

# Adsorption Capacity of Vegetable Stalk Alginate Biosorbent Beads on the Removal of Pb<sup>2+</sup> and Cu<sup>2+</sup> Ions

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**Abstract-** This work concerns with the adsorption capacity of Calcium Alginate, Bagasse-Calcium Alginate and *Jatropha curcas*- Calcium Alginate Beads on the removal of Pb<sup>2+</sup> and Cu<sup>2+</sup> ion in wastewater of Myanmar industry. On the aspect of removal of Pb<sup>2+</sup> and Cu<sup>2+</sup> ions by three types of beads, the removal percent depends on pH, the mass of adsorbent dose and the time contact between adsorbent and adsorbate. The optimal pH for the removal of Pb<sup>2+</sup> was observed at 6 and at pH 5 for Cu<sup>2+</sup> ion. The equilibrium for the removal of Pb<sup>2+</sup> and Cu<sup>2+</sup> ions were reached after 30 minute in contact with the beads and metal solution. So, the prepared biosorbent beads can be used as adsorbent for the removal of heavy metals from wastewater.

**Keywords:** adsorption; three types of beads; lead; copper; adsorbent; adsorbate

## 1. INTRODUCTION

Heavy metals are defined as metals with a specific weight usually more than 5.0 g/cm<sup>3</sup>, which is five times heavier than water. The toxicity of heavy metals occurs even in low concentration of about 1.0 - 10.0 mg/L (Website 1). Heavy metals can pose health hazards to man and aquatic lives if their concentrations exceed allowable limits. Concentration of heavy metals below these limit even have potential for long-term contamination, because heavy metals are known to be accumulative within biological systems (Quek *et al.*,1998) Heavy metal pollution of aqueous streams is a major environmental problem for the modern world.

Adsorption techniques for water treatment have become more popular in recent years with regard to their efficiency in the removal of pollutants, especially heavy metal ions, colour, odor and organic pollution (Lai and Lin,2003, Steinhäuser *et al.*, 2008). Adsorption has advantages over other methods for remediation of heavy metals from contaminated water because its design is simple (Viraraghyam and Rao, 1991).

The utilized materials in this work were sugarcane bagasse and *Jatropha curcas* stalks waste. The vegetable stalk alginate biosorbent beads were found to be effective in its exchange and removal capacity towards the toxic heavy metal such as lead and copper.

Sugarcane Bagasse has high and fast adsorption capacities due to its porous structure and tremendous surface area. The main stem of the *Jatropha curcas* plant should be cut once the tree in 1 m tall. This will lead to increase branching of the tree. The more branches a plant has, the higher the production of fruits and therefore more seeds (Sukarin, Yamada and Sakaguchi.,1987). These removing or cutting branches are agricultural waste which were prepared dry *Jatropha curcas* powder and they can be used as low cost heavy metal adsorbent in this paper.



Figure 1: Photograph of sugarcane plant



Figure 2: Photograph of *Jatropha curcas* plant

## 2. EXPERIMENTAL

### 2.1 Sampling and Preparation of Vegetable Stalk Alginate Biosorbent Beads

Bagasse waste were obtained from Sugarcane plant from Ein Mae Township, Ayeyarwaddy Division and *Jatropha curcas* stalk waste were kindly supplied by Myanmar Five Star Line, Part of Terminal, Thaketa Township, Yangon in October, 2006. The waste were rinsed three times with distilled water, dried, cut and grounded to obtain a fine powder. The fine powder was sieved to get the particle size range of 105-125  $\mu\text{m}$ . These bagasse powder and *Jatropha curcas* stalk powder were stored in separately in tightly sealed bottle and they were ready to use.

In the preparation of three types of beads (calcium alginate beads, bagasse-calcium alginate beads and *Jatropha curcas*-calcium alginate beads) by using the optimum conditions were found to as 2% w/v of vegetable stalk powder, 6% sodium alginate and 10% calcium chloride. The beads were allowed to harden in this solution for 24 hr.

### 2.2 Removal of Metal Ions from Aqueous Solution

#### 2.2.1 Materials

Lead II nitrate, copper II sulphate, sulphuric acid, hydrochloric acid, sodium hydroxide, potassium iodide, sodium thiosulphate, EDTA, starch, Xylenol orange,

#### 2.2.2 Apparatus

A measuring cylinder (100 mL), bulb pipette, Erlenmeyer flasks (250 mL), pH meter (Jenway, England), electric shaker bath (0-200°C) (SBS-30, Bibby UK), balance (Sartorius AG BL 2105)

#### 2.2.3 Pretreatment

The biosorbent composite beads were dried at room temperature (25°C) and these beads (diameter  $1.6 \pm 0.2$  mm) were used in sorption experiments (Ashok, *et al.*, 2002). Stock solution containing 1000  $\text{mgL}^{-1}$  of heavy metals ions were prepared by dissolving lead II nitrate and copper II sulphate respectively. Sample solutions were prepared from the stock solution by diluting appropriate aliquots with distilled water.

The adsorption experimental studies were carried out at room temperature using conical flasks containing 2.00 g dried biosorbent composite beads and 100 mL of heavy metal ion solution.

### 2.3 Effect of pH on the Removal of Lead

#### 2.3.1 Sample

Prepared biosorbent beads (CA, BCA and JCA beads)

#### 2.3.2 Procedure

The standard stock solution (200  $\text{mg L}^{-1}$ ) of lead II nitrate solution at various pH (3, 4, 5, 6) were prepared by adding 0.1 M nitric acid and 0.1 M sodium hydroxide solution. A fixed amount of biocomposite beads, 2 g was added to 100 mL of 200  $\text{mg L}^{-1}$  lead II nitrate solution and the mixture was shaken with a shaker for 1 hr at room temperature. Then the mixture was filtered through filter paper Whatmann No.1 and  $\text{Pb}^{2+}$  ion in the filtrate was determined by complexometric titration using xylenol orange as indicator.

## **2.4 Effect of pH on the Removal of Copper**

### **2.4.1 Procedure**

The standard stock solution ( $200 \text{ mg L}^{-1}$ ) of copper II sulphate solution at various pH (3, 4, 5, 6) were prepared by adding 0.1 M sulphuric acid and 0.1 M sodium hydroxide solution respectively. A fixed amount of biocomposite beads, 2 g was added to  $200 \text{ mg L}^{-1}$  copper II sulphate solution and the mixture were shaken with a shaker for 1 hr at room temperature. Then the mixture was filtered through filter paper and  $\text{Cu}^{2+}$  ion in the filtrate was determined by iodometric titration using starch indicator.

## **2.5 Effect of Contact Time on the Removal of Lead**

### **2.5.1 Procedure**

Accurate weighed prepared biosorbent beads (2 g) were placed in separate flasks. Then 100 mL of lead stock solution ( $100 \text{ mg L}^{-1}$ ), adjusted at pH 6 was added into each flasks. The flasks were placed on an electric shaker and were shaken to reach the equilibrium. The contact time was varied at interval of 10 min, 20 min, 30 min, 40 min, 50 min and 60 min. The sample solution was separated by filtration. The residual lead content in the solution was determined by complexometric titration.

## **2.6 Effect of Contact Time on Removal of Copper**

### **2.6.1 Procedure**

Accurate weighed prepared biosorbent beads (2 g) were placed in separate flasks. Then 100 mL of copper stock solution ( $200 \text{ mg L}^{-1}$ ), adjusted a pH 5 was added into each flasks. The flasks were placed on an electric shaker and were shaken to reach the equilibrium. The contact time was varied at interval of 10 min, 20 min, 30 min, 40 min, 50 min and 60 min. The sample solution was separated by filtration. The residual copper content in the solution was determined by iodometric titration.

## **2.7 Effect of Dosage on the Removal of Lead**

### **2.7.1 Procedure**

Prepared biosorbent beads sample of various masses ranging from 1 g to 5 g were placed in the flasks and 100 mL of standard lead II nitrate solution ( $200 \text{ mg L}^{-1}$ ) was added to each flask at pH 6. And then the loaded flasks were placed on an electric shaker. In order to attain complete equilibrium, the solutions were shaken for one hour at room temperature.

The sample solutions were removed from the adsorbent by filtration. The residual content of lead in the solution was determined by complexometric titration.

## **2.8 Effect of Dosage on the Removal of Copper**

### **2.8.1 Procedure**

Prepared biosorbent beads sample of various masses ranging from 1 g to 5 g were placed in each flasks and 100 mL of standard copper II sulphate solution ( $200 \text{ mg L}^{-1}$ ) was added to each flask at pH 5. And then the loaded flasks were placed on an electric shaker. In order to attain complete equilibrium, the solutions were shaken for one hour at room temperature. These mixtures were separated out by filtration. The residual content of copper in the filtrate was determined by iodometric method (Vogel, 1961).

## **2.9 Characterization of Metal Sorbed Vegetable Stalks Alginate Biosorbent Beads**

### **2.9.3 XRD Analysis**

#### **Materials**

Metal sorbed biosorbent composite beads

#### **Apparatus**

XRD Diffractometer (Rigaku, D-Max – 2200)

### **2.9.4 SEM Analysis**

#### **Materials**

Metal sorbed biosorbent composite beads

#### **Apparatus**

Scanning Electron Microscope (JOEL-JSM-5610-Japan)

Ion Sputter (JFC-1600)

## **2.10 Application of Prepared Biosorbent Beads on the Removal of Toxic Heavy Metal Ions in Industrial Wastewater**

Industrial wastewater were collected from GS Battery Factory in Hlaing Tharyar Township, Yangon Division, Myanmar in 2009 .

### 2.10.1 Procedure

The trace element lead and copper from GS battery wastewater were determined by Atomic Absorption Spectrophotometry.

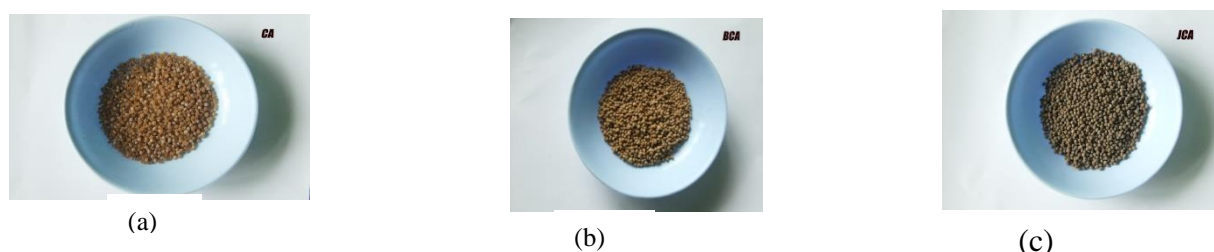
0.2 g of prepared biosorbent beads were placed in each flask and 10 mL of battery wastewater was added into each flask. The flasks were placed on the electric shaker and were shaken for 1 hr at room temperature. The wastewater solution containing heavy metal ions were removed by prepared adsorbent and the residue of lead and copper in the filtrate were determined by atomic absorption spectrophotometry. In such a way, the removal amount of lead and copper from industrial wastewater by vegetable stalk alginate beads could be obtained.

## 3. RESULTS AND DISCUSSION

### 3.1 Preparation of Vegetable Stalks Alginate Biosorbent Beads

Figure 3 show that the calcium alginate beads, bagasse, calcium alginate beads and *Jatropha curcas* calcium alginate beads.

In the preparation of the beads, the optimum condition was selected by mixing 2 g of vegetable stalks powder and 6% w/v of sodium alginate and the mixture was placed drop by drop into 10% CaCl<sub>2</sub> solution to obtain biosorbent beads.



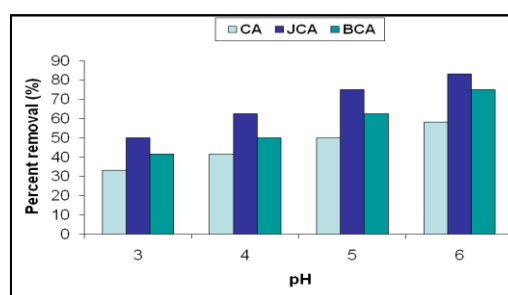
**Figure 3:** (a)calcium alginate beads, (b) bagasse calcium alginate beads (c) *Jatropha curcas* calcium alginate beads

### 3.2 Effect of pH on the Removal of Pb<sup>2+</sup> and Cu<sup>2+</sup> Ions

The pH is an important process parameter on biosorption of metal ions from aqueous solution since it is responsible for protonation of metal binding sites. Table 1 and figure 4 show the percent removal of Pb<sup>2+</sup> ions by prepared biosorbent beads at different pH values. The removal of Pb<sup>2+</sup> ion increased with increasing pH and the highest adsorption was observed at pH 6. Experiments were not performed beyond pH 6 because at higher pH, precipitation of metal was observed and the precipitate may be interfered with the biosorption process. The highest percent removal of Pb<sup>2+</sup> ion by calcium alginate, bagasse-calcium alginate and *Jatropha curcas* –calcium alginate beads were found to be 58.3%, 75% and 83.3% respectively.

**Table 1: Effect of pH on the Removal of Pb<sup>2+</sup> Ion by Biosorbent Beads**

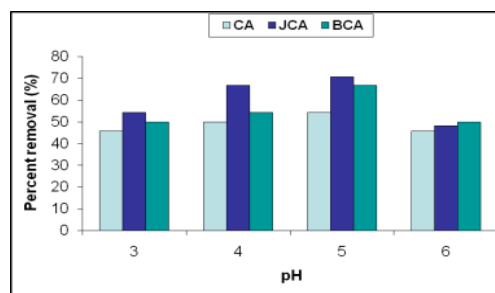
Prepared beads	Initial conc. (mg L <sup>-1</sup> )	Percent removal (%)			
		pH 3	pH 4	pH 5	pH 6
CA	200	33.3	41.6	50.0	58.3
JCA	200	50.0	62.5	75.0	83.3
BCA	200	41.6	50.0	62.5	75.0



**Figure 4: Removal percent of Pb<sup>2+</sup> at different pH**

**Table 2: Effect of pH on the Removal of Cu<sup>2+</sup> Ion by Biosorbent Beads**

Prepared beads	Initial conc. (mg L <sup>-1</sup> )	Percent removal (%)			
		pH 3	pH 4	pH 5	pH 6
CA	200	45.8	50.0	54.2	45.8
JCA	200	54.2	66.7	70.8	58.3
BCA	200	50.0	54.2	66.7	50.0



**Figure 5: Removal percent of Cu<sup>2+</sup> at different pH**

Similarly, Table 2 and Figure 5 show the percent removal of Cu<sup>2+</sup> ion by prepared biosorbent beads. In the case, the removal of Cu<sup>2+</sup> ion increased with increasing pH and the highest adsorption was observed at pH 5. Above the pH 5, the precipitation of metal ion was observed and the precipitate may be interfered in the biosorption process. So the adsorption of Cu<sup>2+</sup> ion by prepared beads was just done at pH 5.

The change in pH may be attributed to the nature of ions in solution and the nature of the adsorbent used. The lower the pH, the more H<sup>+</sup> ions competing with Pb<sup>2+</sup> and Cu<sup>2+</sup> ions for adsorption sites, thus reducing their adsorption. On the other hand, the higher the pH, the less H<sup>+</sup> ions competing with metal ions for adsorption sites, thus increasing their adsorption which explains the obtained results in figures.

### 3.3 Effect of Contact Time on the Removal of Pb<sup>2+</sup> and Cu<sup>2+</sup> Ions

The effect of contact time on the removal of Pb<sup>2+</sup> and Cu<sup>2+</sup> ions on the surface of calcium alginate, bagasse-calcium alginate and *Jatropha curcas*-calcium alginate beads were investigated. It was found that the maximum adsorption occurred within 30 minutes for lead and copper ions on biosorbent beads. This rapid sorption stage indicates that surface sorption on the prepared biosorbent beads. A further increase in contact time has a negligible effect for the removal of Pb<sup>2+</sup> and Cu<sup>2+</sup> ions. Adsorption equilibrium time is defined as the time required for heavy metal concentration to reach a constant value.

Table 3 and figure 6 show the percent removal of Pb<sup>2+</sup> ions by prepared biosorbent beads with various contact time from 10 to 60 minutes. It was found that the maximum percent removal was reached within 30 minutes. After 30 minutes, the removal percent of metal ions were nearly constant. It was found that complete adsorption of Pb<sup>2+</sup> ion by calcium alginate beads (CA beads), bagasse-calcium alginate beads (BCA beads) and *Jatropha curcas*-calcium alginate (JCA beads) were 58.3%, 75% and 83.3%, respectively.

Similarly Table 4 and Figure 7 show the percent removal of Cu<sup>2+</sup> ion by prepared biosorbent beads with various contact time. From the above experiments, the complete adsorption were occurred after 30 minutes. The maximum percent removal of Cu<sup>2+</sup> ion by calcium alginate beads (CA beads), bagasse-calcium alginate beads (BCA beads) and *Jatropha curcas*-calcium alginate (JCA beads) were 54.2 %, 66.7 % and 70.8 %, respectively.

**Table 3: Effect of Contact Time on the Removal of Pb<sup>2+</sup> Ion by Biosorbent Beads**

Contact time (min)	Initial concentration (mg L <sup>-1</sup> )	Percent removal (%)		
		CA	JCA	BCA
10	200	16.7	33.3	25
20	200	41.6	58.3	50
30	200	58.3	83.3	75
40	200	58.3	83.3	75
50	200	58.3	83.3	75
60	200	58.3	83.3	75

**Table 4: Effect of Contact Time on the Removal of Cu<sup>2+</sup> Ion by Biosorbent Beads**

Contact time (min)	Initial concentration (mg L <sup>-1</sup> )	Percent removal (%)		
		CA	JCA	BCA
10	200	25.0	50.0	41.6
20	200	50.0	66.7	62.5
30	200	54.2	70.8	66.7
40	200	54.2	70.8	66.7
50	200	54.2	70.8	66.7
60	200	54.2	70.8	66.7

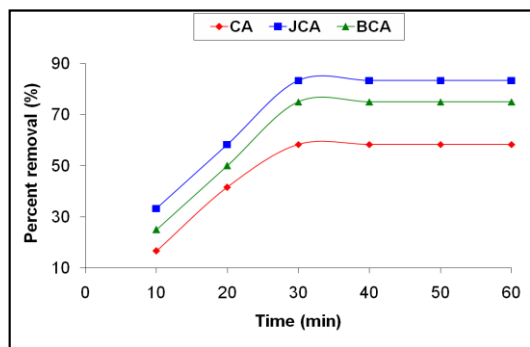


Figure 6: Effect of contact time on the removal of Pb<sup>2+</sup>

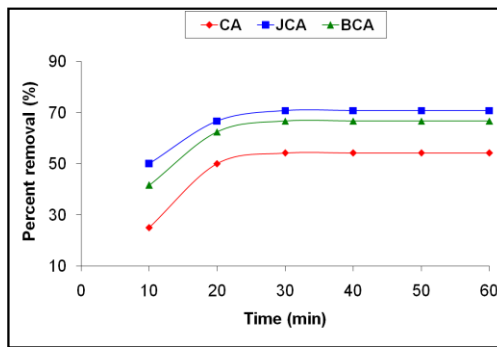


Figure 7: Effect of contact time on the removal of Cu<sup>2+</sup> ion

### 3.4 Effect of Dosage on the Removal of Pb<sup>2+</sup> and Cu<sup>2+</sup> Ions

The effect of dosage on the adsorption of Pb<sup>2+</sup> and Cu<sup>2+</sup> ions on the prepared biosorbent beads were studied. It was found that the removal of Pb<sup>2+</sup> ion from 200 ppm of model lead II nitrate solution increase from 50% to 77.5%, 55.2% to 90.3% and 64.6% to 93.8% with an increase in dose of CA beads, BCA beads and JCA beads from 10 gL<sup>-1</sup> to 50 gL<sup>-1</sup>, respectively. It was also found that the removal of Cu<sup>2+</sup> ion increase from 45.8% to 75%, 54.2% to 81.3% and 58.4% to 83.4% with an increase in amount of CA beads, BCA beads and JCA beads from 10 gL<sup>-1</sup> to 50 gL<sup>-1</sup>, respectively. It was apparent that the percent removal of metal ions increased rapidly with increase in dose due to great availability of the adsorbent. Figure 8 and 9 show the adsorbent dose was increased from 1 to 5 g in 100 mL heavy metal solution, metal uptake capacity was decreased. This may be attributed to reduction of total area of adsorbent, due to probably by aggregation during adsorption of the biomass surface depending on the experimental condition such as pH, ionic strength and temperature.

Table 5: Effect of Dosage on the Removal of Pb<sup>2+</sup> Ion by Biosorbent Beads

Dosage (g L <sup>-1</sup> )	Initial concentration (mg L <sup>-1</sup> )	Percent removal (%)		
		CA	JCA	BCA
10	200	50.0	64.6	55.2
20	200	58.3	83.3	75.0
30	200	66.7	88.6	83.3
40	200	75.0	91.9	87.4
50	200	77.5	93.8	90.3

Table 6: Effect of Dosage on the Removal of Cu<sup>2+</sup> Ion by Biosorbent Beads

Dosage (g L <sup>-1</sup> )	Initial concentration (mg L <sup>-1</sup> )	Percent removal (%)		
		CA	JCA	BCA
10	200	45.8	58.4	54.2
20	200	54.2	70.8	66.7
30	200	62.5	79.2	72.9
40	200	66.7	81.3	79.2
50	200	75.0	83.4	81.3

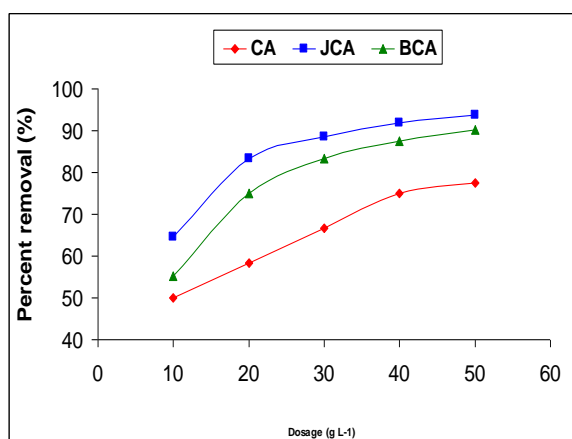


Figure 8: Effect of dosage on the removal of Pb<sup>2+</sup> ion

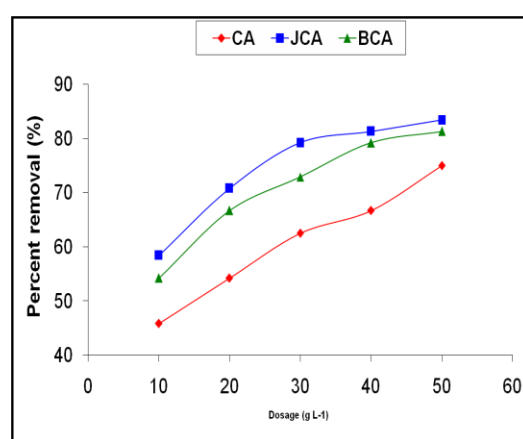


Figure 9: Effect of dosage on the removal of Cu<sup>2+</sup> ion



### 3.5 Characterization of Metal Sorbed Prepared Vegetable Stalks Alginate Biosorbent Beads

#### 3.5.1 On XRD Analysis

Figures 10 (a), (b), (c) and 11 (a), (b), (c) show that XRD spectra of vegetable stalk alginate biosorbent beads before and after adsorption of  $Pb^{2+}$  and  $Cu^{2+}$  ions. These diffractograms changed from amorphous nature to crystalline nature after adsorption of metal ions.

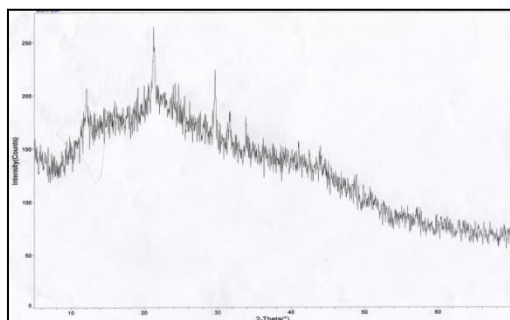


Figure 10(a) XRD diffractogram of BCA beads before adsorption

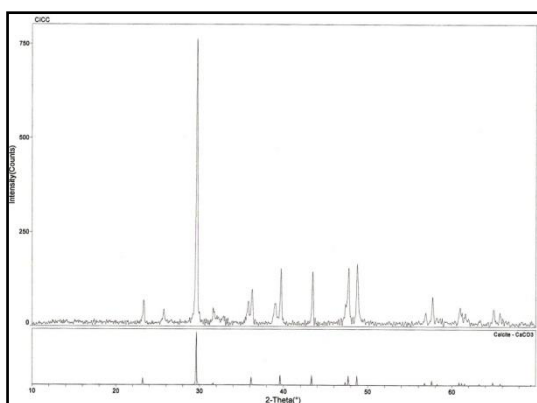


Figure 10 (b) XRD diffractogram of BCA beads after adsorption of  $Pb^{2+}$  ion

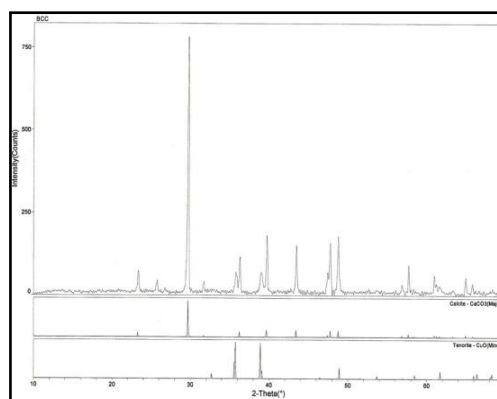


Figure 10 (c) XRD diffractogram of BCA beads after adsorption of  $Cu^{2+}$  ion

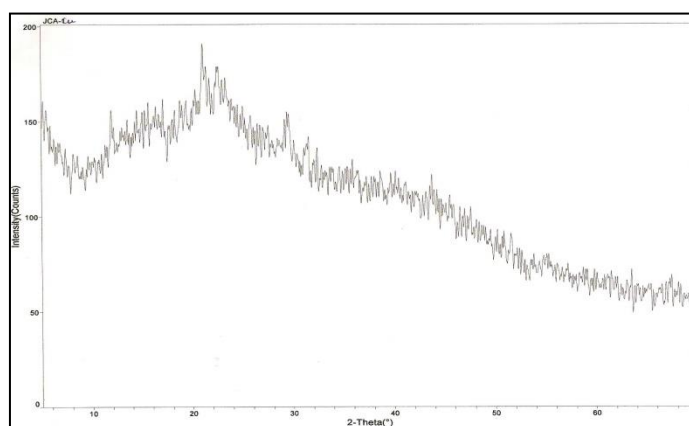
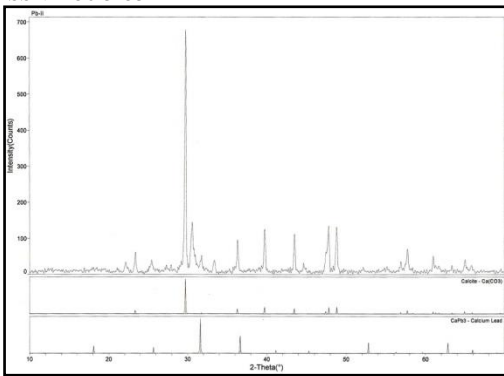
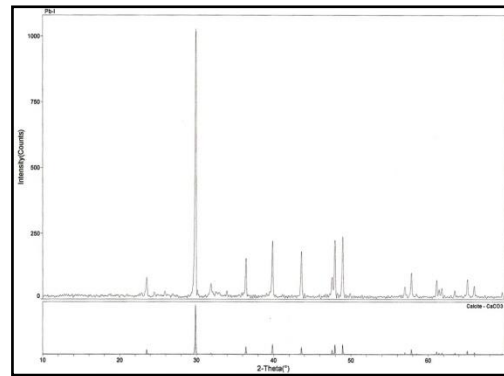


Figure 11(a) XRD diffractogram of JCA beads before adsorption



**Figure 11 (b) XRD diffractogram of JCA beads after adsorption of Pb<sup>2+</sup> ion**



**Figure 11 (c) XRD diffractogram of JCA beads after adsorption of Cu<sup>2+</sup> ion**

### 3.5.2 On SEM Analysis

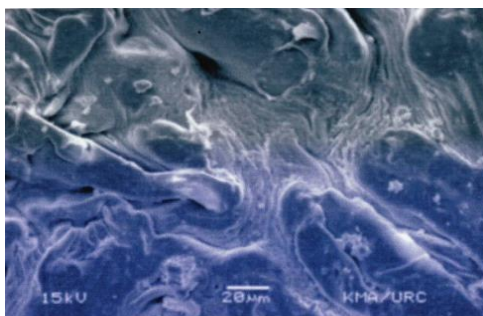
Figures 12 (a), (b), (c) show the surface morphology of bagasse-calcium alginate beads before and after adsorption of lead and copper. Figure 13 (a), (b) and (c) show the surface morphology of *Jatropha curcas* calcium alginate beads before and after adsorption of lead and copper.

After adsorption of lead and copper ion on bagasse-calcium alginate beads, Figure 12 (a), (b), (c) are very different from one another. In all figures cavitated pores were closed by the adsorption of lead or copper. It is one way to explain the nature of images that coprecipitation of specified metal may take place.

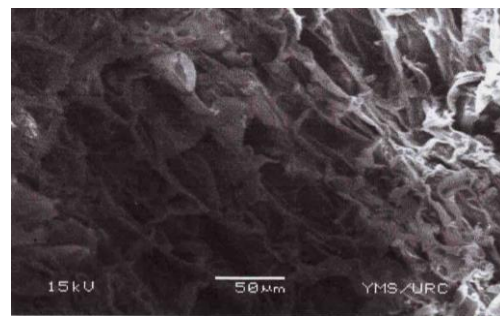
the SEM images of *Jatropha curcas*-calcium alginate beads after adsorption of lead (Figure 13 (a)) and *Jatropha curcas*-calcium alginate beads after adsorption of copper (Figure 13 (b)), firstly, adsorption of specified metal has taken place and secondly coprecipitation of specified meal may occur. Some channels can be seen in the images of JCA beads after adsorption of copper where as no void and channel can be seen in the images of beads and all the cavities are blocked by sorption followed by coprecipitation of the specified metal. Furthermore, after metal uptake, the beads presented a more uniform and organized structure, specially inside (Figures 13 (a), (b), (c)).



**Figure 12(a): SEM micrographs of bagasse-calcium alginate beads before adsorption**



**Figure 12(b) : SEM micrograph of BCA beads after adsorption of Pb<sup>2+</sup> ion**



**Figure 12(b) : SEM micrograph of BCA beads after adsorption of Cu<sup>2+</sup> ion**





Figure 13 (a): SEM micrographs of *jatropa curcas*-calcium alginate beads before adsorption

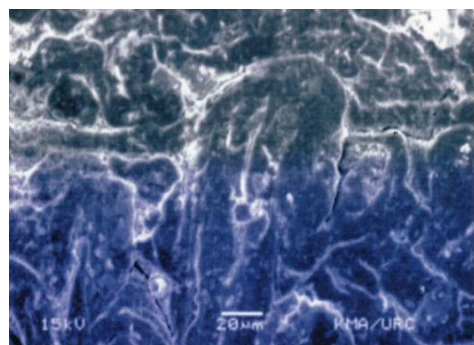


Figure 13(b) : SEM micrograph of JCA beads  
 after adsorption of  $Pb^{2+}$  ion

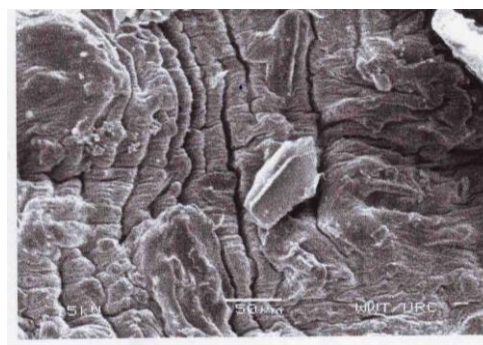


Figure 13(c) : SEM micrograph of JCA beads  
 after adsorption of  $Cu^{2+}$  ion

### 3.6 Application of Bagasse-Calcium Alginate Beads and *Jatropha curcas* Calcium Alginate Beads for the Removal of $Pb^{2+}$ and $Cu^{2+}$ Ions from Industrial Wastewater

In this paper, wastewater samples were collected from GS battery factory, Hlaing Tharyar Industrial Zone, Yangon in 2009. The concentration of lead and copper in industrial wastewater was determined by atomic absorption spectrophotometer.

On the average of lead concentration 3.744 ppm and copper concentration 5.457 ppm were found in industrial wastewater. The contents of these metals in the wastewater sample were found to be reduced significantly by three types of calcium alginate beads. The results are shown in Table 7. According to the resulting data, removal efficiency of Bagasse-calcium alginate beads was 71.55% lead and 70.84% copper. Similarly, removal efficiencies of *Jatropha curcas* calcium alginate beads were 78.5% lead and 74.27% copper respectively. The resulting data are consistent with that found out by working with model solutions consisting of single ionic species. Thus, the removal of lead and copper ion from wastewater sample by prepared vegetable stalks alginate biosorbent beads can be used as an effective adsorbent materials in the treatment of industrial effluents.

Table 7: Metal Removal Efficiency of Beads in Industrial Waste Water by AAS Analysis

Metal ions	Concentration of metal ions in industrial wastewater (ppm)			Removal efficiency (%)	
	Before treatment	After treatment		BCA beads	JCA beads
		BCA beads	JCA beads		
Lead	3.744	1.065	0.804	71.55	78.5
Copper	5.457	1.591	1.404	70.84	74.27

#### 4. CONCLUSION

On the aspect of removal of  $Pb^{2+}$  and  $Cu^{2+}$  ions by three types of beads, the optimal pH for the removal of  $Pb^{2+}$  was observed at 6 and at pH 5 for  $Cu^{2+}$  ion. Sorption experiments were conducted base on the mass of adsorbent dose, it was observed that percent removal of metal ion increase with increase in mass of adsorbent dose however the mass of adsorbent increase with decrease in metal uptake capacity. So, the adsorbent doses  $20\text{ gL}^{-1}$  were used in the whole experiments. It was found that the equilibrium time for the removal of  $Pb^{2+}$  and  $Cu^{2+}$  ions were 30 minute in contact with three types of beads and metal ion solution.

From the above experiments, the highest percent removal of  $Pb^{2+}$  ion by calcium alginate, bagasse-calcium alginate and *Jatropha curcas* –calcium alginate beads were found to be 58.3%, 75% and 83.3% respectively. The highest percent removal of  $Cu^{2+}$  ion by calcium alginate, bagasse-calcium alginate and *Jatropha curcas*–calcium alginate beads were observed that 54.2%, 66.7% and 70.8% respectively. From the above observations, it was found that sorption efficiency of  $Pb^{2+}$  was greater than  $Cu^{2+}$ . Moreover, the metal removal capacity of three types of beads were observed that *Jatropha curcas* –calcium alginate beads were more effective than bagasse-calcium alginate bead and calcium alginate beads. The trend of metal adsorption capacity of prepared biosorbent beads were in the order of JCA beads > BCA beads > CA beads.

#### ACKNOWLEDGMENT

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