CHAPTER 2

EXPERIMENTAL

2.1 Instrument and Apparatus

1) Atomic absorption spectrophotometer, Analyst 800, Perkin-Elmer, U.S.A.
2) Inductively coupled plasma optically emission spectrometer, Optima 3000, Perkin-Elmer, U.S.A.
3) Homogenizer, Unimax 1010, Heidolph, Germany.
4) pH-meter, Russell RL 150, Russell Inc., U.S.A.
5) Grinder, SM1, Retsch GMbH, Germany.
6) Vibratory sieve shaker, Analysett 3, U.K.

2.2 Chemicals

1) Standard solution of Cr(III), Cu(II), Cd(II) and Pb(II), 1,000 mg/l, AAS grade, Merck, Germany.
2) Multielements standard of Cr(III), Cu(II), Cd(II) and Pb(II), 1,000 mg/l, Spectrosol, AAS grade, BDH, U.K.
3) Nitric acid, GR grade, Merck, Germany.
4) Hydrochloric acid, GR grade, Merck, Germany.
5) Sodium hydroxide, AR grade, Ajex Chemicals, Australia.

6) Potassium hydroxide, AR grade, Ajex Chemicals, Australia.
7) Epichlorohydrin, AR grade, Ajex Chemicals, Australia.
8) Acetone, AR grade, Ajex Chemicals, Australia.
9) Ether, AR grade, Ajex Chemicals, Australia.

2.3 Preparation of Standard Solutions and Reagents

2.3.1 Standard solution of metals

For ICP-OES measurement, the mixture of standard solutions containing Cr(III), Cd(II), Cu(II) and Pb(II) of the concentration of 1 mg/l were prepared from multielement stock standard solution (1000 mg/l) by appropriate dilution with deionized water.

For AAS measurement, individual standard solution (100 mg/l) of Cr(III), Cd(II), Cu(II) and Pb(II) at the concentration of 100 mg/l were prepared from the elemental stock standard solution (1000 mg/l) by appropriate dilution with deionized water.

2.3.2 Solution of 5.0 M sodium hydroxide

This solution was prepared by dissolving about 25.0 g of NaOH in deionized water and adjusting to a volume of 100 ml. The solution was standardized against reference standard HCl (previously by standardized) An appropriate volume of this standard NaOH solution was take and diluted to 5.0 M with deionized water.

2.3.3 Solution of 0.2 M sodium chloride

This solution was prepared by dissolving 1.17 g of NaCl (previously dried at 110°C for 180 minutes in an oven) in deionized water and adjusting to 100 ml with deionized water.

2.3.4 Solution of 2.0 M potassium hydroxide

This solution was prepared by dissolving 11.22 g of KOH in deionized water and adjusting to 100 ml with deionized water.

2.3.5 Solution of 0.1 M hydrochloric acid

This solution was prepared by diluting 0.6 ml of concentrated HCl to a total volume of 100 ml with deionized water. The solution was standerdized with 0.1 M NaOH (previously standardized).

2.4 Determination of Pb(II), Cu(II), Cd(II) and Cr(III) ions by FAAS

The initial and residual concentrations of metals were analyzed by using flame atomic absorption spectrophotometer (FAAS). The optimal parameter recommended by analytical methods for atomic absorption spectrophotometry was shown in Table 2.1. It was used throughout this work. The concentrations of metals were directly determined by diluting to the desired concentration ranges. (See appendix)

Table 2.1 Standard atomic absorption conditions

Metal	Wavelength (nm)	Slit (nm)	Characteristic concentration (mg/l)	Characteristic concentration check (mg/l)	Linear range (mg/l)	Relative noise
Pb	217.0	0.7	0.190	9.0	20.0	1.0
Cu	324.8	0.7	0.077	4.0	5.0	1.0
Cd	228.8	0.7	0.028	1.5	2.5	1.0
Cr	357.9	0.7	0.078	4.0	5.0	1.0

2.4.1 Characteristic concentration

Characteristic concentration in atomic absorption (sometimes called "sensitivity") is defined as the concentration of an element (expressed in mg/l) required to produce a signal of 1 % absorption (0.0044 absorbance units). As long as measurements are made in the linear working range, characteristic concentration can be determined by reading the absorbance produced by a known concentration of the element, and solving the following equation:

$$\text{Characteristic Concentration} = \frac{\text{Conc. of Std. x 0.0044}}{\text{Measured Abs}}$$

2.5 Preparation of *Sargassum polyceratium*

2.5.1 Preparation of dried *Sargassum polyceratium*

Dried *Sargassum polyceratium* used as biosorbent in this work was kindly donated by Archan Sirirat Chanvaivit (University of Burapa). After grinding with grinder, the biomass was sieved to select particles between 0.21-2.36 mm.

2.5.2 Preparation of modified *Sargassum polyceratium*

Dried *Sargassum polyceratium* was sieved for particle size of 0.21-0.35 mm. The appropriate size of *Sargassum polyceratium* (50.0 g dry weight) was mixed with 75.0 ml of 0.2 M sodium chloride and 2.75 ml of epichlorohydrin for 10 minutes. Then 20.0 ml of 2.0 M potassium hydroxide was added, and the mixture was stirred slowly at 50 ˚C for 30 minutes. After cooling to room temperature, the mixed solution of 1.0 ml epichlorohydrin and 25.0 ml of deionized water was added and stirred for 16 hours. Subsequently, 125.0 ml of 5.0 M sodium hydroxide and 7.5 ml of carbon disulfide were added and stirred for 2 hours. Finally, the mixture was washed with water, acetone and ether, respectively and allowed to dry at 80°C for 4 hours.

2.6 Procedures

2.6.1 Optimization of adsorption conditions for dried seaweed

The biosorption of Pb(II), Cu(II), Cd(II) and Cr(III) ions from aqueous solutions were studied at room temperature. The initial concentration of Pb(II), Cu(II), Cd(II) and Cr(III) used was 30.00, 7.00, 5.00 and 10.00 mg/l, respectively.

(1) Effect of particle size

The dried seaweed (0.100 g) and 50.0 ml of each metal standard solution were added to polyethylene bottle. The particle size of biomass was varied from 0.21-2.36

mm. Then the suspended solution was shaken for 60 minutes in a shaker at speed of 200 rpm. The supernatant was filtered through filter paper (Whatman No.1) and analyzed for the residual concentration of each metal by AAS.

(2) Effect of solution pH

The dried seaweed (0.100 g, 0.21-0.36 mm) and 50.0 ml of each metal standard solution were added to polyethylene bottle. The initial pH of solution was veried from 2.0-5.0. The solution was adjusted to the required pH using 0.1 M HCl or 0.1 NaOH. Then the suspended solution was shaken for 60 minutes at the speed of 200 rpm. The supernatant was filtered through filter paper (Whatman No.1) and analyzed for the residual concentration of each metal by AAS.

(3) Effect of shaking time

The dried seaweed having a particle size of 0.21-0.36 mm (0.100 g) and 50.0 ml of metal standard solution were added to polyethylene bottle. Then the suspended solution was shaken for 5 minutes at the speed of 200 rpm. The experiments were repeated by variation of shaking time from 5 to 120 minutes.

The supernatant was filtered through filter paper (Whatman No.1) and analyzed for the residual concentration of each metal by AAS.

(4) Effect of other metals

Solutions containing constant amounts of metals of interest along with varying amounts of other metals as described in Tables 2.2-2.5 were prepared. The mixture of 50.0 ml solution was added to 0.100 g of biomass in polyethylene bottle. Then the suspended solution was shaken for 60 minutes at the speed of 200 rpm. The supernatant was filtered through filter paper and the residual concentration of each metal was measured by AAS.

2.6.2 Isotherm character for dried seaweed

The isotherm studies were performed by varying the initial concentrations of Pb(II), Cr(III), Cu(II) and Cd(II) ions over the ranges of 1.00 to 30.00 mg/l at a pH 3.5, 3.0, 4.0 and 3.5, respectively. A 0.100 g of dried seaweed was added to solution. Then the suspended solution was shaken for 60 minutes at the speed of 200 rpm. The residual metal solution was separated from biomass by filter paper. Finally, the residual concentration of the metal was measured by AAS. The 1/qe and 1/Ce for each metal ion were calculated from Table 3.18-3.21. The Langmuir adsorption isotherm for each metal was obtained by plotting 1/qe against 1/Ce. Namely, the log qe and log Ce for each metal ion were calculated from Table 3.18-3.21. The Freundlich adsorption isotherm for each metal was obtained by plotting log qe against log Ce.

2.6.3 Performance characteristics of modified seaweed

The modified seaweed was studied by the same manner as those of dried seaweed (non-modified seaweed). The optimum conditions of adsorption and its isotherm characteristics were carried out as same manner as described in sections 2.6.1.and 2.6.2 For optimization, the initial concentrations of Pb(II), Cr(III), Cu(II), Cd(II) ions were 40.00, 15.00, 15.00 and 25.00 mg/l, respectively.

In the case of isotherm studies, the initial concentrations of Pb(II), Cr(III), Cu (II) and Cd(II) ions were 30.00-80.00, 10.00-30.00, 10.00-30.00 and 20.00-40.00 mg/l, respectively. The 1/qe and 1/Ce for each metal ion were calculated from Table 3.36-3.39. The Langmuir adsorption isotherm for each metal was obtained by plotting 1/qe against 1/Ce. Namely, the log qe and log Ce for each metal ion were calculated from Table 3.36-3.39. The Freundlich adsorption isotherm for each metal was obtained by plotting log qe against log Ce.

2.6.4 Adsorption of metals in wastewater samples by non-modified seaweed and modified seaweed

The non-modified seaweed or modified seaweed (0.100 g, 0.21-0.36 mm) and 50.0 ml of wastewater samples collecting from two industries, i.e Chromium Plating in Nakorn Pathom and wastewater treatment plant in Lamphun were added to a polyethylene bottle. The solution was adjusted to the appropriate pH using 0.1 M HCl or 0.1 M NaOH. Then the suspended solution was shaken for 60 minutes at the speed of 200 rpm. The supernatant was separated from the biomass by filter paper and analyzed for residual concentrations of metals by ICP-OES.

Table 2.2 Experimental design of Pb(II) adsorption in the presence of competitive metals

Target metal : competitive metals	Concentration of metal (mg/l)								
	Ratio (1.0 : 0.5)			Ratio (1.0 : 1.0)			Ratio (1.0 : 1.5)		
	Cu(II)	Cr(III)	Cd(II)	Cu(II)	Cd(II)	Cr(III)	Cu(II)	Cd(II)	Cr(III)
Pb(II) : Cu(II)	5.0	-	-	10.0	-	-	15.0	-	-
Pb(II) : Cr(III)	-	5.0	-	-	10.0	-	-	15.0	-
Pb(II) : Cd(II)	-	-	5.0	-	-	10.0	-	-	15.0
Pb(II) : Cr(III) : Cu(II)	5.0	5.0	-	10.0	10.0	-	15.0	15.0	-
Pb(II) : Cr(III) : Cd(II)	-	5.0	5.0	-	10.0	10.0	-	15.0	15.0
Pb(II) : Cd(II) : Cu(II)	5.0	-	5.0	10.0	-	10.0	15.0	-	15.0
Pb(II) : Cr(III) : Cd(II) : Cu(II)	5.0	5.0	5.0	10.0	10.0	10.0	15.0	15.0	15.0

Initial concentration of target metal = 10.00 mg/l

Table 2.3 Experimental design of Cu(II) adsorption in the presence of competitive metals

Target metal : competitive metals	Concentration of metal (mg/l)								
	Ratio (1.0 : 0.5)			Ratio (1.0 : 1.0)			Ratio (1.0 : 1.5)		
	Pb(II)	Cr(III)	Cd(II)	Pb(II)	Cr(III)	Cd(II)	Pb(II)	Cr(III)	Cd(II)
Cu(II) : Pb(II)	5.0	-	-	10.0	-	-	15.0	-	-
Cu(II) : Cr(III)	-	5.0	-	-	10.0	-	-	15.0	-
Cu(II) : Cd(II)	-	-	5.0	-	-	10.0	-	-	15.0
Cu(II) : Pb(II) : Cr(III)	5.0	5.0	-	10.0	10.0	-	15.0	15.0	-
Cu(II) : Cr(III) : Cd(II)	-	5.0	5.0	-	10.0	10.0	-	15.0	15.0
Cu(II) : Pb(II) : Cd(II)	5.0	-	5.0	10.0	-	10.0	15.0	-	15.0
Cu(II) : Cr(III) : Cd(II) : Pb(II)	5.0	5.0	5.0	10.0	10.0	10.0	15.0	15.0	15.0

Initial concentration of target metal = 10.00 mg/l

Table 2.4 Experimental design of Cd(II) adsorption in the presence of competitive metals

Target metal : competitive metals	Concentration of metal (mg/l)								
	Ratio (1.0 : 0.5)			Ratio (1.0 : 1.0)			Ratio (1.0 : 1.5)		
	Pb(II)	Cr(III)	Cu(II)	Pb(II)	Cr(III)	Cu(II)	Pb(II)	Cr(III)	Cu(II)
Cd(II) : Pb(II)	5.0	-	-	10.0	-	-	15.0	-	-
Cd(II) : Cr(III)	-	5.0	-	-	10.0	-	-	15.0	-
Cd(II) : Cu(II)	-	-	5.0	-	-	10.0	-	-	15.0
Cd(II) : Pb(II) : Cr(III)	5.0	5.0	-	10.0	10.0	-	15.0	15.0	-
Cd(II) : Cr(III) : Cu(II)	-	5.0	5.0	-	10.0	10.0	-	15.0	15.0
Cd(II) : Pb(II) : Cu(II)	5.0	-	5.0	10.0	-	10.0	15.0	-	15.0
Cd(II) : Pb(II) : Cr(III) : Cu(II)	5.0	5.0	5.0	10.0	10.0	10.0	15.0	15.0	15.0

Initial concentration of target metal = 10.00 mg/l

Table 2.5 Experimental design of Cr(III) adsorption in the presence of competitive metals

Target metal : competitive metals	Concentration of metal (mg/l)								
	Ratio (1.0 : 0.5)			Ratio (1.0 : 1.0)			Ratio (1.0 : 1.5)		
	Pb(II)	Cd(II)	Cu(II)	Pb(II)	Cd(II)	Cu(II)	Pb(II)	Cd(II)	Cu(II)
Cr(III) : Pb(II)	5.0	-	-	10.0	-	-	15.0	-	-
Cr(III) : Cd(II)	-	5.0	-	-	10.0	-	-	15.0	-
Cr(III) : Cu(II)	-	-	5.0	-	-	10.0	-	-	15.0
Cr(III) : Pb(II) : Cd(II)	5.0	5.0	-	10.0	10.0	-	15.0	15.0	-
Cr(III) : Cd(II) : Cu(II)	-	5.0	5.0	-	10.0	10.0	-	15.0	15.0
Cr(III) : Pb(II) : Cu(II)	5.0	-	5.0	10.0	-	10.0	15.0	-	15.0
Cr(III) : Pb(II) : Cd(II) : Cu(II)	5.0	5.0	5.0	10.0	10.0	10.0	15.0	15.0	15.0

Initial concentration of target metal = 10.00 mg/l