More than 90% of alginate and polymannuronate in the mixture were precipitated by 5 mM FeCl₃, whereas about 75% of polyguluronate was precipitated by the same concentration of FeCl₃, as shown in Fig. 6. The relative affinity of ferric ion for polyguluronate at low concentration was lower than those for alginate and polymannuronate.

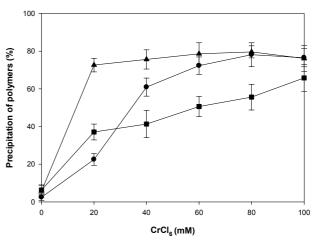


Fig. 4. Precipitation of alginate, polyguluronate, and polymannuronate by CrCl₆ (●; alginate, ■; polyguluronate, and ▲; polymannuronate).

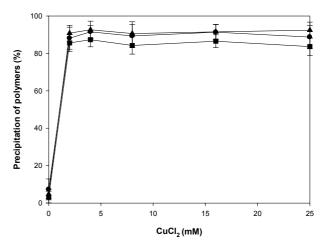


Fig. 5. Precipitation of alginate, polyguluronate, and polymannuronate by CuCl₂ (●; alginate, ■; polyguluronate, and ▲; polymannuronate).

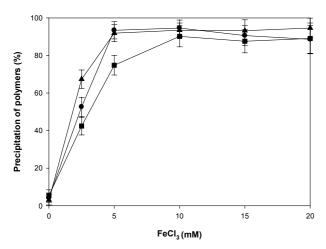


Fig. 6. Precipitation of alginate, polyguluronate, and polymannuronate by FeCl₃ (●; alginate, ■; polyguluronate, and ▲; polymannuronate).

About 80% of polymannuronate, 75% of alginate, and 60% of polyguluronate in the mixture were precipitated by 40 mM MnCl₂, as shown in Fig. 7. The affinity of manganese ions for polyguluronate was higher than those for alginate and polymannuronate. About 80% of alginate, polyguluronate and polymannuronate in the mixture were precipitated by 5 mM PbCl₂, as shown in Fig. 8. Lead ions, as well cadmium and copper ions, showed relatively high affinity for alginate, polyguluronate, and polymannuronate. Similar amounts of alginate, polyguluronate, and polymannuronate were precipitated by the same concentration of lead ions.

More than 80% of alginate, polyguluronate, and polymannuronate in the mixture were precipitated by 10 mM

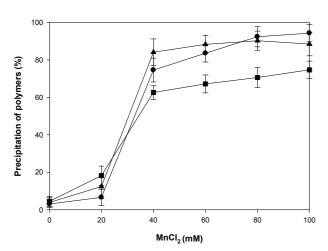


Fig. 7. Precipitation of alginate, polyguluronate, and polymannuronate by MnCl₂ (●; alginate, □; polyguluronate, and ▲; polymannuronate).

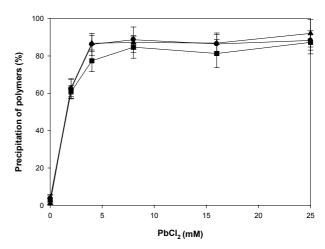


Fig. 8. Precipitation of alginate, polyguluronate, and polymannuronate by PbCl₂ (●; alginate, ■; polyguluronate, and ▲; polymannuronate).

SrCl₂, but the relative ability of strontium ions to precipitate polyguluronate was lower than alginate and polymannuronate at a low concentration of SrCl₂, as shown in Fig. 9. More than 90% of alginate and polymannuronate were precipitated by 10 mM ZnCl₂, whereas less than 70% of polyguluronate was precipitated by the same concentration of ZnCl₂, as shown in Fig. 10. Unlike alginate and polymannuronate, less than 80% of polyguluronate was precipitated by 50 mM ZnCl₂. The ability of HgCl₂, MgCl₂, and RbCl to precipitate alginate, polyguluronate, and polymannuronate was relatively low, unlike other metal ions used in this study. Less than 20% of alginate, polyguluronate, and polymannuronate were precipitated by 100 mM of HgCl₂, MgCl₂, and RbCl, respectively (Data were

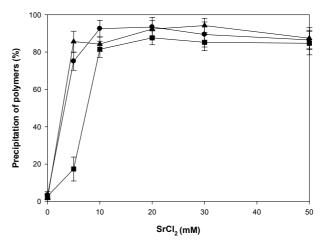


Fig. 9. Precipitation of alginate, polyguluronate, and polymannuronate by SrCl₂ (●; alginate, ■; polyguluronate, and ▲; polymannuronate).

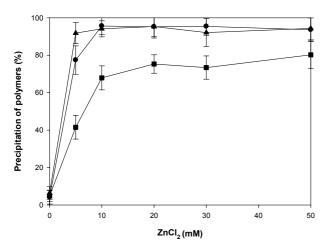


Fig. 10. Precipitation of alginate, polyguluronate, and polymannuronate by ZnCl₂ (●; alginate, □; polyguluronate, and ▲; polymannuronate).

not shown.).

Seaweed alginate isolated from *Laminaria hyperborean*, which is 69% guluronate residues, has a higher affinity for Cd²⁺, Cu²⁺, Pb²⁺, Sr²⁺, and Sr²⁺ than Ca²⁺, whereas it has a lower affinity for Co²⁺, Mg²⁺, Ni²⁺, and Zn²⁺ than Ca²⁺ [6,22]. The mechanism of gel formation is believed to be processed by dimerization of polyguluronate sequences with divalent cations, such as Ca²⁺ chelated between the chains – the so–called Egg–box model [14,19,20]. The affinity of some met–al ions for polymannuronate in this study appears to break the basic rules of Powell's "Egg–Box" theory, because the affinities of Ca²⁺, Co²⁺, Cr⁶⁺, Fe³⁺, Mn²⁺, Sr²⁺, and Zn²⁺ for polymannuronate were slightly higher than those for alginate and polyguluronate.

$P_{1/2}$ of polymers to be precipitated by metal ions

Relative abilities of 13 different metal ions to precipitate alginate, polyguluronate, and polymannuronate were compared with their $P_{1/2}$ values, as shown in Table 1. $P_{1/2}$ is defined as the concentration of each metal ion, expressed in mM, required to precipitate 50% of alginate, polyguluronate, and polymannuronate from 400 μ g/ml of its solution [15]. $P_{1/2}$ was calculated using the Boltzman equation from plots drawn by percentage of alginate, polyguluronate, and polymannuronate precipitated by each metal ion at its different concentration [18]. These values can be used to prospect the relative binding ability of metal ions or affinity of alginate, polyguluronate, and polymannuronate for metal ions.

On the basis of P_{1/2} values, relative affinities of alginate,

Table 1. The precipitation of seaweed alginate, polyguluronate, and polymannuronate by metal ions expressed as a $P_{1/2}^{-1}$

		P1/2 (mM)		
Ions	P1/2 (IIIIVI)			
	Alginate	Polyguluronate	Polymannuronate	
Ca ²⁺	4.1 ± 0.1	5.2 ± 0.4	2.0 ± 0.2	
Cd^{2+}	1.2 ± 0.1	1.1 ± 0.1	1.0 ± 0.1	
Co^{2+}	5.2±1.1	12.9±1.8	4.7 ± 0.5	
Cr^{6^+}	31.1 ± 0.4	56.2±4.1	13.5±0.3	
Cu^{2+}	1.1 ± 0.1	1.2 ± 0.1	1.0 ± 0.2	
$\mathrm{Fe}^{3^{+}}$	1.9 ± 0.2	2.3±0.2	1.5±0.1	
$\mathrm{Hg}^{2^{+}}$	100<	100<	100<	
$\mathrm{Mg}^{2^{+}}$	100<	100<	100<	
$\mathrm{Mn}^{^{2^{+}}}$	32.7±0.3	33.4±1.1	30.3±0.8	
$\mathrm{Pb}^{2^{+}}$	1.6 ± 0.1	1.6 ± 0.1	1.6 ± 0.1	
$\mathrm{Rb}^{^{\scriptscriptstyle +}}$	100<	100<	100<	
Sr^{2^+}	3.3 ± 0.1	6.6 ± 0.4	2.9 ± 0.1	
Zn^{2+}	3.2 ± 0.3	6.6±0.7	2.6±0.1	

1)Concentration of metal ion expressed in mM to precipitate 50% (w/v) of alginate, polyguluronate, and polymannuronate from 400 µg/ml solutions.

polyguluronate, and polymannuronate for metal ions, in order, are as follows; 1) seaweed alginate: $Cu^{2+} > Cd^{2+} > Pb^{2+} > Fe^{3+} >> Zn^{2+} > Sr^{2+} > Ca^{2+} > Co^{2+} >> Cr^{6+} > Mn^{2+} >> Hg^{2+}$, Mg^{2+} , Rb^+ , 2) polyguluronate: $Cd^{2+} > Cu^{2+} > Pb^{2+} > Fe^{3+} >> Ca^{2+} > Sr^{2+}$, Zn^{2+} , $Co^{2+} >> Mn^{2+} > Cr^{6+} >> Hg^{2+}$, Mg^{2+} , Rb^+ , and 3) polymannuronate: Cd^{2+} , $Cu^{2+} > Fe^{3+} > Pb^{2+} > Ca^{2+} > Zn^{2+} > Sr^{2+} > Co^{2+} > Cr^{6+} >> Mn^{2+} >> Hg^{2+}$, Mg^{2+} , Rb^+ . The affinity of alginate prepared from L. digitata (rich in mannuronate residues) for divalent metals was different from that of alginate prepared from L. hyoerborea (rich in guluronate residues) [6]. The following series were obtained; L. digitata: $Pb^{2+} > Cu^{2+} > Cd^{2+} > Ba^{2+} > Sr^{2+} > Ca^{2+} > Co^{2+}$, Ni^{2+} , Zn^{2+} , Zn

Based on their relative abilities to precipitate alginate, polyguluronate, and polymannuronate, metal ions used in this study can be classified into three groups: 1) metal ions having a relatively high ability to precipitate seaweed alginate, polyguluronate, and polymannuronate; Ca²⁺, Cd²⁺, Co²⁺, Cu²⁺, Fe³⁺, Pb²⁺, Sr²⁺, and Zn²⁺, 2) those having a relatively low ability to precipitate them; Mn²⁺ and Cr⁶⁺ and 3) those having no significant ability to precipitate them; Hg²⁺, Mg²⁺, and Rb⁺. Metal ions such as Cd²⁺, Cu²⁺, Fe³, and Pb²⁺ and Zn⁺ showed a higher ability to precipitate alginate, polyguluronate, and polymannuronate than other metal ions used in this study. Some metal ions such as Ca²⁺, Co²⁺, Cr⁶⁺,

Fe³⁺, Mn²⁺, Sr²⁺, and Zn²⁺ showed higher relative affinities for polymannuronate than alginate and polyguluronate. Based on these results, toxic and/or noble metals can be effectively isolated from contaminated environments and/or diluted into aqueous solutions by alginate, polyguluronate, and polymannuronate.

Biosorption of metal ions by biopolymers

Amounts of CdCl₂, CuCl₂, FeCl₃, PbCl₂, and ZnCl₂ bound to 1 g of alginate, polyguluronate, and polymannuronate

were examined. The concentration of cadmium chloride ranged from 0 to 1,000 $\mu g/ml$. The amount of cadmium chloride bound to alginate, polyguluronate, and polymannuronate increased in proportion to the used concentration of cadmium chloride, but it did not increase at above 400 $\mu g/ml$ of cadmium chloride, as shown in Fig. 11A. The concentration of copper chloride ranged from 0 to 500 $\mu g/ml$. The amounts of copper chloride bound to alginate and polymannuronate increased until the concentrations reach to 200 $\mu g/ml$ of copper chloride, whereas that bound to pol-

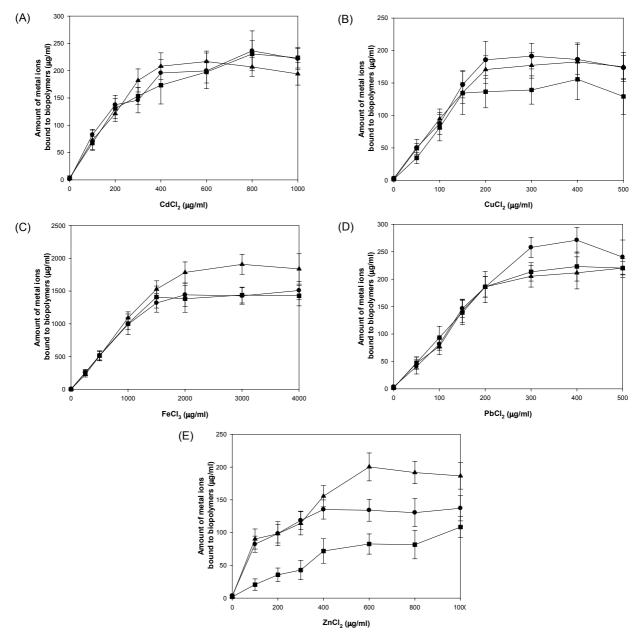


Fig. 11. Amounts of CdCl₂ (A), CuCl₂ (B), FeCl₃ (C), PbCl₂ (D), and ZnCl₂ (E) bound to alginate, polyguluronate, and polyguluronate and polyguluronate (●; alginate, ■; polyguluronate, and ▲; polymannuronate).

yguluronate did not increase at above 150 μ g/ml of copper chloride, as shown in Fig. 11B.

The used concentrations of ferric chloride ranged from 0 to 4,000 µg/ml. The amounts of ferric chloride bound to alginate, polyguluronate, and polymannuronate also increased in proportion to the concentration. However, amounts of ferric chloride bound to polymannuronate did not increase at above 2,000 µg/ml of ferric chloride, whereas those bound to alginate and polyguluronate did not increase at above 1,500 µg/ml of cadmium chloride, as shown in Fig. 11C. The used concentrations of lead chloride ranged from 0 to 500 µg/ml. The amount of lead chloride bound to alginate, polyguluronate, and polymannuronate increased in proportion to the concentration of lead chloride. However, amounts of lead chloride bound to alginate did not increase at above 300 µg/ml of lead chloride, whereas those bound to polyguluronate and polymannuronate did not increase at above 200 μg/ml of lead chloride, as shown in Fig. 11D. When the concentrations of zinc chloride were used from 0 to 1,000 µg/ml, the amount of zinc chloride bound to alginate, polyguluronate, and polymannuronate increased in proportion to the concentration of zinc chloride such as cadmium chloride, copper chloride, ferric chloride, and lead chloride. Amounts of zinc chloride bound to polymannuronate did not increase at above 600 µg/ml of zinc chloride, whereas those bound to alginate and polyguluronate did not increase at above 400 µg/ml of zinc chloride as shown in Fig. 11E.

Amounts of metal ions, Cd²⁺, Cu²⁺, Fe³⁺, Pb²⁺, and Zn²⁺, bound to 1 g of alginate, polyguluronate, and polymannuronate, were calculated using the Boltzman equation from plots drawn by the amount of each metal salt bound to alginate, polyguluronate, and polymannuronate at different concentrations as shown in Table 2 [14]. Amounts of metal ions, Cd²⁺, Cu²⁺, Fe³⁺, Pb²⁺, and Zn²⁺, bound to 1 g of seaweed alginate, were measured in concentrations of 363.5± 45.0, 226.3±9.2, 1,299.4±81.3, 500.7±27.7, and 165.9±11.4 mg, respectively. Amounts of metal ions, Cd²⁺, Cu²⁺, Fe³⁺, Pb²⁺, and Zn²⁺, bound to 1g of polyguluronate, were 354.5±26.5, 177.6±8.7, 1,288.6±60.1, 424.0±7.4, and 140.2±28.5 mg, respectively, whereas those bound to 1 g of polymannuronate were 329.0±10.3, 226.9±1.9, 1,635.6±11.1, 419.8±12.6, and 251.0±49.1 mg, respectively.

It is reported that the uptake of metal ions by microorganisms was mostly dependent on physicochemical adsorption to cell components such as polysaccharides and

Table 2. The maximum amount of metal ions bound to 1 g alginate, polygluronate, and polymannuroante

Ions	Amount of metal ions (mg)			
	Alginate	Polyguluronate	Polymannuronate	
Cd^{2^+}	363.5±45.0	354.5±26.5	329.0±10.3	
Cu^{2+}	226.3±9.2	177.6±8.7	226.0±1.9	
Fe^{3+}	1,299.4±81.3	1,286.6±60.1	1,635.6±11.1	
$\mathrm{Pb}^{2^{+}}$	500.7±27.7	424.0±7.4	419.8±12.6	
$\mathrm{Zn}^{2^{+}}$	165.9±11.4	140.2±28.5	251.0±49.1	

proteins [25]. Cell walls of bacteria, fungi and algae have been used to bind and concentrate some metal ions including cadmium, copper, gold, manganese, mercury, molybdenum, and uranium [1,5,16]. Nonliving biomass of seaweed Ascophyllum nodosum was used to accumulate cobalt to an extent of 160 mg and cadmium exceeding 100 mg per g of biomass [9]. A simple method for isolation of polymannuronate with a low molecular weight from alginate hydrolyzed with acetic acid was developed in our previous study [13]. Amounts of cadmium and lead ions bound to 1 g of polymannuronate in this study were about 330 and 420 mg, respectively. Due to its higher solubility than alginate and a higher affinity for metal ions than polyguluronate, polymannuronate can be used for bioremediation or biosorption of toxic and/or noble metal ions.

Acknowledgement

This study was supported by research funds from Dong-A University.

References

- Darnall, D. W., B. Greene, M. T. Henzl, T. M. Hosea, R. A. McPherson, J. Sneddon, and M. D. Alexander. 1986. Selective recovery of gold and other metal ions from an algal biomass. *Environ. Sci. Technol.* 20, 206–208.
- Cho, K. J., Y. J. Choi, S. C. Ahn, M. Y. Baik, and B. Y. Kim. 2008. Encapsulation with oyster hydrolysate using alginate. J. Life Sci. 18, 708–714.
- 3. Dubois, M., K. A. Gillus, J. K. Hamilton, P. A. Rebers, and F. Smith. 1956. Colorimetric method for sugars and related substances. *Anal. Chem.* **28**, 350–356.
- Gacesa, P., A Squire, and P. J. Winterburn. 1983. The determination of the uronic acid composition of alginates by anion-exchange liquid chromatography. *Carbohydr. Res.* 118, 1–8.
- Greene, B., M. T. Henzl, J. M. Hosea, and D. W. Darnall. 1986. Elimination of biocarbonate interference in the bind-

- ing of U(VI) in mill-waters to freeze-dried *Chlorella vugaris*. *Biotechnol. Bioeng.* **28**, 764-767.
- Haug, A. 1961. The affinity of some divalent metals to different type of alginates. Acta Chem. Scand. 15, 1794–1795.
- Haug, A. and B. Larsen. 1962. Quantitative determination of the uronic acid composition of alginates. *Acta Chem. Scand.* 16, 1908–1918.
- Haug, A. and O SmidsrΦd. 1965. Fractionation of alginates by precipitation with calcium and magnesium ions. *Acta Chem. Scand.* 19, 1221–1226.
- 9. Holan, Z. R., B. Volesky, and I. Prasetyo. 1993. Biosorption of cadmium by biomass of marine algae. *Bioechnol. Bioeng.* **41**, 819–825.
- Jang, L. K., W. R. Brand, M. Resong, W. Mainieri, and G. G. Geesey. 1990. Feasibility of using alginate to absorb dissolved copper. Water Res. 24, 889–897.
- 11. Jang, L. K., S. I. Lopez, S. L. Eastman, and P. Pryfogle. 1991. Recovery of copper and cobalt by biopolymer gels. *Biotechnol. Bioeng.* **37**, 266–273.
- Lee, D. S., H. R. Kim, and J. H. Pyeun. 1998. Effect of low-molecularization on rheological properties of alginate. J. Kor. Fish. Soc. 31, 82-89.
- Lee, D. S., M. K. Shin, J. H. Pyen, and J. W. Lee. 2009. A simple method for isolation of polymannuronate and polyguluronate from alginate hydrolyzed by organic acids. *J. Life Sci.* 19, 34–39.
- 14. Lee, J. W. 1999. Electron microscopic observation of calcium-acetylated seaweed alginate gel. *J. Life Sci.* **9**, 45-49.
- 15. Lee, J. W., R. D. Ashby, and D. F. Day. 1996. Role of acetylation on metal induced precipitation of alginates. *Carbohydr. Polymers* **29**, 337–345.
- 16. Lee, M. G., K. T. Park, and S. K. Kam. 1998. Biosorption of copper by immobilized biomass of marine brown algae

- (Phaeophyta) Hizikia fusiformis. J. Life Sci. 8, 208-215.
- 17. Lin, T. and W. Z. Hassid. 1966. Pathway of alginic acid synthesis in the marine brown alga, *Fucus gardneri* silva. *J. Biol. Chem.* **241**, 5284–5297.
- Maachi, R., M. Abousseoud, and T. Chaabane. 2001. Kinetics of biodegradation of petroleum by *Pseudomonas* sp. *Desalination* 139, 367.
- Morris, E. R., D. A. Rees, and D. Thorn. 1978. Chiroptical and stoichiomtric evidence of specific primary dimerization process in alginate gelation. *Carbohydr. Res.* 66, 145–154.
- 20. Morris, E. R. and D. A. Rees. 1980. Competitative inhibition of interchain interactions in polysaccharide systems. *J. Mol. Biol.* **138**, 363–374.
- 21. Penman, A. and G. R. Sanderson. 1972. A method for the determination of uronic acid sequence in alginates. *Carbohydr. Res.* **25**, 273–282.
- Rees, D. A. 1972. Shapely polysaccharides. *Biochem. J.* 126, 257–273
- 23. SmidsrØd, O. and A. Haug. 1968. Dependence upon uronic acid compositon of some ion-exchange properties of alginates. *Acta Chem. Scand.* **26**, 2563–2566.
- Smidsrød, O. 1974. Molecular basis for some physical properties of alginates in the gel state. Farad. Discuss. Chem. Soc. 57, 263–274.
- 25. Suh, J. H., M. K. Suh, and Y. H. Lee. 2006. Biosorption model and factors for removing lead to *Aureobasidium pullulans* being imperfect fungus. *J. Life Sci.* **16**, 877–883.
- Volesky, B. 1987. Biosorbents for metal recovery. *Tibtech.* 96–101.
- 27. Voragen, A. G. J., H. A. Schols, J. A. De Vries, and W. Pilnik. 1982. High-performance liquid chromatographic analysis of uronic acids and oligogalacturonic acids. *J. Chromat.* **244**, 327–336.

초록: 알긴산, 폴리글루론산 및 폴리만뉴론산에 의한 금속이온의 흡착

정대영 1,2 · 손창우 1 · 김성구 2 · 김이준 3 · 정정한 3,4 · 이진우 3,4 *

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P_{1/2} 값을 참고로 비교한 알긴산, 폴리글루론산 및 폴리만뉴론산의 금속이온들에 대한 상대적인 친화성은 다음과 같다; 1) 알긴산: Cu²⁺>Cd²⁺>Pb²⁺>Fe³⁺>>Zn²⁺>Sr²⁺>Ca²⁺>Co²⁺>>Cr⁶⁺>Mn²⁺>>Hg²⁺, Mg²⁺, Rb⁺, 2) 폴리글루론산: Cd²⁺>Cu²⁺>Pb²⁺>Fe³⁺>>Ca²⁺>Sr²⁺, Zn²⁺, Co²⁺>>Mn²⁺>Cr⁶⁺>>Hg²⁺, Mg²⁺, Rb⁺, 그리고 3) 폴리만뉴론산: Cd²⁺, Cu²⁺>Fe³⁺>Pb²⁺>Ca²⁺>Sr²⁺>Sr²⁺>Co²⁺>Cr⁶⁺>>Mn²⁺>>Hg²⁺, Mg²⁺, Rb⁺, 2리고 3) 폴리만뉴론산: Cd²⁺, Cu²⁺>Fe³⁺>Pb²⁺>Ca²⁺>Sr²⁺>Co²⁺>Cr⁶⁺>>Mn²⁺>>Hg²⁺, Mg²⁺, Rb⁺, 2리고 3) 폴리만뉴론산: Cd²⁺, Cu²⁺, Fe³⁺, Pb²⁺ 및 Zn²⁺의 양은 363.5±45.0, 226.3±9.2, 1,299.4±81.3, 500.7±27.7 및 165.9±11.4 mg이었으며, 폴리글루론산 1g에 흡착하는 Cd²⁺, Cu²⁺, Fe³⁺, Pb²⁺ 및 Zn²⁺의 양은 354.5±26.5, 177.6±8.7, 1,288.6±60.1, 424.0±7.4 및 140.2±28.5 mg이었으나, 폴리만뉴론산 1 g에 흡착하는 Cd²⁺, Cu²⁺, Fe³⁺, Pb²⁺ 및 Zn²⁺의 양은 329.0±10.3, 226.9±1.9, 1,635.6±11.1, 419.8±12.6 및 251.0±49.1 mg이었다. 폴리만뉴론산은 알긴산보다 높은 용해도와 폴리글루론산보다 높은 금속이온에 대한 친화성 때문에 독성이 높은 중금속이나 경제성이 높은 금속을 선택적으로 분리하는데 사용할 수 있을 것이다.