Biosorption of Cu(II), Zn(II), Ni(II) and Pb(II) ions by cross-linked metal-imprinted chitosans with epichlorohydrin

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Cross-linked metal-imprinted chitosan microparticles were prepared from chitosan, using four metals (Cu(II), Zn(II), Ni(II), and Pb(II)) as templates, and epichlorohydrin as the cross-linker. The microparticles were characterized by Fourier transform infrared spectroscopy, solid state $^{13}$C nuclear magnetic resonance spectroscopy, and energy-dispersive X-ray spectroscopy. They were used for comparative biosorption of Cu(II), Zn(II), Ni(II) and Pb(II) ions in an aqueous solution. The results showed that the sorption capacities of Cu(II), Zn(II), Ni(II), and Pb(II) on the templated microparticles increased from 25 to 74%, 13 to 46%, 41 to 57%, and 12 to 43%, respectively, as compared to the microparticles without metal ion templates. The dynamic study showed that the sorption process followed the second-order kinetic equation. Three sorption models, Langmuir, Freundlich, and Dubinin–Radushkevich, were applied to the equilibrium isotherm data. The result showed that the Langmuir isotherm equation best fitted for monolayer sorption processes. Furthermore, the microparticles can be regenerated and reused for the metal removal.

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1. Introduction

Water pollution caused by toxic metals is one of the most pressing environmental problems today. The potential sources of metals from industrial effluents include fertilizers, metal fabrication, paints, pigments, and batteries. Among the many methods available for the removal of metals from industrial effluents, including chemical precipitation, ion exchange, coagulation, membrane separation, and electro deposition, sorption seems to be the most economically feasible alternative (Chen et al., 2002; Ciesielski et al., 2003; Hu et al., 2003; Ning, 2003; Reddad et al., 2003). Attention has been focused on various sorbents that have high metal capacity for the removal of metals from contaminated water at low cost (Bailey et al., 1999; Babel and Kurniawan, 2003).

Chitosan, or poly($\beta$-1,4)2-amino-2-deoxy-$\alpha$-glucopyranose, can be obtained from chitin, a natural biopolymer extracted from crustacean shells by the partial deacetylation of its acetamido groups using strong alkaline solutions. Because chitosan has some unique properties, such as biodegradability, biocompatibility, and bioactivity, it has various potential applications in biomedical products, cosmetics, food processing, and wastewater treatment, among others (Guibal et al., 1998, 1999; Ngah et al., 2002a; Ravi Kumar et al., 2004). It also has both hydroxyl and amino groups that can be chemically modified. Several cross-linking methods with cross-linkers such as glutaraldehyde, epichlorohydrin, ethylene glycol diglycidyl ether, and dihydroxy azacrown ether, and gamma irradiation in the presence of carbontetrachloride have been used to modify both the physical and chemical properties of chitosan to improve its chemical stability in acidic media and resistance to biochemical and microbiological degradation (Ngah et al., 2002b; Yang et al., 2002; Ramnani and Sabharwal, 2006; Baroni et al., 2008; Chen et al., 2008). Although the cross-linking method may enhance the resistance of chitosan against chemicals and acids, it can also reduce the sorption capacity of metals. This may be because the cross-linking reaction decreases the amount of amino and hydroxyl groups, which are thought to play an important role in the sorption process. To overcome this problem, the ion-imprinting method has been used to prepare cross-linked metal-imprinted chitosans using glutaraldehyde (GLA) as a cross-linker via Schiff’s base linkage (Sun et al., 2006; Sun and Wang, 2006b; Chen et al., 2009). Its recognition is based on the specification of the ligand, coordination geometry, coordination number, charge, and size of the metal ion. This may be higher in the sorption capacity of metals, and can be quite stable in an acidic medium.

The objective of this study was to prepare cross-linked metal-imprinted chitosan microparticles from chitosan using the ion-imprinting method. Four metals, Cu(II), Zn(II), Ni(II), and Pb(II), were used as templates and epichlorohydrin (ECH) was used as the
cross-linker. These microparticles were characterized using Fourier transform infrared (FTIR) spectroscopy, solid state $^{13}$C nuclear magnetic resonance ($^{13}$CNMR) spectroscopy, and energy-dispersive X-ray (EDX) spectroscopy. The influences of the molar ratios and cross-linking times on the sorption capacity were investigated. The data were used for comparative biosorption of Cu(II), Zn(II), Ni(II), and Pb(II) ions in an aqueous medium. Furthermore, the sorption equilibrium, sorption kinetics, sorption efficiency, and desorption were examined to gain better insights into the experimental results. This information can be used for further applications in the treatment of waste effluents from the industry.

2. Materials and method

2.1. Chemicals

Chitosan from Sigma–Aldrich Co. (USA) was hydrolyzed with sodium hydroxide to give it a deacetylation percentage of approximately 90% by analysis of FTIR spectroscopy. The average molecular weight of chitosan was 690,000 Da, as measured by a viscometric method. Epichlorohydrin (ECH) with 99.6% purity was purchased from Tedia Co. (USA). Cupric chloride, zinc chloride, nickel chloride, and lead acetate were purchased from Wako Co., (Japan) and were of analytical-reagent grade. All reagents were used without further purification.

2.2. Preparation

A 0.2 M sodium hydroxide solution was gradually added to a 100 mL solution of acetic acid (1%, v/v) containing chitosan (8.8 mmol) and a metal ion (8.8 mmol). The solution was vigorously stirred to obtain a gelatinous metal-complexed chitosan of around pH 6.5–7.0. The isolated gel was then added to a 100 mL aqueous solution of ECH (88.0 mmol). The mixture then stirred for 2 h at 40–50 °C. The process was followed by filtering and intensive washing of the precipitate with distilled water to remove any unreacted ECH. The precipitate was again stirred with a 1.2 M hydrochloric acid solution to remove the metal ion. This stage was monitored by atomic absorption spectrometry (AAS). The precipitate was stirred with 1% (v/v) acetic acid to remove non-cross-linked chitosan. Finally, it was washed in a solution of 0.2 M sodium hydroxide, distilled water, and acetone. The precipitate was dried in a vacuum oven for 8 h. The resulting material was ground and sieved to produce microparticles with diameters between 250 and 500 μm.

The FTIR spectra were obtained with a Perkin Elmer Spectrum One FTIR spectrometer. The EDX spectra were obtained with an Oxford EDS INCA 350 spectrometer. The solid state $^{13}$CNMR spectra were measured on a Brucker Advance 400 NMR spectrometer. The mean pore diameters were obtained using a mercury intrusion porosimeter, Scientek STK019348.

2.3. Sorption study

The microparticles, which were prepared from 5.0, 10.0, and 15.0 molar ratios of ECH/chitosan with different cross-linking times (2, 3, and 4 h), were studied to determine the sorption capacity of the metal ions. This was done by adding 10 mg of each kind of particle into 100 mL of a 10 mg/L metal ion solution at an initial pH of 5.0 while stirring at 25 °C for 4 h. The solutions were filtered and the concentrations of metal ions were measured on a Hitachi 170-30 atomic absorption spectrophotometer. The sorption capacity (Q) (mg/g) of the metal ion was calculated from Equation (1),

$$ Q = \frac{(C_i - C_f)V}{W} $$

where $C_i$ is the initial concentration of metal ion (mg/mL), $C_f$ is the final concentration of metal ion (mg/mL), $V$ is the volume of metal ion solution (mL), and $W$ is the weight of the microparticles (g).

2.4. Kinetic study

The kinetic study was accomplished by adding 10 mg of each kind of the microparticles into 100 mL of a 10 mg/L metal ion solution at an initial pH of 5.0 while stirring at 25 °C. Then, 10 mL aliquots of these solutions, at intervals of 20 min, were filtered to measure the concentrations of metal ions by AAS. The sorption capacities of the metal ions were calculated using Equation (1).

2.5. Equilibrium study

Equilibrium studies of the microparticles were conducted in 100 mL of metal ions with initial concentrations in the range of 0–10 mg/L at an initial pH of 5.0 while stirring for 4 h at 25 °C. Solutions were filtered and the concentrations of the metal ions were measured by AAS. The amounts of metal ion adsorbed were calculated based on the differences of the concentration in the aqueous solution before and after sorption and on the weight of the cross-linked chitosan particles according to Equation (1).

3. Results and discussion

3.1. Characterization

The cross-linked metal-imprinted chitosan microparticles were prepared using the ion-imprinting method (Fig. 1). Chitosan was first chelated with a metal ion through ion–dipole and ion–ion interactions, followed by chemical cross-linking with ECH via an ether linkage (Sun et al., 2006). Finally, the metal ion was removed to obtain microparticles, which were found to be insoluble in distilled water, alkaline media, and even in acidic media (pH 1.0). The FTIR spectra (Fig. 2) of the microparticles showed absorption peaks at 3607 and 3382 cm$^{-1}$, assigned to amide N–H stretching vibrations, 1658 cm$^{-1}$, assigned to C=O stretching vibrations, 1566 cm$^{-1}$, assigned to amide N—H bending vibrations, and 1148, 1055, and 1023 cm$^{-1}$, all of which were assigned to C—O—C and C—O stretching vibrations (Silverstein et al., 2005). In addition, the solid state $^{13}$CNMR spectra of the microparticles showed three additional peaks at 61.6, 85.0, and 97.6 ppm due to the existence of residual amino and hydroxyl groups of chitosan after the cross-linking reaction. Cross-linked Zn(II)-imprinted chitosan microparticles were larger than the cross-linked chitosan microparticles prepared from a 10.0 molar ratio of ECH/chitosan had 26% and 19% higher sorption capacity than Zn(II) microparticles prepared from a 5.0 molar ratio of ECH/chitosan.
from 5.0 and 15.0 molar ratios of ECH/chitosan, respectively (Fig. 3).
In addition, the sorption capacity of the Zn(II) ion on the micro-
particles prepared from a 10.0 molar ratio of ECH/chitosan with 2 h
of cross-linking was found to be 25% and 32% larger than the
microparticles prepared with 3 and 4 h of cross-linking, respec-
tively (Fig. 4). The three other cross-linked Cu(II)-, Ni(II)-, and Pb
(II)-complexed chitosan microparticles showed the same results.
Thus, the optimal conditions for the preparation of these metal-
imprinted microparticles included 10.0 molar ratio of ECH/chitosan
and 2 h of cross-linking. The increase in adsorption capacity from
5.0 to 10.0 molar ratios of ECH/chitosan may be attributed to that
the low level of cross-linking prevents the formation of closely
packed chain arrangement without a significant decrease in the swelling capacity. In addition, it cannot only reduce the opening of chitosan network by supplementary linkages between chitosan chains, but also expands the chitosan network, resulting in a reduction of steric hindrance effect and a significant increase of adsorption capacities of metal ions. At higher molar ratio of ECH/chitosan (e.g. 15.0), the declining swelling and decreasing accessibility of the higher cross-linking chitosan are due to a more extensive three-dimension network and an increase in the hydrophobic character, thus resulting in the decrease of adsorption capacity (Sun et al., 2006). At the same time, the shorter reaction time may involve in forming the flexible structure of the cross-linked chitosan, resulting in better for adsorption of metal ions.

### 3.3. Sorption kinetics

Fig. 5 presented the sorption kinetics for the metal ions in the microparticles at an initial pH of 5.0 from 0 to 300 min. For all the metal ions, a rapid initial rate of metal sorption on the microparticles was observed during the first 60 min and gradually approached the limiting sorption after 180 min. This may be attributed to the fact that the sorption sites on the microparticles were gradually covered by the metal ions, causing the sorption processes to become dependent on the rate of the metal ions adsorbed from the bulk liquid phase to the actual sorption sites.

In order to investigate the mechanism of the metal sorption on the microparticles, the dynamic experimental data were applied to the first-order equation (2),

\[
\ln(Q_e - Q_t) = \ln Q_e - k_1 t,
\]

and the second-order kinetic equation (3),

\[
t/Q_t = 1/(k_2 Q_e^2) + t/Q_e,
\]

where \(Q_e\) and \(Q_t\) are the sorption capacities of the metal ion (mg/g) at equilibrium and at a given time \(t\), respectively; and \(k_1\) (min\(^{-1}\)) and \(k_2\) (g/(mg min)) are the first-order and second-order rate constants, respectively. According to Equation (2), the plot of \(\ln(Q_e - Q_t)\) against \(t\) gives a straight line with a slope of \(-k_1\) and an intercept of \(\ln Q_e\). From Equation (3), the plot of \(t/Q_t\) against \(t\) gives a straight line with a slope of \(1/Q_e\) and an intercept of \(1/(k_2 Q_e^2)\).

The dynamic experimental data were listed in Table 1. The first-order rate constants were from 0.91 \(\times\) 10\(^{-2}\) to 1.48 \(\times\) 10\(^{-2}\) min\(^{-1}\) with correlation coefficients from 0.9508 to 0.9842. The second-order rate constants were from 2.98 \(\times\) 10\(^{-3}\) to 5.42 \(\times\) 10\(^{-3}\) g/(mg min) with correlation coefficients from 0.9989 to 0.9995. In addition, the calculated equilibrium adsorption capacities, \(Q_{e,cal}\), from the second-order kinetics fitted well with the experimental data, \(Q_e\). This indicated that the second-order kinetic equation appeared to fit the dynamic experimental data better than that of
the first-order kinetics. The second-order kinetic equation is also more likely to show that the sorption behavior may involve valency forces through electron-sharing between the metal ions and sorbents (Niu et al., 2007; Chen et al., 2008, 2009).

3.4. Sorption equilibrium

The metal ions, Cu(II), Zn(II), Ni(II), and Pb(II), are precipitated as hydroxides at pH >6, >8, >8, and >6, respectively. The initial pH effects on the sorption of the ions were determined only in pH <6. The results showed that the sorption capacities of the metal ions followed the pH order of: 1.0 < 3.0 < 5.0. Sorption was likely to be lower at lower pH because of the electrostatic repulsion between the metal cations and the protonated amino groups. As pH values increased, amino groups were freed from protonation for the sorption behavior in the chelation mechanism of metal ions. Thus, the optimum pH for the adsorption of Cu(II), Zn(II), Ni(II) and Pb(II) was at pH 5.0. The experimental equilibrium isotherms for the sorption of Cu(II), Zn(II), Ni(II), and Pb(II) ions on the cross-linked microparticles at an initial pH of 5.0 and 25 °C were shown in Fig. 6. They were applied to three different sorption isotherms: Langmuir isotherm equation (4) (Langmuir, 1918; Ng et al., 2003),

\[
\frac{C_e}{Q_e} = \frac{C_e}{Q_m} + \frac{1}{(Q_m K_L)},
\]

Freundlich isotherm equation (5) (Freundlich, 1906; Niu et al., 2007),

\[
\ln Q_e = \frac{1}{n} \ln C_e + \ln K_F,
\]

and Dubinin–Radushkevich isotherm equation (6) (Ramnani and Sabharwal, 2006).
formation between the metal ions and amino group and/or hydroxyl group of chitosan.

3.5. Comparative sorption

Four microparticles were used as sorbents to investigate the comparative sorption of the four-metal ions in 50 mg/L aqueous solutions of the metal ions at an initial pH 5.0 and 25 °C for 4 h. The sorption capacity (mmol/g) of the Cu(II) ion showed in the following order: Cu(II) as template (0.33) > Zn(II) as template (0.29) > Ni(II) as template (0.28) > Pb(II) as template (0.24). The sorption capacity of the Cu(II) ion indicated an increase from 25 to 74% compared to the microparticles without metal templates. The sorption capacities (mmol/g) of the Zn(II) ion were as follows: Zn(II) as template (0.22) > Ni(II) as template (0.21) > Pb(II) as template (0.18) > Cu(II) as template (0.17). The sorption capacity of the Zn(II) ion increased from 13 to 46% compared to the microparticles without metal ion templates. For the Ni(II) ion, the sorption order was as follows: Ni(II) as template (0.51) > Pb(II) as template (0.41) > Zn(II) as template (0.38) > Cu(II) as template (0.37). The sorption capacities of the Ni(II) ion increased from 41 to 57% compared to the microparticles without metal ion templates. Finally, the order of the sorption capacity (mmol/g) of the Pb(II) ion was: Pb(II) as template (0.36) > Cu(II) as template (0.35) > Ni(II) as template (0.34) > Zn(II) as template (0.28). Only the sorption capacity of the Pb(II) ion increased from 12% to 43% compared to the microparticles without metal ion templates.

The maximum sorption capacities (Q_m) of Cu(II), Zn(II), Ni(II) and Pb(II) ions on various templated cross-linked-chitosans in the literatures were listed in Table 3. The sorption capacity increased when the initial concentration of the metals increased. In this study, the ECH-cross-linked metal-imprinted chitosan microparticles showed higher sorption capacities of Cu(II), Zn(II), and Ni(II) ions compared to Cu(II) single imprinted chitosan-succinate and Cu (II)/sol–gel double imprinted chitosan-succinate. It also indicated to be of higher sorption capacities of Zn(II) and Pb(II) ions compared to the GLA-cross-linked carboxymethyl-chitosan with a Cu(II) template. Furthermore, the sorption of metal ions on the imprinted microparticles was significantly dependent on the kinds of cross-linkers, metal ion templates, and the metal species used.

3.6. Regeneration

A regeneration study helped to elucidate the nature of the sorption process and removed metal ions from the microparticles. EDTA was used in the desorption study because of its high stability constants with metal ions (Ngah et al., 2002). The templated microparticles were regenerated in three different concentrations of EDTA solutions, 0.25, 0.50, and 1.00 mM, under continuous stirring for 8 h at 25 °C. As shown in Fig. 7, desorption of the microparticles was significantly dependent on the kinds of metals used as templates and concentrations of the EDTA solution. The desorption percentage of Cu(II) ion on microparticles with a Cu(II) template was larger than those of the other metal ions. This may be due to the higher stability constant of Cu(II) ion with EDTA compared to the three other metal ions (Furia, 1972). In addition, desorption was more efficient in 1.00 mM EDTA solution than in acidic solutions, e.g. desorption (%) of Cu(II), Zn(II), Ni(II) and Pb(II): 30, 15, 16, 30 and 25, 12, 13, 27 for 0.1 M HCl and for 0.1 M HNO_3, respectively. It may be attributed to the high stability constants of EDTA with metal ions (Ngah et al., 2002). The microparticles were twice reused for the sorption of Cu(II), Zn(II), Ni(II), or Pb(II). As shown in Fig. 8, the resorption ratios of Cu(II), Zn(II), Ni(II), and Pb(II) ions after and before regeneration were about 0.48, 0.21, 0.25, and 0.40, respectively. Thus, the microparticles can be regenerated and reused for the sorption of metals.

### Table 2
Langmuir, Freundlich, and Dubinin–Radushkevich isotherm parameters for sorption of Cu(II), Zn(II), Ni(II) and Pb(II) ions on cross-linked Cu(II)-, Zn(II)-, Ni(II)- and Pb(II)-imprinted chitosan microparticles, respectively.

<table>
<thead>
<tr>
<th>Metal ion</th>
<th>Cu(II) ion</th>
<th>Zn(II) ion</th>
<th>Ni(II) ion</th>
<th>Pb(II) ion</th>
</tr>
</thead>
<tbody>
<tr>
<td>Langmuir isotherm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Q_m (mg/g) (S.D.)</td>
<td>19.80 (0.29)</td>
<td>14.74 (0.24)</td>
<td>29.23 (0.65)</td>
<td>74.07 (1.60)</td>
</tr>
<tr>
<td>R_L (L/mg) (S.D.)</td>
<td>22.27 (2.81)</td>
<td>3.57 (0.28)</td>
<td>12.28 (0.26)</td>
<td>1.12 (0.12)</td>
</tr>
<tr>
<td>R^2</td>
<td>0.9987</td>
<td>0.9984</td>
<td>0.9983</td>
<td>0.9839</td>
</tr>
<tr>
<td>Freundlich isotherm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>K (mg/g)</td>
<td>16.67 (0.29)</td>
<td>14.47 (1.20)</td>
<td>31.86 (2.20)</td>
<td>51.11 (6.26)</td>
</tr>
<tr>
<td>1/n (S.D.)</td>
<td>0.17 (0.02)</td>
<td>0.21 (0.07)</td>
<td>0.03 (0.01)</td>
<td>0.19 (0.07)</td>
</tr>
<tr>
<td>R^2</td>
<td>0.8581</td>
<td>0.9205</td>
<td>0.9401</td>
<td>0.7505</td>
</tr>
<tr>
<td>Dubinin–Radushkevich isotherm</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Q_m (mg/g) (S.D.)</td>
<td>18.28 (0.0%)</td>
<td>16.69 (0.48)</td>
<td>31.77 (1.04)</td>
<td>75.15 (2.14)</td>
</tr>
<tr>
<td>K (kJ/mol) (S.D.)</td>
<td>10.16 (1.45)</td>
<td>14.80 (2.65)</td>
<td>9.62 (0.01)</td>
<td>12.19 (1.99)</td>
</tr>
<tr>
<td>E (kJ/mol) (S.D.)</td>
<td>0.9681</td>
<td>0.9418</td>
<td>0.8273</td>
<td>0.8898</td>
</tr>
</tbody>
</table>

Note: S.D. in the parentheses is the standard deviation.

### Table 3
The maximum sorption capacity (Q_m) of Cu(II), Zn(II), Ni(II) and Pb(II) ions on various templated cross-linked-chitosans in the literatures.

<table>
<thead>
<tr>
<th>Adsorbent</th>
<th>Concentration of metal ion, pH, temperature, and time</th>
<th>Q_m (mmol/g)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cu(II)</td>
<td>Zn(II)</td>
<td>Ni(II)</td>
<td>Pb(II)</td>
</tr>
<tr>
<td>ECH-cross-linked metal-imprinted chitosan</td>
<td>50 mg/L, pH 5.0, 25 °C and 4 h</td>
<td>0.33</td>
<td>0.22</td>
</tr>
<tr>
<td>GLA-cross-linked N-succinyl-chitosan with Pb(II) template</td>
<td>0.02 mol/L, pH 5.0, 25 °C and 24 h</td>
<td>1.85</td>
<td>0.25</td>
</tr>
<tr>
<td>GLA-cross-linked metal-imprinted chitosan</td>
<td>50 mg/L, pH 5.0, 25 °C and 4 h</td>
<td>0.56</td>
<td>0.24</td>
</tr>
<tr>
<td>GLA-cross-linked chitosan with Cu(II) template</td>
<td>0.08 mol/L, 35 °C, pH 5–6, and 6 h</td>
<td>2.36</td>
<td>2.12</td>
</tr>
<tr>
<td>GLA-cross-linked carboxymethyl-chitosan with Cu(II) template</td>
<td>0.02 mol/L, pH 5.0, 25 °C and 24 h</td>
<td>2.45</td>
<td>0.03</td>
</tr>
<tr>
<td>Cu(II) single imprinted chitosan-succinate</td>
<td>20 mg/L and 25 °C</td>
<td>0.01</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>Cu(II)/sol–gel double imprinted chitosan-succinate</td>
<td>20 mg/L and 25 °C</td>
<td>0.35</td>
<td>&lt;0.01</td>
</tr>
</tbody>
</table>
Cross-linked metal-imprinted chitosan microparticles were prepared from chitosan using four metals, Cu(II), Zn(II), Ni(II), and Pb(II), as templates and epichlorohydrin as the cross-linker. The comparative sorptions of Cu(II), Zn(II), Ni(II), and Pb(II) ions from the aqueous solution were found in the following order of metal-as-template sorption capacities: Cu(II) > Zn(II) > Ni(II) > Pb(II), Zn(II) > Ni(II) > Pb(II) > Cu(II), Ni(II) > Pb(II) > Zn(II) > Cu(II), and Pb(II) > Cu(II) > Ni(II) > Zn(II), respectively. In addition, the results showed that the sorption capacities of the Cu(II), Zn(II), Ni(II), and Pb(II) templates on the microparticles increased from 25 to 74%, 13 to 46%, 41 to 57%, and 12 to 43%, respectively, compared to microparticles without metal ion templates. The dynamic study followed the second-order kinetic equation in the adsorption process. Equilibrium adsorption studies showed that the Langmuir isotherm equation best fitted the monolayer adsorption processes. Furthermore, microparticles can be regenerated and reused for the sorption of metal ions.

References