



## SORPTION OF Pb(II), Cu(II) AND Cd(II) BY BIOMASS OF THE DIFFERENT ACTIVATED SLUDGE

Nguyen Dinh Trung<sup>1</sup>, Truong Dong Phuong<sup>1</sup> and Ning Ping<sup>2</sup>

<sup>1</sup>Institute of Environmental research, Dalat University, Vietnam

<sup>2</sup>Kunming University of Science and Technology Faculty of environmental Science and Engineering Kunming 650093, Yunnan, China

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### ABSTRACT

Biomass of digested sludge and resting sludge can be easily applied as a cheap bio-adsorbent for heavy metal ions such as  $Pb^{2+}$ ,  $Cu^{2+}$  and  $Cd^{2+}$ . Langmuir model was found to fit the experimental adsorption and desorption data of the heavy metal onto the biomass of sludge. There was competition uptake between the difference metal with another when the adsorption system has the di-metal, and tri-metal combination. The maximal adsorption capacity of resting sludge was 0.517 mM  $Pb^{2+}$ /g; 0.552 mM  $Cu^{2+}$ /g; 0.389 mM  $Cd^{2+}$ /g, and the maximal adsorption capacity of the digested sludge was 0.478 mM  $Pb^{2+}$ /g, 0.43 mM  $Cu^{2+}$ /g and 0.549 mM  $Cd^{2+}$ /g. In the absorption system, it was found that, di-metal combine such as ( $Cu^{2+}$  -  $Cd^{2+}$ ), ( $Pb^{2+}$  -  $Cd^{2+}$ ), and ( $Cu^{2+}$  -  $Pb^{2+}$ ), the metal uptake  $Cu^{2+} \approx 3Cd^{2+}$ ,  $Pb^{2+} > 3Cd^{2+}$ , and  $Cu^{2+} \approx Pb^{2+}$  respectively. When tri-metal  $Cu^{2+}$  -  $Cd^{2+}$  -  $Pb^{2+}$  combines the number of complex formation varies in the following order:  $Pb^{2+} > Cu^{2+} > Cd^{2+}$ . Extracellular polymeric substances (EPS) were extracted by sonication method. Humic substances were greater than protein and polysaccharide for digested sludge, for resting sludge the protein is greater than humic. The highest ratio protein/polysaccharide varied from 1.25 to 2.7 for resting sludge. Affinity absorption of  $Cu^{2+}$  and  $Pb^{2+}$  was higher than  $Cd^{2+}$ , which have the competition metal uptake with functional groups such as hydroxyl, carbonyl, amide on the biomass surface of the sludge.

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## 1 INTRODUCTION

Bio-sorption is potentially an attractive technology for treatment of municipal or industrial wastewaters for the separation and recovery of heavy metal ions. Although the traditional role of municipal treatment plants was to remove the soluble and colloidal organic matter, heavy metals are also frequently present in the municipal wastewater (Patterson *et al.*, 1975). The activated sludge process is the most commonly used in wastewater

treatment. Activated sludge flocs are composed of micro-organisms, organic polymers, colloids, mineral particles and ionic components e.g. divalent cations (Erikson *et al.*, 1992; Urbain *et al.*, 1993). Colloidal materials (Extracellular polymeric substances (EPS), bacteria and mineral particles) have the capacity to aggregate and to form flocs. Their structure is based on the relation between micro-organisms, EPS and divalent cations. EPS have a complex biochemical composition; they result from bacterial excretion, cell lysis or are contained in the

effluents. Their composition has been shown to play an important role in flocculation (Sobeck *et al.*, 2002) and in heavy metal adsorption onto activated sludge (Brown *et al.*, 1982; Guibaud *et al.*, 1999). In general, heavy metals at trace concentrations are known to have no detrimental effect on micro-organisms in the activated sludge process, and are up taken by microbial cells as essential micronutrients, involving ion exchange, adsorption and complexation (Bagby *et al.*, 1981; Yetiş *et al.*, 1989; Gökçay *et al.*, 1991). Chua *et al.* (1999) reported that adsorption capacity and rate of copper, chromium, lead and zinc on microbial flocs were much faster than that of organic matter.

Effluents are often contaminated by heavy metals, originate from industries, rainwater or domestic effluents. Toxic metal compounds are frequently used in industrial processes and are widely distributed in the environment. Due to their extended persistence in biological systems and tendency to bio-accumulate as they move up the food chain, which represent important environmental and occupational hazards.

The removal of toxic contaminants from industrial wastewaters is one of the most important environmental issues. Adsorption is a well-established technique for heavy metals removal. Activated carbon is the most efficient and popular adsorbent and has been used with great success for the removal of heavy metal, but due to its high regeneration cost and losses in the application processes, it cannot be used on a great scale (Lee *et al.*, 1998).

At present, there is growing interest in using low cost, non-conventional alternative materials, including yeast biomass, clays, sawdust, etc., instead of activated carbon for heavy metals removal from wastewaters (Li *et al.*, 2003; Tsai *et al.*, 2004). The activated sludge biomass was used as adsorbent also approved of a potential alternative to existing methods for heavy metal removal. Resting part sludge and digested part sludge were used as a bio-sorbent for the removal of  $Pb^{2+}$ ,  $Cu^{2+}$  and  $Cd^{2+}$  ions from aqueous solution, the relation between their composition to be complex to heavy metals were investigated.

## 2 MATERIALS AND METHODS

### 2.1 Materials

Activated sludge was collected from Lam Dong Hospital wastewater treatment plants which was divided into two parts:

Resting part: the activated sludge has just been collected from Lam Dong Hospital wastewater treatment plants, which was placed in an aerated

basin, keeping dissolved oxygen  $> 2$  mg/L for 1 month with no external nutrients at 25°C.

Digested part: another equal part of original sludge was placed in an anaerobic basin kept at 25°C for 1 month without adding any nutrients.

All different types of sludge were concentrated using the centrifuge at 2000 rpm for 15 minutes, that bio-sorbent will be used by research below.

Analytical grades of  $HNO_3$ ,  $NaOH$ ,  $Pb(NO_3)_2$ ,  $Cu(NO_3)_2$  and  $Cd(NO_3)_2$  (Merck) were used. Heavy metals in solutions were measured by Atomic Absorption Spectrometer (AAS) (AA 7000 Shimadzu).

UV-colorimeter was used for determining the biochemical composition of the EPS.

### 2.2 Extraction and determination of EPS

The dry weight content (DW) at temperature 105°C was determined in accordance with Standard Methods for the Examination of Water and Wastewater (APHA, 1992).

Prior to extraction, the biomass of sludge was concentrated using the centrifuge at 2000 rpm for 15 minutes. The residues were recovered and suspended again in their original volume (300 mL) in a buffer solution. The buffer consisted of 2 mM  $Na_3PO_4$ , 4 mM  $NaH_2PO_4$ , 9 mM  $NaCl$  and 1 mM  $KCl$  at pH 7. The sludge (300 mL) samples were then sonicated at 40 W for 2 minutes, repeated two times. The extracted EPS in the supernatant was harvested by centrifugation at 7500 rpm for 15 minutes. Extraction of EPS from sludge was carried out using sonication.

The biochemical composition of the EPS was determined using the following colorimetric methods. The carbohydrate content is measured using the phenol-sulfuric acid method; glucose was used as standard (APHA, 1992). The protein content is measured using the Lowry method, bovine albumin serum was used as standard (Dubois, 1956) and humic substances are measured using a modified Lowry method, humic acid was used as standard (Lowry, 1951; Frolund *et al.*, 1995; Frelund *et al.*, 1995).

### 2.3 Mono-heavy metal $Pb^{2+}$ , $Cu^{2+}$ and $Cd^{2+}$ absorption

#### 2.3.1 Determination of adsorption isotherms

Bio-sorbent (4 g) was re-suspended in solutions containing heavy metal concentrations of 0-500 mg/L. After 24 h, samples were taken from the solutions, and the heavy metal concentration in supernatants was measured by AAS.

The Langmuir isotherm, equation (1), was used to describe the adsorption equilibrium of three metal ions shown in equation (1):

$$q = \frac{q_m b C_e}{1 + b C_e} \quad (1)$$

Where q is the amount of metal adsorbed, mg/g (dry weight);  $q_m$  is the maximum metal uptake value corresponding to sites saturation, mg/g (dry weight);  $C_e$  is the equilibrium metal concentration in solution, mg/L; and b is the ratio of adsorption/desorption rates.

### 2.3.2 Time-course of bio-sorption

The bio-sorbent (4 g wet biomass) was re-suspended in 100 mL of heavy metal solution in a glass container, which was gently shake (60 rpm/min) at 25°C. Samples were taken from the solution at desired intervals 60 minutes each time and subsequently centrifuged at 7500 rpm for 10 minutes. The heavy metals concentration in the resulting supernatant was determined.

The amount of adsorbed metal was calculated following equation (2):

$$q = \frac{V(C_i - C_e)}{B} \quad (2)$$

Where q is the metal uptake or sorption capacity of biomass (in mg or mM/g of biomass);  $C_i$  and  $C_e$  are the metal concentrations before and after adsorption, respectively, B is the mass of bio-sorbent used and V is the used solution volume.

### 2.3.3 Time-course desorption

After bio-sorption experiments, the metal-loaded bio-sorbents were harvested from the biomass/metal solutions initially containing 250 mg  $Pb^{2+}/L$ , 180 mg  $Cu^{2+}/L$  and 180 mg  $Cd^{2+}/L$ , respectively. The bio-sorbents were then washed and re-suspended with distilled water. Amounts of 0.1

M HCl were added into solutions of metal-loaded biomass to adjust the pH value to 2.0. After gentle shake (60 rpm), samples (2 mL) were taken from the suspensions at designated time intervals. The samples were centrifuged and the metal concentration in the supernatant was determined.

### 2.3.4 Adsorption/desorption cycle

The bio-sorbents (4 g) of two types biomass of sludge were shook with the solution contain 250 mg  $Pb^{2+}/L$ , 180 mg  $Cu^{2+}/L$  and 180 mg  $Cd^{2+}/L$ , respectively. After 24 h, the metal-loaded biomass was harvested, washed with distilled water and re-suspended in distilled water. The pH of the suspensions was adjusted to 2.0 by HCl (0.1 M) in order to recover the metal ions from the cells. After being gently shake (60 rpm) for 1-12 h, the bio-sorbent suspensions were centrifuged, and the metal concentration in the supernatant was measured. The regenerated bio-sorbents were again suspended in metal containing solutions for the next adsorption run. The adsorption/desorption steps described above were repeated four times.

## 2.4 Binary and trinary metals sorption test

Wet bio-sorbents (4 g) was transferred into a 250 mL conic flask containing 150 mL of binary, trinary metal-bearing solution, (the heavy metal concentrations were 250 mg  $Pb^{2+}/L$ , 180 mg  $Cu^{2+}/L$  and 180 mg  $Cd^{2+}/L$ , respectively) after 24 h sorption at 25°C, the heavy metals concentration in the resulting supernatant was determined.

## 3 RESULTS AND DISCUSSION

### 3.1 The biomass composition of the different activated sludge

Extraction of EPS from the different sludge samples was carried out with sonicating method, the carbohydrate, protein content and humic substances were measured.

**Table 1: Component of EPS**

Sludge	Protein (mg/gDW)	Polysaccharide (mg/gDW)	Humic (mg/gDW)	PN/PS
Digested sludge	97.8 ± 0.8	78.08 ± 0.3	181.6 ± 1.7	1.25
Resting sludge	377 ± 1.6	137.9 ± 1.2	171.2 ± 6.6	2.7

The result shown that, the humic substance was greater than protein and polysaccharide for digested sludge, on the contrary, for resting sludge, the protein was greater than humic (Table 1). This is in agreement with many researchers that used this method to extract EPS. The highest ratio protein/polysaccharide (PN/PS) varied from 1.25 to 2.70 found in resting sludge. Guibaud *et al.* (1999) showed a correlation existing between the parame-

ters of complexation and the composition in proteins, in humic substances and in polysaccharides. There was no correlation between lipids, nucleic acids and uronic acids contents and their affinity for metals. The lipid content of EPS may be too low to have a significant influence on metal complexation. Fukushi *et al.* (2001) proved the important role of the proteins in the fixation of metals ions. There is relation between humic substances

contained in the EPS and their affinity for the metals investigated. Humic substances were shown to be strongly implicated in metal complexation, for divalent and trivalent cations, particularly, in several environmental elements such as soils, sediments and natural water (Varrault *et al.*, 2002). Goodwin and Forster (1989) showed the affinity sorption for these metals by of polysaccharides.

### 3.2 Mono-heavy metal Pb<sup>2+</sup>, Cu<sup>2+</sup> and Cd<sup>2+</sup> absorption

#### 3.2.1 Bio-sorption isotherms

The correlation between adsorption isotherms and the biomass of the sludge for Pb<sup>2+</sup>, Cu<sup>2+</sup> and Cd<sup>2+</sup> are shown in Figure 1. The result of bio-sorption of Pb<sup>2+</sup>, Cu<sup>2+</sup> and Cd<sup>2+</sup> by biomass of sludge are shown in the Table 2.

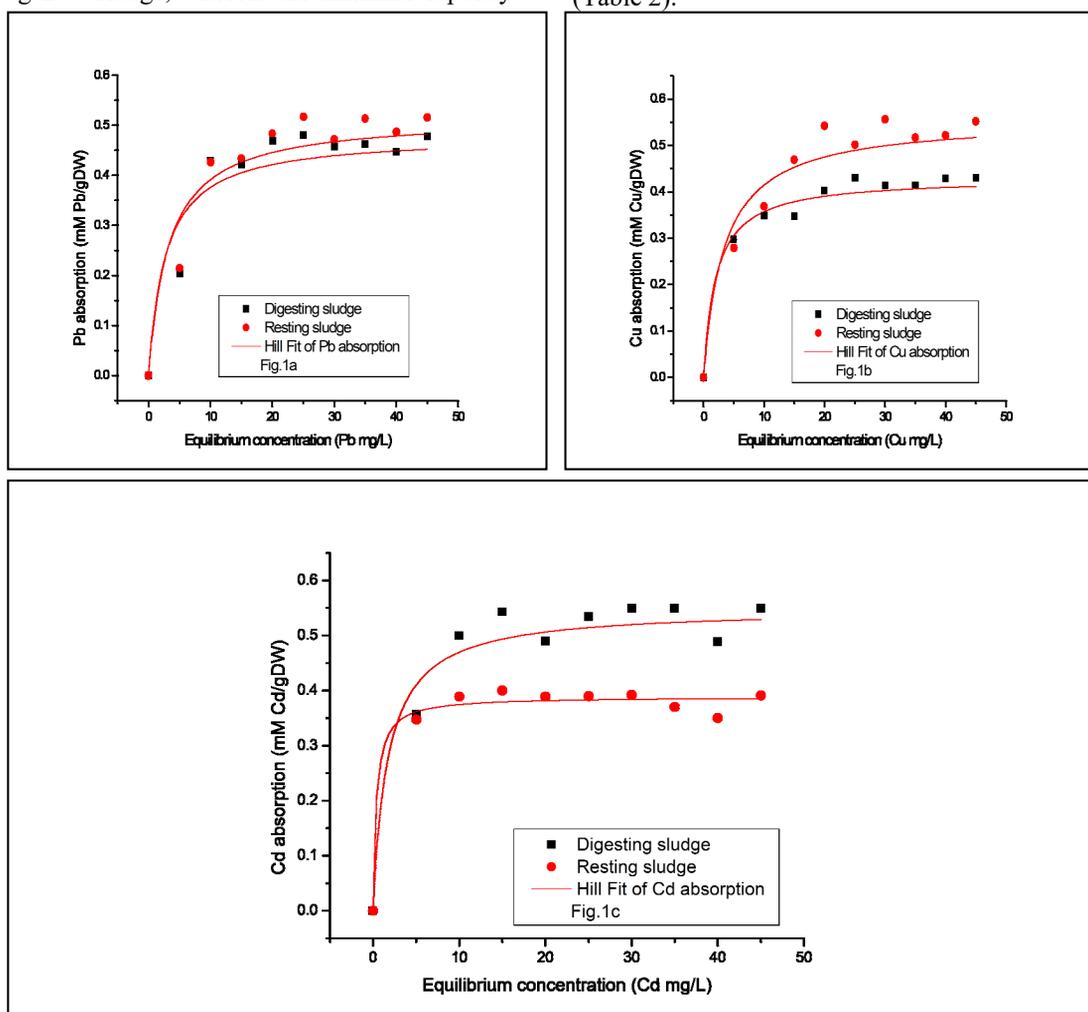
**Table 2: Parameters of the Langmuir isotherms for bio-sorption of Pb<sup>2+</sup>, Cu<sup>2+</sup> and Cd<sup>2+</sup> by biomass of sludges**

Metals	Digested sludge		Resting sludge			
	q <sub>max</sub> (mg/gDW)	q <sub>max</sub> (mM/gDW)	b	q <sub>max</sub> (mg/gDW)	q <sub>max</sub> (mM/gDW)	b
Pb <sup>2+</sup>	99.1	0.478	0.055	107	0.517	0.047
Cu <sup>2+</sup>	27.3	0.430	0.087	35.1	0.552	0.046
Cd <sup>2+</sup>	61.7	0.549	0.108	43.7	0.389	0.411

#### Lead:

As indicated in Figure 1a, the maximal adsorption capacity of resting sludge was nearly 0.517 mM Pb<sup>2+</sup>/g DW sludge, whereas the maximal capacity

of digested sludge was 0.478 mM Pb<sup>2+</sup>/g DW sludge. The metal-biomass affinity for two types of the sludge was not much different, indicated the constant of the adsorption/desorption b value (Table 2).



**Fig. 1: The Langmuir isotherms for bio-sorption of Pb<sup>2+</sup>, Cu<sup>2+</sup> and Cd<sup>2+</sup> by biomass of sludge**

**Copper:**

Isotherms for the adsorption of  $\text{Cu}^{2+}$  are illustrated in Figure 1b as the bio-sorption of  $\text{Pb}^{2+}$ , maximal  $\text{Cu}^{2+}$  adsorption capacities for resting sludge was higher than digested sludge. The maximal  $\text{Cu}^{2+}$  adsorption was 0.43 mM/g DW and 0.552 mM/g DW for digested sludge and resting sludge respectively.

**Cadmium:**

Isotherms for the adsorption of  $\text{Cd}^{2+}$  are shown in Figure 1c, unlike the bio-sorption of  $\text{Pb}^{2+}$  and  $\text{Cu}^{2+}$ , a maximal  $\text{Cd}^{2+}$  adsorption capacity for resting sludge was lower than digested sludge. The maximal adsorption of  $\text{Cd}^{2+}$  appears to be 0.549 mM/g DW for digested sludge and 0.389 mM/g DW for resting sludge.

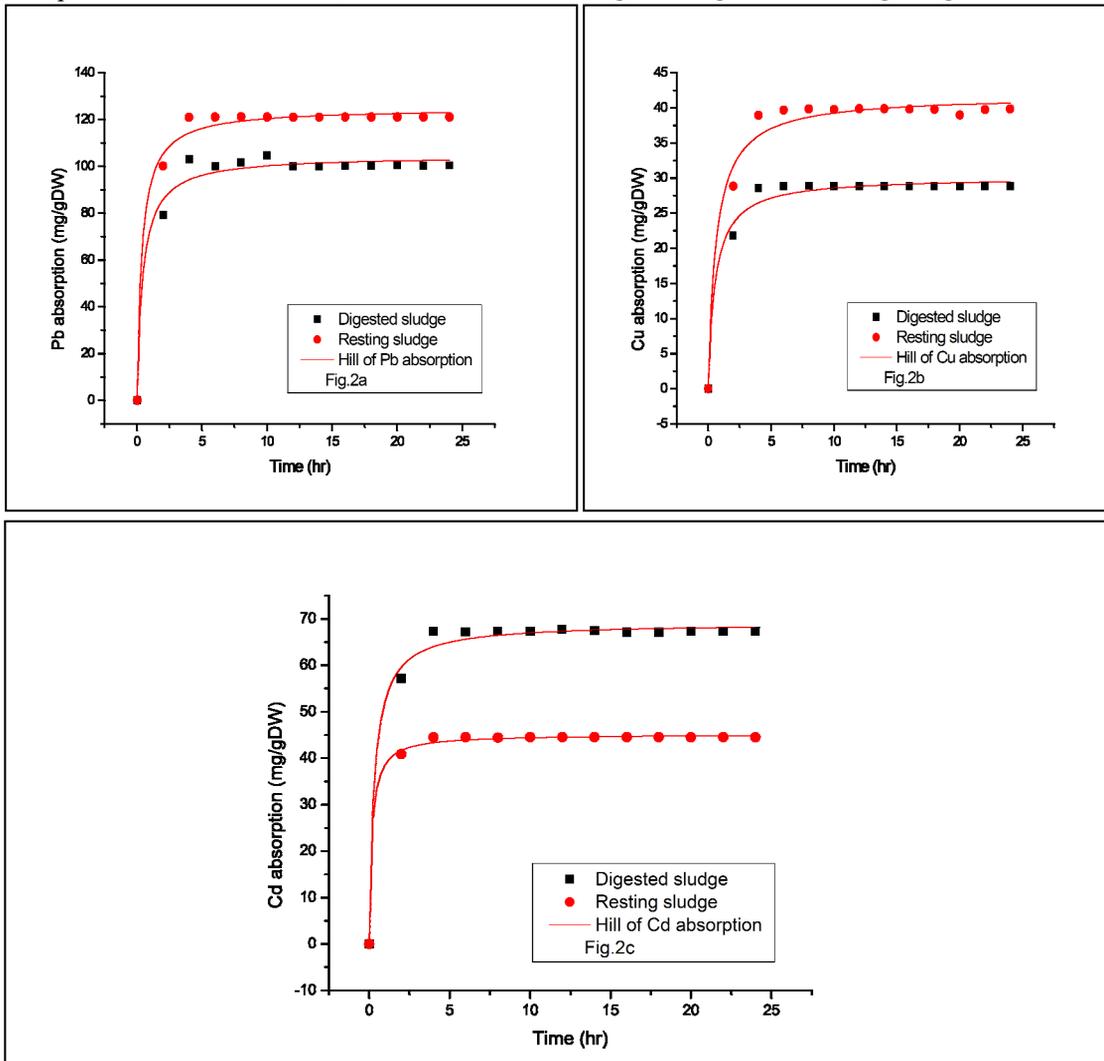
$\text{Cd}^{2+}$ -biomass reached the highest affinity when it was compared with  $\text{Pb}^{2+}$  and  $\text{Cu}^{2+}$ , illustrated that

adsorption/desorption rate was high or the  $\text{Cd}^{2+}$  bio-sorption changed a lot during the bio-sorption process.

The bio-sorption of  $\text{Cd}^{2+}$ ,  $\text{Cu}^{2+}$  and  $\text{Pb}^{2+}$  by the biomass of two sludges, when each ion metal was present alone, the number of complex formation varied in the following order:  $\text{Cu}^{2+} > \text{Pb}^{2+} > \text{Cd}^{2+}$  for resting sludge but in contrary with digested sludge.

*3.2.2 Time-course of bio-sorption*

Time-course profiles for the adsorption of  $\text{Pb}^{2+}$ ,  $\text{Cu}^{2+}$  and  $\text{Cd}^{2+}$  by digested sludge and resting sludge are shown in Figure 2. In all cases, the ion metals adsorbed rapidly during the first hour and remained nearly constant after 2 h. The metal capacity adsorption for resting sludge higher than that of digested sludge for  $\text{Pb}^{2+}$  and  $\text{Cu}^{2+}$  (Fig. 2a, Fig. 2b). On the contrary,  $\text{Cd}^{2+}$  adsorption in digested sludge was higher than resting sludge.



**Fig. 2:** Time-course profiles of bio-sorption of  $\text{Pb}^{2+}$ ,  $\text{Cu}^{2+}$  and  $\text{Cd}^{2+}$  by digested and resting sludges

The constituent both of digested and resting sludges are mainly proteins carbohydrates and phenolic compounds which contain function groups such as carboxyl, hydroxyl and amine (Table 1), in Figure 2c, Cd<sup>2+</sup> sorption was attributed to complexation of Cd<sup>2+</sup> with functional groups of proteins, polysaccharides, lipids and humic acids of biomass. The results agree with Guibaud *et al.* (2003), who demonstrated that the number of binding sites and the complexation constant were strongly linked to proteins, polysaccharides and humic substances content.

### 3.2.3 Time-course desorption

The kinetics of desorption of Pb<sup>2+</sup>, Cu<sup>2+</sup> and Cd<sup>2+</sup> from the heavy-metal-loaded biomass are demonstrated in Figure 3. It can clearly be seen that metals desorbed rapidly, and desorption reached equilibrium around 5-10 minutes. The efficiency desorption was 95% for Pb<sup>2+</sup> and 89% for Cu<sup>2+</sup>, whereas only 80% of Cd<sup>2+</sup> was recovered. The results agree with Al-Qodah *et al.* (2006) for Cd<sup>2+</sup> and Cu<sup>2+</sup>.

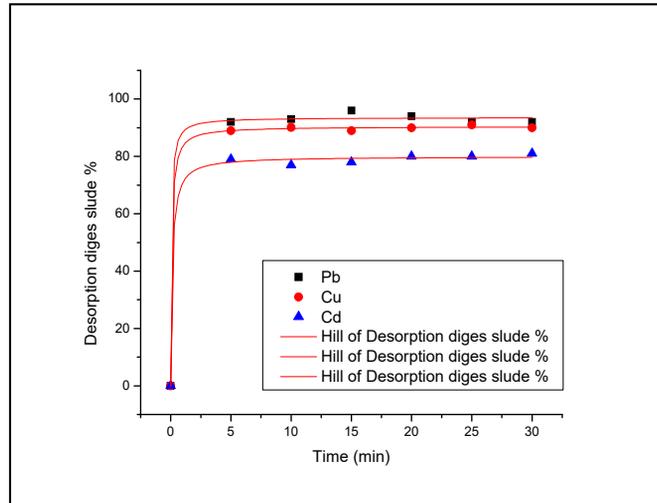


Fig. 3: Time-course profiles for the desorption of Pb<sup>2+</sup>, Cu<sup>2+</sup> and Cd<sup>2+</sup> from metal-loaded

### 3.2.4 Adsorption/desorption cycle

The result in Figure 4 indicated the loss of bio-sorption capacity by resting sludge was faster than

digested sludge, which could be due to the surface biomass of the resting sludge changed more than after each cycle.

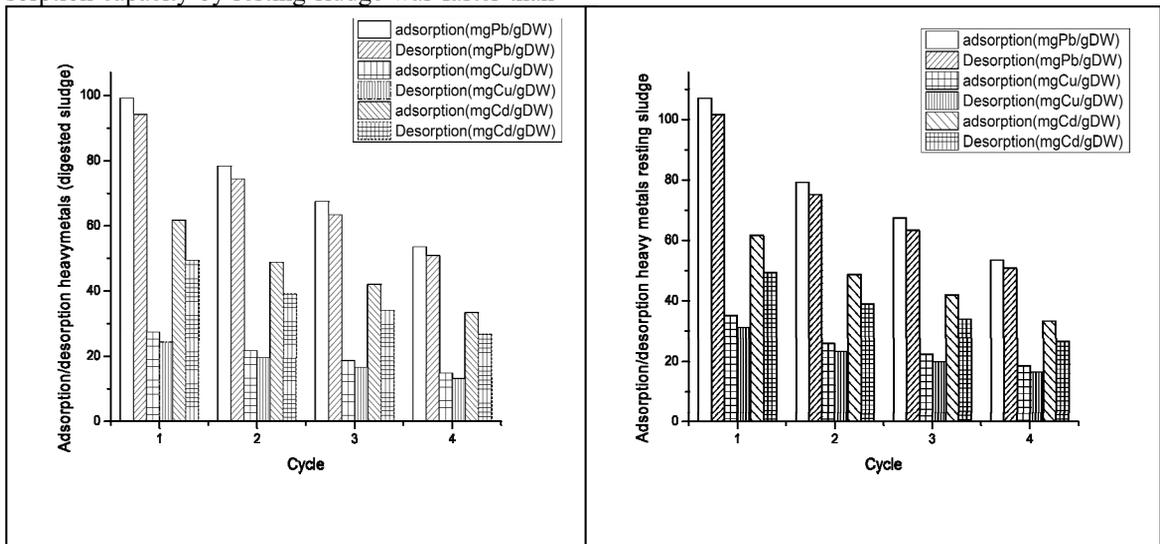


Fig. 4: Adsorption/desorption heavy metals versus cycles

Whatever type of sludge, the loss bio-sorption capacity after four cycles can be the interaction between the heavy metals and the functional groups of the biomass changed, and there was the loss of the functional groups after each cycle.

It was impossible to reuse the biomass of the sludge in a new sorption-desorption cycle due to the lack of bio-sorbent capacity. This fact points

out that hydrochloric acid exerts on molecules of biological tissues of the biomass, so they will be destroyed.

### 3.3 Binary and trinary metals sorption test

The competition uptake between the difference metal with another when the adsorption system has the di-metal, and tri-metal combination, the results are shown in Table 3.

**Table 3: Binary and trinary metals sorption**

Heavy metals	M.W	Digested sludge (mM/g DW)	Resting sludge (mM/g DW)
<b>Pb<sup>2+</sup> - Cu<sup>2+</sup></b>			
Pb <sup>2+</sup>	207.12	0.318	0.369
Cu <sup>2+</sup>	63.54	0.304	0.280
<b>Pb<sup>2+</sup> - Cd<sup>2+</sup></b>			
Pb <sup>2+</sup>	207.12	0.450	0.459
Cd <sup>2+</sup>	112.4	0.149	0.119
<b>Cu<sup>2+</sup> - Cd<sup>2+</sup></b>			
Cu <sup>2+</sup>	63.54	0.452	0.488
Cd <sup>2+</sup>	112.4	0.151	0.144
<b>Cu<sup>2+</sup> - Cd<sup>2+</sup> - Pb<sup>2+</sup></b>			
Cu <sup>2+</sup>	63.54	0.271	0.296
Pb <sup>2+</sup>	207.12	0.266	0.337
Cd <sup>2+</sup>	112.4	0.092	0.109

M.W: Mol weight

#### 3.3.1 Di-metal combination

##### Cu<sup>2+</sup> - Pb<sup>2+</sup> system

When Pb<sup>2+</sup> presented in the system, the Cu<sup>2+</sup> uptake decreased, the maximal adsorption of Cu<sup>2+</sup> changed from 0.430 to 0.304 and 0.552 to 0.280 mM/g DW for digested and resting sludge respectively (Table 2, Table 3). In the absorption system, there was competition uptake between Cu<sup>2+</sup> and Pb<sup>2+</sup>.

##### Pb<sup>2+</sup> - Cd<sup>2+</sup> system

When Pb<sup>2+</sup> presented in the system, the Cd<sup>2+</sup> uptake decreased from 0.546 to 0.149 mM/g DW for digested sludge and 0.389 to 0.119 mM/g DW for resting sludge, whereas Pb<sup>2+</sup> did not significantly decrease (Table 2, Table 3). With high levels of the overall metal concentration present in the solution, the bio-sorbent easily reached the saturation level, but there was the competition uptake between Cd<sup>2+</sup> and Pb<sup>2+</sup> in the system; about 75% of the total metal uptake due to Pb<sup>2+</sup>.

##### Cu<sup>2+</sup> - Cd<sup>2+</sup> system

At equilibrium, the Cd<sup>2+</sup> uptake decreased from 0.546 to 0.151 mM/g DW and 0.389 to 0.144 mM/g DW for digested sludge and resting sludge respectively, whereas Cu<sup>2+</sup> uptake decreased from 0.552 to 0.488 mM/g DW for resting sludge, but for digested sludge, the Cu<sup>2+</sup> uptake was not

changed (Table 2, Table 3). The result indicated that, when di-metal combined for example Cu<sup>2+</sup>-Pb<sup>2+</sup>, the amount of Pb<sup>2+</sup> complexation was greater than Cu<sup>2+</sup> for both types of sludges. The amount heavy metal absorption was Cu<sup>2+</sup> ≈ 3Cd<sup>2+</sup> and Pb<sup>2+</sup> > 3Cd<sup>2+</sup> with Cu<sup>2+</sup>-Cd<sup>2+</sup> system and Pb<sup>2+</sup>-Cd<sup>2+</sup> system respectively. The results showed that the difference between the bio-sorption abilities of the type of sludge toward metals caused by its component. With high levels of overall metal concentration present in the solution, the bio-sorbent easily reached the saturation level, but there was competition uptake between Cd<sup>2+</sup> and Cu<sup>2+</sup> in the system; about 75% of the total metal uptake was due to Cu<sup>2+</sup>.

#### 3.3.2 Tri-metal combination (Cu<sup>2+</sup> - Pb<sup>2+</sup> - Cd<sup>2+</sup>)

From the result in Table 3, there was competition between three metals with each other in the system, but affinity absorption of Cu<sup>2+</sup> and Pb<sup>2+</sup> was higher than Cd<sup>2+</sup>, so the Cd<sup>2+</sup> uptake decreased from 0.546 to 0.092 mM/g DW and 0.389 to 0.19 mM/g DW for digested sludge and resting sludge respectively. Affinity absorption of Cu<sup>2+</sup> and Pb<sup>2+</sup> was higher than Cd<sup>2+</sup>, there may be a competition metal uptake with functional groups such as hydroxyl, carbonyl, amide on the biomass surface of the sludge. Those functional chemistry groups played the most important role in binding Cu<sup>2+</sup> and Pb<sup>2+</sup> (Xuejiang *et al.*, 2006).

#### 4 CONCLUSIONS

Biomass of digested and resting sludges can be easily applied as a cheap adsorbent for heavy metal ions such as  $\text{Cd}^{2+}$ ,  $\text{Cu}^{2+}$  and  $\text{Pb}^{2+}$ . Langmuir model was found to fit the experimental adsorption and desorption data of the heavy metals onto the biomass of two types sludge. The maximal adsorption capacity of (each g DW) resting sludge was nearly  $0.517 \text{ mM Pb}^{2+}$ ;  $0.552 \text{ mM Cu}^{2+}$ ;  $0.389 \text{ mM Cd}^{2+}$ , whereas, the maximal adsorption capacity of the digested sludge was  $0.478 \text{ mM Pb}^{2+}$ ,  $0.43 \text{ mM Cu}^{2+}$  and  $0.549 \text{ mM Cd}^{2+}$ . When have di-metal, tri-metal combination appeared in adsorption systems, there was competition uptake between the one metal with another in the system.

The results indicate that the main uptaken metals were  $\text{Cu}^{2+}$ ,  $\text{Pb}^{2+}$ . When there was competition between three metals with each other In the system, affinity absorption of  $\text{Cu}^{2+}$  and  $\text{Pb}^{2+}$  was higher than  $\text{Cd}^{2+}$ . There may be a competition of metals with functional groups such as hydroxyl, carbonyl, amide on the biomass surface of the sludge. Those functional groups played the most important role in binding  $\text{Cu}^{2+}$  and  $\text{Pb}^{2+}$  and  $\text{Cd}^{2+}$ . The results obtained indicate that it is possible to use resting sludge and digested sludge as an adsorbent, the heavy metals after uptake on the biomass of sludge can be reused.

#### REFERENCES

- APHA. 1992. Standard methods for the examination of water and wastewater, 18th edn. American Public Health Association, Baltimore, Md.
- Bagby, M.M., Sherrard, J.H., 1981. Combined effects of Cd and Ni on the activated sludge process. *J. Wat. Pollut. Control. Fed.* 53: 1609–1619.
- Box, J.D., 1983. Investigation on the Folin - Ciocalteu phenol reagent for determination of polyphenolic substances in natural waters. *Water Res.* 17: 511– 525.
- Brown, M.J., Lester, J.N., 1982. Role of bacterial extracellular polymers in metal uptake in pure bacterial culture and activated sludge: effect of metal concentration. *Water Res.* 16: 1539–1548.
- Chua, H., Yu, P.H.F., Sin, S.N., Cheung, M.W.L., 1999. Sub-lethal effects of heavy metals on activated sludge microorganisms. *Chemosphere.* 39: 2681–2692.
- Dubois, M., Giles, K.A., Hamilton, J.K, Rebers, P.A., Smith, F., 1956. *Anal. Chem.* 28: 350-356.
- Erikson, L., Steen, I., Tendaj, M., 1992. Evaluation of sludge properties at an activated sludge plant. *Water Sci. Technol.* 25: 251–265.
- Frolund, B., Palmgren, R., Keiding, K., Nielsen, P.H., 1995. Extraction of activated sludge exopolymers by a cation exchange resin. *Water Res* (in press).
- Frolund, B., Gribe, T., Nielsen, PH., 1995. Enzymatic activity in the activated sludge floc matrix. *Appl Microbiol Biotechnol* (in press).
- Fukushi, K., Kato, S., Antsuki, T., Omura, T., 2001. Isolation of copper binding proteins from activated sludge culture. *Water Sci. Technol.* 44: 453–459.
- Gökçay, C.F., Yetiş, Ü., 1991. Effect of chromium (VI) on activated sludge. *Wat. Res.* 25: 65–73.
- Goodwin, J.A.S., Forster, C.F., 1989. A further examination into the composition of activated sludge surfaces in relation to their settlement characteristics. *Water Res.* 20: 1427–1528.
- Guibaud, G., Tixier, N., Bouju, A., Baudu, M., 2003. Relation between extracellular polymers composition and its ability to complex Cd, Cu and Pb. *Chemosphere.* 52: 1701–1710.
- Guibaud, G., Baudu, M., Dollet, P., Condat, M.L., Dago, C., 1999. Rôle des polymères extracellulaires dans l'adsorption du cadmium par les boues actives. *Environ. Technol.* 20: 1045–1054.
- Lee, M.Y., Shin, H.J., Lee, S.H., Park, J.M., Yang, J.W., 1998. Removal of lead in a fixed-bed column packed with activated carbon and crab shell. *Separation Science and Technology.* 33: 1043–1056.
- Li, J.Y., Shukla, S.S., Dorris, K.L., Shukla, A., Margrave, J.L., 2003. Adsorption of chromium from aqueous solutions by maple sawdust. *Journal of Hazardous Materials.* 100 (1–3): 53–56.
- Lowry, O.H., Rosebrough, N.J., Farr, A.L., Randall, R.J., 1951. Protein measurement with the Folin phenol reagent. *J Biol Chem.* 193:265-275.
- Patterson, J.W., 1975. *Wastewater Treatment Technology.* Ann Arbor Science, New York.
- Qodah, Al.Z., 2006. Biosorption of heavy metal ions from aqueous solutions by activated sludge. *Desalination.* 196: 164–176.
- Sobeck, D.C., Higgins, M.J., 2002. Examination of three theories for mechanisms of cation-induced bioflocculation. *Water Res.* 36: 527–538.
- Tsai, W.T., Lai, C.W., Hsien, K.J., 2004. Adsorption kinetics of herbicide paraquat from aqueous solution onto activated bleaching earth. *Chemosphere.* 55: 829–837.
- Urbain, V., Manem, J., Block, J.C., 1993. Bioflocculation in activated sludge: an analytical approach. *Water Res.* 27: 829–838.
- Varrault, G., Bermond, A., 2002. Trace metal sorption ability of insolubilized humic acids. *Environ. Technol.* 22: 421–428.
- Xuejiang, W., Ling, C., Siqing, X., Jianfu, Z., Chovelon, J-M., Renault, N.J., 2006. Biosorption of Cu(II) and Pb(II) from aqueous solutions by dried activated sludge, *Minerals Engineering.* 19: 968–971.
- Yetiş, Ü., Gökçay, C.F., 1989. Effect of nickel(II) on activated sludge. *Wat. Res.* 23: 1003–1007.