The paper provides important information on the use of fungal biotechnology in the treatment of wastewaters containing heavy metals. The authors reported that pretreating *Aspergillus oryzae* by acid washing increased copper removal. The results of our experiments on biosorption of heavy metals such as lead, cadmium, copper and nickel on *Aspergillus niger* showed that metal removal can be significantly increased or decreased depending on the pretreatment step and the heavy metal involved. We studied the effect of acid pretreatment (by using glacial and acetic acid) on biosorption of heavy metals on *A. niger*. The pretreatment steps used in our experiments are as follows: 10% v/v solution of o-phosphoric and glacial acetic acid was used. The live biomass was boiled for 15 min in acid solution and then upon cooling, the solution was centrifuged to separate the biomass. The biomass was then generously washed with deionized water till the pH of the wash solution was in the near neutral range (pH 7.0 ± 0.2). We would like to point out that pretreatment conditions of our experiments were more aggressive than those used by the authors. The results of our experiments are shown in Table 1: Biosorption of lead and copper was found to increase by acid pretreatment, but biosorption of cadmium and nickel decreased in comparison to metal biosorption by live cells. Galun *et al.* (1987) also observed that hydrochloric acid pretreatment of *Penicillium digitatum* biomass resulted in a decrease in biosorption of heavy metal such as Ni, Cu, Zn and Cd. Ting and Teo (1994) observed that nitric acid pretreatment resulted in increased biosorption of Cd and Zn on *Saccharomyces cerevisiae*. We further observed that boiling *A. niger* biomass in 0.5 N NaOH solution, 15% formaldehyde solution and solution of commercial laundry detergent for 15 min resulted in significant improvement in biosorption of copper, cadmium and lead in all cases (up to three times that of live biomass). It can be concluded that a particular pretreatment method may be specific to a particular strain and heavy metal under consideration when enhancement of biosorption potential is considered.

**REFERENCES**


**AUTHOR'S REPLY**

We agree with the conclusion drawn by Kapoor and Viraraghavan. That is a particular pretreatment method may be specific to a particular fungal species as well as heavy metal. Actually, we have some experimental results relative to their results as listed in Table 1. In our experiments, the biomass were washed by $5 \times 10^{-3}$ M perchloric acid for 15 min twice. It shows that treatment by perchloric acid results in a positive effect on the biosorption of those four metals. This evidence, however, was not found in the species of *Rhizopus oryzae*. Except for a little decrease on Pb(II) biosorption, the acid-treated *R. oryzae* biomass possessed the similar adsorption capacity of other three metals to the native biomass. As described in the paper of Huang and Huang (1996) by referring the X-ray EDA spectra, *A. oryzae* biomass...
biomass after being acid treated contains a higher percentage of surface nitrogen, and the surface components of *R. oryzae*, however, remain unchanged after acid pretreatment. These spectral results are direct evidence for the changes of metal adsorption caused by acid pretreatment.

Viraraghavan's comment also mentioned they used more aggressive methods to treat live biomass by boiling for 15 min in acid solution. We respond as follows: Boiling could cause a loss of amino-functional groups on the fungal surface through the non-enzymic browning reaction (also called as Maillard reaction) (Whistler and Daniel, 1985). Actually, we have a similar result showing that the decrease of cadmium adsorption by *R. oryzae* after heat pretreatment (Huang and Chiu, 1994). Hence, the different results obtained from our experiments and Viraraghavan’s experiments are mostly due to whether the heating has been applied or not.

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