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Article

Biosorptive capacity of Cd(II) and Pb(II) by lyophilized cells of *Pleurotus eryngii*

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The discharge of heavy metals into aquatic ecosystems has become a matter of concern in over the world the last few decades. In this study, the lyophilized cells of *Pleurotus eryngii* (mushroom) were used as an inexpensive biosorbent for Cd(II) and Pb(II) removal from aqueous solutions. The effect of various physicochemical factors on Cd(II) and Pb(II) biosorption such as pH (2.0-7.0), initial metal concentration (0.0-300 mg L^{-1}), temperature, fungal biomass and contact time (0-120 min) were studied. Optimum pH for removal of Cd(II) and Pb(II) was 6.0, and the contact time was 45 min at room temperature. The nature of biosorbent and metal ion interaction was evaluated by Infrared (IR) spectroscopic technique. IR analysis of mushroom biomass revealed the presence of amino, carboxyl, hydroxyl and methyl groups, which are responsible for biosorption of Cd(II) and Pb(II). The maximum adsorption capacities of *P. eryngii* for Pb(II) and Cd(II) calculated using Langmuir adsorption isotherm were 82.0 and 16.13 mg g^{-1}, respectively. The adsorption isotherms for two biosorbed heavy metals were fitted well with Freundlich isotherm as well as Langmuir model with correlation coefficient (r^2>0.99). Thus, this study indicated that the *P. eryngii* is an efficient biosorbent for the removal of Cd(II) and Pb(II) from aqueous solutions.

Key words: Biosorption, *Pleurotus eryngii*, Mushroom, Heavy metals, Adsorption isotherms

Introduction

Current technologies of many industries usually produce hazardous materials. Heavy metals in soils may come from various human activities, such as industrial and energy production, construction, vehicle exhaust, waste disposal, as well as coal and fuel combustion. Heavy metals, such as Pb and Cd are common pollutants in soils mainly due to traffic emissions and leaching from mining locations. Lead and cadmium have no essential biological function and are highly toxic to plants, animals and humans (Smirjdkova et al., 2005).

Environmentally ubiquitous fungi are structurally unique organisms that contribute to the significant removal of metal ions from wastewater than other microbes. This is because of their great tolerance towards heavy metals and other adverse conditions such as low pH, and their intracellular metal uptake capacity (Gadd, 1987). Currently several bio-adsorbents are used including seaweeds, molds, yeast, bacteria, crabshells, agricultural products which are by-products of agriculture and industries. It has been demonstrated that some fungal species are typically associated with heavy metal rich substrata and can be even considered as hyper accumulators of heavy metals (Purvis and Halls, 1996). Among the groups of fungi, basidiomycetes (wood rotting fungi) are useful source of mycelial biomass for biosorption of metal ions, being easy to be cultivated, acquire high yield and is generally regarded as safe. Therefore, biosorbents made from these fungi can be easily accepted by the public when applied practically. The potential of wood rotting fungi to remove metal ions hadn't been studied enough in comparison with micromycetes and the role of macromycetes in this field has been known only for few decades (Gonen et al., 2008; Jarosz-Wilkolazka et al., 2006; Veit et al., 2005; Vimala and Das, 2009). In some cases, their ability to bind metals is same or even better, than that of microfungi and yeast (Javaid, 2008). The ability of the basidiomycetes to adsorb and accumulate metals together with excellent mechanical properties of fungal mycelia provides an opportunity to utilize such candidates in selective sorption of industrial heavy metal ions from

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polluted waters (Bayramoglu et al., 2005; Razmovski & Šćiban, 2008). The mechanisms of heavy metal uptake may include simple physicochemical binding to certain cellular components and extracellular molecules (Gadd, 1997). *Pleurotus ostreatus* accumulated 20% of Cd(II) intracellularly in the presence of 150 mg L^{-1} of Cd(II) in the medium (Favero et al., 1991).

This study aims to determine the possibility of using *P. eryngii* biomass in biosorption of cadmium Cd(II) and lead Pb(II), which are of the most frequent heavy metals found in industrial waste water and acid mine drainage. Our investigation was designed to understand the capacity of *Pleurotus eryngii* (a wood rotting fungus) to remove Cd(II) and Pb(II) from aqueous solution. The influence of initial pH, contact time, temperature, fungal biomass and initial metal ion concentration on biosorption were evaluated.

Materials and Methods

Characterization of *Pleurotus eryngii* *Pleurotus eryngii* has a good viability, (also known as king trumpet mushroom) sold generally as edible mushrooms in every supermarket. An effective cultivation method was introduced to Japan around 1993 and has become popular there in a variety of dishes. *P. eryngii* has high fiber and glucan contents, and contains sterols, proteins, and micro-elements. The protein, carbohydrate, total lipid, crude fiber, and total mineral contents were ranged from 18 g to 38 g; 9 g to 50 g; 1 g to 12 g; 8 g to 52 g; and 5 g to 12 g, respectively, per 100 g of the mushroom species (Khan et al., 2009). Identification of our isolated mushroom was confirmed by 18S rDNA sequence analysis as *Pleurotus eryngii* (Fig. 1). White-rot fungi are capable of accumulating high levels of heavy metals from the environment (Baldrian, 2003). *Pleurotus ostreatus* accumulated 20% of Cd(II) intracellularly in the presence of 150 mg L^{-1} of Cd(II) in the medium (Favero et al., 1991). *Pleurotus ostreatus* easily tolerated up to 5 mM Cd in straw or soil (Baldrian and Gabriel, 2003). *Pleurotus eryngii* showed the strong abilities to accumulate Zn and Co (Drzewiecka et al., 2010).

Biosorbent material A commercial edible mushroom '*Pleurotus eryngii*' was used in this study. *P. eryngii* was bought from the market in Chuncheon, Gangwon-do, Republic of Korea. The mushroom was rinsed three times with sterile water then freeze-dried using lyophilizer (FD5505 Ilshin, Korea). Identification of the mushroom as *Pleurotus eryngii* was confirmed by 18S rDNA sequence analysis.

Pb(II) and Cd(II) solutions A stock Pb(II) solution of 1,000 mg L^{-1} was prepared by dissolving 1.61 g $Pb(NO_3)_2 \cdot 3H_2O$ in 1,000 mL of deionized water. Stock Cd(II) solution of 1,000 mg L^{-1} was prepared by dissolving 2.3709 g $CdSO_4 \cdot 8/3H_2O$ in 1,000 ml of deionized water. The chemicals used for this study were of analytical grade and they were supplied by Sigma Aldrich (Sigma Aldrich, St. Louis, MO). The heavy metals stock solutions were sterilized by autoclaving at a pressure of 1.5 atm and a temperature of 121℃ for 15 min.

Biosorption methodology The freeze-dried cells (100 mg) were inoculated to a series of Erlenmeyer flasks (9 flasks for each metal) containing the diluted solution (30 mL) with 50 mg L^{-1} of each heavy metal studied. The flasks were shaken (150 rpm) at 30℃ for a certain time (0-60 min). The fungal biomass was separated from the solutions at the time intervals (0, 5, 10, 15, 20, 25, 30, 45 and 60 min) using centrifuge at 10,000 rpm for 10 minutes at room temperature. To study the effect of different concentrations of metal and pH on biosorption; initial concentrations (50, 100, 150, 200, 300 mg L^{-1} of each metal) and pH (2.0, 3.0, 4.0, 5.0, and 6.0) at initial concentration (100 mg L^{-1}) were carried out using 100 mg of lyophilized cells at 30℃ at the equilibrium time. All the biosorption experiments were repeated three times to confirm the results. Also, blank experiments were conducted to ensure that no adsorption had taken place on the walls of the apparatus used.

Biosorption equilibrium models The biosorption isotherm curve represents the equilibrium distribution of metal ions

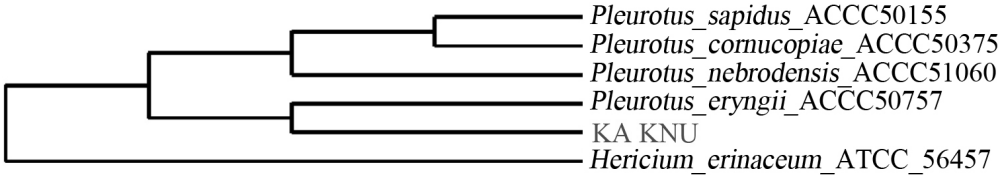

Fig. 1. Phytogenetic tree of *Pleurotus eryngii*.

between the aqueous and solid phase. Several isotherm models are available to describe this equilibrium distribution. The two most common types for describing this type of system are the Langmuir and Freundlich. The Freundlich isotherm describes a more complicated adsorption model where interactions occur between adsorbed molecules. It describes monolayer adsorption on a heterogeneous surface. It takes the following mathematical form (Eq. 1) (Volesky 1990).

$$q_{eq} = K_f C_e^{1/n} \quad (1)$$

where, K_f and n are the adsorption capacity and the intensity of adsorption, respectively. Freundlich parameters can be determined from the linear form of the equation (1) by plotting the ln q versus ln C_e, the slope is the value of 1/n and the intercept is equal to ln K_f.

The linear form of this model is given in equation (2)

$$\ln q_{eq} = 1/n \ln C_e + \ln K_f \quad (2)$$

The Langmuir isotherm model was chosen to estimate the maximum adsorption capacity corresponding to complete monolayer coverage on the biomass surface. The mathematical formula of Langmuir equation can be expressed as

$$q_{eq} = \frac{q_{\max} b C_e}{1 + b C_e} \quad (3)$$

The linear form of Langmuir is

$$C_{eq}/q_{eq} = 1/q_{max}b + C_{eq}/q_{max} \quad (4)$$

where q_{max} is the Langmuir constant (mg g^{-1}) reflecting the maximum adsorption capacity of the metal ion per unit weight of fungal biomass to form a complete monolayer on the surface bound at high C_{eq}. C_{eq} is the unadsorbed metal concentration remaining in the solution. The value of Langmuir constant b (L mg^{-1}) represents a ratio of adsorption rate constant to desorption rate constant, which also gives an indication of the affinity of the metal for binding sites on the biosorbent. q_{max} and b can be determined from the linear form of Langmuir equation (3) by plotting C_{eq}/q_{eq} vs. C_{eq}.

FT-IR spectra Fourier Transform Infrared Spectroscopy (FT-IR spectra) for the samples was performed in order to give a qualitative and preliminary characterization of the main functional chemical groups present on the fungal biomass which are responsible for heavy metal biosorption. A raw sample of fungal biomass and fungal biomass loaded with Cd(II) and Pb(II) were analyzed using FT-IR (Bio-Rad, FTS, 3000 MX) adopting KBr disk technique.

Data evaluation The specific metal biosorption q was calculated using the following equation [5]:

$$q_e(mg/g) = \left[\frac{C_i - C_e}{M}\right] * V \quad (5)$$

where q_e is the specific metal biosorption (mg metal g biomass^{-1}), V is the volume of metal solution (l), C_i and C_e are the initial and equilibrium concentration of metal (mg metal L^{-1}), respectively, and M is the dry weight of the biomass (g).

Results and Discussion

The investigation of the efficacy of the metal uptake by the microbial biomass is essential for the industrial application of biosorption, as it gives information about the equilibrium of the process which is necessary for the design of the equipment.

The major factors affecting the biosorption process are (1) the contact time (2) initial metal ion concentration, (3) temperature, (4) pH, and (5) biomass concentration in solution.

Equilibrium contact time Firstly, sorption experiments were performed for different contact time with fixed concentration, temperature and pH. Our data revealed that the optimum contact time was 45 min at room temperature for biosorption of both Pb(II) and Cd(II) by *Pleurotus eryngii*. Increasing time more than 45 min did not show any increase in the biosorption of heavy metal. However, Pb(II) showed higher biosorption capability to be biosorbed by *Pleurotus eryngii* comparing to Cd(II) (Fig. 2). This favorable type of adsorption may be explained by the difference in their ionic radii (Joo et al., 2010). The ionic radius of Pb(II) is 1.20 A° while for Cd(II) is 0.97 A°. The smaller the ionic radius, the greater its tendency to be hydrolyzed, leading to reduced biosorption. Passive uptake is thought to be physical adsorption or ion exchange at the cell surface, reaching the adsorption equilibrium within 30-40 min (Das et al., 2008). Equilibrium adsorption

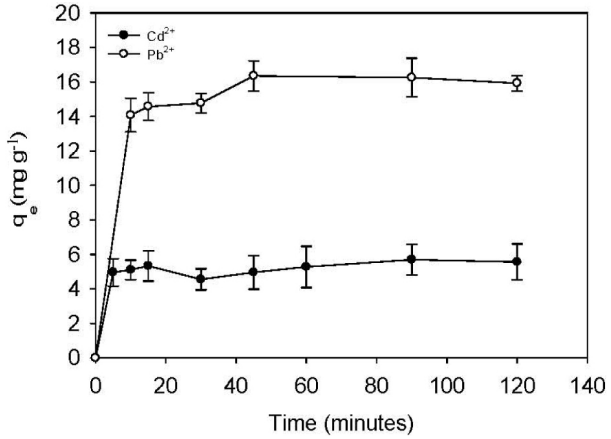

Fig. 2. Effect of contact time on the biosorption of Cd(II) and Pb(II) by lyophilized cells of *Pleurotus eryngii* (biomass dosage 3 g L^{-1}, heavy metal (100 mg L^{-1}), pH (6.0) and temperature (30℃)).

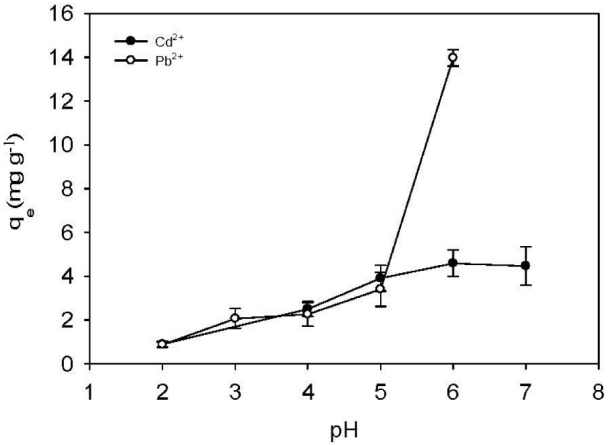

Fig. 3. Effect of pH on the biosorption of Cd(II) and Pb(II) by lyophilized cells of *Pleurotus eryngii* (biomass dosage 3 g L^{-1}, heavy metal (100 mg L^{-1}), contact time (30 min), and temperature (30℃)).

levels of Pb(II) and Cu (II) by filamentous fungi present on the corncob surfaces were attained after about 90 minutes of exposure (Jonglertjunya, 2008). This appears to be an energy-independent surface binding process that occurs at a rate of about 2 mg Cd(II) per gram of mycelial dry weight. *Rhizopus* sp. biosorbed 80% of Ni(II) within 2 hours at an initial concentration of 10 mg L^{-1} (Mogollon et al., 1998).

Effect of initial pH on adsorption capacity The pH seems to be the most important parameter in the biosorption process. It affects the solution chemistry of the metals, the activity of the functional groups in the biomass and the competition of the metalic ions (Friis and Keith, 1998; Galun et al., 1987). The results presented in Fig. 3 also showed that the amount of adsorbed heavy metal ions was dependent on the initial pH of the solution and the type of biosorption media. The rate of biosorption was markedly influenced by the levels of initial pH in the solution. In this study, optimum pH for removal of Cd(II) and Pb(II) was 6.0. Below pH 6.0 the biosorption process activity decreased gradually (Fig. 3). Low pH compared to the optimum pH 5.5 reduced Zn sorption because of the strong competition from hydrogen ions for binding sites on fungi (Zhou, 1999). In similar findings by earlier investigators it has been attributed to protonation or poor ionization of acidic functional group of cell wall at low pH, inducing a weak complexation affinity between the cell wall and the metal ions (Chergui et al., 2007). Low pH (4.0 and below) limits the biosorption of Cu(II), Ni(II) and Zn(II) ions on fungal biomass surfaces, probably due to the ion exchange between metallic species and competition effects with oxonium (hydronium) ion to some extent in the biosorption mechanism (Fourest and Volesky, 1997; Yin et al., 1999).

Effect of metal concentration The results of present findings clearly indicate that the sorption capacity increased and reached a saturation value as the metal ion concentration increased in aqueous medium (Fig. 4). However, Pb(II) showed higher ability than Cd(II) to be biosorbed by *P. eryngii* biomass. This assessment is in line with previously reported data on metal ion sorption by many other similar studies (Javaid et al., 2010; Sheng et al., 2007). There is evidence that at high metal ion concentration the number of ions sorbed is more than at low metal concentration, where more binding sites were free for interaction (Mukhopadhyay et al., 2007). Biosorption is mainly used to treat wastewater where more than one type of metal ions would be present; the removal of one metal ion may be influenced by the presence of other metal ions. The effect of metal ion concentration on the biosorption activity in this experiement of Pb(II) and Cd(II) was carried out separately (Fig. 4). Lead, copper, and cadmium display a lower adsorptive capacity when present together in solution than when present as individual metal ions in solution (Bai and Abraham, 2002). However, uranium uptake by biomass of bacteria, fungi and yeasts was not affected by the presence of manganese, cobalt, copper, cadmium, mercury and lead in solution (Sakaguchi and Nakajima, 1991).

Effect of fungal biomass Fungal biosorbents can sorb heavy metals such as Cu, Al and Sb from aqueous solution

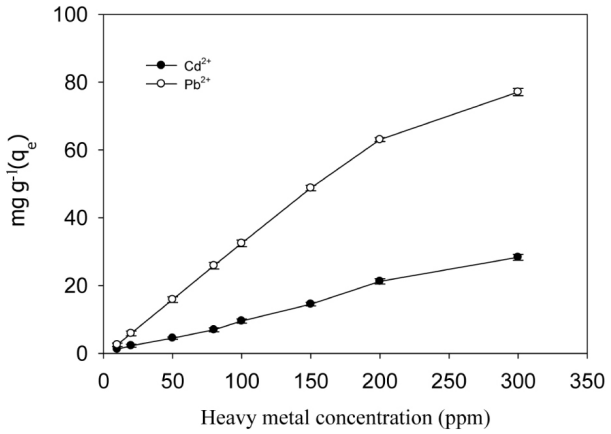

Fig. 4. Effect of heavy metal concentration on the biosorption of Cd(II) and Pb(II) by lyophilized cells of *Pleurotus eryngii* (biomass dosage 3 g L^{-1}, pH (6.0), contact time (30 min), and temperature (30℃)).

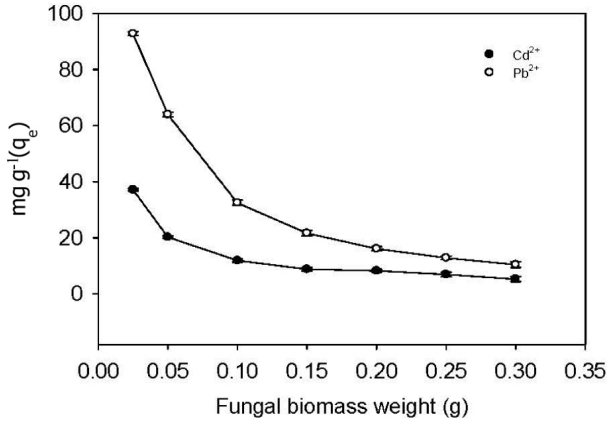

Fig. 5. Effect of the fungal biomass weight on the biosorption of Cd(II) and Pb(II) by lyophilized cells of *Pleurotus eryngii* (heavy metal (100 mg L^{-1}), pH (6.0), contact time (30 min), and temperature (30℃)).

and the process is dependent on fungal species, biosorbent size and concentration, solution pH and ionic composition (Tomko et al., 2006). The effect of the fungal biomass weight on the biosorption of Cd(II) and Pb(II) was investigated at heavy metal concentration of 100 mg L^{-1}, pH 6.0, contact time 30 min, and temperature 30℃. According to our results the biosorptive capacity was decreased by increasing the biosorbent dosage (Fig. 5). That may be due to an increase in biomass concentration which leads to crowding of the binding sites and decreases the specific adsorption. This assessment is in agreement with previously reported data. Biosorption generally decreased with increase in biosorbent particle size and its concentration (Zhou, 1999). Fourest and Roux (1992) invalidated this hypothesis attributing the responsibility of the specific uptake decrease due to metal concentration shortage in solution. Biomass concentration

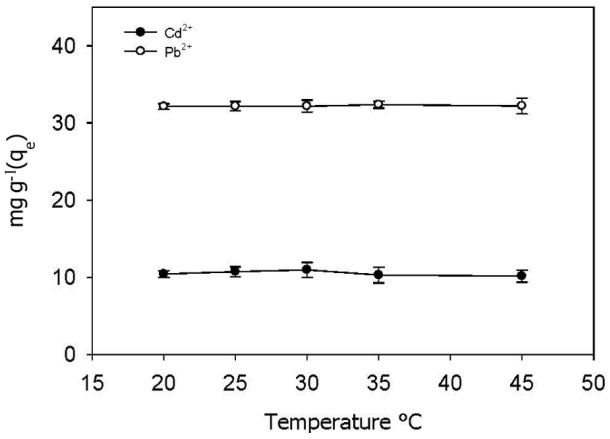

Fig. 6. Effect of temperature on the biosorption of Cd(II) and Pb(II) by lyophilized cells of *Pleurotus eryngii* (biomass dosage 3 g L^{-1}, heavy metal (100 mg L^{-1}). pH (6.0) and contact time (30 min)).

in the solution seems to influence the specific uptake; for lower values of biomass concentration lead to interference between the binding sites (Das et al., 2008). Ultimately, the biosorption results not only in metal removal but also provides an eco-friendly environment. Hence, this factor needs to be taken into consideration in any application of microbial biomass as biosorbent. (Ahalya et al., 2003).

Effect of temperature Temperature has important effects on the adsorption process. As the temperature increases, the rate of diffusion of adsorbate molecules across the external boundary layer and interval pores of the adsorbent particle increases (Ho and McKay, 1998). The temperature range of 20-45℃ apparently exhibited no significant influence on biosorption potential of test fungal species (Javaid et al., 2010). The findings are in conformity with our experiment in which temperature is not seemed to influence the biosorption performances in the range of 20-45℃ (Fig. 6). The negated effect of temperature on biosorption performance within this range was probably due to exothermic reaction (Saglam et al., 2002). Aksu et al. (1992) reported that temperature did not influence the biosorption processes in the range of 20-35℃.

Biosorption isotherm - assessment of sorption performance The equilibrium distribution is important in determining the maximum biosorption capacity. The two widely accepted and linearized equilibrium adsorption isotherm models for single solute system are given by the following:

$$C_{eq}/q_{eq} = 1/q_{max}b + C_{eq}/q_{max}$$

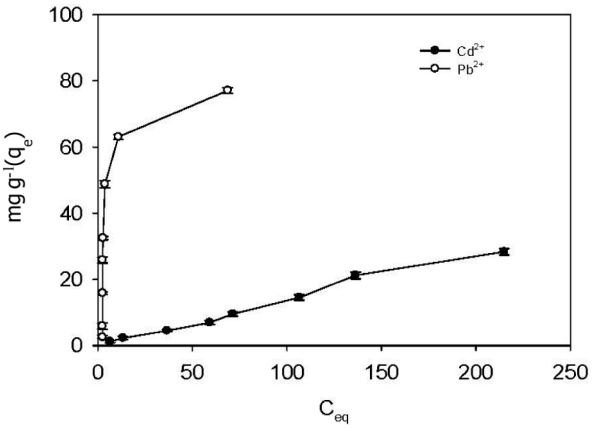

Fig. 7. Biosorption isotherm of Cd(II) and Pb(II) by lyophilized cells of *Pleurotus eryngii* (biomass dosage 3 g L^{-1}, heavy metal (100 mg L^{-1}), temperature 30℃, pH (6.0), and contact time (30 min)).

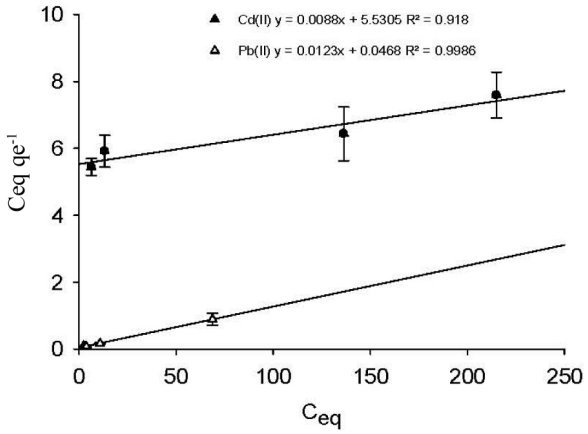

Fig. 8. The linear form of Langmuir adsorption isotherm of Cd(II) and Pb(II) by lyophilized cells of *Pleurotus eryngii*.

where q is milligrams of metal accumulated per gram of the biosorbent material; C_{eq} is the metal residual concentration in solution; q_{max} is the maximum specific uptake corresponding to the site saturation and b is the ratio of adsorption and desorption rates. This is a theoretical model for monolayer adsorption. Another empirical model for monolayer adsorption is

$$\ln q = 1/n \ln C_e + \ln K_f$$

These models can be applied at a constant pH. These models are used in literature for modeling of biosorption equilibrium in the presence of one metal. These values are plotted in a 2D line where the specific uptake q is reported as a function of the metal concentration C_{eq}.

But the above stated adsorption isotherms may exhibit an irregular pattern due to the complex nature of both the sorbent material and its varied multiple active sites, as well as the complex solution chemistry of some metallic compounds. Various applications are available for biomass immobilization. The principal techniques that are available in literature for the application of biosorption are based on adsorption on inert supports, on entrapment in polymeric matrix, on covalent bonds in vector compounds, or on cell cross-linking.

In this investigation, biosorption of Cd(II) and Pb(II) by lyophilized cells of *P. eryngii* at biomass dosage 3 g L^{-1}, heavy metal 100 mg L^{-1}, temperature 30℃, pH 6.0, and contact time 30 min. The initial sharp decrease in heavy metal ion's concentrations in the liquid-phase implied higher rate of biosorption (Fig. 7). At lower concentrations, all metal ions present in the solution could interact with the binding sites and thus the biosorption percentage was higher than those at higher ion concentrations. The high magnitude of the correlation coefficients (Table 1) shows that both Freundlich and Langmuir models were fitted well to illustrate the biosorption pathway of Pb(II) and Cd(II) by *P. eryngii*.

Langmuir Adsorption Isotherm The equilibrium adsorption isotherm is one of the main parameters in the design of adsorption systems. The Langmuir isotherm equation is used in this study since the Freundlich isotherm is not consistent to detect the maximum biosorptive capacity of *P. eryngii* to remove Cd(II) and Pb(II) from aqueous solution with the present data. The maximum adsorption capacities for Pb(II) and Cd(II) biosorption by *P. eryngii* calculated from Langmuir adsorption isotherm were 82.0 and 16.13 mg g^{-1}, respectively. Fig. 8 shows the linear form of Langmuir adsorption isotherm of Cd(II) and Pb onto the lyophilized cells of *P. eryngii*. Table 1 indicates very high regression correlation coefficients (r^2>0.99) and (r^2>0.92) for Pb(II) and Cd(II), respectively.

Freundlich Adsorption Isotherm The magnitudes of intercept Kf and *n* (Freundlich constants) were calculated from Freundlich plots. The value of *n*, which is related to the distribution of bonded ions on the sorbent surface, was greater than unity. The magnitude of Freundlich constant expresses easy separation of metal ions from aqueous medium and indicates favorable adsorption. The adsorption isotherm for two biosorbents fitted well with Freundlish isotherm than Langmuir model with correlation coefficient (r^2>0.99). Table 1 indicates very high regression correlation coefficients (>0.80) and (>0.98) for Pb(II) and Cd(II),

Table 1. Langmuir and Freundlich isotherm parameters for Pb(II) and Cd(II) biosorption by *Pleurotus eryngii*.

	q_{max}	b	r^2	n	K_f	r^2
Pb(II)	82	0.25	0.998	4.34	31.5	0.80
Cd(II)	16.13	0.011	0.92	1.1	5.1	0.99

K_f sorptive capacity, n sorptive intensity, r^2 correlation coefficient, b Langmuir constant, q_{max} maximum adsorption capacity.

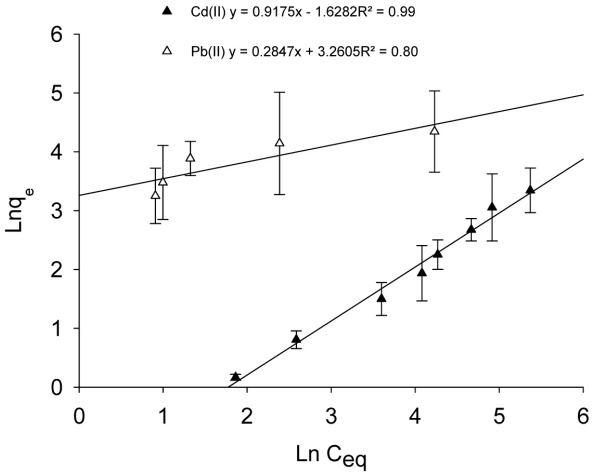

Fig. 9. The linear form of Freundlich adsorption isotherm of Cd(II) and Pb(II) by lyophilized cells of *Pleurotus eryngii*.

respectively. Figure 9 shows the linear form of Freundlich adsorption isotherm of Cd(II) and Pb(II) by lyophilized cells of *Pleurotus eryngii* respectively.

FT-IR Spectra of Cadmium and Lead-loaded *Pleurotus eryngii* Another technique related to the fungal biosorption phenomenon was carried out using FT-IR. FTIR spectroscopy method was used to show the functional groups present on the surface of the adsorbent. Fungi as biosorbent have been proved effecient and economical for the removal of toxic metals from dilute aqueous solutions (Horikoshi et al., 1981).

Figure 10 shows the FTIR spectra of heavy metal free lyophilized cells of *Pleurotus eryngii* and after loading of heavy metal (biosorption of Cd(II) and Pb(II)) on the fungal biomass at room temperature. The fungal biomass was dried to nil moisture before and after biosorption and sent for FT-IR analysis. This analysis had eventually confirmed the difference between functional groups in relation to biosorption of cadmium and lead. Figure 10 shows the changes in the spectrum of the biomass after sorption of Pb(II) and Cd(II). An interesting phenomenon was the sharp decrease in the band intensity at 500-600

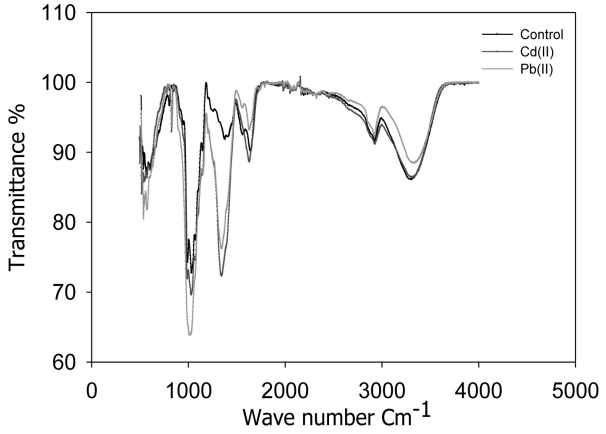

Fig. 10. FTIR spectra of heavy metal free lyophilized cells of *Pleurotus eryngii* and after loading of heavy metal (biosorption of Cd(II) and Pb(II)) on the fungal biomass at room temperature.

cm^{-1} and 1040-1100 cm^{-1} corresponding to C-H and C=O stretching, respectively, after metal binding. On the basis of the change of the band, it was reasonable to assume that the peak value suggested the metal chelating. FT-IR spectrum of *P. eryngii* thallus before and after biosorption (Fig. 10) shows the difference in peak at 1330-1430 cm^{-1} corresponding to O-H bond and little change in the region correspond to C-H bond (600-700 cm^{-1}). There may be shifting of functional group and the change in this region suggested metal bonding during the adsorption in *P. eryngii* also. The very strong adsorption band around 3200-3400 cm^{-1} found in these samples may be due to presence of N-H stretching of amines and amides and polymeric association which was normally found in hydroxyl compounds. The IR analysis of biosorbent specifically the 1650-1620 cm^{-1} band indicated the existence of the amide band of amide bond in poly-N-acetyl glucosamine (chitin) and the protein peptide bond present in biomass considered to be due to combined effect of double bond stretching vibrations (mainly C=O) and hydrogen bonding (Li et al., 2008). The adsorption band around 2900-2850 cm^{-1} correspond to C-H stretching of CH_2 groups. The stretching of peak from 1030-1110 cm^{-1} could be due to the involvement of the C=O of polysaccharides in the biosorption process. Fungi offer a wide range of chemical groups that can attract and sequester the metals in biomass. Cell walls are composed of structural polysaccharides, proteins and lipids that offer metal-binding functional groups (Veglio and Beolchini, 1997). FT-IR analysis of acid treated immobilized *A. niger* was used for a qualitative and preliminary analysis of chemical functional groups present on its cell wall which

provided the information on nature of cell wall and Cr (VI) interaction during the process of biosorption (Chhikara et al., 2010).

The FT-IR spectra of *P. eryngii* dried biomass recorded before and after heavy metal biosorption has shown some changes in the peak patterns, which were finally analyzed and was found that chemical interaction such as ion-exchange between methyl (-CH_3), amide (-NH_2) and hydroxyl (-OH) group of biosorbent and heavy metal ion were mainly involved in biosorption of heavy metals onto *P. eryngii* cell wall surface. In biosorption research, there is a lack of information about the sorption sites and mechanisms responsible for the capture of metallic ions by the biomasses (de Carvalho, 2003). Fungal melanins of filamentous fungi also contribute to the removal of metals, and their interactions with metals have been explained by Fogarty and Tobin (1996). Na, K, Ca and Mg ions were released from the biomass after biosorption of Pb, Cd, Ni and Zn, indicating that ion exchange was a key mechanism in the biosorption of metal ions by *Mucor rouxii* biomass (Yan and Viraraghavan, 2008). Scanning electron microscopy coupled with x-ray energy dispersion analysis indicated that K(I) and Ca(II) are replaced by Pb(II) on the cell wall of *Mucor rouxii* (Lo et al., 1999). Among various biosorbents, chitin is the second most abundant natural biopolymer after cellulose. However, more important component than chitin is chitosan, which has a molecular structure similar to cellulose. Presently, chitosan is attracting an increasing amount of research interest, as it is an effective scavenger for heavy metals. However, a major difference results from the fact that fungal chitin is associated with other polysaccharides which do not occur in the exoskeleton of arthropods. Furthermore, the occurrence of chitosan is apparently restricted to fungi (Peter, 2005).

Conclusion

In this study, the lyophilized cells of *Pleurotus eryngii* were used as an inexpensive biosorbent for Cd(II) and Pb(II) removal from aqueous solutions. *Pleurotus eryngii* is common macrofungus so it is very available in all markets and cheap in comparing to other biosorpent e.g. bacteria or micro-fungi which need higher cost and facilities for cultivation in sufficient amount. Factors affecting biosorption e.g. initial pH, contact time, temperature, fungal biomass dose and initial metal ion concentration were studied. The data revealed that the optimum contact time was 45 min at room temperature for biosorption of both Pb(II) and Cd(II) by *P. eryngii*. Optimum pH for removal of Cd(II) and Pb(II) was 6.0. Below pH 6.0, the biosorption process acivity decreased gradually. The biosorptive capacity decreased as increasing the biosorbent dosage. Pb(II) showed higher ability than Cd(II) to be biosorbed by *P. eryngii* biomass. The equilibrium experimental data and the Langmuir type adsorption isotherms showed that the dried biomass of *P. eryngii* as a biosorbent material have high affinity and large sorption capacity for Pb(II) and Cd(II). Langmuir isotherm equation indicated very high regression correlation coefficients (>0.99) and (>0.92) for Pb(II) and Cd(II), respectively. The maximum adsorption capacities for Pb(II) and Cd(II) biosorption by *P. eryngii* from Langmuir adsorption isotherm were 82 and 16.13 mg g^{-1}, respectively. The magnitude of Freundlich constants K_f and n expressed easy separation of metal ions from aqueous medium and indicated favorable adsorption. The adsorption isotherm for were fitted well with both of Freundlich and Langmuir adsorption isotherm models. FTIR spectra confirmed the changes in the functional groups and the surface properties of fungal biosorbent after loading of heavy metal, and imply the role of hydroxyl, carbonyl, amid, and methyl groups in metal chelating. Use of *P. eryngii* (mushroom) biomass instead of resins and activated carbon or conventional adsorbents is good alternative for metal ion removal from industrial effluent.

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