A study on the biosorption of lead by living biomass of *Rhizopusarrhizus*(Fisher)

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***Abstract*:** Pollution of water bodies such as lakes, river, and ponds etc. due to the presence of heavy metals increases upto a greater extent and become a key environmental problem throughout the entire world. Conventional methodsfor the removal of metal ions from aqueous system are very expensive, time taking, less effective and are not eco- friendly because they produced huge amount of toxic chemical compounds. Biosorption is better alternative technique to conventional treatment method for the biodegradation and removal of metal ions from aqueous system. Biological materials (Biosorbent) have been used as for the biosorption of metal ions from wastewater. In the present investigation, the capability of living biomass of *Rhizopusarrhizus*for the biosorption of lead metal was studied. Living biomass of *Rhizopusarrhizus* (100 mg and 200 mg) and contact time of 15 minutes indicates the most appropriate dose and time period for adsorption of lead metal ions. For the adsorption of lead metal solution, maximum adsorption capacity of 100 mg *Rhizopusarrhizus*biomass was observed as 94.70% at the 80ppm concentration. In case of 200 mg fungal biomass, 98.47 % metal removal was noticed at the 80ppm metal concentration.The maximum specific uptake (Q value) was 37.88 at 80ppm concentration in case of 100 mg biomass. In case of 200 mg biomass, the maximum Q value 19.694 was noticed at 80ppm concentrationlead. Present study showed that 100 mg and 200 mg living biomass of *Rhizopusarrhizus*acts as a effective biosorbent for the adsorption of lead metal ions.

Keywords: *Rhizopusarrhizus*, biosorption, contact time, heavy metals, lead metal, specific metal uptake (Q value)

1. INTRODUCTION

The degradation and removal of heavy metal ions and the recovery of valuable ions from wastewaters and soils are the most important environmental and economic problems. Heavy metals andother metal ions exist as contaminants in wastewater of many industries. Some metals associated with these activities are cadmium, chromium, copper, mercury and lead. Poisonous metals ions are released into the environment by means of coal combustion, sewage wastewaters, automobile exhaust, battery manufacturingindustries, mining activities, andthe utilization of fossil fuels. These metals are non-degradable, persistent and accumulate continuously. Lower concentrations of these heavy metals arebeneficial but in higher concentration they become highly toxicant and causes various health hazard to the living components.

Lead is a general metabolic poison, enzyme inhibitor and causes mental retardation andbrain damage in young children (Asgari, 2008). Leadhas the ability to replace calcium content of bone and hence its removal from the environment is highly essential.The Environment protection Agency indicated the permissible limit of lead in drinking water it is 0.05 mg/l and in wastewater is 0.1mg/l (El-Said, 2010). In comparison to the various water treatment methods, biosorption is generally preferred forthe removal of metalcontaminants due to its high capability, cost effectiveness, easy handling and availability of different biosorbents (Guoet al.2009).

Therefore, some types of biomass are often chosen to serve as biosorbents to remove heavy metals(Matheickal, and Q. Yu,1996; Chang and Hong,1994; Chu et al.1997; Abollino et al.2003).

Khan et al. (1997) reported that Bioremediation is the method by which inorganic and organic waste biologically degraded or transformed into less toxic forms, can serve as the alternative methods for removing the heavy metals from the contaminated soils or waters. Various methods such as bioventing, biosorption, bioaugmentation, bioreactor, land farming and biostimulation are included in bioremediation. Biosorption method describes the removal of metals ions by their passive binding to biosorbentsfrom an aqueous solution, is considered as an alternative to conventional methods. Although freely suspended biomass may have better contact with adsorbant during biosorption process.

Sari and Tuzen (2008) performed biosorption characteristics of Cd(II) ions using the red alga (*Ceramiumvirgatum*) and found that the red alga is an effective and alternative biomass for the removal of Cd ions from aqueous solution. Anayurt et al.(2009) performed equilibrium, thermodynamic,and kinetic studies on biosorption of Pb(II) and Cd(II) from aqueous solution using macrofungus (*Lactariusscrobiculatus*) biomass and concluded that the recovery of themetal ions from this biomass was found higher than

95%using 1M HCl and 1M HNO3. Biomass of microbes can be used for the decontamination of metal bearing wastewaters (Umrania, 2006).Keskinkan et al. (2003) reported the biosorption of copper, zinc and lead on submerged aquatic plant *Myriophyllumspicatum.*

Transportation of metal ions across the cell membrane depends on the metabolism of cell. This type of biosorption may take place only with viable cells. Cell walls of microbial biomass, mainly composed of polysaccharides, proteins and lipids have abundant metal binding groups such as carboxyl, sulphate, phosphate and amino groups (Kuyucak and Volesky, 1988). Basically biosorption by living organisms comprises of two steps: First,metabolism independent binding of metals to the cell walls and second, metabolism dependent intracellular uptake, whereby metal ions are transported across the cell membrane. (Costa, et.al., 1990, Gadd et.al., 1988, Huang et.al.,1990).Holan and Volesky (1995) reported the capability of *Rhizopus* sp. in removing heavy metals from different substrate. *Rhizopusarrhizus* has been used for the removal of metal ion from environment either by complex formation of metal ions with organic acids produced by the fungi or by the adsorption of metal ions to the component of fungal cell wall. Therefore, the present study is focused on the following two objectives:

(a) The ability of living biomass of *Rhizopusarrhizus*to adsorb lead metal solutions;
(b) The effect of different concentrations of lead (as lead nitrate) metal solutions on the biosorptive capacity of living biomass of *Rhizopusarrhizus.*

2.Materials and methods

Approximately, 1 kg of soils were collected (apparently free from pollution) from agriculture fields. The upper layer of soil was removed and then soil samples were taken into the laboratory in fresh sterile polythene bags. Dilution plate method was used for the isolation of soil fungi from soil samples.

2.1 Preparation of fungal biomass

Rhizopusarrhizusisolated from the soil samples was selected for the biosorption study of lead. Pure culture of Rhizopus*arrhizuswas* prepared on PDA (potato dextrose agar) medium.

2.2 Preparation of lead solution of different concentrations

Lead (as lead nitrate) was used to study the efficiency of fungal (*Rhizopusarrhizus)* biomass to adsorb metal. Stock solutions of metal were prepared in a manner so as to obtain following different concentrations of metal solution i.e., 10ppm, 20ppm, 40ppm, 80ppm.

3. Biosorption of lead using living biomass of*Rhizopusarrhizus*

The method followed by Bhole et al. (2004) was adopted with slight modification to determine the efficiency of living biomass of*Rhizopusarrhizus* for the biosorption of lead as lead nitrate.

50 ml of 10ppm solution of lead was taken into each of 6 flasks. Similarly, a set of 6 flasks were used for each of the 20ppm, 40ppm, and 80ppm concentrations of lead. Fungal biomass was then added into these flasks as under:

1. 10ppm lead nitrate solution (control i.e., without fungal biomass) (3 flasks)
2. 10ppm lead nitrate solution + 100mg fungal biomass (3 flasks)
3. 10ppm lead nitrate solution + 200mg fungal biomass (3 flasks)
4. 20ppm lead nitrate solution (control i.e., without fungal biomass) (3 flasks)
5. 20ppm lead nitrate solution + 100mg fungal biomass (3 flasks)
6. 20ppm lead nitrate solution + 200mg fungal biomass (3 flasks)
7. 40ppm lead nitrate solution (control i.e., without fungal biomass) (3 flasks)
8. 40ppm lead nitrate solution +100mg fungal biomass (3 flasks)
9. 40ppm lead nitrate solution + 200mg fungal biomass (3 flasks)
10. 80ppm lead nitrate solution (control i.e., without fungal biomass) (3 flasks)
11. 80ppm lead nitrate solution + 100mg fungal biomass (3 flasks)
12. 80ppm lead nitrate solution + 200mg fungal biomass (3 flasks)

Now all these flasks were placed on a rotatory shaker for 15 minutes. After a contact period of 15 minutes, the biomass of fungi was separated by filtering the mixture using Whatman filter paper No. 40 to prevent the probable interference of turbidity. The filtrate was then further processed to examine the concentration of lead remaining in the solution. Now the content of 3 flasks of each set were mixed together to get a composite solution for visible spectrophotometry. With the help of visible spectrophotometer, the concentration of remaining lead in the supernatant was examined at wavelength of 640-650nm.

Biosorbent + 100 ml of metal solution
↓
Shaking on rotatory shaker for 15 min.
↓
Filtered through Whatman filter paper No. 40
↓
Collect the supernatant
↓
O.D. at 640-650 nm

The amount of metal bound by the biosorbent was calculated as Follows (Hussein et al. 2004)

$$Q = V (Ci - Cf) / m$$

Where,
Q is the metal (Lead) uptake (mg lead per g biosorbent),
V is the liquid sample volume (ml),
Ci is the initial concentration of lead in solution (mg/l),
Cf is the final concentration of lead in solution (mg/l),
m is the amount of the added biosorbent on the dry basis (mg),

Similarly, biosorption efficiency (R %) of the particular biomass can be calculated as:

$$R = [(Ci - Cf)/ Ci] \times 100 \%$$

The biosorptive efficiency of a particular biomass of test fungi and its particular quantity was interpreted as under:

1.	0-10	Very poor
2.	10-20	Poor
3.	20-40	Moderate
4.	40-60	Good
5.	60-80	Very good
6.	80-100	Excellent

4. Result and discussion

The present investigation was conducted to examine the biosorptive efficiency of living biomass of *Rhizopusarrhizus*to adsorb lead from the solutions of different concentrations of lead(as lead nitrate).Different amount of living biomass of fungi as biosorbent i.e., 100 mg and 200 mg were allowed the lead from solutions of test metal solutions. After the contact period of 15 minutes, the observations weretaken. The results obtained are presented in the Table 1.This study indicates that the living biomass of *Rhizopusarrhizus*is quite effective material for the biosorption of lead metal.

A glance at the table 1 and fig. 1 reveals that:

As much as 94.70 % could be adsorbed in 100 mg biomass after the contact period of 15 minutes at the higher concentration of metal in solution i.e., 80 ppm. While at the lower concentrations, from 10 ppm to 40ppm the biosorption of lead by living biomass of *Rhizopusarrhizus* increased effectively from 25.180 % at 10ppm, 63.80 % at 20ppm and 88.947 % at 40ppm.

The increase in living biomass of *Rhizopusarrhizus*from 100 mg to 200 mg led to increase in the biosorption percentage up to 98.470 %. In case of 100 mg biomass, the maximum specific uptake (Q value) was noticed at 80ppm concentration i.e., 37.88, followed by 17.789 at 40ppm, 6.38 at 20ppm and 1.259 at 10ppm concentrations of metal (Table 1, fig.1).In case of 200 mg biomass of *Rhizopusarrhizus*, the pattern of increase in biosorption was also noticed as in case of 100 mg biomass of fungi. About 98.470 % metal removal was observed from the 80ppm metal concentration by 200 mg fungal biomass. From 10 ppm to 40ppm the biosorption of lead by living biomass of *Rhizopusarrhizus* increased effectively from 67.96 % at 10ppm, 68.99 % at 20ppm and 87.815 % at 40ppm.

In case of 200 mg biomass, Table 2, fig. 2, showed that the maximum Q value 19.694 was observed at 80ppm lead concentration, followed by 8.7815 at 40ppm, 3.449 at 20ppm and 1.699 at 10ppm concentrations of metal solution. The Q values indicated that increase in biomass caused a great decrease in the specific metal uptake per mg of biomass. Some worker noticed that the initial concentrations of test metal in the solution have a great effect on the sorption rate of metals by fungal biomass and it was also found that the adsorption of metal ions increase upto a greater extent with the increase in initial concentration of metal ions. These observations clearly showed that the biomass amount for the biosorption of all the concentrations of test metal at a particular incubation period can be adjusted.Chauhan et al. (2002) reported higher concentration of biomass in the solution reduces the distance between the biosorbent particles.Jaikumar and Ramamurthi (2009) reported that increasing dose of biosorbentdecreases the biosorption capacity.Prabha et al (2014) reported that percentage removal of metal ion increase as the adsorbent dose increase.

Table 1: Biosorption of lead (as lead nitrate) from aqueous solution of different concentrations by living biomass (100 mg) of *Rhizopusarrhizus.*

Amount of Biomass	Initial Conc. of lead in the solution (ppm)	Final Conc. of lead in the solution (ppm)	Amount of lead adsorbed (ppm)	Percentage of lead adsorbed	Q-value
100 mg	10	7.482	2.518	25.180%	1.259
	20	7.240	12.760	63.805%	6.380
	40	4.421	35.579	88.947%	17.789
	80	4.420	75.760	94.70%	37.880

Table 2: Biosorption of Lead (as lead nitrate) from aqueous solution of different concentrations by living biomass (200 mg) of*Rhizopusarrhizus*

Amount of Biomass	Initial Conc. of lead in the solution (ppm)	Final Conc. of lead in the solution (ppm)	Amount of lead adsorbed (ppm)	Percentage of lead adsorbed	Q-value
200 mg	10	3.204	6.796	67.960%	1.699
	20	6.202	13.798	68.99%	3.449
	40	4.874	35.126	87.815%	8.7815
	80	1.224	78.776	98.470%	19.694

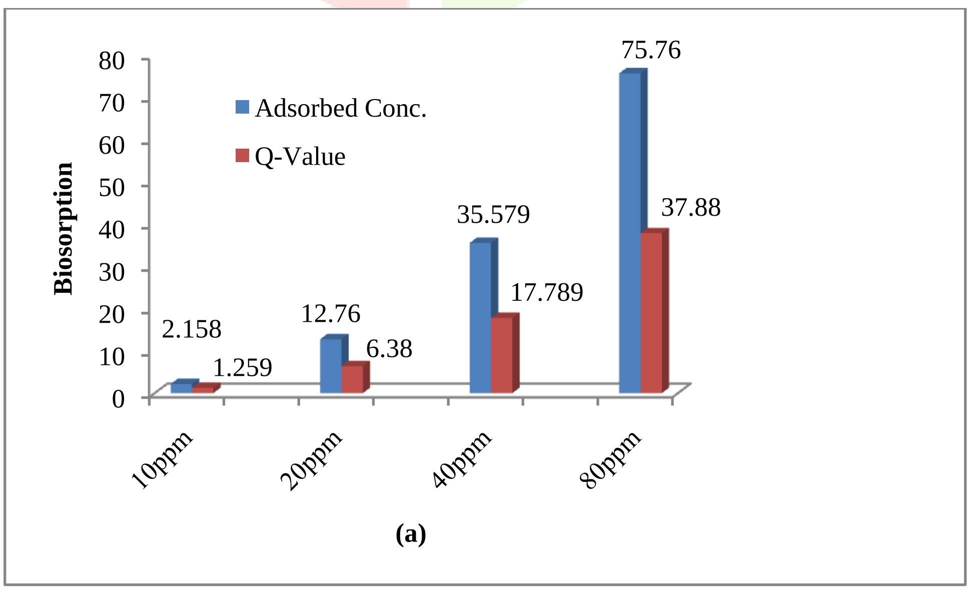

Figure 1: (a) Biosorption of lead

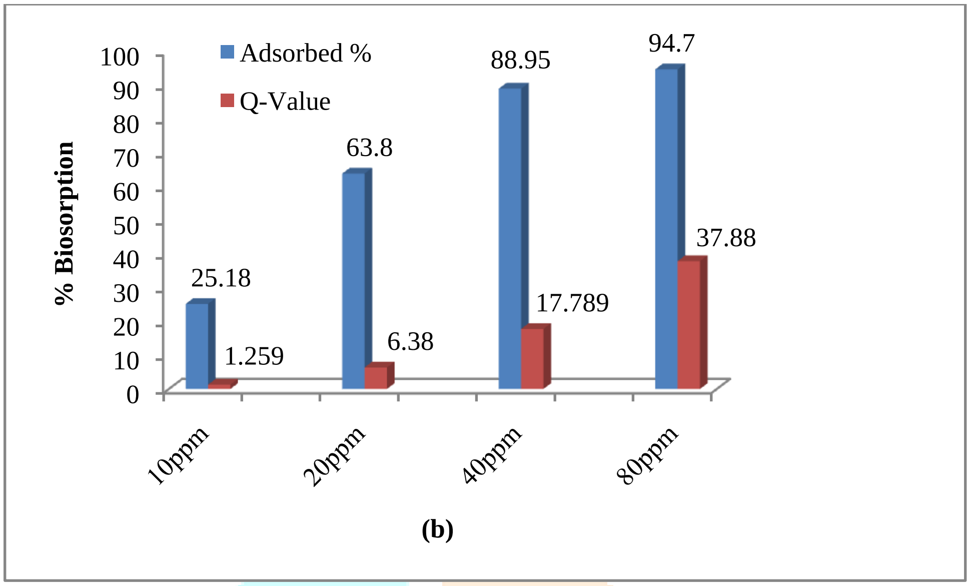

Figure 1: (b) % Biosorption profile of lead by living biomass (100 mg) of *Rhizopusarrhizus*

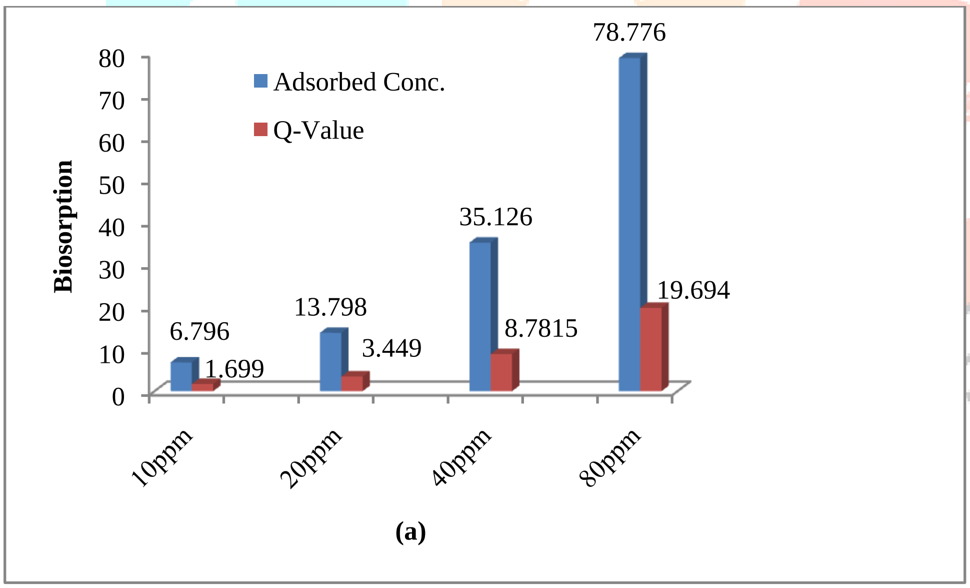

Figure 2: (a) Biosorption of lead

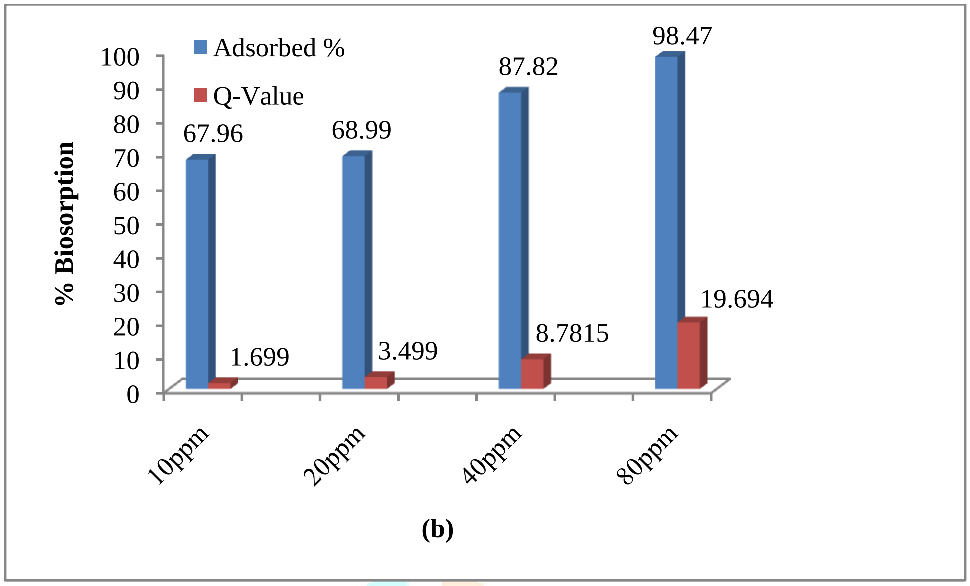

Figure 2: (b) % Biosorption profile of lead by living biomass (200 mg) of *Rhizopusarrhizus*

5. Conclusion

The present work showed that the living biomass of *Rhizopusarrhizus*is a good biosorbent for lead metal and it can be used for adsorption based metal ions treatment system. Present investigation indicated that the 200 mg (maximum dose of fungal biomass) was the most effective concentration for the leadbiosorptionthan the 100 mg fungal biomass. Present work clearly showed that biosorption efficiency increase as the amount of biomassincreases andbiosorption of lead metal ions by living biomass of *Rhizopusarrhizus*is a cost-effective and environmental friendly technology. The living biomass of*Rhizopusarrhizus* can be used as aneffective component for the managing the heavy metal pollution. Conventional treatment methods for the removal of metal contaminant from aqueous system are time consuming and are not eco-friendly. An alternative to the conventional method, biosorptionis quite effective alternative to the conventional methods highly efficient for the bio-detoxification of heavy metal pollutants from aqueous solution and for controlling the environmental pollutants.

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