



Trade Science Inc.

# Environmental Science

An Indian Journal

---

*Current Research Paper*

ESAIJ, 3(2), 2008 [194-196]

## *In-vitro* biosorption of lead and zinc by using living biomass of *aspergillus oryzae*

H.S.Ravikumar Patil, H.K.Makari\*, H.Gurumurthy

Department of Biotechnology, G.M.Institute of Technology, Davangere-577006 Karnataka, (INDIA)

Tel : +918192-233345; Fax: +918192-233344

E-mail : mac.gmit@gmail.com

Received: 25<sup>th</sup> September, 2007 ; Accepted: 30<sup>th</sup> September, 2007

### ABSTRACT

Bioremediation for elimination of heavy metals gaining much importance through microorganisms. This study was aimed to *in-vitro* elimination of lead and zinc by using of filamentous fungi *Aspergillus oryzae*. Biosorption of Lead and Zinc by *Aspergillus oryzae* was tested at 5 different initial concentrations ranging from 20ppm to 100ppm . The samples were analyzed for decrease in concentrations after 5 days incubation using Atomic Absorption Spectrophotometer (Chemito AA-203). *Aspergillus oryzae* showed the maximum percentage removal of Pb and Zn at 20ppm concentration and minimum adsorption at 100ppm. It was also observed that removal efficiency of *Aspergillus oryzae* decreases with increasing concentrations. © 2008 Trade Science Inc. - INDIA

### KEYWORDS

Biosorption;  
Lead;  
Zinc;  
Fungi;  
Heavy metals.

### INTRODUCTION

Heavy metals are among the most toxic contaminants present in the environment. Contamination of the aqueous environment by heavy metals is a worldwide environmental problem and as a result, their removal from waste water has attracted much attention from researchers in the past 20 years. Pollutant metals including Cu, Zn, Cd, Pb, Fe, Ni, Ag, Th, Ra and U released into the environment persist indefinitely, circulating and eventually accumulating throughout the food chain becoming serious threat to the environment and pose health problems.

Heavy metals traditionally removed by physical-chemical processes; Ion exchange, reverse osmosis, precipitation, solvent extraction, membrane technologies,

electrochemical treatments. These techniques have significant disadvantages including incomplete metal removal, the need of expensive monitoring equipments and some physical methods not suitable to remove heavy metal concentration in the order of 1-100mg<sup>[24]</sup>. The use of microorganisms to remove metals is an emerging technology and gaining attention among environmental research communities. Microorganisms do not degrade metals but may immobilize metal precipitation from polluted environment. Recent works have revealed the potential of using microorganisms for the reduction of metals. The interaction of microorganisms and many pollutant metals has not been fully understood.

Zinc is one of the most common elements in the Earth crust. Zinc is found in the air, soil and water and is present in all foods. Atomic Weight of zinc is 65.38.

## Current Research Paper

Zinc has a melting point of 419.58°C, boiling point of 907°C, with a valence of 2. In its pure elemental (or metallic) form, zinc is a bluish-white. Recommended Dietary Allowances for zinc is 11mg/day for men and 8 mg/day for women. If large doses of zinc are taken by mouth even for a short time stomach cramps, nausea, and vomiting may occur. Ingesting high levels of zinc for several months may cause anemia, damage the pancreas, and decrease the levels of high-density lipoprotein (HDL) cholesterol. Zinc may be taken up by animals from soil or drinking water.

Lead occurs naturally in the environment and it is one out of four metals that have the most damaging effects on human health. Atomic number of lead is 82. Its melting point and boiling point are 327°C and 1755°C respectively. Lead is a bluish-white lustrous metal. Lead fulfils no essential function in the human body. It can enter the human body through uptake of food, water and air. It can cause several unwanted effects, such as rise in blood pressure, kidney damage, disruption of nervous systems, brain damage and declined fertility of men through sperm damage.

Some biomasses of fungi types are very effective in accumulating heavy metals, such as *Aspergillus niger*, *Aspergillus terreus*, *Rhizopus oryzae*, *Penicillium chrysogenum*, *Metarrhizium anisopliae var. anisopliae* and *Penicillium verrucosum*. Yeast such as *Saccharomyces cerevisiae* and *Rhodotorula mucilaginosa*, Algae such as *Chlorella vulgaris* and bacteria such as *Bacillus subtilis* and *Pseudomonas aeruginosa*<sup>[13]</sup>. Fungi group has shown better accumulation of nickel and chromium by physico-chemical and biological mechanisms including extra cellular binding by metabolism-dependent accumulation<sup>[24]</sup>.

Filamentous fungi may be better suited for this purpose than other microbial groups, because of their high tolerance towards metals, cell wall binding capacity and intracellular metal uptake capabilities. With all these considerations our present research intended find out more information on biosorption models with reference to best combination of metals, biomass types and assessment of biosorption efficiency.

### MATERIALS AND METHODS

Stock solutions of lead and Zinc were prepared in different initial concentrations ranging from 20ppm, 40ppm, 60ppm, 80ppm and 100ppm and added to

sterilized each 50 ml of SD broth in separate conical flasks. Then inoculated with two loopful of *A.oryzae* spores. Experimental set up was incubated for five days at room temperature. Observation was made for colour changes in fungal mat.

The fungal mat was removed by filtration method by using A1 filter paper and collected the filtrate from each conical flask. The filtrate samples were subjected to atomic absorption analysis to determine the residual concentration of metal in the medium was analysed by Atomic Absorption Spectrophotometer (Chemito AA 203)

### RESULTS AND DISCUSSION

Studies on biosorption of Lead and Zinc from stock solutions by *A.oryzae* was incubated for a period of 5 days at five different initial concentrations of 20, 40, 60, 80 and 100ppm

**TABLE 1 : The absorption capacity of *A.oryzae* for different concentration of lead(Pb) at 5day incubation**

Initial conc. of Pb in medium (ppm)	Residual conc. of Pb in medium (ppm)			Amount of Pb absorbed by fungal mat (ppm)	% of Pb absorbed by fungal mat (ppm)
	Test 1	Test 2	Avg		
20	1.855	1.802	1.828	18.172	90
40	7.699	7.100	7.399	32.601	81
60	28.756	31.106	29.931	30.069	50
80	50.703	58.297	54.5	25.5	26
100	73.936	80.558	77.247	22.753	22

**TABLE 2 : The absorption capacity of *A.oryzae* for different concentration of zinc (Zn) at 5day incubation**

Initial conc. of in Zn medium (ppm)	Residual conc. of Zn in medium (ppm)			Amount of Zn absorbed by fungal mat (ppm)	% of Zn absorbed by fungal mat (ppm)
	Test 1	Test 2	Avg		
20	0.889	0.823	0.856	19.114	95
40	2.244	2.35	2.297	37.703	94
60	8.56	10.55	9.555	50.445	84
80	13.24	19.402	16.326	63.674	79
100	45.271	54.225	49.748	50.252	50

### Colour morphology

**TABLE 3 : The color morphology of *A.oryzae* for different concentration of lead at 5day incubation**

Initial conc. of Pb in the medium ( ppm)	Color of the mycelial biomass of <i>A.oryzae</i>
Control	Green
20	Parrot green
40	Parrot green
60	Creamish white
80	Creamish white
100	White

## Current Research Paper

**TABLE 4 : The color morphology of *A.oryzae* for different concentration of zinc at 5day incubation**

Initial conc. of Zn in the medium (ppm)	Color of the mycelial biomass of <i>A.oryzae</i>
Control	Green
20	Green
40	Green
60	Green
80	Slight greenish
100	Pale green

### CONCLUSION

This study leads to the conclusion that *A.oryzae* have the capacity to accumulate Lead and Zinc. It showed higher sorption for Zinc than Lead. This high Zinc absorption capacity made them well suited for removal of heavy metal from contaminated water, bioleaching, bioremediation of polluted sites and effluent treatment.

Biosorption is highly economical and ecofriendly as this generate no further waste into the environment. However there are still much uncertainty associated with the development of treating waste water by living fungi and more future work is necessary.

### REFERENCES

- [1] Ahmet, Semra, Figen; Turk.Journal of Biology., **26**, 23-28 (2005).
- [2] Z.Asku, T.Kutsal, S.Gun; Environmental Technology, **12**, 915-921 (1991).
- [3] Z.Aksu; 'The Biosorption of Copper(II)', C.vulgaris, Z.Ramigera(Eds.); Environmental Technology., **13**, 579-58 (1992).
- [4] D.Brady, J.R.Ducan; Applied Microbial Biotechnology, **34**, 149-154 (1994).
- [5] B.Bina, M.Kermani, H.Mohavahedian, Z.Khazaei; Pakistan Journal of Biological Sciences, **9(8)**, 1525-1530 (2006).
- [6] C.L.Brierley; Geomicrobiology Journal, **8**, 201-223; G.M.Gadd; 'Fungi and Yeasts for Metal Accumulation', In L.Ehrlich, Brierly (Eds); 'Microbial Mineral Recovery', McGrawHill, New York, 249-276 (1989).
- [7] C.Bruno, L.Pablo, H.Robert, E.S.Manuel; Environmental Chemistry., **1**, 180-187 (2004).
- [8] M.J.Brown, J.N.Lester; Water Research, **16**, 1539-1548 (1982).
- [9] T.J.Butter, L.M.Evison, Hancooh, K.A.Matis, A.Zouboulis; Water Resource., **32(2)**, 400-406 (1998).
- [10] M.A.Dias, I.C.A.Lacerda, P.F.Pimentel Castro, C.A.Rosa; Letters in Applied Microbiology, **34**, 46-50 (2002).
- [11] E.Fourest, C.J.Roux; Applied Microbiology and Biotechnology, **37(3)**, 399-403 (1992).
- [12] G.M.Gadd, C.White; Trends in Biotechnology, **11**, 353-359 (1999).
- [13] N.C.M.Goomes, L.C.S.Mendonca-Hagler, I.Savvadis; Brazilian Journal of Microbiology, **29**, 85-92 (1998).
- [14] H.Hussein, S.F.Ibrahim, K.Kandeel; Electronic Journal of Biotechnology, **56**, 197-103 (2005).
- [15] A.Hammami; Minerals Engineering. **16**, 723-729 (2003).
- [16] R.Jalali, H.Ghafourian, Y.Asef, S.J.Davarpanah, S.Sepehr; Journal of Hazardous Materials, **92(3)**, 253-262 (2002).
- [17] A.Kapoor, T.Viraraghavan; A review.Bioresource Technology, **53**, 195-206 (1995).
- [18] S.Karthikeyan, R.Balasubramanian; Bioresource Technology, **98**, 452-455 (2007).
- [19] L.Norton; Advances in Environmental Research, **56**, 156-162 (2003).
- [20] F.Pagnanelli, L.Toro, F.Veglio; Waste Management, **22**, 901-907 (2002).
- [21] E.S.Shumate, W.G.Strandberg; Comprehensive Biotechnology, **13**, 235-247 (1985).
- [22] M.Tsezos, A.A.Deutschmann; Journal of Chemical Technology and Biotechnology, **53**, 1-12 (1992).
- [23] K.Vijay Raghavan, J.R.Jegan, Palanivelu, Velan; Electronic Journal of Biotechnology, **22**, 61-71 (2004).
- [24] B.Volesky, Holan; Biotechnology and Bioengineering Symposium, **16**, 121-126 (1995).
- [25] B.Volesky; 'Removal and Recovery of Heavy Metals by Biosorption In: Biosorption of Heavy Metals', Boston, USA, CRC Press, 7-43 (1990).
- [26] G.Yekta, S.Uren, U.Guvenc; Turk Journal of Biology., **27**, 23-29 (2003).
- [27] J.Yang, B.Volesky; Water Research, **33(15)**, 3357-3363 (1999).
- [28] Yi-Tin, Wang, M.Evans, Chirwa, H.Shen; Journal of Environmental Engineering, **21**, 300-312 (2000).