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Biosorption of Lead by *Pleurotus florida* and *Trichoderma viride*

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Authors' contributions

This work was carried out in collaboration between all authors. Equal contribution was given by four authors. All authors read and approved the final manuscript.

Research Article

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ABSTRACT

Aims: The objective of the work is to remove Pb^{2+} by *Pleurotus florida* and *Trichoderma viride* in batch studies and to study the kinetics and adsorption isotherm of Pb^{2+} adsorption by fungal species and to determine the desorption performance by suitable desorbing agents.
Study Design: Experimental study.
Place and duration of the study: This work was carried out at Department of Biotechnology, Government College of Technology, Coimbatore, Tamil Nadu, and India for a period of five months.
Methodology: The polluted sample was collected from Valankulam lake, Coimbatore. The biomass of *Pleurotus florida* and *Trichoderma viride* were used as adsorbents. Atomic Absorption Spectrophotometer was used to quantify Pb^{2+} concentration. The optimum conditions of pH, adsorbent dose and contact time for biosorption were determined.
Results: Maximum adsorption of Pb^{2+} for *Trichoderma viride* and *Pleurotus florida* were observed at a pH of 6 and 7 respectively. The optimum quantities of adsorbent required for the removal of Pb^{2+} were 0.2g for both the organisms. Adsorption of Pb^{2+} was found to reach equilibrium in 1 h and 1.5 h for *Pleurotus florida and Trichoderma viride* respectively.
Conclusion: Hence, *Pleurotus florida* and *Tricoderma viride* are suitable adsorbents for the removal of Pb^{2+} from effluents. This methodology can be used for the removal of lead

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from waste water before its disposal.

Keywords: Biosorption; toxic heavy metals; adsorbent; effluent.

1. INTRODUCTION

Heavy metals released by a number of industrial processes are major pollutants in marine, ground, industrial and even treated wastewaters. The most important characteristics of these metals are that they are non degradable and can accumulate in living tissue [1]. Therefore, for reducing lead mediated pollution in environment, Pb^{2+} should be removed from wastewater before its disposal [2]. The conventional methods for heavy metal removal from industrial effluents are precipitation, coagulation, ion exchange, cementation, electro dialysis, electro winning, electro coagulation and reverse osmosis [1,3,4].

Biosorption is a promising method for removal of toxic metal ions by living and dead microbial cells from aqueous solutions [5,6,7,8]. It can be efficiently used for the treatment of large volumes of effluents with low concentration of pollutants. So, the process does not depend on the viability of the biomass [9]. The applicability of fungi as biosorbent has some advantages due to their small size, ubiquity, ability to grow under controlled conditions and resilience to a wide range of environmental situations.

Biosorption is the binding and concentration of heavy metals from aqueous solutions (including very dilute concentrations) by certain types of inactive, dead and live microbial biomass [10,11]. Fungi, in common with other microbial groups, can accumulate metals from their external environment by means of physico-chemical and biological mechanisms [10]. Biosorption is a promising method for removal of toxic metal ions from waste water. Its advantage is especially in the treatment of large volumes of effluents with low concentration of pollutants. The major advantages of biosorption over conventional treatment methods include: Low cost, good efficiency, minimization of chemical and biological sludge, regeneration of biosorbent, and possibility of metal recovery [9]. The Pb^{2+} concentration in the sample was analyzed by Atomic Absorption spectrophotometer [10,12].

Fungi are chosen for biosorption because of their special physiology and adsorbing capacity. Chitin and chitosan present in fungi are well known metal ion adsorbers due to the presence of both carboxyl and amine groups.

2. MATERIALS AND METHODS

2.1 Media Used and Biomass Production

Oyster mushroom (*Pleurotus florida*) and *Trichoderma viride* were obtained from Tamil Nadu Agricultural University, Coimbatore. Potato Dextrose Agar and Molasses Yeast liquid medium were used for the maintenance and growth of *Trichoderma viride* and *Pleurotus florida* respectively [10,13]. 70ml of molasses medium was transferred to the 250 ml conical flask and autoclaved for 15 minutes. The mycelia disc of *Trichoderma viride* [14] was transferred to the flask and incubated for 10 days at 25ºC. After three days, the growth of the fungus was observed in the form of pellicles, which increased in diameter on subsequent days. The mycelial mat was collected by filtering through muslin cloth after 10 days of growth and washed thoroughly with deionized water to remove the growth medium sticking on its surface [15].

2.2 Preparation of Standard and Adsorbents

Standard solutions of Pb^{2+} nitrate were prepared in the range of 0ppm to 20ppm using double distilled water. For regeneration studies, Hydrochloric acid and Sodium hydroxide of different concentrations of 0.01 M, 0.1M and 1M were used [10]. The obtained biomass of *Pleurotus florida* and *Trichoderma viride* were cleansed with distilled water and dried at 60ºC for a period of 24h. The biomasses were ground into fine particles using electric grinder and mortar and pestle respectively. The particles were sieved using 300µm sieve and used for biosorption studies.

2.3 Pb^{2+} Analysis

The presence of Pb^{2+} in the sample was analysed by Atomic Absorption Spectro photometer (GBG 901, Australia). The samples for Pb^{2+} analysis were collected from Valankulam Lake (Coimbatore Corporation).

2.4 Batch Adsorption

The biosorption studies were conducted in batch process to evaluate the effect of pH, initial metal concentration and contact time at different concentrations of biomass on removal of Pb^{2+} ions. All biosorption experiments were carried out in 250ml of Erlenmeyer flasks in rotary incubator shakers at 140rpm and 30ºC.

Metal uptake (q) can be determined by: [11,10]

$$Q = V \times (Ci - Ce) / S \qquad (1)$$

where,
Q (metal uptake, mg/ g) is the amount of metal ions adsorbed on the biosorbent.
V (ml) is the volume of metal containing solution in contact with the biosorbent.
Ci and Ce (mg /l) are the initial and equilibrium (residual) concentrations of metals in the solution, respectively and
S (g) is the amount of added biosorbent on dry basis.

Effect of pH on biosorption rate of the fungal biomass was investigated in the initial pH range from 3.0 to 10.0 at 30ºC. The pH of the solution was adjusted using 1N HCl and 0.1N NaOH. 0.2g biomass of *Pleurotus florida* and *Trichoderma viride* were transferred to the flasks and the reaction mixture was shaken in rotary incubator shaker at 140 rpm for 3h. The initial concentrations of Pb^{2+} ions were varied from 4 mg/l to 20 mg/l at optimum pH, for 3h contact time. Similarly the contact time was varied from 30 min to 3h to determine optimum biosorption time at optimum pH. Effects of biosorbent dose were also investigated. Biomass dose were varied from 0.04g to 0.24g/100ml of 10mg/l Pb^{2+} solutions. Pseudo first order rate kinetics was applied and value of rate constant k_{ad} were derived from Lagergren plots.

2.5 Adsorption Isotherm

The adsorption isotherm is the initial experimental step to determine the feasibility of adsorption treatment. It is a batch equilibrium test, which provides data relating adsorbate per unit weight to the amount of adsorbate remaining in the solution. Adsorption data for a wide range of adsorbate concentrations are most conveniently described by various adsorption isotherms, namely Langmuir or Freundlich isotherm [15].

The Langmuir model can be described as [4,17,18]

$$q_e = Q_0 \, b \, C_e / 1 + bC_e \quad (2)$$

where,
q_e is the uptake of metal ions per unit weight of the adsorbent.
Q_0 is the moles of solute sorbed per unit weight of adsorbent.
b is the constant relates the affinity between the biosorbents and biosorbate.
C_e equilibrium concentration of ions [15,16].

The constants, Q_0 and b are evaluated from the linear plot of the logarithmic equation.

$$1/q_e = 1/\, Q_0 + 1/bQ_0 \, x \, 1/C_e \quad (3)$$

The Langmuir model is based on the assumption that maximum adsorption occurs, when a saturated monolayer of solute molecule is present on the adsorption surface. The energy of adsorption is constant and there is no migration of adsorbate molecule in the surface plane.

The Freundlich isotherm is of the form: [4,17]

$$q_e = k \, C_e 1/n \quad (4)$$

The logarithmic form of the above equation is as follows:

$$\log q_e = \log k + 1/n \log C_e \quad (5)$$

where,
q_e is the uptake of metal ions per unit weight of biosorption.
C_e is the equilibrium concentration of metal ions in solution.
k is the Freundlich constants denoting adsorption capacity.
n is the empirical constants, is a measure of adsorption intensity [15,16].

The value of k and 1/n were found by plotting the graph between log q_e and log C_e. The value of log k is the intercept and value of 1/n is the slope of the plot. The value of k is determined from the antilog of the intercept value. A high value of 'k' and 'n' indicates high adsorption throughout the concentration range and vice-versa. A low value of 'n' indicates high adsorption at strong solute concentration [19].

3. RESULTS AND DISCUSSION

3.1 Results

Biosorption of Pb^{2+} by *Pleurotus florida* and *Trichoderma viride* are influenced by several factors like pH, initial Pb^{2+} ion concentration, adsorbent dose and contact time.

3.1.1 Effect of pH

The effect of pH on the biosorption of Pb^{2+} by *Pleurotus florida* (A) and *Trichoderma viride* (B) were studied with at pH 3.0 to 10.0. The results are presented in Fig (3.1) and Fig (3.2) respectively. The maximum biosorption of Pb^{2+} by A and B were observed at neutral and acidic pH respectively. It was found that the removal of Pb^{2+} was 100% with *Pleurotus florida* at pH 7.0 and 90% with *Trichoderma viride* at pH 6.0.

3.1.2 Effect of adsorbent dose

The influence of the amount of adsorbent dose on Pb^{2+} adsorption was studied at 30ºC and pH 7.0 for A and pH 6.0 for B respectively, by varying the adsorbent dose from 0.04 g to 0.24 g, while keeping the volume and concentration of the metal solution constant. This is shown in Fig (3.3) & Fig (3.4). But it is apparent that the percentage removal of Pb^{2+} increases rapidly with increase in the dose due to greater availability of the biosorbent. Adsorption is maximum with 0.2 g of biosorbent, when the biosorption concentration was increased from 0.04 g to 0.2 g in 100ml,the Pb^{2+} ion concentration were decreased from 10 mg/g to 5 mg/g for *Pleurotus florida* and 7.5 mg/g to 4.5 mg/g for *Trichoderma viride.*

3.1.3 Effect of contact time

Maximum Pb^{2+} removal with fungal biosorbents was in initial periods of 60 min for *Pleurotus florida* and 90 min for *Trichoderma viride*, after which no significant removal of Pb^{2+} was observed with both the biosorbents. It is shown in Fig (3.5) & Fig (3.6). This rapid sorption stage indicates that surface sorption occur on the fungal cell surface. The kinetics of metal adsorption on the surface is usually rapid during initial period of time. In order to analyze whether sorption of Pb^{2+} follows pseudo first order reaction, kinetic experiments were carried out for a regular interval of 5 min for A and 10 min for B at 10 ppm of 0.2 g of adsorbent [15,1].

Lagergren Pseudo First Order Kinetics [20]

$$\log (q_e - q_t) = \log q_e - k_{ad} / 2.303 \times t \tag{6}$$

where, k_{ad}(min) is the rate constant of adsorption; q_e and q_t are the amount of Pb^{2+} adsorbed(mg/g) at equilibrium and any time t min. This is shown in Fig (3.11) & Fig (3.12). The k_{ad} values are calculated from the graph plotted between log (q_e-q_t) and time t (min) [8].The straight lines and the value of R_2 confirm that the adsorption process follow first order rate kinetics in each case. The value of pseudo first order rate constant was found to be 2.23×10^{-2} for A and 3.385×10^{-2} for B.

3.1.4 Adsorption experiment

The Pb^{2+} adsorption followed Freundlich and Langmuir model. The value of Freundlich constant (k and n) and Langmuir constants (Q_0 and b) were evaluated from Fig (3.7) & Fig (3.8) for A and Fig (3.9) & Fig (3.10) for B. The high value of Langmuir constant Q_0 (12.195 mg/g) and b (0.058) with *Pleurotus florida* than *Trichoderma viride* Q_0 (10.989 mg/g) and b (0.057) indicated better adsorption capacity. Higher values of k and n, and lower values of b indicate better affinity of the biomass.

3.1.5 Regeneration studies

The applicability of fungal biomass for metal ion recovery from waste stream requires that, the biomass be regenerated efficiently. Thus the bound metal can be recovered in concentrated form and the biomass can be reused. Regeneration of adsorbed Pb^{2+} on to biomass was performed by taking different concentration of Hydrochloric acid and Sodium hydroxide at 0.01 M, 0.1 M, and 1 M of 100 ml solution. Metal adsorbed biomass was separated from equilibrium solution by means of centrifugation. It is found that the acidic solution desorption capacity was higher when compared to alkaline solution. For *Pleurotus florida* the order of desorption was 0.1 M HCl> 0.01 MHCl> 1 M HCl> 0.1 M NaOH > 0.01 M NaOH > 1 M NaOH. At 1 M NaOH there is no desorption. In *Trichoderma viride* the order of desorption was 1 M HCl> 0.01 M HCl >0.1 M HCl > 1 M NaOH > 0.1 M NaOH > 0.01 M NaOH. The results are shown in Fig (3.13) & Fig (3.14).

3.2 Discussion

The effect of pH on biosorption is due to interaction of Pb^{2+} cations with the carboxyl groups of chitosan or chitin present on the fungal surface. Effect of adsorbent dose is attributed to reduction of total area of biosorbent due to aggregation during biosorption. The rapid sorption stage indicates that biosorption occurs only on the fungal surface. The extent of adsorption efficiency increases with time and attains equilibrium at 90 minutes for both the adsorbents.

3.3 Tables and Figures

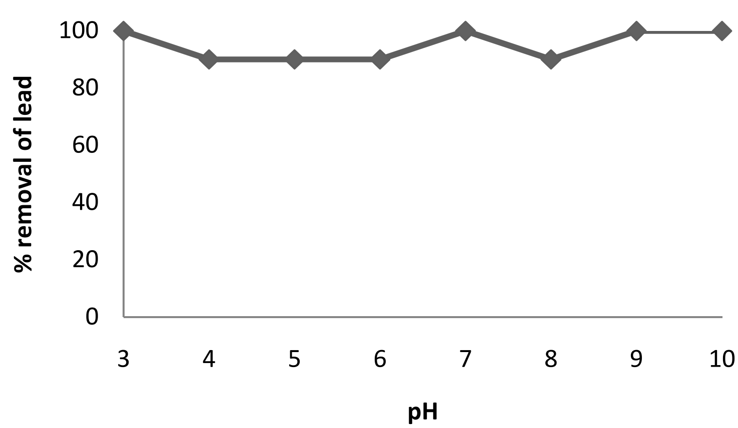

Fig. 3.1 Effect of pH on biosorption of Pb^{2+} by *Pleurotus florida*

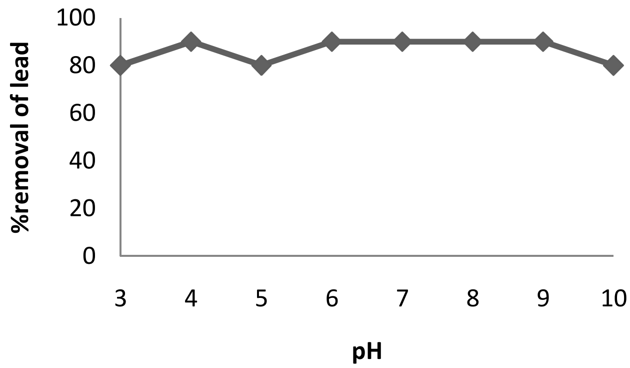

Fig. 3.2 Effect of pH on biosorption of Pb^{2+} by *Trichoderma viride*

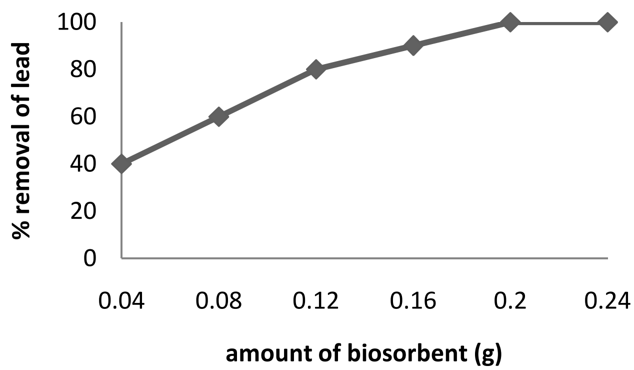

Fig. 3.3 Effect of biosorbent dose on Pb^{2+} biosorption by *Pleurotus florida*

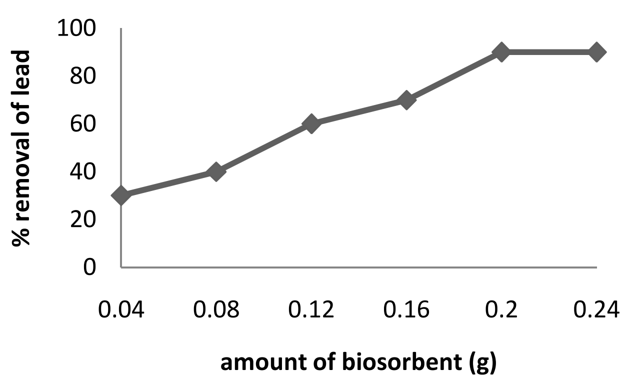

Fig. 3.4 Effect of biosorbent dose on Pb^{2+} biosorption by *Trichoderma viride*

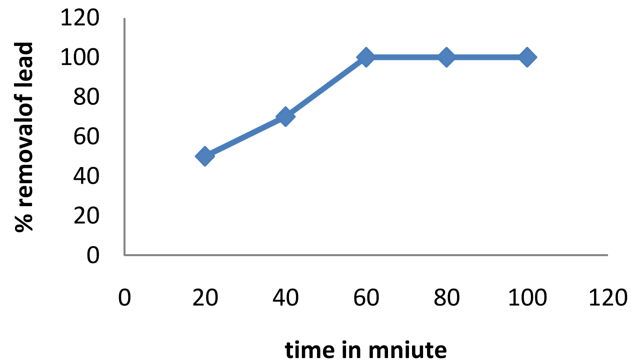

Fig. 3.5 Effect of time on Pb^{2+} biosorption by *Pleurotus florida*

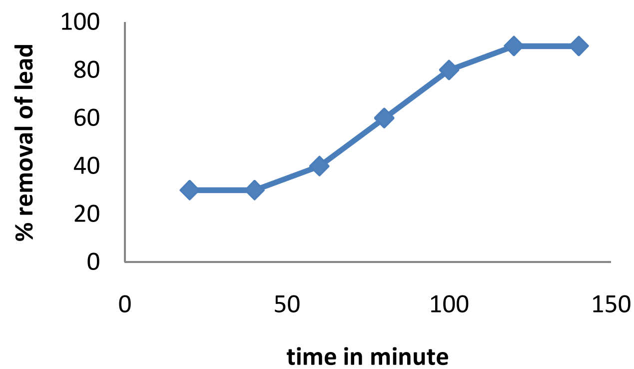

Fig. 3.6 Effect of time on Pb^{2+} biosorption by *Tricoderma viride*

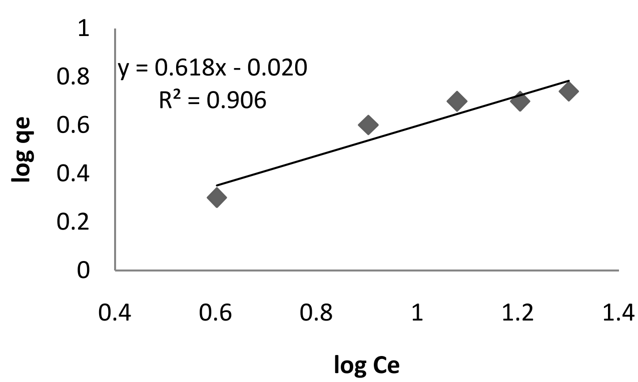

Fig. 3.7 Freundlich isotherm for Pb^{2+} removal by *Pleurotus florida*

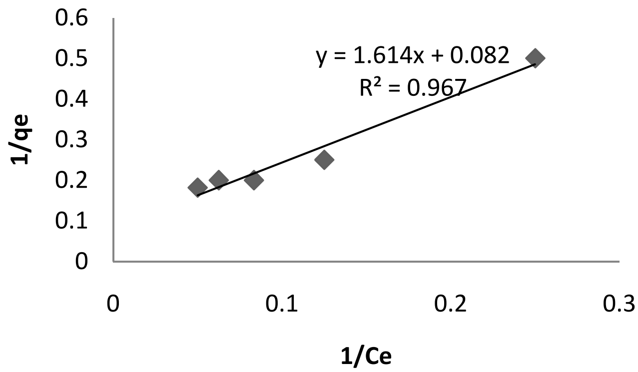

Fig. 3.8 Langmuir isotherm for Pb^{2+} removal by *Pleurotus florida*

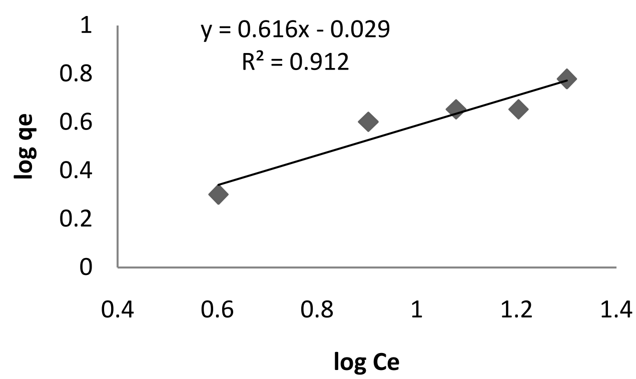

Fig. 3.9 Freundlich isotherm for Pb^{2+} removal by *Trichoderma viride*

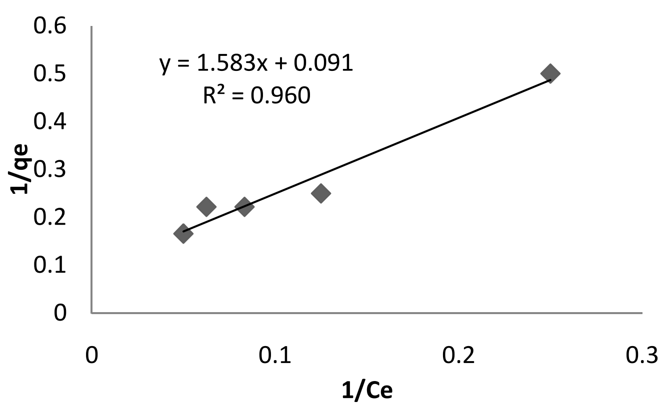

Fig. 3.10 Laungmuir isotherm for Pb^{2+} removal by *Trichoderma viride*

Table 3.1 Langmuir isotherm and Freundlich isotherm constants, coefficient of correlation

Biosorbent	Freundlich constants			Langmuir constants		
	K(mg/g)	N	r^2	Q_0(mg/g)	b(l/mg)	r^2
Pleurotus florida	0.954	1.610	0.906	12.195	0.058	0.967
Trichoderma viride	0.935	1.623	0.912	10.989	0.057	0.960

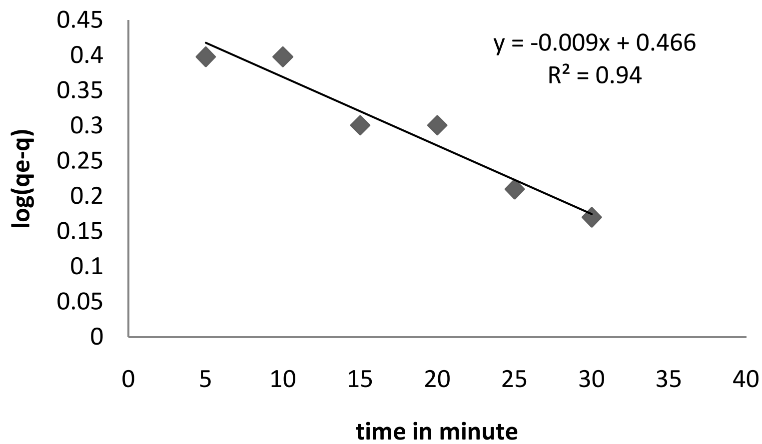

Fig. 3.11 Lagergren Pseudo first order kinetics for Pb^{2+} removal by *Trichoderma viride*

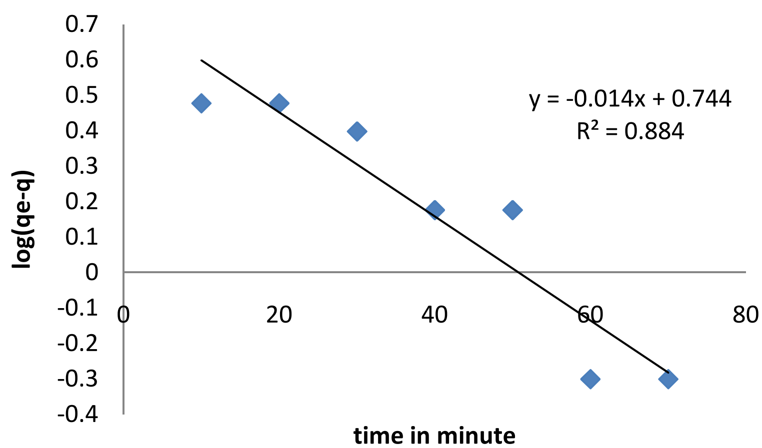

Fig. 3.12 Lagergren Pseudo first order kinetics for Pb^{2+} removal by *Pleurotus florida*

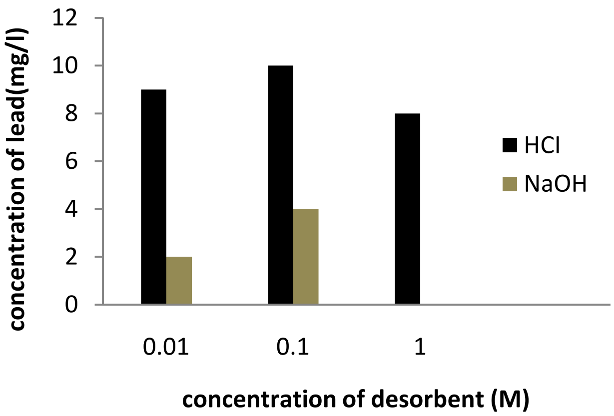

Fig. 3.13 Regeneration of Pb^{2+} from *Pleurotus florida*

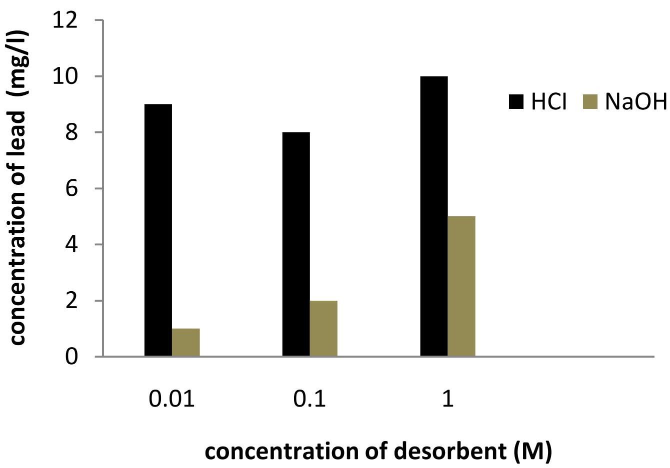

Fig. 3.14 Regeneration of Pb^{2+} form *Trichoderma viride*

4. CONCLUSION

Pleurotus florida and *Trichoderma viride* are suitable for the removal of Pb^{2+} from polluted sample. The adsorption is strongly dependent on pH and adsorbent dose. A maximum of about 90% Pb^{2+} removal was achieved at pH 6 for *Trichoderma viride* and 100% at pH 7 for *Pleurotus florida*. Removal of Pb^{2+} by batch studies showed that 0.2 g of *Pleurotus florida* and *Trichoderma viride* were the optimum quantities required for the removal of Pb^{2+} from 100mL of 10 mg/l Pb^{2+} nitrate solution. Adsorption of Pb^{2+} is fairly rapid in first 20 min and increased slowly to reach equilibrium in 1 h for *Pleurotus florida* and 1.5 h for *Trichoderma viride*. The regression analysis of equilibrium data fitted more to Langmuir adsorption isotherm than Freundlich isotherm. The adsorption of Pb^{2+} follows pseudo first order kinetics. Regeneration study revealed that exhausted biosorbent may be regenerated and used for nearly 6 cycles without significant loss of sorption capacity indicating that fungal biosorbent possesses good attritional characteristics. Over the last few decades, the huge increase in the use of heavy metals has resulted in an increased flux of metallic substances in aquatic environment. So, taking into consideration the health issues of living organisms it is mandatory to remove Pb^{2+} from waste water.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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