

Chapter 11

Biosorption of Uranium for Environmental Applications Using Bacteria Isolated from the Uranium Deposits

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Abstract Attempts were made to recover uranium (U) occurring in nuclear fuel effluents and mine tailings using bacteria isolated from U deposits in Canada, the United States, Australia, and Japan. To establish which microorganisms accumulate the most U, hundreds of strains of microorganisms were screened. Extremely high U accumulating ability was detected in some bacteria isolated from North American U deposits. *Arthrobacter* and *Bacillus* sp. accumulated approx. 2,500 $\mu\text{mol U/g}$ dry wt. of microbial cells within 1 h. Cells removed U from refining wastewater with high efficiency. Cells also accumulated thorium with high efficiency. *Lactobacillus* cells isolated from Japanese U deposits removed more U from seawater than the other bacteria that had superior U removal capacity from nonsaline U solutions. Cells immobilized with polyacrylamide gel had excellent handling characteristics and can be used repeatedly in U adsorption–desorption cycles. These bacteria from U deposits can be used as an adsorbing agent for the removal of the nuclear fuel elements, which may be present in nuclear effluents, mine tailings, seawater, and other waste sources.

11.1 Introduction

The recoveries of nuclear fuel elements, such as uranium (U) and thorium (Th), from aqueous systems have become a focus of interest for exploitation of undeveloped energy resources. The removal of radioactive elements and toxic heavy

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metals from contaminated sources is also a worthwhile priority for environmental protection initiatives. In this regard, efforts have concentrated on studying the accumulation of U by microorganisms, including bacteria (Andres et al. 1993; Byerley et al. 1987; Friess and Myers-Keith 1986; Gorab et al. 1991; Hu et al. 1996; Marques et al. 1991; Strandberg et al. 1981), fungi (Byerley et al. 1987; Galun et al. 1983a, b; Tsezos and Volesky 1981; White and Gadds 1990), and yeasts (Strandberg et al. 1981; Shumate et al. 1978).

We have investigated U accumulation from aqueous systems using bacteria isolated from U mines, among which some strains were found to possess extremely high U accumulating ability (Sakaguchi et al. 1996). Microbial biomass may thus be considered for use as a removal agent for the recovery of U from metallurgical effluents, mine tailings, seawater, and other waste sources.

In U deposits, it can be presumed that some microorganisms having a high accumulating ability for U and different species having an ability to leach U from ore may exist in mine soil and aqueous systems. It would, therefore, be beneficial to isolate microorganisms having an enhanced ability to accumulate U from mines.

Recently, we screened hundreds of types of microorganisms existing in U deposits located in North America, Australia, and Japan for their ability to accumulate significant quantities of U, and identified new strains which accumulated large quantities of U, such as *Bacillus subtilis* in Australia, *Arthrobacter* and *Bacillus* sp. in North America, and *Lactobacillus* and *Bacillus* sp. in Japan (Sakaguchi 1998). In this chapter, new strains of bacteria identified in North American, Australian, and Japanese U deposits, especially *Arthrobacter*, *Lactobacter*, and *Bacillus* sp., are discussed for their potential for the removal of nuclear fuel elements such as U from U refining wastewater and seawater.

11.2 Screening of Microorganisms Isolated from U Deposits for Their U Accumulating Ability

To determine the ability of microorganisms isolated from U deposits in Canada, the United States, Australia, and Japan to accumulate U, hundreds of strains of microorganisms were screened.

The medium for growing microorganisms contained 3 g/L beef extract, 5 g/L peptone, and 5 g/L NaCl in deionized water. The microorganisms were maintained on agar slants and grown in 300 mL of the medium in a 500-mL flask with continuous shaking (120 rpm) for 72 h at 30°C. Cells were collected by centrifugation, washed thoroughly with deionized water, and then used in the following accumulation experiments.

U was supplied as $\text{UO}_2(\text{NO}_3)_2$. The pH of the solution was adjusted to 5.8 with 0.1 M HNO_3 . Resting microorganisms (15 mg dry wt.) were suspended in 100 mL solution (pH 5.8) containing 84 μM U and the suspension was shaken for 1 h at 25°C. Cells were collected by filtration through a membrane filter (pore size 0.2 μm). The quantity of U accumulated by the cells was determined by measuring

U in the filtrate using an inductively coupled plasma quantometer (ICPS8000; Shimadzu Corporation, Kyoto, Japan). The microbial strains were identified by our coworker (Sakaguchi 1998).

The quantities of U accumulated by the cells ranged from a minimum of 10.9% to a maximum of 98.3% (Table 11.1). Of special interest to this discussion is the wide range of effectiveness with which different species of microorganisms accumulate U.

Of these microorganisms tested, extremely high U accumulating ability was found in *Arthrobacter* (96.4%) and *Bacillus* sp. (95.9%) found in US, *Lactobacillus* (97.8%) and *Bacillus* sp. (97.6%) found in Japan, and *Bacillus* sp. (98.3%) found in Australia (Sakaguchi 1998), which accumulated large quantities of U per gram dry wt. of microbial cells within 5 min.

11.2.1 Factors Affecting U Accumulation by Bacteria

In order to obtain basic information regarding the removal of U using strains of bacteria found in U deposits, some factors affecting U accumulation were investigated in detail using *Arthrobacter* sp., US-10 isolated from United States U deposits.

11.2.2 Effect of pH on U Accumulation

The pH of a test solution was adjusted to the desired value with 0.1 M HNO₃ or 0.1 M NaOH. Resting *Arthrobacter* cells (15 mg dry wt.) were suspended in 100 mL solution containing 84 μM U for 1 h at 25°C.

Additionally, Zeta potential was measured by the electrolysis method using 30–50 bacteria (bacterial concentration was 2.5×10^8 cells/mL) at pH 2–9 in 0.01 M NaNO₃ solution (ZC-2000; Microtec-Nichion, Chiba, Japan).

The effects of pH on U absorption using *Arthrobacter* sp., US-10 and *Lactobacillus* sp., JPN-10 are shown in Fig. 11.1. The amounts of absorbed U are highest at approx. pH 5–8 and decrease with increasing acidity below pH 4 using *Arthrobacter* cells. On the other hand, the amounts of absorbed U using *Lactobacillus* were highest at pH 6, and rapidly decreased below pH 5 and above pH 7. Low pH is the result, of course, of higher proton concentrations, which can compete for binding with U, and *Arthrobacter* cells clearly remained less affected than *Lactobacillus* by the decreasing pH until about pH 4. Conversely, the high pH is, of course, the result of reduced proton concentration and increased hydroxide ions, which compete with microbial cells. Again, *Arthrobacter* exhibited a higher pH range for binding capacity than *Lactobacillus*, achieving good absorption at pH values as high as 8, while *Lactobacillus* performed well only at pH 6.

The effects of pH on Zeta potential of the cell surface are shown in Table 11.2. The quantities of U accumulated are highest at approx. pH 5 and decrease below pH 4.

Table 11.1 Accumulation of uranium using microorganisms from soil or water at uranium deposit

Strain number	Uranium accumulated			Uranium accumulated			
	(%)	Strain number	Uranium accumulated (%)	Strain number	Uranium accumulated (%)	Uranium accumulated (%)	
US-1	14	CAN-1	12.6	AUS-1	19.4	JPN-1	10.9
US-2	17.1	CAN-2	23.7	AUS-2	25.4	JPN-2	21.2
US-3	23.9	CAN-3	33.9	AUS-3	36.6	JPN-3	32.2
US-4	38.7	CAN-4	49.8	AUS-4	46.2	JPN-4	44.7
US-5	43.9	CAN-5	55.7	AUS-5	53.3	JPN-5	54.9
US-6	58.9	CAN-6	67.9	AUS-6	68.8	JPN-6	65.2
US-7	72	CAN-7	74.2	AUS-7	74.7	JPN-7	76.8
US-8	84.4	CAN-8	85.7	AUS-8	85.1	JPN-8	86.2
<i>Bacillus</i> sp., US-9	95.9	CAN-9	95.4	AUS-9	96.9	<i>Bacillus</i> sp., JPN-9	97.6
<i>Arthrobacter</i> sp., US-10	96.4	CAN-10	96.9	<i>Bacillus</i> sp., AUS-10	98.3	<i>Lactobacillus</i> sp., JPN-10	97.8

Fig. 11.1 Effects of pH on the amount of uranium absorbed using *Arthrobacter* and *Lactobacillus* cells. Symbols: closed circles, uranium (VI) accumulated ($\mu\text{mol/g}$ dry wt. cells) using *Arthrobacter* cells; open circles, uranium (VI) absorbed ($\mu\text{mol/g}$ dry wt. cells) using *Lactobacillus* cells

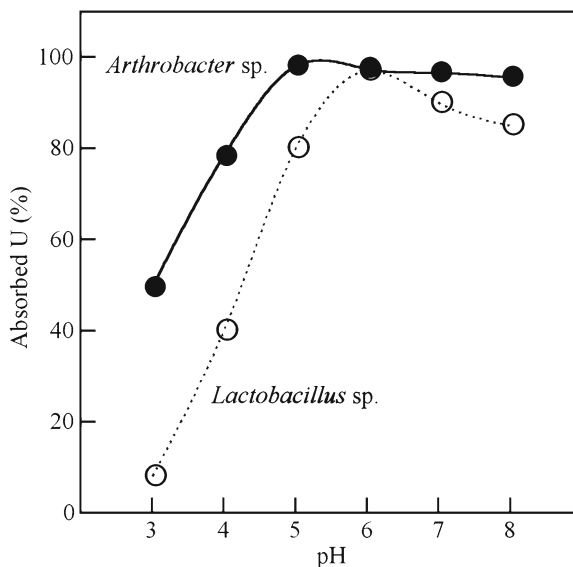


Table 11.2 Effect of pH on Zeta potential of *Arthrobacter* cell

pH	2.07	3.00	4.04	5.09	6.07	6.97	8.06
Zeta potential (mV)	3.69	-1.50	-7.33	-21.01	-23.18	-22.11	-23.06

Thus, the accumulation of U by *Arthrobacter* is markedly affected by solution pH. Zeta potential of the cell surface increased with increasing acidity of the solution.

The main U species at pH 6 includes some cations, such as $(\text{UO}_2)_3(\text{OH})_5^+$ and UO_2OH^+ (Tsuruta 2006). On the other hand, Zeta potential of the cell surface was -23.18 mV at the same pH. Therefore, cationic uranyl species readily bond with negatively charged cell surfaces of *Arthrobacter* sp.

11.2.3 Effect of U Concentration on U Absorption

Resting *Arthrobacter* and *Lactobacillus* cells (15 mg dry wt.) were suspended in 200 mL of solution (pH 5.8) containing a specified quantity of U for 1 h at 25°C .

Quantities of absorbed U using *Arthrobacter* and *Lactobacillus* sp. ($\mu\text{mol U/g}$ cells) increased as U concentration increased (Fig. 11.2). *Arthrobacter* and *Lactobacillus* cells accumulated 2,480 and 2,120 $\mu\text{mol U/g}$ of cells, respectively.

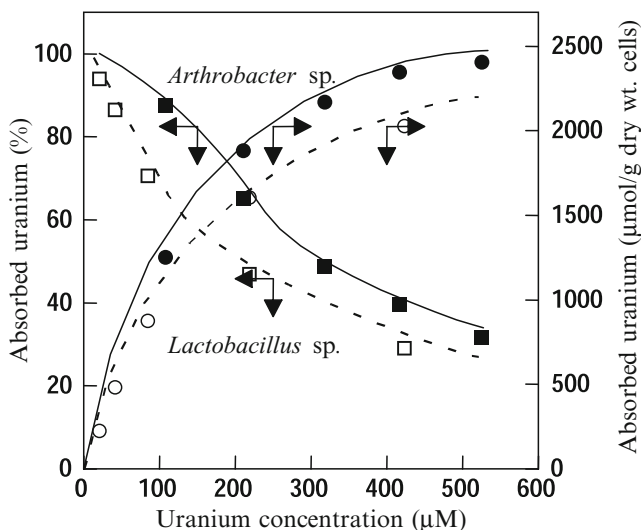


Fig. 11.2 Effect of uranium concentration on uranium absorption using *Arthrobacter*, US-10 and *Lactobacillus*, JPN-10 cells. Symbols: circles, accumulated uranium ($\mu\text{mol/g}$ dry wt. cells); squares, accumulated uranium (%); closed, absorbed uranium using *Arthrobacter* sp.; open, absorbed uranium using *Lactobacillus* sp.

Additionally, U absorption using these cells, especially *Lactobacillus* sp., does not obey the Langmuir isotherm over the entire U concentration range tested. It appears that the experimental data show a dual pattern. The dotted line was calculated using less than a $21.8\text{-}\mu\text{M}$ residual U concentration (as the initial uranium concentration was $85.0\text{ }\mu\text{M}$) using *Lactobacillus* cells. The solid line was separately calculated using residual concentrations below and above $21.8\text{ }\mu\text{M}$. When the initial U concentration increased beyond $85.0\text{ }\mu\text{M}$ (with a $21.8\text{-}\mu\text{M}$ residual U concentration), the absorbed quantity increased to values far greater than those calculated based on the relationship in the low concentration range. On the other hand, the dotted line in Fig. 11.3 for *Arthrobacter* cells was calculated using a residual U concentration below $153\text{ }\mu\text{M}$ (and an initial U concentration of $318\text{ }\mu\text{M}$). The solid line was separately calculated for *Arthrobacter* cells using residual concentrations below and above $153\text{ }\mu\text{M}$. When the initial U concentration increased beyond $318\text{ }\mu\text{M}$ (with a residual U concentration of $153\text{ }\mu\text{M}$), the absorbed quantity increased to values a little greater than those calculated based on the relationship in the low concentration range. A similar result was obtained (Epstein 1966) from the absorption of potassium using barley roots. The estimated m , n , and maximum accumulated U capacity $Q(U)_{\text{max}}$ ($=1/m$) are summarized in Table 11.3. A high $Q(U)_{\text{max}}$ value of $2,580$ and $2,370\text{ }\mu\text{mol/g}$ dry wt. cells is estimated from the high U concentration region using *Arthrobacter* and *Lactobacillus* cells, respectively.

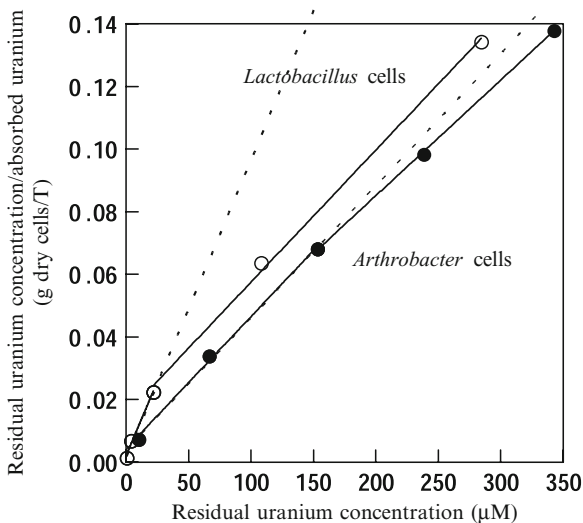


Fig. 11.3 Equilibrium isotherm of uranium absorption using microbial cells. Symbols: *closed*, absorbed uranium using *Arthrobacter* sp.; *open*, absorbed uranium using *Lactobacillus* sp. $C_e(U)/Q_U = mC_e(U) + n$, where Q_U indicates the amount of absorbed uranium (μmol uranium/g dry wt. cells), C_e is the residual uranium in the solution (μM), and m and n are constants

Table 11.3 Estimated Langmuir constants and $Q(U)_{\text{max}}$ from the Langmuir isotherm

Strains	Initial uranium concentration (μM)	Residual uranium concentration (μM)	m	n	$Q(U)_{\text{max}}$ ($\mu\text{mol/g}$ dry cells)
<i>Arthrobacter</i> sp.	107–318	10.0–153	4.22×10^{-4}	4.32×10^{-3}	2,371
	318–525	153–343	3.69×10^{-4}	1.13×10^{-2}	2,710
	107–525	10.0–343	3.88×10^{-4}	6.45×10^{-3}	2,580
<i>Lactobacillus</i> sp.	21.0–85.0	0.483–21.8	9.51×10^{-4}	2.05×10^{-3}	1,050
	85.0–423	21.8–284	4.22×10^{-4}	1.53×10^{-2}	2,370
	21.0–423	0–284	4.56×10^{-4}	769×10^{-3}	2,200

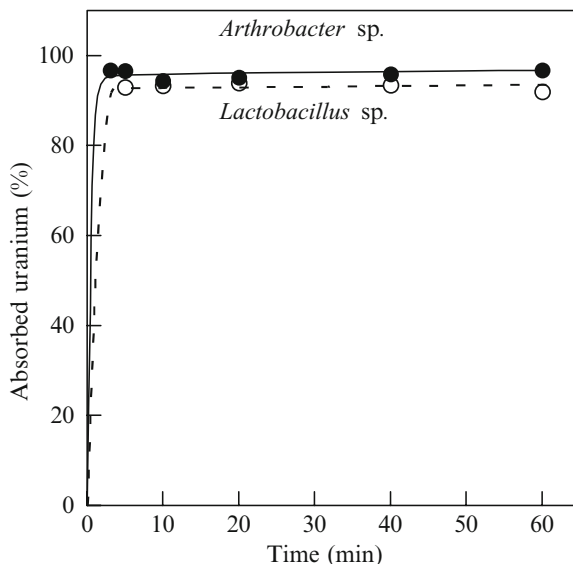
See the legend of Fig. 11.3

11.2.4 Time Course of U Accumulation

Resting *Arthrobacter* and *Lactobacillus* cells (15 mg dry wt.) were suspended in 100 mL solution (pH 5.8) containing 84 μM U at 25°C.

Quantities of U accumulated by *Arthrobacter* and *Lactobacillus* cells increased rapidly during the first 5 min following the application of U (Fig. 11.4).

Fig. 11.4 Time course of uranium absorption using *Arthrobacter* and *Lactobacillus* cells. Symbols: closed, absorbed uranium using *Arthrobacter* sp.; open, absorbed uranium using *Lactobacillus* sp.



11.2.5 Release of U from Cells by Washing with EDTA

Resting cells (15 mg dry wt.) were suspended in 100 mL solution (pH 6.0) containing 84 μM uranium for 1 h at 25°C. Cells with accumulated U were washed three times with 10 mL of 10 mM EDTA solution.

When cells of *Arthrobacter* sp., US-10, *Bacillus* sp., US-9; and *Lactobacillus* sp., JPN-10 were washed with EDTA, approx. 70, 80, and 53% of the accumulated U was desorbed from the resting cells, suggesting that most U is coupled with ligands that are easily substituted by EDTA (Fig. 11.5). However, 30, 20, and 47% of accumulated U in *Arthrobacter*, *Bacillus*, and *Lactobacillus* sp. were therefore not substituted by EDTA washing that incorporated within cell membranes. The ratio of released U (%) occurred in the following order: *Bacillus* sp. > *Arthrobacter* sp. > *Lactobacillus* sp.

11.2.6 Distribution of U in Microbial Cells

The present experiments were undertaken to determine which parts of the cells had accumulated U in *Arthrobacter*, US-10 and *Lactobacillus*, JPN-10 cells.

Resting cells of *Arthrobacter* and *Lactobacillus* (640 mg fresh weight) were suspended in 1,000 mL solution (pH 5.8) containing 500 μM of U for 1 h at 25°C. The cells were fractionated as described in Fig. 11.6. The freeze-dried *Arthrobacter*

Fig. 11.5 Release of uranium from *Lactobacillus* sp., JPN-10; *Arthrobacter* sp., US-10; or *Bacillus* sp., US-9 cells by washing with EDTA

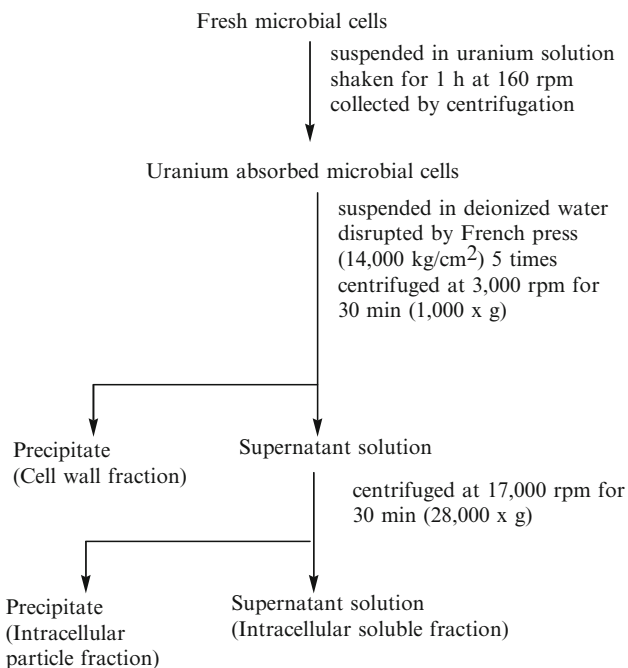
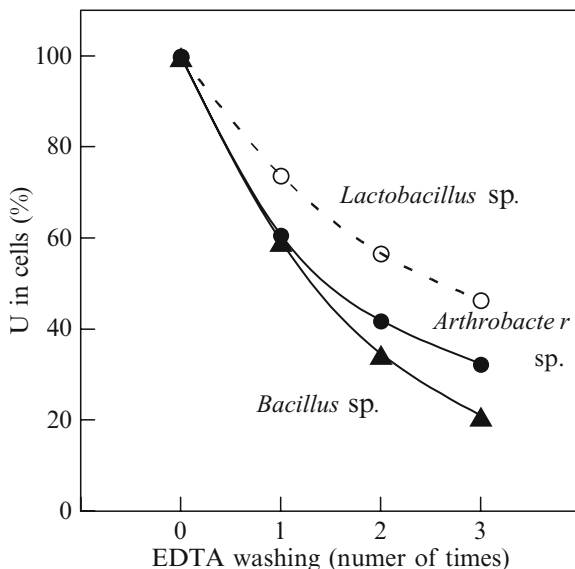


Fig. 11.6 Fractionation of *Lactobacillus* sp., JPN-10, *Arthrobacter* sp., US-10 absorbed uranium

Table 11.4 Distribution of uranium in microbial cells

Fractions	Absorbed uranium			
	<i>Arthrobacter</i> sp.		<i>Lactobacillus</i> sp.	
	(μmol)	(%)	(μmol)	(%)
Whole cells	237	100	220	100
Cell wall fraction	140	59	209	95
Intracellular particle fraction	51	21	11	5
Intracellular soluble fraction	32	13	4	2

and *Lactobacillus* cells, cell wall, and intracellular particle fractions were digested in the mixed solution of same volume of conc. $\text{HNO}_3/\text{H}_2\text{SO}_4$.

The quantities of accumulated U in each fraction were recorded in the following order (Table 11.4)

Cell wall > intracellular particle > intracellular soluble.

Abundant U was determined in the cell wall fraction and small amounts occurred in the intracellular fractions. In the case of *Arthrobacter*, this result coincides with the results noted above for EDTA washing. On the other hand, the results using *Lactobacillus* suggested that most accumulated U was coupled with the cell wall fraction; half of the accumulated U was not released by washing with EDTA. Therefore, the bond of U with cell wall of *Lactobacillus* cells appears to be strong.

On the basis of these findings, it seems reasonable to postulate that the accumulated U using the cells, especially *Lactobacillus* sp. is mostly dependent on the physical–chemical binding relationships with cell wall components.

11.2.7 Selective Accumulation of U Using *Arthrobacter*, US-10 Cells

To determine which heavy metal ions are most readily accumulated by bacterial cells, the selective accumulation of ions using *Arthrobacter*, US-10 cells from a solution containing six metal cations and UO_2^{2+} was examined. Resting cells (15 mg dry wt.) were suspended in 100 mL of a solution (pH 5.0) containing 4×10^{-5} M of Mn^{2+} , Co^{2+} , Ni^{2+} , Cu^{2+} , Zn^{2+} , Cd^{2+} , and UO_2^{2+} for 1 h at 25°C .

The relative degree of heavy metal ions accumulated using *Arthrobacter*, US-10 cells was (Fig. 11.7) $\text{UO}_2^{2+} > \text{Cu}^{2+} > \text{others}$.

11.3 Accumulation of Th and Selective Accumulation of Th and U by Bacteria

As described above, bacteria found in U deposits accumulated U with high efficiency. In this course of our study, the question was raised as to whether these strains had the ability to accumulate Th, another common waste from nuclear processing (and an environmental contaminant), as well as U.

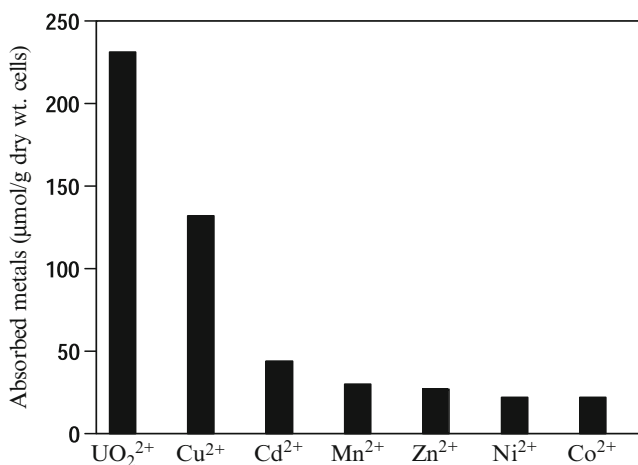


Fig. 11.7 Selective accumulation of heavy metals using *Arthrobacter*, US-10 cells

Table 11.5 Accumulation of thorium and/or uranium from the solution containing thorium and/or uranium

Strains	Metal accumulated from the solution containing Th or U only (μmol/g dry wt. cells)		Metals accumulated from the mixed solution containing Th and U (μmol/g dry wt. cells)	
	Th	U	Th	U
<i>Arthrobacter</i> sp.	98.8	98.9	98.5	52.1
<i>Bacillus</i> sp.	94.0	99.7	98.3	15.8
<i>Lactobacillus</i> sp.	47.1	64.9	46.7	17.9

As thorium hydroxide is precipitated in a solution containing thorium at pH 4.0, the accumulation of Th is examined at pH 3.5. Resting cells (15 mg dry wt.) were suspended in 100 mL solution (pH 3.5) containing 50 μM Th (as Th(NO₃)₄) and/or U for 1 h at 25°C.

Both *Arthrobacter* sp., US-10 and *Bacillus* sp., US-9 can also accumulate Th with high efficiency. However, the quantities of U and Th accumulated by *Lactobacillus* from the solution containing one metal only were lower than those using *Arthrobacter* and *Bacillus* sp. These results appear reasonable, because solution pH strongly affects the accumulation of both elements using *Lactobacillus* cells. The quantity of accumulated U from the solution containing both elements at pH 3.5 by *Arthrobacter* was half that from the solution containing U only; however, that accumulated by *Bacillus* from the solution containing both elements was far lower than that from the U-only solution. Accordingly, the effect of Th on U accumulation by *Bacillus* is greater than that by *Arthrobacter*. Thus, the *Arthrobacter* sp. appears to be the most efficient choice for a mixed solution of Th and U (Table 11.5).

11.3.1 Recovery of U by Immobilized Bacteria

As described above, bacteria such as *Arthrobacter*, *Bacillus*, and *Lactobacillus* sp. can accumulate large quantities of U from aqueous systems. However, the free cells of these bacteria are not reusable because of their mechanical instability and susceptibility to cell degradation. Furthermore, free cells are not suitable for use in column systems, because they cause plugging. To overcome these deficiencies with free cells, the cells of *Arthrobacter* sp., US-10 having high U accumulating ability were immobilized with polyacrylamide.

Five grams of precultured *Arthrobacter* cells were suspended in 4.5 mL isotonic NaCl solution and 680 mg acrylamide monomer. A total of 34 mg *N,N'*-methylenebis(acrylamide), 0.3 mL 3-dimethylaminopropionitrile solution (5%), and 0.34 mL potassium persulfate solution (2.5%) were added to the suspension. After solidification, the gel was crushed into small pieces (50–100 mesh), washed thoroughly with isotonic NaCl solution followed by deionized water, and then used for adsorption experiments.

To obtain basic information on the recovery of U using immobilized microbial cells, U adsorption–desorption cycle tests were carried out. It was previously shown (Sakaguchi et al. 1996) that the U retained on the adsorbent can easily be desorbed with dilute Na_2CO_3 solution, so 0.1 M Na_2CO_3 solution was used as the desorbent in this experiment.

Fifteen milliliters of a solution (pH 5.8) containing 42 μM U was adsorbed on a column (bed volume, 2 mL) of immobilized *Arthrobacter* cells at a space velocity of 20 h^{-1} . Adsorbed uranium was desorbed with 10 mL of 0.1 M Na_2CO_3 solution. The test was replicated five times.

The ability of the immobilized *Arthrobacter* cells to adsorb U did not decrease after six repetitions of adsorption–desorption cycles (Fig. 11.8). Thus, immobilized microbial cells appear to have excellent handling characteristics and can be used repeatedly in adsorption–desorption cycles.

11.3.2 Removal of U from U Refining Wastewater by Bacteria

As mentioned above, some microbial species have a high U accumulating ability, which suggests the possibility that they may be used for the removal of U from U mine tailings, U refining wastewater, and other waste sources.

We attempted to remove U from U refining wastewater sampled at the Ningyotoge Environmental Engineering Center of the Japan Atomic Energy Agency using bacteria exhibiting a significant ability to accumulate U. Resting cells (15.0 mg dry wt.) were suspended in 100 mL of a solution (pH 6.0) of wastewater containing 21.0 μM U for 1 h at 25°C.

Lactobacillus and *Bacillus* sp. isolated from Japanese U deposits removed 88.1 and 74.4% U, respectively (Table 11.6), when solution pH was adjusted initially to

Fig. 11.8 Test of repeated uranium adsorption-desorption using immobilized *Arthrobacter*, US-10 cells

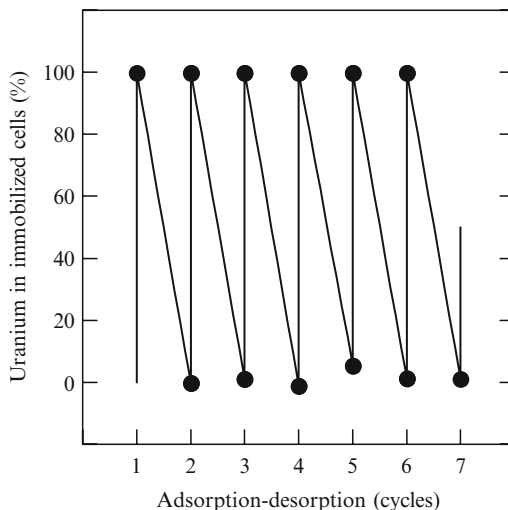


Table 11.6 Uranium removal from uranium refining wastewater using microbial cells isolated from Japanese uranium mine

Strains	Removed U (%)	
	pH adjusted only started at pH 6.0	pH adjusted continuously at pH 6.0
<i>Lactobacillus</i> sp.	88.1	99.5
<i>Bacillus</i> sp.	74.4	95.5

Table 11.7 Uranium removal from uranium refining wastewater using immobilized microorganisms isolated from uranium mines

Strains	Adsorbed uranium U (%)
<i>Arthrobacter</i> sp.	100
<i>Bacillus</i> sp.	100

6.0. Solution pH gradually decreased, with *Bacillus* cells being more adversely affected by pH change than *Lactobacillus* cells. However, both strains quantitatively removed U when the pH was maintained at 6.0. These species can thus remove U from U refining wastewater with a high efficiency.

Attempts were also made to remove U from U refining wastewater using immobilized microorganisms having a high ability to adsorb U. Uranium refining wastewater (100 mL, pH 6.0) supplemented with 2.1 mM of U were adsorbed on a column (bed volume 2 mL) of immobilized bacterial cells at a space velocity of 10 h⁻¹ at 25°C.

Immobilized bacterial cells isolated from U mines in the United States can also remove U from the U refining wastewater with high efficiency (Table 11.7).

Table 11.8 Accumulation of uranium using microorganisms isolated from uranium mines

Solutions	Accumulated U (%)		
	<i>Lactobacillus</i> sp.	<i>Arthrobacter</i> sp.	<i>Bacillus</i> sp.
Uranium only solution (pH 8)	94.7	94.2	94.6
Natural seawater	36.2	0.8	0.9
Decarbonated seawater	70.2	6.1	6.0

11.3.3 Removal of U from Seawater by Bacteria

The removal of U from seawater supplemented with 4.2 μM U using the bacteria isolated from U deposits was examined. The concentration of carbonate in seawater is $\sim 2.34 \times 10^{-3}$ M (Ogata et al. 1971). The amount of U removed by *Chlorella* cells from solutions containing 1.196×10^{-3} M sodium hydrogen carbonate was less at pH values above 6 than at pH 5 (Nakajima et al. 1979). The decrease in the amount of removed U from solutions containing carbonate was estimated from the amount of the UO_2CO_3 formed at pH 6 and of $\text{UO}_2(\text{CO}_3)_3^{4-}$ formed at pH values greater than 7 (Nakajima et al. 1979, 1981). Although *Lactobacillus* sp. removed 36.2% of U from seawater, it removed nearly twice as much (70.2%) when the seawater was decarbonated (Table 11.8). *Arthrobacter* and *Bacillus* cells, which can remove large amounts of U from nonsaline water, removed far less U from either seawater or decarbonated seawater than did *Lactobacillus*. Accordingly, *Lactobacillus* has great potential in applications to remove significant quantities of U from seawater.

11.4 Conclusion

In U deposits located in Canada, the United States, Australia, and Japan, we isolated strains of bacteria such as *Arthrobacter*, *Bacillus*, and *Lactobacillus* sp. having a significant ability to accumulate U. These species could accumulate approx. 2,500 μmol U/g dry wt. of microbial cells within 1 h. These strains accumulated U selectively from solution containing six other heavy metals in solution. Cells also accumulated Th as well as U with high efficiency. These species removed U from U refining wastewater with high efficiency. *Lactobacillus* also accumulated U from seawater more effectively than other microbial cells which have high accumulating capacities, from nonsaline U solution.

Cells immobilized with polyacrylamide gel have excellent handling characteristics and can be used repeatedly in U adsorption–desorption cycles.

These strains of *Arthrobacter*, *Bacillus*, and *Lactobacillus* can be used as an adsorbing agent for the removal of nuclear fuel elements which may be present in nuclear fuel processing effluents, mine tailings, seawater, and other environmental sources.

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