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Removal of lead and cadmium in batch and packed bed column system by using PVA-alginate immobilized fungal sorbent

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ABSTRACT

"Mucor heimalis", a fungal biomass immobilized in PVA- alginate, was utilized as potential biosorbent for removal of Pb(II) and Cd(II) from aqueous solution in column system. The experiments were conducted to study the effect of important design parameters such as bed height, flow rate, and initial metal ion concentration. At a bed height of 13 cm, flow rate 1.0 mL/min, and initial metal ion concentration 15 mg/L, the maximum Pb(II) and Cd(II) uptake capacity of Mucor heimalis was found to be 2.53 and 1.83mg/g respectively. The bed depth service time (BDST) model was in good agreement with experimental data with high correlation coefficient (> 0.999 for Pb(II) and > 0.998 for Cd(II)). The bed sorption capacity (N_o) at 1 and 3 mL/min were found to be 410.64 and 415.54 mg/L for Pb(II) and 353.34 and 386.91mg/L for Cd(II) and rate constant (K_a) were 0.0960 and 1.296 L/mg/h for Pb(II) and 0.0874 and 0.299 L/mgh for Cd(II), respectively. The column regeneration studies were carried out using 10% HCl as eluant for three sorption-desorption cycles. The high Pb(II) and Cd(II) removal ability and regeneration efficiency of this biosorbent suggest its applicability in industrial processes and data generated would help in further up scaling of the sorption process.

Key words: Lead; Cadmium; *Mucor heimalis (*MHB); Column system; Bed Depth Service Time Model.

INTRODUCTION

The intensification of industrial activity in the recent years is greatly contributing to an increasing discharge of toxic compounds in the natural environment, particularly in aquatic systems. Among the widely found toxic compounds, heavy metals bear a special significance because of their highly toxic nature even at very low concentrations. Increased usage of these heavy metals has even resulted in their depletion from their natural resources. Moreover, stricter environmental regulations have led to an increased and urgent need for controlling heavy metal discharge into the environment. The major industries that are responsible for the discharge of wastewater containing metals include mining, metallurgy, electroplating, and refining industries [1]. There are several conventional techniques, utilized for removing heavy metals from aqueous streams such as chemical precipitation as synthetic coagulants, solvent extraction, ion exchange and reverse osmosis. The application of such traditional treatment techniques however, needs enormous cost and continuous input of chemicals, which becomes impracticable and uneconomical and causes further environment damage [2, 3]. Hence, the search for easy, effective, economic and eco-friendly technique is underway which is required for the fine tuning of effluent/wastewater treatment.

The use of biological material for removing and recovering heavy metals from contaminated industrial effluents has emerged as a potential alternative method to the conventional ones. Many microorganisms have been shown to be capable of concentrating heavy metals from their aqueous environment, and the term biosorption is used to describe for such passive, non-metabolically mediated process of metal binding by living or dead biomass [4]. Fungi may be better suited for this purpose than other microbial groups owing to their high tolerance towards metals, intracellular metal uptake, and wall-binding capacities [5]. Fungal species such as *Rhizopus arrhizus, Rhizopus nigricans*

Aspergillus niger, Mucor roxii, Mucor racemosus etc. have been extensively studied for heavy metals biosorption, and the process mechanism seems to be species specific [6-12]. However, relatively only a few studies have been reported with *Mucor heimalis* [13].

The ultimate success of these studies, however, lies in developing engineering systems employing these microbes in treating large quantities of contaminated water on a continuous basis. Among the available reactor types for biosorption, packed bed columns offer several advantages, viz., simple to operate, high process yield, and easy scale-up[1]. For this, the use of free microorganisms is technically and economically not feasible for small and medium-flow treatment application due to high costs involved in the process. In such cases, immobilized microbial cell system in fixed bed columns could provide additional advantages over freely suspended cells. The advantages include ease of regeneration and reuse of the biomass, easy solid–liquid separation operation, and minimal clogging in the fixed bed [4].

Therefore, the present work was aimed to investigate the regeneration and reuse of the immobilized biomass of *Mucor heimalis* in the biosorption of lead and cadmium in individual packed bed columns operated under continuous mode. To analyze the performance of these columns in the removal of the metals, several operating parameters investigated. In addition to this BDST model was also analyzed for scale up the sorption process.

EXPERIMENTAL SECTION

Preparation of microorganism

The strain used in this study was *Mucor heimalis (*NCIM 873) and taken from National Chemical Laboratory Pune, India. The culture was routinely maintained at 4^0C on potato dextrose agar medium slant (200 g of peeled potatoes; 20g of dextrose 0.1 g of yeast extract and 20 g of agar) and aerobically cultivated in potato dextrose broth. The flasks were incubated at 30^oC in orbital shaker at 120 rpm for 48 h. The growing cells from the culture broth were separated from the liquid by filtration and washed several times with double distilled water. The wet cell biomass was dried for 24 h at 60°C in an oven. Dried cells were powdered by blender in uniform size and used for sorption experiment.

Immobilization of MHB in Poly Vinyl Alcohol (PVA) and alginate

Slurry of PVA and alginate was prepared by dissolving 2.0 g PVA (Himedia chemical) and 1.0 g of sodium alginate (Himedia chemical) in 100 mL of hot double distilled water and mixed with 5.0g of biomass powder and stirred. A 100 ml aliquot of this suspension was then extruded into 500 mL of 2 mM $CaCl_2.2H_2O$ (Qualigens Chemicals) solution in a drop-wise manner by syringe for polymerization and bead formation. The suspension drops gets precipitated upon contact with solution $CaCl_2.2H_2O$, forming gel beads of nearly 0.3 cm diameter. These beads were then soaked in $CaCl_2.H_2O$ solution for 8 h for the complete gelling. After 8 h of curing, the beads were subjected to three cycles of freezing and thawing to get spherical beads. Blank PVA-alginate beads were also prepared without adding biosorbent and used as control.

Preparation of standards and reagents

All chemicals and reagents used were of analytical grade and were used without further purification (purchased from E. Merck, India Ltd., Mumbai, India). Stock solutions of Cd(II) and Pb(II) of 1000 mg/L were prepared from 2.74g of $Cd(NO_3)_2.4\ H_2O$ and 1.59 g of $Pb(NO_3)_2$ in 1000 mL of de-ionized, double distilled water containing a few drops of concentrated HNO_3 to prevent the precipitation of Cd(II) and Pb(II) by hydrolysis. Required initial concentration of Cd(II) and Pb(II) samples were prepared by appropriate dilution of the above stock standard solution. Standards for calibration of AAS for Cd(II) and Pb(II) were prepared from standard solution of cadmium and lead purchased from E. Merck, India Ltd. Mumbai, India.

Batch sorption experiments

Biosorption experiments were performed in 250ml conical flasks previously rinsed with HNO_3 in order to remove any metal that remained unabsorbed on the glass wall. The pH of the metal solutions in the conical flask was initially adjusted to desired values by using 0.1 M/HNO_3/NaOH; the sorbents (free biomass and immobilized biomass) were added to each flask and were agitated on the shaker until the equilibrium was reached. The sorbent (free biomass and immobilized biomass), separated by centrifugation/ filtration at 15000 rpm for five minutes, was analyzed for remaining Cd(II) concentration in the sample. The biosorption capacity of the metal ion was calculated by the equation:

$$q = (C_o - C_{e)}\ XV/M \qquad (1)$$

Where q is the metal uptake (mg/g), V is the volume (L), W is the amount of biomass (g) and C_o & C_e are the initial and equilibrium metal concentrations (mg/L) respectively. All experiments in this work were conducted in triplicate and concurrent values were taken.

Column design and Experimental procedure

Experiments were carried out in borosilicate glass column of 30 cm height and 2.0 cm internal diameter. It was filled with different amount of immobilized beads i.e. 4.9g, 7g and 9.1g in order to achieve different bed height of 7, 10 and 13cm respectively. In the column, 0.5 mm stainless steel mesh and 1.0 cm glass wool were kept at the bottom and the at top of the column respectively to support the beads in the column and to minimize the effects of air bubble at the inlet and outlet regions of a packed column and ensure a closely packed arrangement. A 3.0 cm layer of glass beads was placed at the column base for providing a uniform inlet flow and good Pb(II) and Cd(II) solution distribution into the column. The reactor was operated in an up flow mode and the flow rates (1, 2 and 3 ml/min) were regulated with a peristaltic pump (Miclins India, Model No. PP-10). Immobilized beads without addition of biomass were used as control. The treated Pb(II) and Cd(II) solution was collected from the top with same flow rate of feed stream and was estimated for the Pb(II) and Cd(II) concentration. Operation of the column was stopped when effluent metal concentration exceeded a value of 98% of the initial metal ion concentration. The feed Pb(II) and Cd(II) concentration was varied in the range of 5 to 15 mg/l. The reactor system was operated at room temperature (±28^{o}C).

Desorption was carried out by passing selected desorbent through the column bed in upward direction at a flow rate of 2.0 mL/min. The effluent metal solution was collected and analyzed for Pb(II) and Cd(II) content. On the completion of desorption cycle, the column was rinsed with deionized double distilled water in the same manner as for biosorption till the eluting distilled water attains pH between 5 to 7. The desorbed and regenerated column bed was reused for next cycle. Another cycle of sorption-desorption was repeated in the same manner as above mentioned. All the experiments were performed in triplicates.

Modeling and analysis of column data

To analyze the dynamic removal of metal ion in up-flow fixed bed column breakthrough curves (C_t/C_0 vs. time t) were drawn and the data was evaluated with the help of following equations as previously used by [14, 15]:
Effluent volume:

$$V_{eff} = F.t_e \quad (2)$$

Total amount of metal ion sent to column:

$$m_{total} = C_o Ft_e / 1000 \quad (3)$$

Total percentage removal of metal ion

$$Total\ metal\ removal\ (\%) = m_{ad} / m_{total} x100 \quad (4)$$

The metal desorbed (m_d) can be calculated from the area below the desorption curve (outlet concentration vs. time) multiplied by the flow rate. The desorption efficiency can be calculated from

$$Desorption\ efficiency\ (\%) = m_d / m_{ad} x100 \quad (5)$$

Where, t_b is the breakthrough time at which the outlet concentration reached 1 mg/L, t_e is the exhaustion time at which the outlet concentration exceeded 98% of that at the inlet concentration. The total quantity of metal biosorbed in the column (m_{ad}) was calculated from the area above the breakthrough curve (outlet metal concentration vs. time) multiplied by the flow rate (F). Dividing the m_{ad} by the biosorbent mass (M) leads to the uptake capacity (Q) of the biomass.

Modelling of breakthrough curves

Bed Depth Service Time (BDST) approach based on Bohart and Adams equation is widely used [16, 17]. BDST is the simple model for predicting the relationship between bed height (Z) and service time (t) in term of process concentration and adsorption parameters. Hutchins proposed a linear relationship between bed height and service time given by equation

$$t = \frac{N_o Z}{C_o u} - \frac{1}{k_a C_o} \ln(\frac{C_o}{C_b} - 1) \quad (6)$$

Here t is the service time, N_o is the dynamic bed capacity (mg/L), Z is the bed height of the column (cm), u is the influent linear velocity (cm/h) defined as the ratio of the volumetric flow rate F (mL/h) to the cross sectional area of the bed S_c (cm^2), C_b is the breakthrough concentration of solute (mg/L), C_o the initial concentration of solute in the liquid phase (mg/L) and k_a is the rate constant in BDST model (L/mgh). Eq (6) is rewritten in the form of straight line.

$$t = aZ - b \quad (7)$$

$$\text{where,} \quad \text{slope, } a = \frac{N_o Z}{C_o u}, \quad (8)$$

$$\text{intercept, } b = \frac{1}{k_a C_o} \ln(\frac{C_o}{C_b} - 1). \quad (9)$$

Thus, N_o and K_a can be evaluated from slope (a) and the intercept (b) of the plot of t_b versus Z, respectively.

The critical bed depth (Z_o) is the theoretical depth of the sorbent sufficient to ensure that the outlet solute concentration does not exceed the breakthrough concentration C_b at time t=0, Z_o can be calculated by Eq (10) [16, 18]:

$$Z_o = \frac{u}{N_o K_a} \ln(\frac{C_o}{C_b} - 1) \quad (10)$$

Analysis of Pb(II) and CdII) in aqueous solution

The analysis of lead and cadmium in sample solution was done using Atomic Absorption Spectrophotometer, AAS (Shimadzu AA-6300, Japan). The hollow cathode lamp was used as light source and was set at 283.3 nm wavelength for Pb(II) and 228.8 nm wavelength for Cd(II), using 10 mA lamp current and 0.7 nm slit width, and with deuterium lamp for background correction. In order to generate flame instrument grade (98%) acetylene, delivered at 4.0 L/min at a pressure of 0.9 kg/cm2, together with compressed air supplied at 17.5 L/min flow rate and 3.5 kg/cm^2 gas pressure. The instrument was calibrated from 0.1 to 10.0 mg/L for Pb(II) and Cd(II). Other range samples were diluted until results within the calibration range were obtained for the metal ion.

RESULTS AND DISCUSSION

Batch study

Effect of initial pH and removal efficiency of Pb(II) and Cd(II) by free and immobilized biomass

Fig. 1 shows the biosorption of Pb(II) and Cd(II) by free and PVA- alginate immobilized *Mucor heimalis* biomass as a function of pH (blank beads were used as control). This study were carried out by varying the pH from 1.0 to 6.5 for Pb(II) and 1.0 to 8.0 for Cd(II) a pH below the onset of metal hydrolysis and precipitation [19, 20]. The experiment was performed in Erlenmeyer flask containing 50 mL of 15 mg/L of initial cadmium and lead solution at room temperature (±28°C). It was clear from figure that Pb(II) and Cd(II) sorption were very low at pH below 2.0 for both free and immobilized biomass. The maximum uptake of Pb(II) 14.3 and 13.2 mg/g were achieved at 5.0 for both the free and immobilized biomass respectively. While in case of Cd(II) the maximum uptake of 13.9 and 11.5 mg/g were achieved at pH 6.0 for both the free and immobilized biomass respectively. It is also clear from figure that an increase in uptake of Pb(II) and Cd(II) was observed with increasing pH from 2 to 5 for Pb(II) and 2 to 6 for Cd(II) for the free and immobilized biomass respectively. At pH around 6 and 6.5, the Pb(II) and Cd(II) biosorption capacity leveled off at a maximum value for both types of biomass respectively. It can be explained that at low pH the functional group on the surface of biosorbent got protonated which restrict the approach of metal cations as a result of the repulsive force whereas high pH increases, more of the functional group would be exposed which will carrying negative charges that subsequently attracted to the metal ions. In addition to this, the uptake capacity of free biomass was greater than that of the PVA- alginate immobilized biomass in both the cases. This variation could be due to many reasons. First, the binding of cationic and anionic metal species to the fungal cell wall is assumed to occur predominantly through surface adsorption. Second, the mass transfer of the metal ions from aqueous phase to the solid sorbent sites is dependent on porosity of the sorbent. Third, the native biomass in contact with the metal

ions under the conditions of moderate agitation has its binding sites freely exposed to the sorbate. However, in immobilized systems, the sorbent particles entrapped and retained at the interior may not have accessibility to the metal ions [21].

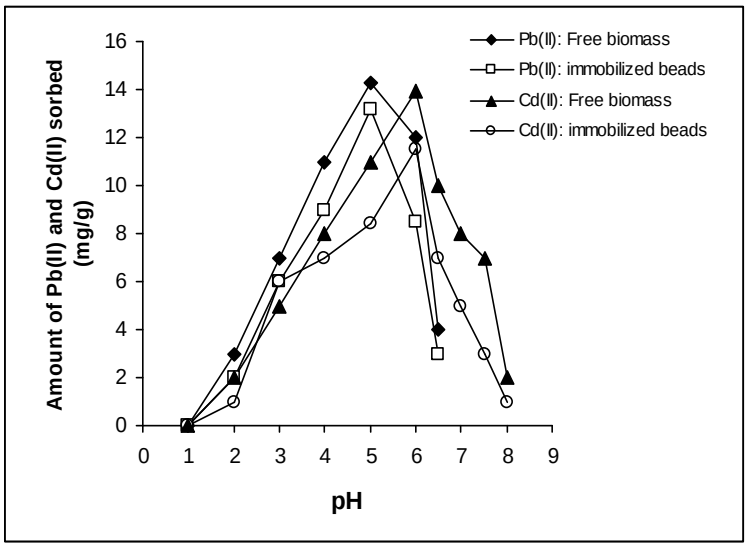

Fig. 1 Effect of pH: pH range= 1 to 6.5 for Pb(II) and 1 to 8.0 for Cd(II), C_o=15 mg/L, room temp. = ±28°C

Column study

Effect of bed height

In order to optimize bed height for the maximum Pb(II) and Cd(II) removal, experiments were carried out by varying the bed height 7, 10 and 13 cm at flow rate of 1 mL/min and 15mg/L Pb(II) and Cd(II) concentration. The plot of effluent concentration versus time (C_t/C_o vs. Time) at different bed heights were plotted (Fig. 2 for Pb(II) and Fig. 3 for Cd(II)) and data calculated from the breakthrough curves are presented in Table 1. Result indicates that the breakthrough time, exhaustion time, uptake capacity, percentage removal and volume treated increased with the rise in bed height from 7 to 13 cm for both the metal ion. This displacement of the front of adsorption with increase in bed depth can be explained by mass transfer phenomenon that takes place in this process. When the bed depth reduced, axial dispersion phenomenon predominates in the mass transfer and reduces the diffusion of metallic ions. The solute (metallic ions) has not enough time to diffuse into the whole adsorbent mass. Consequently, an important reduction in the volume treated (1539.6 to 759.6 mL for Pb(II) and from 1140 to 600 mL for Cd(II)) was observed when the bed height decreased from 13 to 7 cm. Further, it was also observed that the sorption capacity was increased with the increase in bed height for both Pb(II) and Cd(II). This increase in adsorption capacity with an increase in the bed height can be due to the increase in the specific surface of the adsorbent, which supplies more fixation binding sites. In addition to this, breakthrough time and exhaustion time were increased with the increasing bed height. Since breakthrough time is the determining parameter of the process, the larger it is, the better the intraparticulate phenomenon and the bed adsorption capacity are.

Effect of flow rate

The effect of flow rate on Pb(II) and Cd(II) sorption was studied by varying the flow rate (1.0, 2.0 and 3 mL/min) at bed height of 13 cm and 15 mg/L of initial Pb(II) and Cd(II) concentration. The plot of C_t/C_0 vs. time at different flow rate is also shown in Fig. 2 for Pb(II) and Fig. 3 for Cd(II). The breakthrough time, exhaustion time, treated volume effluent, percentage removal and uptake capacity with respect to flow rate were evaluated from the sorption data and presented in Table 1. It was found that breakthrough time, exhaustion time, percentage Pb(II) removal and uptake capacity decreases as the flow rate increased, whereas the volume of treated effluent increased (1539.6 to 1710 mL for Pb(II) and 1140 to 1198.8 mL for Cd(II)) with increase in flow rate from 1.0 mL/min to 3 mL/min. At lower flow rate, the contact time of the Pb(II) solution and immobilized biomass in the column was longer and hence Pb(II) ions got more time to diffuse onto the immobilized biomass and better adsorption capacity and percentage removal were obtained. Actually, when the flow rates were low, the external mass transfer controlled the process and it was ideal for intra-particle diffusion systems. Thus, lower the flow rates; more effective was the diffusion process and higher was the residence time of the sorbate, which ultimately resulted in higher sorption capacity [22, 14]. At a higher flow rate, the biosorbent got saturated early (certainly because of reduced contact time); a larger amount of ions was adsorbed onto the immobilized biomass and a weak distribution of the liquid was there into the column. This led to a lower diffusivity of the solute onto the immobilized biomass. It was also

observed that at higher flow rate, the immobilized biomass got saturated easily. This resulted in higher volume treated at higher flow rate, i.e. improper utilization of the sorption capacity of immobilized biomass.

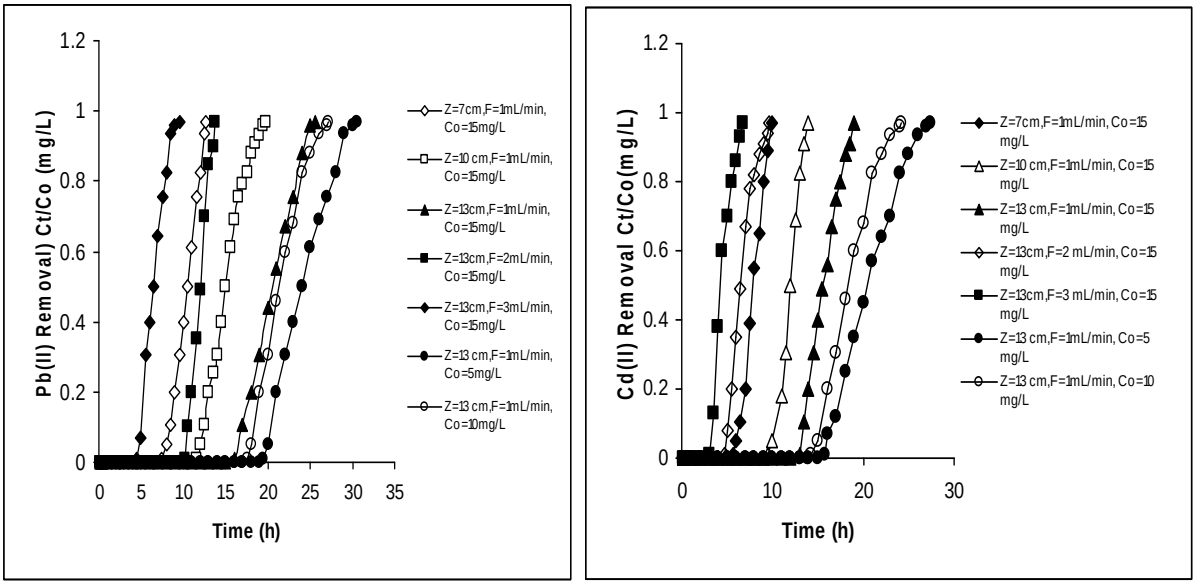

Fig.2 Breakthrough curve for Pb(II) sorption: Z= 7,10 and 13 cm, F=1,2 and 3 mL/min and C_o=5, 10 and 15mgL, pH = 5, temp. = ±28 ºC
Fig. 3 Breakthrough curve for Cd(II) sorption Z= 7,10 and 13 cm, F=1, 2 and 3 mL/min and C_o=5, 10 and 15mgL, pH = 6, temp. = ±28ºC

Effect of initial metal concentration

In order to optimize initial metal concentration for maximum Pb(II) and Cd(II) removal, experiments were carried out by varying the initial Pb(II) and Cd(II) concentration viz. 5, 10 and 15 mg/L at constant bed height of 13 cm and flow rate 1 mL/min respectively. The plot of effluent Pb(II) and Cd(II) concentration versus time (C_t/C_o vs. Time) at different initial Pb(II) concentration were plotted (Fig. 2 for Pb(II) and Fig. 3 for Cd(II)) and data calculated from the breakthrough curves are presented in Table 1. Results indicate that immobilized beads of MHB get saturated early at high concentration for both the metal ion. The breakthrough time considerably

Table 1 Column data for packed bed immobilized beads column for biosorption of Pb(II) and Cd(II) onto immobilized MHB at different process parameter

Process Parameter	t_b (h)	t_e (h)	V_{eff} (mL)	% removal	Q (mg/g)
Pb(II)					
Bed height, Z (cm): C_o= 15 mg/L, F= 1mL/min					
7 (4.9g)	7.4	12.66	759.6	94.0	2.18
10 (7.0g)	11.5	19.8	1188	95.9	2.44
13 (9.1g)	16.0	25.66	1539.6	99.5	2.53
Flow rate, F (mL/min): C_o= 15 mg/L, Z= 13cm					
1	16	25.66	1539.6	99.5	2.53
2	10.1	13.7	1644	90.5	2.19
3	4.5	9.5	1710	76.3	2.12
Initial Pb(II) concentration, C_o (mg/L): Z= 13 cm, F= 1mL/min					
5	19.4	30.6	1836	86.8	0.88
10	17.5	27.2	1632	90.9	1.63
15	16.0	25.66	1539.6	99.5	2.53
Cd(II)					
Bed height Z (cm): C_o= 15 mg/L, F= 1mL/min					
7 (4.9g)	5.6	10	600	92.5	1.69
10 (7.0g)	9.5	14	840	95.2	1.71
13 (9.1g)	13.0	19	1140	97.2	1.83
Flow rate, F (mL/min): C_o= 15 mg/L, Z= 13cm					
1	13.0	19	1140	97.2	1.83
2	9.5	9.66	1159.2	90	1.71
3	7.5	6.66	1198.8	73	1.44
Initial Cd(II) concentration C_o (mg/L): Z= 13 cm, F= 1mL/min					
5	15.8	27.4	1644	84	0.758
10	14.3	24.2	1452	89	1.42
15	13.0	19	1140	97.2	1.83

C_o = Initial concentration, t_e = exhaustion time (h), t_b = breakthrough time (h), V_{eff} = Volume effluent (mL), Q= Column Uptake capacity, %= Percent decreases from about 30.6 to 25.66h for Pb(II) and 27.4h to 19 for Cd(II) as the concentration increases from 5 to 15 mg/L respectively. A rise in the inlet metal concentration reduced the treated volume before the fixed bed sorption bed got saturated. A high metal concentration may saturate the immobilized beads more quickly, thereby decreasing the breakthrough time and exhaustion time. Also, it is clear that the maximum bed capacity of Pb(II) and Cd(II) increased with the increase in the initial Pb(II) and Cd(II) concentration. The maximum bed capacities at 5, 10 and 15 mg/L initial concentrations were 2.18 to 2.53 mg/g for Pb(II) and 0.758 to 1.83 mg/g for Cd(II) respectively. Decreasing the initial Pb(II) and Cd(II) concentration increased the treated volume that could be processed, and shifted the breakthrough curve to the right. The driving force for adsorption is the concentration difference between the solute on the sorbent and the solute in the solution. A high concentration difference provides a high driving force for the adsorption process and this may explain why higher adsorption capacities were achieved in the column fed with a higher Pb(II) and Cd(II) concentration.

Application of BDST Model

For this purpose the service time i.e. breakthrough time of the column corresponding to bed height 7, 10 and 13 cm at two flow rates 1.0 and 3.0 mL/min at constant initial Pb(II) concentration of 50 mg/L, was recorded. Thereafter the graph was plotted between service time and bed depth and is given in Fig. 4. From the slope and intercept of the BDST plot, the BDST parameter viz. sorption rate constant (k_a) and bed sorption capacity (N_o) calculated. The good values of correlation coefficient (> 0.999 for Pb(II) and > 0.998 for Cd(II)) showed that the variation of the service time with the bed depth is linear at both the flow rates for both the metal ion respectively, thus, indicating the validity of the BDST model when applied to the continuous column studies. The values of rate constant, k_a were 0.0960 and 1.296 L/mg/h for Pb(II) and 0.0874 and 0.299 L/mgh for Cd(II) at 1 and 3mL/min of flow rate respectively. The rate constant, which is calculated from the intercept of BDST plot, characterizes the rate of solute transfer from the liquid phase to solid phase. It was found that the values of the rate constant was influenced by flow rates and showed an increasing trend with the increase in flow rate indicating that the overall system kinetics was dominated by external mass transfer in the initial part of the sorption in the column. In general, if ka is large, a short bed is required to avoid breakthrough, but as k_a decreases a progressively longer bed is required to avoid breakthrough. The computed values of bed sorption capacity were 410.64 and 415.54 mg/L for Pb(II) and 353.34 and 386.91mg/L for Cd(II) at 1 and 3 mL/min flow rate respectively. The critical bed depth was also calculated with the Eq.10. It represent the sufficient height of the column bed in order to avoid breakthrough at $t_b = 0$. The critical bed depth was found to be 1.88 and 0.414 for Pb(II) and 2.40 and 1.927 for Cd(II) at 1 and 3 mL/min respectively. These result indicated that the critical bed depth decreased with an increase in the flow rate of the solute through the column. And it represents a sufficient length of the adsorption zone to attain a satisfactory effluent [23]. Thus the finding, that with the increase in flow rate the theoretical bed depth (Z_o) decreased correlated well with the observed performance in the breakthrough curves and thus explained the experimental results for poor performance of the column at higher flow rates.

The simplicity and advantage of using the BDST model is that it can applied for prediction of the slope for any unknown flow rate with a known slope at a given flow rate without any further experimental run. Thus, the values of constants obtained from the experimental plot can be extrapolated for alternative flow rates, by modifying the equation. A simplified form of the Bohart-Adams model is: t = aZ – b, where a is the slope, $a = N_o / C_o u$ and b is the intercept, $b = (1/k_a C_o)\ln(C_o / C_t - 1)$. When a new flow rate, other than the one used in the development of constants, is used to the column system, the equation can be modified by utilizing the new slope:

$$a' = a\frac{u}{u'} = a\frac{F}{F'} \qquad (11)$$

Where a and u are the old slope and influent linear velocity, respectively, and a` and u` are the new slope and influent linear velocity. As the column used in experiment has the same diameter, the ratio of original (u) and the new influent linear velocity (u`) and original flow rate (F) and new flow rate (F`) will be equal.

For the present study the BDST model parameters were calculated experimentally at two flow rates of 1 and 3 mL/min for Pb(II) and Cd(II). And the equation thus obtained for Pb(II) were y = 1.43333x + 2.7 and y = 0.4833x + 0.2 with r^2 value of 0.999 and 0.999 at 1 and 3 mL/min respectively (Fig. 4). For Cd(II) equations were y = 1.23333x + 2.9667 and y = 0.45x – 0.8667 with r^2 value of 0.999 and 0.997 at 1 and 3 mL/min respectively. In order to show validity of application of BDST model in predicting the column design at a new flow rate, the BDST equations were predicted at the flow rate of 3 mL/min for both the metal ion using sample flow rate of 1 mL/min, so

that direct comparison can be done between the experimental and predicted analysis. The results obtained after the prediction at 3 mL/min flow rate is 0.4777x + 2.7 with r2 value of 1.000 for Pb(II) and 0.4111x -0.8667 with r2 value of 1.000 for Cd(II) respectively . It can be seen from the result that the predicted (0.4777 for Pb(II) and 0.4111 for Cd(II)) and experimental (0.4833 for Pb(II) and 0.45 for Cd(II)) values of slopes for flow rate of 3mL/min were in good agreement for both the metal ion.

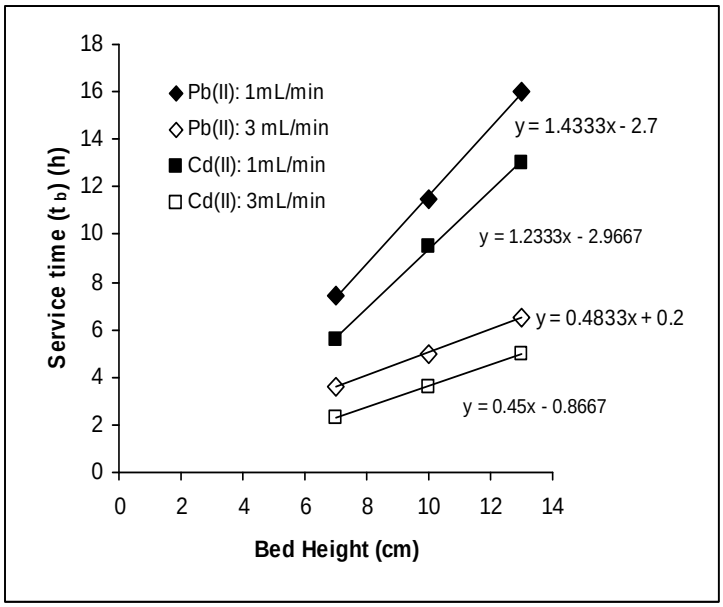

Fig. 4 Bed Depth Service Time (BDST) Plot

Regeneration and Reuse of biosorbent

A successful desorption process requires the proper selection of eluants, which strongly depends on the type of biosorbent and the mechanism of biosorption. Also, the elutant must be (i) non damaging to biomass, (ii) less costly, (iii) environmental friendly and, (iv) effective [4]. In the present section an attempt has been made to regenerate the exhausted biosorbent and to use it for various sorption-deorption cycle.

The column regeneration studies was carried out for various sorptions-desorption cycles. For this purpose, the column was initially packed with 9.1 g of immobilized beads for initial bed height of 13 cm and flow rate of to 1 mL/min at constant lead and cadmium concentration of 15 mg/L and temperature ±28^{o}C with 10% HCl for Pb(II) and Cd(II) metal ion. The pH of the solution was initially adjusted at 5 and 6.0 for Pb(II) and Cd(II) respectively. The breakthrough curves thus obtained for three sorption cycles for Pb(II) and Cd(II) was represented in Fig. 4a. The various parameter viz. volume of effluent, breakthrough time, exhaustion time, uptake capacity, critical bed height, desorption time and desorption efficiency were calculated with the help of above mentioned figures and data therefore are given in Table 3. It observed from Fig.4a and Table 3 that breakthrough time decreases from 16 to 14.6 h for Pb(II) and 13h to 10.9 h for Cd(II) and exhaustion time increased from 25.66h to 27.60h for Pb(II)and 19 to 20.4h for Cd(II) respectively, as the regeneration cycle progressed from first to third cycle. Which resulting a broadened mass transfer zone. This behavior may primarily have been due to the gradual deterioration of the biosorbent because repeated usage and due to previous elution processes which affected the biomass binding sites .11 The percentage removal of were found to be 99.5% to 95.9% for Pb(II) and 97.2 to 93.0% for Cd(II) for first to third cycle respectively. Thus, the overall performances of the immobilized beads in all the cycles were very satisfactory so the removal efficiency was very high.

The desorption experiment were performed with already established desorbing agent HCl. The flow rate in desorption process was maintained at 2.0 mL/min to avoid the over contact of the desorbing agent. Desorption curves for all the cycles are presented in Fig. 4b. The curves observed in all the cycles exhibited a similar trend; a sharp increase at the beginning, followed by a gradual decrease. The desorbent performed very well and percent desorption efficiency of 99.9, 99.5 and 99% for Pb(II) and 99.1, 97.5 and 97.0% for Cd(II) for first to third cycle respectively (Table 3). The desorption process was carried out for desorption time of 90, 80 and 72 min for Pb(II) and 95, 90 and 84 min for Cd(II) as compared to exhaustion time of 25.66, 26.50 and 27.60h for Pb(II) and 19, 19.8 and 20.4h for Cd(II) for first, second and third cycle respectively for the sorption process, which resulted in highly concentrated Pb(II) and Cd(II) solutions in only a small volume of the desorbent and time. For instance, in cycle 1 at t = 10 min. and 20 min for Pb(II) and Cd(II) the effluent concentration was 24.8 and 18.3mg/L respectively. The

total volume of Pb(II) and Cd(II) bearing solution (15 mg/L) treated during this regeneration study was around 4.7856 and 3.552 L in three cycles and total volume of 0.1N HCl utilized for desorption process was nearly 0.240 and 0.270 L which corresponds to approximately 3.49 and 2.65 days of continuous operation respectively. HCl is the most efficient and inexpensive desorbent for Cd(II) metal ion desorption in this study.

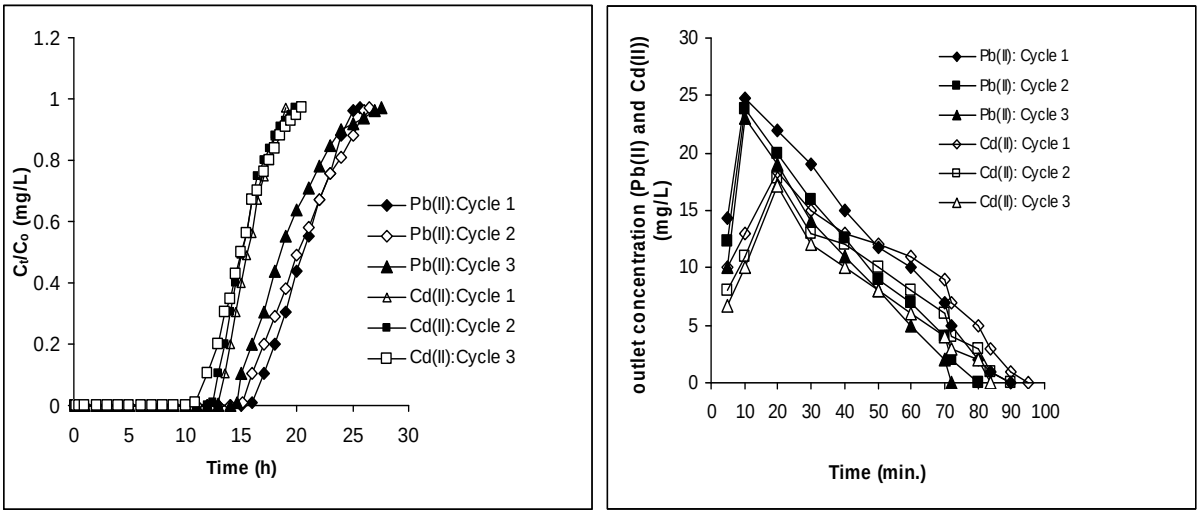

Fig. 5(a) Sorption breakthrough curve for Pb(II) and Cd(II) during three sorption cycle: F= 1 mL/min, C_o = 15mg/L, Z = 25 cm)
Fig. 5 (b) Column desorption curve for Pb(II) and Cd(II) during three desorption cycle : Desorbing agent = 10% HCl, F = 2 mL/min

Table 3 Sorption-Desorption process parameter for various sorption- desorption cycle

Cycle No.	t_b (h)	t_e (h)	Veff (mL)	Q (mg/g)	% Removal	D.T. (min.)	% D.E.
Pb(II): C_o=15 mg/L, F=1mL/min, Z=13 cm							
I	16	25.66	1539.6	2.53	99.5	90	99.9
II	15.1	26.50	1590	2.54	97.1	80	99.5
III	14.6	27.60	1656	2.61	95.9	72	99.0
Cd(II): C_o=15 mg/L, F=1mL/min, Z=13 cm							
I	13	19.0	1140	1.83	97.2	95	99.1
II	12.4	19.8	1188	1.86	95.0	90	97.5
III	10.9	20.4	1224	1.87	93.0	84	97.0

D.T.(h) = Desorption Time; %D.E.= Desorption efficiency

CONCLUSION

The following conclusions can be drawn from the present study:
(1) This study identifies the immobilized microbial biomass of "*Mucor heimalis(*MHB*)*" as a suitable biosorbent to be utilized for continuous removal of Pb(II) and Cd(II) ions from aqueous solution.

(2) The sorption of lead and cadmium is strongly dependent on the bed height, flow rate, and initial metal ion concentration. An increase in bed height resulted in improved sorption performance. With the increase in flow rates, however, the uptake capacity was found to decrease. The increase in initial metal ion concentration resulted in higher uptakes. The maximum uptake of 2.53 and 1.83 mg/g for Pb(II) and Cd(II), respectively, was observed at 13 cm bed height, 1.0 mL/min flow rate, and 15mg/L initial metal ion concentration.

(3) The BDST model was used to predict the relationship between service time and bed height, which is essential in column process design. The bed sorption capacity (N_o) at 1 and 3 mL/min were found to be 410.64 and 415.54 mg/L for Pb(II) and 353.34 and 386.91mg/L for Cd(II) and rate constant (K_a) were 0.0960 and 1.296 L/mg/h for Pb(II) and 0.0874 and 0.299 L/mgh for Cd(II), respectively.

(4) The sorption performance of immobilized MHB for lead and cadmium removal was successfully evaluated for three sorption-desorption cycles using 10%HCl. Thus, using MHB as sorbent provides the opportunity to extract the sorbed lead and cadmium from biosorbent to recycle the biosorbent. Apart from this it being a much cheaper and easily handled decontamination method. The regenerated biosorbent can be utilized again for the removal purpose and then can be disposed of without any harm to the environment.

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