Jr. of Industrial Pollution Control 24 (2)(2008) pp 133-137

REMOVAL OF CHROMIUM (VI) FROM AQUEOUS SOLUTION BY CHEMICALLY MODIFIED BIOMASS OF *ASPERGILLUS NIGER*

SNEHA NARVEKAR AND VARSHA K.VAIDYA

Department of Microbiology, Institute of Science, 15, Madame Cama Road, Mumbai 400 032, India

Key words : *Aspergillus niger*, Biosorption, Hexavalent Chromium, Functional groups.

ABSTRACT

Sporulated biomass of *Aspergillus niger* was evaluated for its ability to remove Chromium (VI) by biosorption both prior to and after subjecting it to various physical treatments such as autoclaving and chemical treatments with acids, alkali, chelating agent and cross-linker. Pre-treatments with acetic acid (5%), phosphoric acid (10%), gluteraldehyde (2%), oxalic acid (0.001M and 0.01M) and hydrochloric acid (1M, 5M) after autoclaving enhanced the adsorption capacity of *Aspergillus niger* biomass, which ranged from 2.16% to 86.88% in comparison with the untreated biomass. Comparison between the sporulated and mycelial biomass indicated effectiveness of the mycelial biomass in sorption of Chromium (VI). The mycelial biomass treated with 5M HCl combined with autoclaving proved to be the most successful in the removal of Chromium (VI).

INTRODUCTION

Chromium (VI) contamination is not uncommon, especially near industries involved in leather tanning, chrome painting, metal cleaning and processing, wood preservation and alloy preparation. Tannery effluent is a major source of aquatic pollution in India. Of the total Chromium used in the processing of leather, 40% is retained in the sludge; disposal of which onto land and water bodies has led to increased Chromium levels reaching as high as 30,000 mg / kg. In Ludhiana and Chennai, Chromium levels in underground water have been recorded at more than 12 mg / L and 550–1500 mg / L, respectively (Chandra and Kulshreshtha, 2004). Another study carried out by Central Pollution Control Board on the groundwater quality in Kanpur revealed Chromium (VI) levels at 6.2 mg / L. Detailed assessment of the tannery waste contaminated sites in Tamil Nadu, India and Mount Barker near Adelaide, Australia revealed extensive contamination of soil and surface or ground water that ranged between 100 to 70,000 mg / kg of Chromium (VI) in surface and subsurface soils (Kamaludeen *et al*. 2003).

Hexavalent Chromium is one of the toxic elements whose concentration in natural environment can profoundly disrupt biological processes. The mutagenic and carcinogenic properties of Chromium (VI) necessitate the use of effective remedial methods. In addition to higher costs involved, difficulties associated with chemical and physical techniques in remediation of a Chromium (VI) contaminated site to

Corresponding author : Dr. K. Vaidya, 2, Dnyandevi, Sahitya Sahawas, Bandra (East), Mumbai 400 051.
Email - varshavaidya@gmail.com

EPA recommended levels (50 ppm), assert the need for economical and eco-friendly bioremedial measures.

Biosorption is a passive, non-metabolically-mediated process of metal binding by the biosorbent. It offers several advantages including cost effectiveness, high efficiency, minimization of chemical / biological sludge and regeneration of biosorbent with the possibility of metal recovery. Polysaccharides and proteins of the fungal cell walls contain many functional groups such as carboxyl, hydroxyl, phosphates and amino groups that can bind with metal ions (Awofolu *et al*. 2006). Modifications of the cell wall and biomass of microorganisms have been explored extensively in attempts to increase the sorption efficiency of metal ions by the biomass using several modifying agents (Awofolu *et al*. 2006; Loukidou *et al*. 2003; Bai and Abraham, 2002; Galun *et al*. 1986 and Park *et al*. 2005).

This study was undertaken to investigate the effect of physical and chemical pretreatments on biosorption of Chromium (VI) by *Aspergillus niger* biomass.

MATERIALS AND METHODS

Media and chemicals

The media chemicals were of analytical grade, obtained from Hi Media Laboratories, India. Other chemicals were obtained from Merck, India, Qualigens Fine Chemicals, India and Sigma Aldrich, USA.

Preparation of hexavalent Chromium solution

A stock solution (1000 mg / L) of Chromium (VI) was prepared by dissolving 2.828 g of K2Cr2O7 (AR grade) in 100 ml distilled water. The stock solutions were then appropriately diluted to get the test solutions of desired strength.

Preparation of the adsorbent

Pure strain of Aspergillus niger was obtained from (NRRL 326) National Chemical Laboratory, Pune. It was cultured in Czapeck Dox broth containing 0.25% Tween 80 (to prevent sporulation), while sporulated biomass was produced in the medium without Tween 80 under static conditions at 28±2oC. The biomass harvested after 7 days was washed thoroughly with distilled water and dried at 80oC in an oven for 24 hours. Dried biomass was ground in a mortar and pestle, and sieved through a 150 mesh sieve. The biomass processed in this manner was used as a biosorbent in metal uptake experiments to evaluate the biosorption capacity.

Analysis of Chromium (VI) ions

The concentration of the Chromium (VI) ions was determined spectrophotometrically after complexation of the metal ion with 1, 5-diphenylcarbazide. The reaction determines only hexavalent Chromium and is very sensitive with the molar absorptivity based on Chromium being about 40,000 L / g / cm at 540 nm (American Public Health Association, 1986). Treatment methods used for enhancing the biosorption by the biomass

Different physical and chemical treatments were given to the biomass to examine their effect on the sorption capacity of the fungus.

1. Treatment with Oxalic acid, Malic acid and Ethylenediamine tetraacetic acid (EDTA) (Awofolu, *et al*. 2005) : 1 g of powdered biomass each, separately placed in clean 10 mL beakers was slightly moistened with distilled water. 7 mL of 0.05M EDTA, 0.1M, 0.01M and 0.001M oxalic acid and 0.1M Malic acid were separately added to each beaker. The slurry was dried overnight in an oven at 50ºC.
2. Treatment with H_2SO_4, HCl, HNO_3 and NaOH (Bai and Abraham. 2002): 5 grams of biomass was mixed with 500 mL of 0.5M, 1M and 5M HCl, 1M H_2SO_4, 1M HNO_3 and 1M NaOH with agitation (120 rpm) for 24 h.
3. Treatment with acetic acid and phosphoric acid (Cabuk *et al*. 2005): 30 grams of biomass was boiled separately for 15 min with 200 mL of 10% (v / v) acetic acid and 10% (v / v) phosphoric acid.
4. Treatment with gluteraldehyde (2%) (Cabuk *et al*. 2005): 30 grams of biomass was boiled for 15 min in 500 mL of 2% (v / v) gluteraldehyde.
5. Washing, drying and autoclaving the biomass. The modified dried biomass obtained after each treatment was then washed with distilled water to remove unreacted reactant until the pH of the filtrate was near neutral. The modified biomass was again dried at 80oC for 6 h. The biomass obtained after all the above treatments was further autoclaved (Cabuk *et al*. 2005) at 15 lbs for 15 min. It was then filtered and dried at 80oC for 6 h.

The effect of each pretreatment was studied by carrying out biosorption studies in conical flasks at pH 2.0 using 50 mg / L of biosorbent in 50 mg / L of Chromium (VI) solution. The flasks contents were agitated on a rotary shaker at 120 rpm for 1 h after which the solution was centrifuged at 3,000 rpm for

10 min and the supernatant solution was analyzed for Chromium (VI) concentration.

Effect of the type of the biomass (sporulated and mycelial) of Aspergillus niger on the Chromium (VI) biosorption

The potential of the untreated and treated mycelial biomass of Aspergillus niger (0.05g / L) in the removal of Chromium (VI) from the solution (50 mg / L) was studied at an optimum pH of 2.0. The treatments that increased the sorption capacity of the raw sporulated biomass were given to the mycelial biomass.

RESULTS AND DISCUSSION

The ability of fungal biomass in sequestering the metallic species has mainly been traced to the cell wall though it is not necessarily the only site where the sequestered metals are located. Biosorption of heavy metals in fungal biomass occurs as a result of ionic interactions and complex formation between metal ions and functional groups present on the fungal cell wall. The importance of any given group for biosorption of a certain metal by a certain biomass depends on number of factors. Besides the number of sites available in the biosorbent, it also depends on the accessibility and the chemical state of the site (i.e. availability) and on the affinity between the site and the metal (i.e. binding strength). Numerous chemical groups have been suggested to contribute to biosorption by facilitating the metal ion to bind on them.

Pretreatment methods used for enhancing the biosorption by the dead sporulated biomass of *A. niger*.

In order to enhance the sorption capacity of the biomass, pretreatment methods aiming at modification of the functional groups were used. These included chemical pretreatments such as the use of acids, alkali, chelating agent and cross linker.

Treatment with acids

According to Tunali *et al*. (2005), the effect of acid pretreatments on the biosorption ability of organisms varies from high to none depending on the type of microorganisms used and the type of heavy metal ions studied. Generally acid treatment enhances Chromium adsorption than the untreated biomass control systems. This could be attributed to the fact that acid hydrolysis yields relatively pure amino sugar, D-glucosamine, which is more easily protonated at adsorption pH. As the pH is lowered, the overall surface charge on the biomass becomes positive or less negative, which promotes the approach of negatively charged metal ions like CrO42-. The amino and carboxyl groups and the nitrogen and oxygen of the peptide bonds can be available for characteristic coordination bonding with metallic ions. At low pH values (below pH 2.0) the protonation of the functional groups (e.g. carboxyl and amino groups) has been known to give an overall positive charge to the biomass, which is able to adsorb negatively charged (anionic) metal ions (Park *et al.* 2005). Thus, acid treatment might expose more binding sites; and therefore, the accessibility of the anionic Chromium (VI) to the sorption sites might be increased.

In the present study, the sporulated biomass of Aspergillus niger was treated with different concentrations of HC_l, H_2SO_4, HNO_3, oxalic acid, malic acid and phosphoric acid. Amongst all the acids tested only 0.001M oxalic acid and 5M HCl increased the sorption capacity of the biomass by 18.20% and 0.02% respectively relative to the untreated biomass as shown in Table 1. 5% acetic acid showed a negligible decrease in the biosorption capacity of the biomass in comparison with the untreated biomass. The remaining acids showed a drastic reduction in the sorption efficiency ranging from 11.34% (0.5M HCl) to 93.98% (15% acetic acid) relative to the untreated biomass.

Conflicting reports of the effect of the acid on the biosorption have been reported in the literature. Kapoor and Viraraghavan (1998) reported that acid pretreatment decreased the biosorption capacity of *Aspergillus niger*. Park *et al.* (2005) have reported that pretreatments with HNO_3 and H^2SO^4 were superior to HCl treatment in the removal of Chromium (VI). However, in the present investigation contradictory results were obtained showing that HNO_3 and H_2SO_4 pretreatments did not increase the sorption while 5M HCl treatment showed the highest sorption.

Awofolu *et al*. (2006) have shown that malic acid and oxalic acid pretreatments showed better sorption of lead compared to the untreated biomass. However, in the present study, malic acid treatment did not increase the Chromium (VI) sorption capacity while the decreasing concentrations of oxalic acid and acetic acid increased the sorption capacity of the sporulated biomass of *Aspergillus niger*. This can be attributed to simpler structures of oxalic / acetic acids which tend toward some form of linear molecular orientation that could facilitate easy bonding with binding sites (especially amino groups) on the biom-

ass thus increasing the number of sites for metal bonding (Awofolu, *et al*. 2005). Reuse of the mycelial mat for dye decolorization can be cost effective, time saving and advantageous for effluent treatment. Xinjiao *et al.* (2003) showed 91.1%, 87.8% and 86.4% efficiencies of dye decolorization for the first, second and third cycle respectively. In the present work higher decolorization efficiencies of 99.00± 0.75%, 95 ± 1.20%, 87± 0.90 % and 71± 1.45% for first to fourth cycles were observed respectively. The results of the present investigation show extensive potentiality of white rot fungi in treatment of dye containing effluents.

Pretreatments with chelating agent and cross linker

The biomass was treated with chelating agent such as EDTA and cross linker such as 2% solution of gluteraldehye, in order to observe the effect of pretreatments on the sorption capacity of the biomass. Both the pretreatments viz. gluteraldehyde and EDTA caused a drastic decrease of 77.84% and 93.73% respectively in the biosorption capacity of the biomass as compared to the untreated biomass, as shown in Table 1.

Gluteraldehyde and EDTA pretreatments have been cited in the literature as effective pretreatments for increase in biosorption of lead ions (Cabuk *et al*. 2004; Awofolu, *et al*. 2005). However, the decrease in the sorption of Chromium ions observed in the present study, could be attributed to bulkiness of the EDTA molecule and to the orientation of the hydroxyl and the amine groups within the EDTA structure during the modification (Awofolu, *et al*. 2005).

Pretreatment with alkali

Treatment with 1M NaOH decreased the efficiency of removal of Chromium (VI) by the biomass of Aspergillus niger by about 77.84% compared to the untreated biomass of Aspergillus niger, as shown in Table 1. Alkali pretreatments cause hydrolysis of protein constituents and also deacetylation of chitin. However, it also brings about drastic effects like swelling of the biomass, probably due to the polymer chain breakage thereby hindering the operational stability. Tunali *et al*. (2005) and El-Sayed *et al*. (2004) have reported favorable effects of alkali pretreatments for cation biosorption. In the present investigation, Chromium (VI) in aqueous solution, being in a negatively charged oxo anion complex form, was bound to the chitin rich surface by a different mechanism and therefore exhibited a negative trend (Bai & Abraham, 2002).

Effect of autoclaving on the chemically treated biomass

Cabuk *et al*. (2004) reported that autoclaving the biomass leads to exposure of some more active metal binding sites embedded in the cell wall of the biomass. Hence, in the present investigation Chromium (VI) removal efficiency of the chemically pretreated biomass was compared with their autoclaved counterparts as shown in Fig 1.

Except for a negligible increase seen in case of untreated and HCl (0.5M) treated biomass, autoclaving enhanced the effect of all the chemical treatments as shown in Fig 1. Thus, the coupling of the physical and chemical treatments proved to be highly beneficial showing a sharp increase in the biosorption efficiency ranging from 39.50% to 86.88% in comparison with the untreated biomass as shown in Table 1. The most pronounced effect was seen in case of the biomass treated with 5M HCl. It was observed that with increasing concentrations of HCl from 0.5M to 5M, the Chromium (VI) sorption capacity of the biomass increased from 8.55 to 17.88%, which could be attributed to increased protonation of the biomass.

Autoclaving increased the sorption capacity of both the EDTA and glutaraldehyde pretreated biomass, though a striking effect was observed with gluteraldehyde pretreated biomass. A huge rise of 435.45% in the biosorption capacity than the treated unautoclaved biomass was recorded. Despite this stupendous rise, the sorption capacity was only 15.69% more in comparison with the untreated biomass. Autoclaving of the 1M NaOH treated biomass also showed a similar trend with 147.64% increase in the sorption efficiency than the unautoclaved treated biomass. However, even with this enhanced capacity, the autoclaved NaOH treated biomass proved to be an inferior biosorbent as compared to the untreated biomass.

Loss in biomass after pretreatments

As seen in Table 2, different pretreatments caused a loss in the biomass. Park *et al*. (2005) have also reported loss in biomass after various chemical pretreatments. These pretreatments result in the cleaning of the cell wall thereby replacing the natural mix of ionic species bound on the cell wall with various functional groups. Thus, the weight of the biomass was decreased through the pretreatments as shown in table 2. The loss in the biomass ranged from 2.5% to 66.1%.

Selection of the pretreatment

Economic perspective plays an important role in the selection of the biosorbent for industrial applications. A comprehensive analysis of the cost of the chemicals, percentage loss incurred in the biomass after the pretreatments and the percentage increase in the biosorption capacity achieved, led to the selection of the pretreatments for further work.

In the present study, it was observed that the enhancement of biosorption capacity achieved was offset to some extent by considerable loss in the biomass. This criteria was used for eliminating pretreatments by H_2SO_4 (1M), Malic acid (0.1M), Acetic acid (10% and 15%), HCl (0.5M), Oxalic acid (0.1M), phosphoric acid (5% and 15%), HNO_3 (1M) and EDTA (0.05M). These pretreatments not only decreased the biosorption but also led to 24.9% to 66.1% loss of biomass as shown in Table 2.

Pretreatment using acetic acid (5%), Phosphoric acid (10%), Gluteraldehyde (2%), Oxalic acid (0.001M and 0.01M), HCl (1M and 5M) after autoclaving enhanced the adsorption capacity of *Aspergillus niger* biomass which ranged from 2.16% to 86.88% in comparison with the untreated biomass leading to loss in biomass to a varying degree.

The negligible increase in biosorption when compared with the loss incurred due to phosphoric acid (10%) suggested that the negligible gain in biosorption did not warrant the use of chemical pretreatment. 2% gluteraldehyde though caused a meager 2.5% loss in the biomass after pretreatment, its cost was a hindrance in it choice as a pretreatment chemical. Hence, only acetic acid (5%), oxalic acid (0.001M and 0.01M), HCl (1M and 5M) pretreatments were chosen for further work using mycelia, on the basis of the gains in sorption efficiency and acceptably less loss in biomass after the pretreatments.

Sorption of Chromium (VI) ions by the mycelial biomass

Biosorption of untreated sporulated biomass was compared with the sorption by mycelial biomass. It was observed that the mycelial culture was more effective in sorption than the sporulated one. The mycelial biomass was 11.28% more effective than the sporulated biomass. This increase can be attributed to the exposed cell surface of the mycelial biomass.

The mycelium consists of multinucleated mass of cytoplasm enclosed within a rigid, much branched system of tubes which are fairly uniform in diameter. Cell walls of fungi present a multi-laminate architec-

Table 1. Effect of various chemical pretreatments on the dead sporulated biomass of *A. niger*

Treatments	% Increase in sorption before autoclaving	% Increase in sorption after autoclaving
Untreated	0	1.589
Acids		
Acetic acid 5%	-3.7636	44.565
Acetic acid 10%	-24.815	-0.754
Acetic acid 15%	-93.985	-60.072
H_2SO_4 1M	-87.473	-72.257
HCl 0.5M	-11.343	-10.653
HCl 1M	-13.649	54.915
HCl 5M	0.0229	86.889
HNO_3 1M	-90.605	-87.670
Malic acid 0.1M	-81.210	-41.620
Oxalic acid 0.1M	-38.457	-15.349
Oxalic acid 0.01M	-18.163	39.509
Oxalic acid 0.001M	18.208	42.141
Phosphoric acid 5%	-93.985	-84.855
Phosphoric acid 10%	-76.143	2.165
Phosphoric acid 15%	-63.911	-47.681
Alkali		
NaOH 1M	-77.847	-45.999
Chelating agent and cross-linker		
EDTA 0.05M	-93.736	-87.670
Gluteraldehyde 2%	-77.847	16.760

Table 2. Comparative analysis of loss in biomass after treatments and % sorption of Chromium (VI) by the dead sporulated biomass of *A. niger*

Treatment	% Loss in biomass%	Change in biosorption
Acetic acid 5%	25.7	44.565
Acetic acid 10%	32.4	-0.754
Acetic acid 15%	36.8	-60.072
HCl 0.5M	20.1	-10.652
HCl 1M	20.4	54.915
HCl 5M	25.3	86.889
Oxalic acid 0.1M	48.2	-15.349
Oxalic acid 0.01M	34.9	39.508
Oxalic acid 0.001M	50.1	42.141
Phosphoric acid 5%	26.4	-84.855
Phosphoric acid 10%	25.5	2.164
phosphoric acid 15%	26.8	-47.681
$H SO_4$ 1M	62.4	-72.256
HNO_3 1M	24.9	-87.669
Malic acid 0.1M	66.1	-41.620
NaOH 1M	30.4	-45.998
EDTA 0.05M	39.3	-87.669
Gluteraldehyde 2%	2.5	16.759

Effect of various treatment of sorption of chromium (VI)

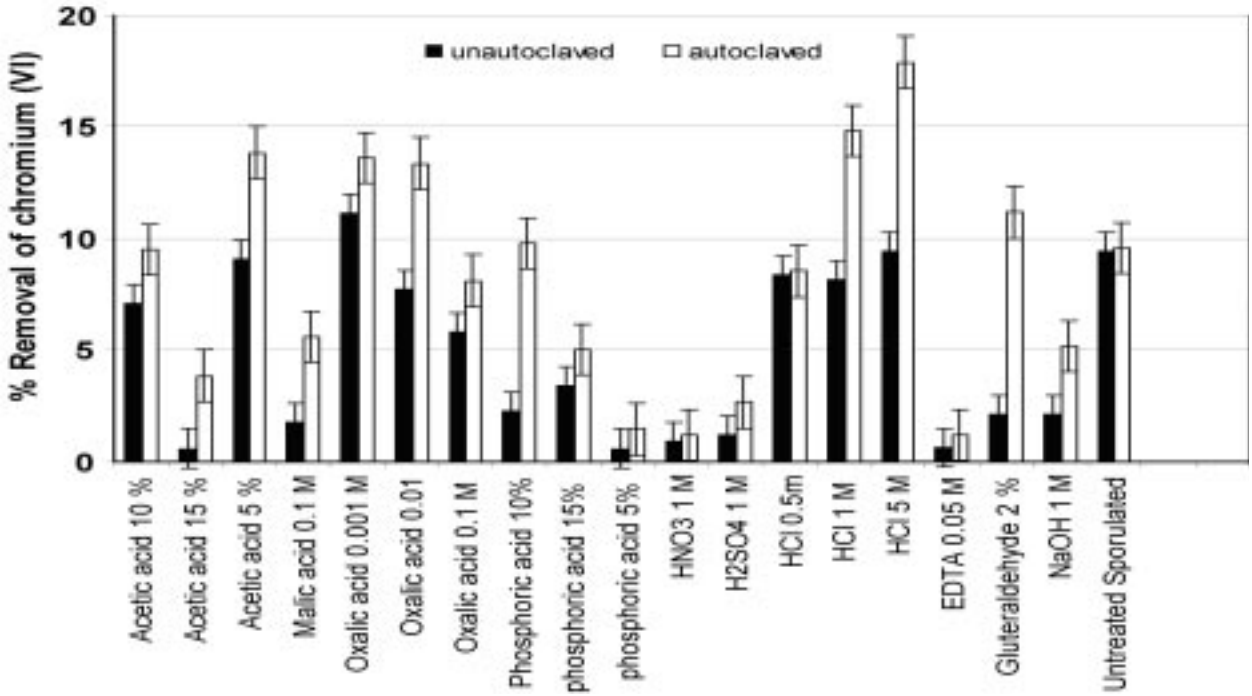

Fig. 1 Effect of autoclaving on chemically treated dead sporulated biomass of *A.niger* in the sorption of Chromium (VI) ions

Effect of physico-chemical treatment on the mycelial culture

Fig. 2 Effect of the pretreatments on the Chromium (VI) sorption by mycelial biomass of *A. niger*

ture where up to 90% of their dry mass consists of amino or non-amino polysaccharides. The fungal cell walls can be considered as a two phase system consisting of chitin framework embedded on an amorphous polysaccharide matrix. The cell walls are rich in polysaccharides and glycoproteins such as glycans (ß-1-6 and ß -1-3 linked D-glucose residues), chitin (ß -1-4 linked N-acetyl-D-glucosamine), chitosan (ß -1-4 linked D-glucosamine), mannans (ß -1-4 linked mannose) and phosphormannans (phosphorylated mannans). Various metal binding groups, viz amine, imidazole, phosphate, sulphate, sulfhydryl and hydroxyl are present in the polymers (Alluri *et al*. 2007). Pigments, polyphosphates and inorganic

ions are also found in the fungal cell wall. The cell wall of *Aspergillus niger*, consists of neutral carbohydrate (73 to 83%) and hexomine (9 to 13%), with smaller amounts of lipid (2 to 7%) and phosphorus (less than 0.1% of wall weight). The acetyl content is 3 to 3.4%, which corresponded to 1 mol / mol of hexosamine. This suggests the involvement of other components besides chitin in the metal sequestering (Sag, 2001).

Effect of the pretreatments on the Chromium (VI) sorption by the mycelial biomass

The mycelial biomass was subjected to those pretreatments that increased the biosorption capacity of the sporulated biomass. It was observed that all the pretreatments enhanced the sorption to a greater extent in mycelial biomass. Although a noticeable increase of 31.59% increase in the sorption than the sporulated biomass was seen in case of treatment with 5% acetic acid, the highest biosorption capacity was observed in case of biomass treated with 5M HCl as seen in Fig 2. 5M HCl treatment of the mycelial biomass showed maximum biosorption (20.17% removal) of Chromium (VI) ions. This increase in the sorption, after the treatment of mycelial biomass may be due to further enhancement of the increased metal sorption sites presented by the mycelial biomass.

Thus, the treatment with 5M HCl, combined with autoclaving proved to be the ideal treatment, striking a balance between cost effectiveness and increased biosorption efficiency, compensating for the loss in the biomass caused due to the pretreatment.

CONCLUSION

The ability of the sporulated and mycelial biomass of A. niger to sorb Chromium (VI) ions from the aqueous solution was demonstrated by batch experiments at pH 2.0. The results indicated the effectiveness of the physico-chemical treatments (5M HCl followed by autoclaving) in enhancing the efficiency of Chromium (VI) removal. The modification protocol was simple, fast and efficient in sorption of Chromium (VI) ions. The modified biomass of Aspergillus niger demonstrated its potential for industrial applications. More information is required to determine the best combinations of metals, biomass type and other conditions.

ACKNOWLEDGEMENT

The investigators gratefully acknowledge the help received from various Scientists from National Institute of Oceanography, Goa during the course of these investigations. We thank Dr. P.V. Narvekar, Chemical Oceanography, for helping in many ways and especially Dr. N.B. Bhosle, Marine Corrosion and Materials Research, for providing us with the most of the facilities required to carry out our study. Special thanks are due to Dr. Shyamprasad & Mr. Khedekar, V.D., Geological Oceanography, for providing the SEM facilities and to Dr. Solimabi Wahidullah and Dr. P.S. Parameswaran, Chemical Oceanography, for providing the FTIR and other instrumental facilities.

REFERENCES

Alluri, H. K., Srinivasa Reddy S. R., Settalluri, V.S., Singh, J., Suryanarayana, V. and Venkateshwar P. 2007. Biosorption: An eco-friendly alternative for heavy metal removal. *Afr. J. Biotechnol.* 6 (25) : 2924-2931.

APHA 1986. In: *Standard Methods for Examination of Water and Wastewater*, 16th ed. American Public Health Association, Washington, DC.

Awofolu, O.R., Okonkwo, J.O., Merwe, R.R.D., Badenhorst, J. and Jordaan, E. 2006. A new approach to chemical modification protocols of *Aspergillus niger* and sorption of lead ion by fungal species. *Electronic J. Biotechnol.* 9 (4) : 340-348.

Bai, R.S. and Abraham, T.E. 2002. Studies on enhancement of Cr (VI) biosorption by chemically modified biomass of *Rhizopus nigricans. Water Res.* 36 : 1224-1236.

Cabuk, A., Ülhan, S., Fülük, C. and Caliskan, F. 2005. $Pb2^{+}$ biosorption by pretreated fungal biomass. *Turkish J. Biol.* 29 : 23-28.

Chandra, P. and Kulshreshtha, K. 2004. Chromium accumulation and toxicity in aquatic vascular plants. *The Botanical Rev.* 70 (3) : 313-315.

El-Sayed, M. and El-Morsy, 2004. *Cunninghamella echinulata* a new biosorbent of metal ions from polluted water in Egypt. *Mycologia.* 96 (6) : 1183-1189.

Galun, M., Galun, E., Siegel, B. Z., Keller, P., Lehr, H. and Siegel, S.M. 1987. Removal of metal ions from aqueous solutions by *Penicillium* biomass: Kinetic and uptake parameters. *Wat. Air Soil Poll.* 33: 359-371.

Kamaludeen, S.P.B., Megharaj, M., Juhasz, A.L., Sethunathan, N. and Naidu, R. 2003. Chromium-microorganism interactions in soils: remediation implications. *Rev. Environ.Contam. Toxicol.* 178 : 93-164.

Kapoor, A. and Viraraghavan, T. 1997. Heavy metal biosorption sites in *Aspergillus niger*. *Bioresource Technol.* 61 : 221-227.

Loukidou, M.X., Matisa, K.A., Zouboulisa, A.L. and

Kyriakidoub, M.L. 2003. Removal of As (V) from wastewaters by chemically modified fungal biomass *Water Res.* 37 : 4544-4552.

Park, D., Yun, Y. and Park, J. M. 2005. Studies on hexavalent chromium biosorption by chemically-treated biomass of Ecklonia sp., *Chemosphere.* 60 : 1356-1364.

Sað, Y. 2001. Biosorption of heavy metals by fungal biomass and modeling of fungal biosorption: A review, *Separation and Purification Rev*. 30 (1): 1-48.

Tunali, S., Kiran, I. and Akar, T. 2005. Chromium (VI) biosorption characteristics of *Neurospora crassa* fungal biomass. *Minerals Eng.* 18 : 681-689.