Int J Pharm Bio Sci 2012 Oct; 3(4): (B) 1145 - 1149

Research Article **Microbiology**

International Journal of Pharma and Bio Sciences **ISSN 0975-6299**

URANIUM AND THORIUM UPAKE BY LIVE AND DEAD CELLS OF BACILLUS SP.

SUJIN JEBA KUMAR.T.,* AND BALAVIGNESWARAN.C.K

Department of Bio-Technology,Udaya School of Engineering, Vellamodi, Kanyakumari District, TN.

ABSTRACT

Bacillus Sp. was isolated near the mining area and was studied for biosorption U and Th. This study presents uptake of uranium (U) and thorium (Th) by live and dead cells of Bacillus Sp. Increasing concentration of U and Th showed decrease in absorption by Bacillus Sp. Dead cells of Bacillus Sp. exhibited same or more uptake of U and Th than living cells. Increasing temperature promotes uptake of U and Th by Bacillus Sp.

KEYWORDS : Bacillus Sp., Thorium, Uranium, Biosorption

SUJIN JEBA KUMAR.T
Department of Bio-Technology, Udaya School of Engineering, Vellamodi, Kanyakumari District, TN.

Corresponding author

INTRODUCTION

Radionuclides [uranium (U) and thorium (Th)] have high toxicity, causing severe ecological and public health hazards. Bio-adsorbents have emerged as promising alternatives to conventional heavy metal management stragies.[1] Uranium contamination poses a threat in some surface and ground waters.[2] This has prompted the development of various technologies for its removal from wastes produced from nuclear power programs and nuclear fuel reprocessing activities. Alongside developments from traditional chemical treatment processes, interest has also centered on the application of biomaterials technology for the removal of trace amounts of toxic metals from dilute aqueous wastes. Uranium and Thorium adsorption by different living and non living biomass are reported in the literature[3,4,5,6,7,8] Conventional treatments of meal effluents are either ineffective, costly, complicated or have sludge disposal problems. Biosorption is regarded as an innovative technology to remove metals from aqueous solution because it has some obvious advantages[9] high efficiency and selectivity for absorbing metals in low concentrations, energy-saving, broad operational range of pH and temperature, easy reclamation of metals, and easy recycling of the biosorbent. Identifying microbial ligands/cellular processes involved in metal sequenstration has lead to development of engineered organizations with various cell surface displays that facilitate their applications as industrial catalysts, biosorbents, bioremediation, biofuel and biosensors.[10] This study presents an inexpensive alternative method to evaluate efficiency of isolated Bacillus Sp. from mining area to absorb U and Th from solutions.

MATERIALS AND METHODS

(i) Isolation of Bacillus Sp.

Isolation Bacillus Sp. was isolated from contaminated water of mining area of Indian Rare Earth Ltd., Manavalakurichy, TN, India. Selected bacterium was indentified based on Bergey's Manual of systematic bacteriology. Isolated *Bacillus Sp*. were maintained in nutrient agar and cultivated on LB medium.

(ii)Preparation of Uranium (U) and Thorium (Th)solutions and their uptake experiment

All U and Th solutions were prepared from uranyl nitrate (Merck) and thorium nitrate (Merck) respectively. Bacterial cells were incubated at 28^{o}C in LB medium. Mid-log phase cells of test bacterium were harvested by centrifuging at 5000 rpm for 10 min. Cells were then washed in sterile distilled water 3 times and re-suspended in 10 ml distilled water. A known volume of Isolated bacterial cell suspension (1 mg dry mass/ml of U solution) was added, mixed and incubated at 28^{o}C. After 12 h of shaking (150 rpm, 28^{o}C), biomass was centrifuged at 5000 rpm for 10 min. Supernatant was used for U estimation. Th was added in place of U to check uptake ability. Different cell concentration (0.2 – 3.0 mg dry mass of Bacillus Sp.) was added to solutions (1 ml each) containing U (30 (μg ml^{-1}) and Th (20 (μg ml^{-1}).

(iii)Different Concentration of Uranium (U) and Thorium (Th) Uptake by Live and Dead cells of Bacillus Sp.

Live cells were harvested by centrifuging at 500 rpm for 10 min. Cells were then washed three times in sterile distilled water and re-suspended in 10 ml distilled water. Cell mass (1 mg) was used to uptake different concentration of metals [U (10, 15, 20, 25 and 30 (μg ml^{-1})] from solutions (1 ml each). Dead cells (1 mg) were used to uptake U and Th from solutions. Bacterial suspensions were heated at 100^{o}C for 15 min to kill cells.

(iv) Temperature Effect on uptake of Uranium (U) and Thorium (Th)

In order to study temperature effect, a known volume of each cell suspension (1 mg dry

mass / ml of U and Th solution) was added. Mixtures were incubated at different temperatures (20, 30, 40 and 50°C). After 12 h of shaking at 150 rpm, biomass was centrifuged at 5000 rpm for 10 min.

(v) Measurement of metal uptake

The uranium uptake was calculated by the simple concentration difference method. The initial concentration, Co (mg/1) and metal concentration at any time, Ct (mg/l) were determined using UV-Vis spectrophotometer at 650 nm (Hitachi Model 330, Japan) and the metal uptake q was calculated from the mass balance as follows:

$$q = \frac{(Co - C1),V}{w.1000},$$

where *V* is the volume of solution in millimeter and *w* the mass of adsorbent in grams. The measurement of metal uptake was indicated in percentage. The mean of five experimental values were considered as an optimum value.

RESULT AND DISCUSSION

Table -1
Concentrations of uranium and thorium uptake by live cells of Bacillus

Sl.No.	**Uranium conc.** (μg ml^{-1})	**Uranium adsorbed** (%)	**Thorium conc.** (μg ml^{-1})	**Thorium adsorbed** (%)
1.	5	100	4	84
2.	10	90	8	72
3.	15	78	12	60
4.	20	64	16	45
5.	25	52	20	33
6.	30	40	24	21

1) Concentrations of uranium and thorium uptake by live cells of Bacillus

Bacillus strains absorbed highest U at low concentration (5mgml^{-1}) and U uptake decreased as U concentration increase. Also, Th uptake decreased as Th concentration increased and highest Th uptake was achieved at low concentration of (4(μg Th ml^{-1}). Thus U and Th uptake decreased as metal concentration increased; lowest absorption was at highest concentration increased; lowest absorption was at highest concentration used (U, 30(μg U ml^{-1} ; Th, 20 (μg ml^{-1} · It showed composition of cell wall of *Bacillus Sp.* to entrap U and Th ions as metal ion uptake biomass is believed to occur through interactions with functional groups native to proteins, lipid, and carbohydrates that make up the cell wall.[11]

Table -2
Concentrations of biomass of Bacillus Sp. to uptake Uranium and Thorium

Sl. No.	Cell Concentration (mg)	Uranium adsorbed (%)	Thorium adsorbed (%)
1.	0.4	30	20
2.	1.0	45	30
3.	1.6	60	40
4.	2.2	70	60
5.	2.8	85	70
6.	3.2	90	80
7.	4.0	100	100

2) Different concentration of Biomass of Bacillus Sp. to uptake metals

Effect of cell biomass (0.4 – 4 mg, dry basis) of Bacillus Sp. on metal uptake was studied using solutions containing U (30 ($\mu g\ ml^{-1}$) and Th (20 ($\mu g\ ml^{-1}$) maximum uptake (100%) of U and Th was achieved (Table 2) at highest cell concentration used (4.0 mg). The result may be due to the saturation of the adsorption sites and increase in the number of ions competing for he available binding sites in the biomass for complexation of U at higher concentration.[12]

Table - 3
Concentrations of uranium and thorium uptake by dead cells of Bacillus

Sl. No.	Uranium conc. ($\mu g\ ml^{-1}$)	Uranium adsorbed (%)	Thorium conc. ($\mu\ gml^{-1}$)	Thorium adsorbed (%)
1.	5	100	2	100
2.	10	95	4	95
3.	15	90	8	85
4.	20	83	12	75
5.	25	78	16	68
6.	30	70	20	55

3) Different concentration of Uranium (U) and Thorium (Th) uptake by dead cells Bacillus Sp.

Dead cells absorbed more U and Th than living cells (Table 3). It was found that dead cells could accumulate heavy metals to same or even greater extent than living cells. Since same concentration of living and dead cells was used, increased removal by dead cells may be due to death process itself.

Table - 4
Effect of temperature on uptake of uranium and thorium by Bacillus Sp.

Sl. No.	Temperature ^oC	Uranium adsorbed (%)	Thorium adsorbed (%)
1.	20	50	10
2.	30	70	30
3.	40	80	40
4.	50	90	60

4) Temperature Effect on uptake of U and Th by Bacillus Sp.

Metal uptake increased with increase of temperature (Table 4). At 50^{o}C was maximum uptake of U (90%) and Th (60%) as the result of study of the effect of the temperature on the phenomenon of sorption for different metal-biomaterials systems are disparate.[13] In this case, the increasing temperature mainly resulted in increase in biosorption.

CONCLUSION

Increase in biomass concentration of Bacillus Sp. uptake more amounts of U ant Th. Dead cells exhibited same or greater uptake of U and Th than living cells. Maximum Th (100%) was absorbed by Bacillus Sp. at 0.4 mg dry mass per ml solution. Increasing temperature uptake of U and Th by Bacillus Sp. Future studies include biosorption studies of U and Th using chemically modified and immobilize Bacillus for efficient adsorption.

REFERENCES

1. Shumate, E.S. & Strandberg, W.G.,.1935. Accumulation of metals by microbial cells. *Comprehensive Biotechnol*, 13. 235-247.
2. Laul, J.C., 1992. Natural radionuclides in groundwaters. J. Radioanal. Nucl. Chem. 156, 235-242.
3. Guibal,E., Roulph,C., Le Cloirec, P., 1992. Uranium biosorption by a filamentous fungus Mucor michei pH effect on mechanisms and performances of uptake. Water Res. 26. 1139-1145.
4. Hu, M.Z.C., Norman.J.M., Faison, N.B., Reeves, M., 1996. Biosorption of uranium by Bacillusaeruginosa strain CSU. characterization and comparison studies. Biotechnol. Bioeng.51, 237-247.
5. Byerley, J.J., Scharer, J.M., Charles, A.M., 1987. Uranium (VI) biosorption from process solutions. Chem. Eng. J. 36, 46-49.
6. Horikoshi, T., Nakajima, A., Sakaguchi, T., 1979. Uptake of uranium by Chlorella regularis. Biol. Chem. Tokyo 43. 617-623.
7. Yang.J., Volesky.B., 1999. Biosorption of uranium on Sargassum biomass. Water. Res. 33, 3357-3363.
8. Tsezos, M., Volesky, B., 1981. Biosorption of uranium and thorium. Biotechnol. Bioeng. 23. 583-604.
9. Chen, M., Gan, Y.R., 1999. Biosorption of heavy metal. Chemical Industry and Engineering 16(1), 19-25.
10. Mandal, D., Bolander M.E., Mukhopadhyay D., Sarkar G & Mukherjee, P., 2006. The use of microorganisms for the formation of metal nanoparticles and their applications, *Appl. Microbiol Biotechnol.* 69. 485-492.
11. Chen, X.C., et. al. 2007. Determination of copper binding in Bacillusputida CZ1 by chemical modifications and x-ray absorption spectroscopy. Appl. Microbial. Biotechnol. 74. 881-889.
12. Bai, S.R., Abraham, T.E., 2001a. Biosorption of Chromium (VI) from aqueous solution and electroplating waste water using fungal biomass. Chemical Engineering Journal 135, 202-208.
13. Benguella, B., Benaissa, H., 2002. Cadmium removal from aqueous solution by chitin: kinetic and equilibrium studies. Water Res. 36. 2463-2474.