

MICROBIAL LEACHING OF LOW-GRADE SANDSTONE URANIUM ORES: COLUMN LEACHING STUDIES

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ABSTRACT

Microbial leaching studies on a low-grade sandstone uranium ore from Baghalchur Ore Deposits, D. G. Khan, Pakistan, containing 0.027 % U_3O_8 for extraction of uranium, were conducted in columns. Baghalchur sandstone uranium ore which is alkaline in nature, contained 5.0 % calcite [$CaCO_3$], 2-3 % Fe_2O_3 and pyrite [FeS_2] less than 0.1%. The ore amended with sulfur and/or sulfur slag as external energy source was found to leach with indigenous microflora mostly belonging to the genus *Thiobacillus* which are present in the uranium mine water. Column leaching studies revealed that when the ore was amended with elemental sulfur and irrigated with mine water (pH 3.5) 53% U_3O_8 could be solubilized from it. However, when the natural mine water was used as such (pH 7.4) the solubilization of uranium was decreased to 41 % U_3O_8 in 90 days under similar conditions of percolation rate and temperature. The addition of $(NH_4)_2SO_4$ (3.0 g/L) in mine water was found to enhance the uranium leaching to 70 % U_3O_8 from the columns containing ore amended with sulfur slag.

INTRODUCTION

The applications of biotechnological principles to recover metals from low-grade resources offer a number of advantages over conventional hydrometallurgical techniques. The major benefits perceived are such as low-cost, ease of maintenance and safety of environment. Heap, dump and *in-situ* leaching operations for recovering uranium from ores are presently being carried out on commercial scale in several countries (Torma 1986). These leaching processes are aided by natural populations of acidophilic iron- and sulfur- oxidizing *Thiobacillus* bacteria which have readily been isolated from uranium mine waters and leach liquors several years ago in the Elliot Lake Area, Ontario, Canada (Gow et al 1971; Harrison et al 1966), where *in-situ* leaching was practiced during the 1960's (MacGregor 1966).

Uranium recovery by bacterially-assisted leaching was began during the 1960's from acid leaching of mining wastes and of working and worked out stopes of mines (Fisher 1966 ; Fletcher 1970 ; Duncan and Bruynestyne 1971). At the Stanrock mine, Canada, a monthly recovery of 7.5 tonnes U_3O_8 was produced in this way (Sasson 1975) and recently, over 0.84×10^6 lbs of U_3O_8 has been recovered from stopes of worked out mines at the Denison Mines Ltd., Canada (Marchbank 1987). Exploitation of bacterial leaching enables the

recovery of uranium from low-grade ores (0.01-0.05% U_3O_8), uneconomic to process by conventional methods. The process can be applied to even poorer and of course to high grade material, such as uranium rich pillars supporting the roof of mine (Kelly 1976).

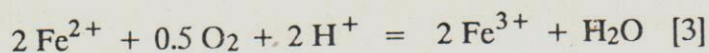
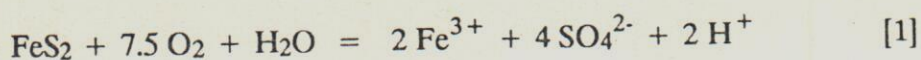
The well-established bacterial leaching techniques have been applied to sulfide ores or to the ores containing the iron sulfide minerals. Since these ores forms of oxidizable sulfur and iron, which can be used in degradation reactions carried out by microorganisms. In contrast the microbial degradation of silicate minerals required the availability of the external energy source (Ralph 1985). However, little informations are available in literatures about the biohydrometallurgy of uranium from sandstone type ore deposits. Some work on sedimentary type uranium ores from Fortsua, Austria and Ningyo-Toge, Japan, have also been reported (Bosecker and Wirth 1980; Tomizuka and Takahara 1972).

Uranium in nature does not exist as sulfides but usually occurs as insoluble tetravalent oxides. Uranium minerals are often associated with metal sulfides such as pyrite (FeS_2), pyrrhotite (FeS) and chalcopyrite ($CuFeS_2$). Over 100 minerals of uranium have been described in literature, but minerals of economic importance include, among many others, uraninite and pitchblende, which represent different crystalline forms of UO_2 in vein type ore deposits (Elevatroski 1977; Gow et al 1971). According to the data published jointly by the OECD Nuclear Energy Agency and International Atomic Energy Agency (IAEA) in 1983 the world's reasonably assured resources of uranium which are recoverable at US \$ 130/Kg U, to be 2.293×10^6 tonnes of U, with additional two and three million tonnes estimated. The major uranium resources of the world can be assigned on the basis of their geological setting to the following six categories of ore types are given in Table # 1 (Barthel 1980; Gow 1985).

Table 1: Major types of uranium ore deposits in the World

Deposit Type	Distribution of Uranium (%)
Sandstone	39
Quartz-pebble Conglomerate	17
Proterozoic Unconformity Related	15
Disseminated	14
Vein	7
Others:	8
a) Uraniferous shales	
b) Phosphates	
c) Calcretes	
d) Porous limestones.	
e) Lignites.	

The solubilization of metals depends on the activity of *Thiobacillus ferrooxidans* and *Thiobacillus thiooxidans*. Both species are chemolithotrophic bacteria which derive their energy from the oxidation of inorganic compounds. *Thiobacillus ferrooxidans* oxidizes both, reduced sulfur or iron compounds, whereas *Thiobacillus thiooxidans* oxidizes elemental sulfur and reduced sulfur compounds (Tuovinen 1986). In the case of metal sulfides, the production of sulfuric acid and soluble ferric iron is one of mechanism by which *T. ferrooxidans* indirectly accelerates the leaching of uranium ores:



Reactions [1] and [3] are catalyzed by the *T. ferrooxidans* at low pH value. The rate of bioleaching of uranium is directly related to the rate at which the bacterium oxidizes Fe^{2+} to provide energy for its growth requirement (DiSpirito and Tuovinen 1982; Ferroni et al 1986). Many laboratory studies have been demonstrated that these acidophilic bacteria can accelerate, with varying success, leaching rates and can increase the uranium recovery under controlled conditions (Lundgren and Silver 1980). Extraction in columns, either with or without circulation of the leaching medium, simulates heap and dump leaching (Derry et al 1976; Manchee 1979).

Present studies were aimed to find out the possibilities of microbial leaching of low-grade sandstone uranium ores, overburden and waste residues, and to ascertain the role of microorganisms in this process.

MATERIALS AND METHODS

Sandstone Uranium Ore

A 1000 kg sample of low-grade sandstone uranium ore was collected from mine site Baghalchur, Dera Ghazi Khan and was well mixed with by repeated coning and quartering technique. The homogenized ore was separated into different size fractions by screening and each fraction was weighed to determine the particle size and uranium content distribution at different fractions. Different fractions of minerals were obtained using heavy liquid separation technique. Heavy minerals were separated by Frantz Isodynamic Magnetic Separator. Mineral identification was carried out by examining polished grain mounts in reflected light using a petrographic microscope and loose grains were examined under a binocular microscope. Grain mounts of bulk samples and of mineral separates were prepared and examined by Frantz Isodynamic Magnetic Separator and Heavy Liquid Separation techniques.

External Energy Source

Elemental sulfur and sulfur slag (a waste of sulfuric acid plant) containing 20-40% sulfur and iron contents were used as external energy source for acidophilic *Thiobacillus* bacteria.

Microorganisms

Indigenous microflora closely resembling to *Thiobacillus ferrooxidans* and *Thiobacillus thiooxidans* present in mine water and ore samples were isolated by standard enrichment techniques. For column leaching experiments, no extra inoculum was made and only these bacteria present in the leaching system, were propagated and employed further in these investigations.

Media Used

Mineral salts medium 9K as described by Silverman and Lundgren (1959) was used for isolating *Thiobacillus ferrooxidans*. The medium contained (g/L); $(\text{NH}_4)_2\text{SO}_4$ 3.0; KH_2PO_4 0.5; $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ 0.50; KCl 0.10; $\text{Ca}(\text{NO}_3)_2$ 0.01; $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$ 44.20. pH was adjusted to 2.0 with 5 M H_2SO_4 . Medium used for isolating *Thiobacillus thiooxidans* was 9K mineral salts medium in which ferrous sulfate was replaced by elemental sulfur (10g/L). For isolating *Thiobacillus acidophilus* the medium described by Guay and Silver (1975) was used. It contained (g/L); glucose 10.0; KH_2PO_4 and $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ 0.50 and pH was adjusted to 4.0 - 4.5 with dilute H_2SO_4 . Medium used for isolating *Thiobacillus thioparus* was Medium # 36 as described in the DSM Catalogue of Strains (1989).

Growth of Bacteria

Isolation of *Thiobacillus ferrooxidans* and *Thiobacillus thiooxidans* was carried out in 250 mL Erlenmeyer flasks containing 90 mL of 9KFe^{2+} and 9KS^0 media respectively; with 10 mL of mine water was used as inoculum. The flasks were incubated at 30°C on an orbital shaking incubator at 100 rpm for several days. Mine water samples from various seepage and drainage at different locations in the mine area were examined for the presence of bacteria growing on ferrous iron (*Thiobacillus ferrooxidans*) or sulfur (*Thiobacillus thiooxidans* and *T. ferrooxidans*) or on glucose at pH 4.0-4.5 (*T. acidophilus*) and thiosulfate (*Thiobacillus thioparus*) at pH 6.6. Mine water samples were centrifuged at $12,000\times g$ at 4°C for 10 min to obtain cell pellet, which was resuspended in 10 mL of autoclaved distilled water. It was used for isolation of different types of bacteria.

Column Leaching Studies

Four polyvinyl chloride (PVC) columns 25 cm in diameter and 2.0 meter height, were used during these studies. The columns had flat acrylic bottoms which were fitted with 10 mm dia outlets in the centre and were labeled as A, B, C and D. The ore was manually charged in the columns. Each column was loaded with 100 Kg of low-grade sandstone uranium ore (0.027% U_3O_8) amended with 5-7% w/w elemental sulfur except column D in which sulfur slag 10-12% w/w was used as external energy source for acidophilic *Thiobacillus* bacteria. Each column was fed from separate reservoirs which contained 200 L mine water with and without certain treatments as lixiviant. The column A was only fed from mine water as collected from mine site without any treatment, while column B was from mine water whose pH was adjusted with H_2SO_4 to a value of 3.5. Other two columns (C and D) were fed with mine water of adjusted pH 3.50 and containing 3.0 g/L $(\text{NH}_4)_2\text{SO}_4$ as nitrogen source for bacteria as well. The mine water was circulated at a rate of 1.5 L/hr in each column. Periodically, the column effluent samples were taken from each column and analyzed for pH, U_3O_8 solubilized and iron content. Autotrophic sulfur and iron-oxidizing bacteria in the column effluents, were also enumerated by employing the liquid 9KFe^{2+} and 9KS^0 media as described by Silverman and Lundgren (1959). The presence of some heterotrophs like *Thiobacillus acidophilus* were also monitored by using liquid and solid media described by Guy and Silver (1975). Bacterial population was determined by direct count using bacterial counting chamber (Petroff-Hauser Bacterial Counter, Arthur and H. Thomas Company, Philadelphia; depth 0.02 mm, area 0.04 mm^2). Total bacterial population was enumerated by placing the chamber under phase-contrast microscope.

Analytical Techniques

Uranium content of the ore was analyzed by spectrophotometric method using dibenzoyl-methane as chromogenic reagent (Nagi et al 1974). The concentration of solubilized uranium content was determined using arsenazo-111 spectrophotometric method (Bhatti et al 1989)

and total iron content by using 1,10-Phenanthroline spectrophotometric method (Christian 1986).

RESULTS AND DISCUSSION

Mineralogy of Ore

Sandstone uranium ore deposits occur in the foothills of the Sulaiman Range in the Dera Ghazi Khan District, Pakistan, and the best exposure of uranium mineralization containing 0.05 - 0.30 % U_3O_8 content, is at Baghalchur. The nature of uranium minerals are tyuyamunite, carnotite and uranophane in the oxidized zone and uraninite, pitchblende and coffinite in the unoxidized zones. The unoxidized ore is grey to black in color. It is present below the water table while the oxidized ore which is of yellowish to brown color, exists above it. The uranium minerals are irregularly mixed with the matrix and occurs mainly as a thin coating on pebbles, grains and clay balls (Moghal 1974). About 40 % proportion of the world's reasonably assured resources of uranium exists in the sandstone type ore deposits (Table # 1) and uranium minerals in such deposits occur in the grain-binding material and usually upto 5 % calcite and minor amounts of pyrite are present (Gow 1985).

The ore loaded into the columns had an average uranium content of 0.027 % U_3O_8 on dry matter basis. The Baghalchur sandstone is light grey, mostly medium to fine grained, soft, friable and poorly sorted. Erratic pebbles and cobbles occur sporadically throughout the section. These can be quartzite, limestone or calcareous clay balls. The sandstone also contains abundance of heavy minerals which frequently outline cross-bedding. The major constituents of the ore were quartz, clay, calcite, feldspar, hornblende, chlorite, epidote, muscovite and the refractory fragments while the pyrite was among minor constituents. Magnetite constitutes the major portion of the heavy minerals. Uranium mineral is present as black coated grains. Clay and calcite were found to be the major acid consuming materials of the ore (Table #2). The particle size distributions of the ore charged in the columns are given in Table #3. Sieve analysis results revealed that it contained about 50% of ore fraction of particle size +250 microns and the ore grade increased in the finest range. It was higher (0.032 % U_3O_8) in the particle size of -64 microns and lower in the +250 microns and above (Table # 4). Evidently it contained a little amount of pyrite content which is the main source for bacterial leaching process. So the ore has to be amended with elemental sulfur and sulfur slag as external energy source for *Thiobacillus ferrooxidans* and *Thiobacillus thiooxidans* for bioleaching process.

Leaching of Ores

During the microbial leaching of low-grade sandstone uranium ore, the elemental sulfur and sulfur slag supplied as external energy source oxidized to sulfuric acid and ferric sulfate solution by the action of iron- and sulfur-oxidizing acidophilic thiobacillus bacteria. Both metabolites sulfuric acid and ferric sulfate solution act as lixiviants for hydrometallurgical processes.

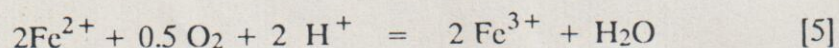


Table 2: Stereomicroscopic mineral analysis of Baghalchur low-grade sandstone uranium ore.

Minerals	Mineral present (%)	Minerals	Mineral present (%)
Quartz	59.00	Epidote	3.00
Feldspar	5.00	Limonite	0.05
Calcite	3.10	Rutile	0.02
Pyrite	0.10	Hematite	0.50
Chlorite	2.90	Zircon	Traces
Muscovite	2.00	Magnetite	0.21
Biotite	1.82	Clay	4.83
Actinolite	0.16	Black coated grains	3.21
Hornblende	3.17	Refractory fragments	9.14
Tourmaline	Traces	Black Uranium bearing grains	
Pyroxene	0.50		2.60

Table 3: Particle size distribution of uranium loaded into columns

Fraction Size No	ASTM Size (um)	Weight of Ore Fraction (%)
+30	+500	5.32
- 30 to + 60	+250	46.90
- 60 to + 80	+177	19.14
- 80 to + 100	+149	8.68
-100 to + 120	+125	5.04
-120 to + 200	+ 74	3.00
-200 to + 230	+ 63	2.30
-230 to + 300	+ 53	1.92
-300	+ 45	4.20

Table 4: Distribution of uranium content at different size fraction of uranium ore loaded into columns

ASTM Fraction No.	ASTM Particle Size (um)	U ₃ O ₈ (%)
Ore	-	0.027
+30	+595	0.024
- 30 to + 40	+420	0.023
- 40 to + 60	+250	0.022
- 60 to +100	+149	0.025
-100 to +150	+105	0.029
-150 to +200	+ 74	0.032
-200 to +230	+ 63	0.032

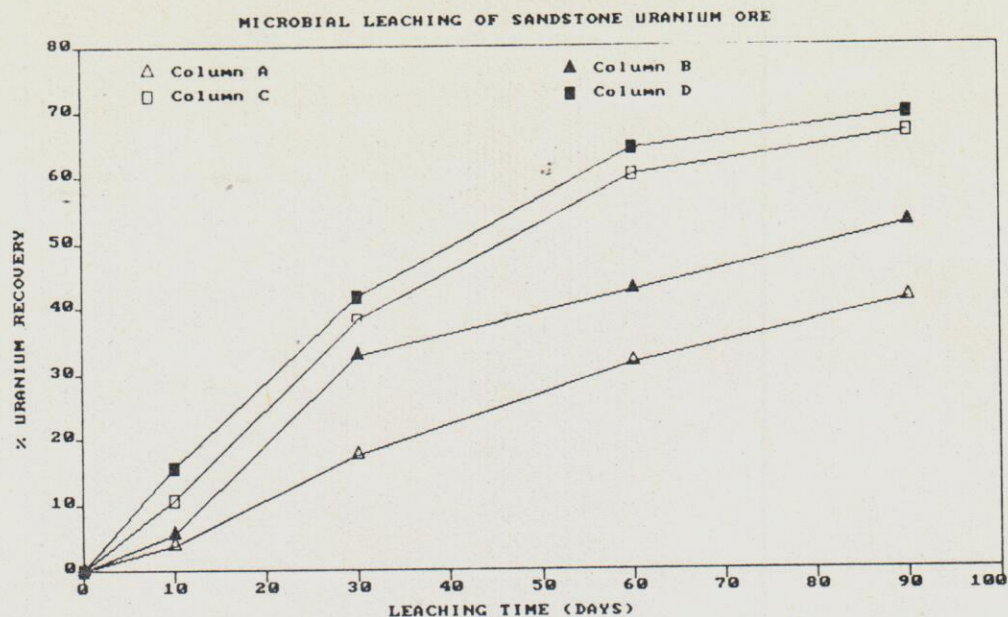
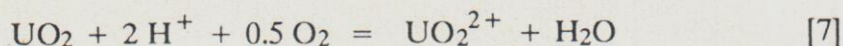
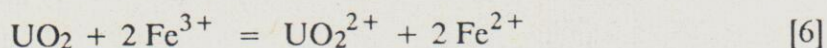
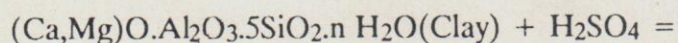
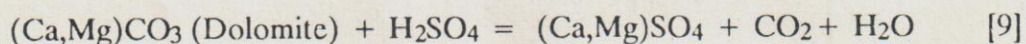
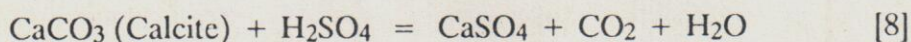


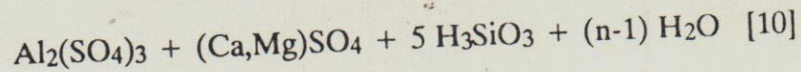
Figure 1. Microbial leaching of sandstone uranium ore in PVC columns.



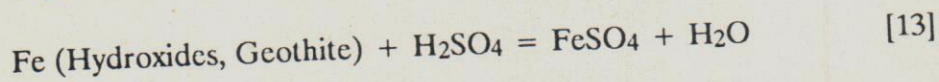
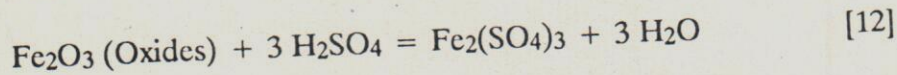
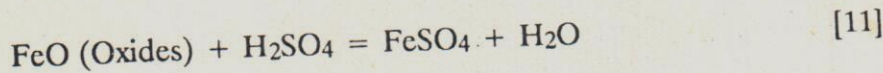
Reactions [4] and [5] are catalyzed by the action of bacteria. The solubilization of uranium from ores involves its oxidation from U^{4+} to U^{6+} which is generally affected through indirect action by bacteria forming acidic ferric sulfate solutions (Guay et al 1977; DiSpirito and Tuovinen 1982).

The results of the column leaching experiments on low grade sandstone uranium ores are presented in Figure # 1. The maximum amount of uranium was leached out 66.50% and 70.0% from columns C and D respectively in 90 days. In both these columns initial pH 3.5 of the feeding mine water (inoculum) was adjusted with sulfuric acid and 0.3% w/v $(\text{NH}_4)_2\text{SO}_4$ was dissolved as nitrogen source for chemolithotrophic *Thiobacillus ferrooxidans*. The recovery of uranium from Baghalchur sandstone ore was correlated to a substantial consumption of sulfuric acid due to the high CaCO_3 content which is present as limestone and dolomite in the ore matrix. The iron oxides minerals present in the ore also consumed H_2SO_4 . Therefore, the first sulfuric acid produced from biochemical oxidation of sulfur and/or sulfur slag react with these minerals according to the following reactions:

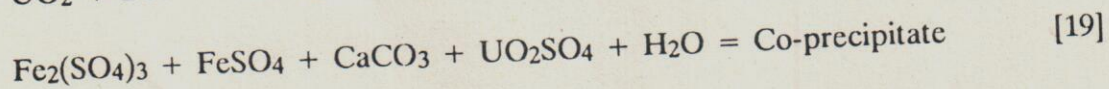
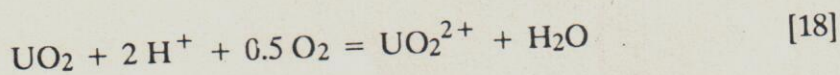
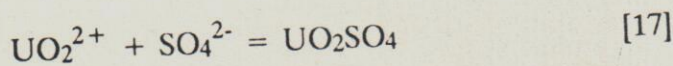
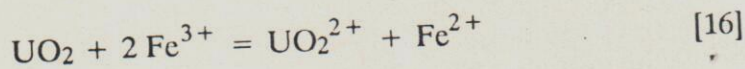
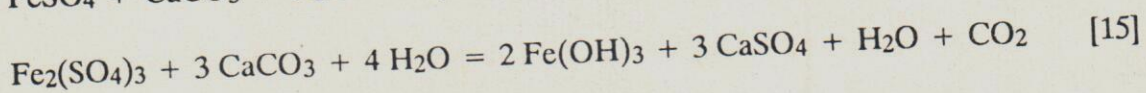
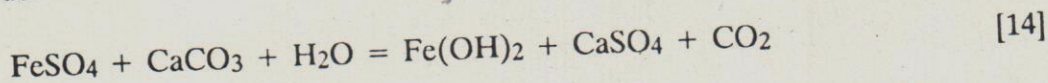




Calcium carbonate is the major acid consuming constituent which is responsible for generation of CO_2 as well as the formation of very fine precipitate of CaSO_4 (Equations 8 & 9). Therefore, the pH values of the columns effluents continuously increased and CO_2 thus produced could be utilized by the chemolithotrophic bacteria. The calcium sulfate formed blocked capillaries of liquid percolation to some extent. The colloidal suspension passed out of the columns into the collection reservoirs. Similarly, the acid also reacts with the clay mineral present in the ore matrix [Equation 10]. The water soluble sulfates are leached out with the formation of colloidal silica and alumina. Their behaviour is similar to that of calcium sulfate. Iron that is present both in ferrous and ferric states in the ore minerals also undergo a chemical reaction with sulfuric acid and produced ferrous and ferric sulfate solutions according to the following equations:



FeSO_4 produced in equations [11], [12] and [13] are oxidized to $\text{Fe}_2(\text{SO}_4)_3$ at low pH by the action of iron-oxidizing thiobacilli, resulted as lixiviant for uranium solubilization. However, sometimes when iron bearing solution recycled and percolated through columns, colloidal iron is precipitated as ferrous or ferric hydrated oxides, where CaCO_3 content of the ore matrix was not completely neutralized [Equation 18]. These irons hydrated oxides are gelatinous in nature and responsible for rendering the sections of the ore bed totally impermeous.



hydrated oxides

It was observed during the microbial column leaching studies that when uranium bearing solution was recycled through the columns, when calcite was not completely neutralized, co-precipitation of uranium and iron occurred, sometimes resulting in the loss of uranium in the column ore bed. Sulfuric acid added to adjust the pH 3.5 of feeding mine water provided better conditions for the growth of *Thiobacillus ferrooxidans* and *Thiobacillus thiooxidans*. This

was the reason, that more uranium was leached out from columns B,C and D as compared to column A (Figure I). It was found that about 41% U_3O_8 was leached out from column A which was loaded with ores amended with elemental sulfur and irrigated with mine water (pH 7.4) as inoculum of indigenous microflora of thiobacilli. However, when the initial pH of this mine water was adjusted to 3.5, the uranium leaching was found to have increased to 53% U_3O_8 , after 90 days. Since a further fortification of irrigation medium by ammonium sulfate (3.0 g/L) could also augment the uranium leachability from 53% to 66% U_3O_8 , it indicated that with mine water the process operated under nitrogen limiting conditions. The physical appearance of the bacterial leach liquors was also found to be turbid and dark yellow in column C. In addition, soluble and as well as Fe(III)-precipitate was also noted in the column effluents. The column containing ore amended with sulfur slag (column D) and irrigated with mine water of initial pH 3.5 and containing ammonium sulfate (3.0 g/L) as nitrogen source for *Thiobasillus ferrooxidans* and *Thiobacillus thiooxidans*, exhibited 70% U_3O_8 leachability which was the highest. The presence of soluble ferric iron as well as Fe(III)-precipitate (Jarosite) was also observed in the column effluents.

Table 5: Variations of pH at various depths in columns after 100 days of leaching process

Depth (cm)	pH of ore slurries			
	Column No.			
	A	B	C	D
0-30	2.4	2.2	2.1	2.0
30-60	4.4	3.0	2.9	2.2
60-90	6.6	4.7	4.6	3.2

Furthermore, a drastic change in pH of the orebed at various depths in all columns was observed (Table # 5). It was less than 2.5 at 0-30 cm depths in all these columns, which was probably due to the availability of maximum oxygen at the top surface of column. Thus bacterial activity enhanced the rate of biooxidation of elemental sulfur and sulfur slag to produce H_2SO_4 . As the depth of the ore bed increased the availability of air/oxygen was limited. The color of ore in columns A and B was similar to the ore feed (grey color). It was light yellow in column C, whereas dark brown/brick red in the column D. The pH values of ore was found to increase with the increase of depths in all the columns. Similarly, the pH of the effluents obtained from column leaching studies was found to be not less than 3.0 in any of the columns throughout these studies. It was due to the formations of microcysts in the leaching systems in which bacteria produced the sulfuric acid, resulting the solubilization of uranium at low pH but increased as it passed through the un-neutralized portion of calcite of the ore matrix, thereby increasing its pH which was flushed out as column's effluents.

CONCLUSIONS

This study has indicated that uranium can be leached out, microbially from sedimentary type-silicate ores. This process is simple, economically viable and free of environmental hazards. Initial results are very encouraging, yielding high recovery from these ores.

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REFERENCES

- Barthel F. 1980. Worldwide review of uranium deposits and their geological surroundings. Paper presented at the Seminar on Uranium Geology, Nov. 10-13, 1980, Lahore, Pakistan.
- Bhatti TM Mateen A Amin M Khalid ZM and Malik KA. 1989. Spectrophotometric determination of uranium (VI) in bacterial leach liquors using Arsenazo-111 (Direct Method). Proceedings of the 1st National Conference on Chemistry. Oct 8-10, 1989 (M. Irfan and MA Khawaja eds.), pp 482-412, Peshawar University, Pakistan.
- Bosecker K and Wirth G. 1980. Bacterial leaching of carbonate bearing uranium ore. Biogeochemistry of Ancient and Modern Environment (PA Trudinger, MR Walter and BJ Ralph eds.), pp 557- 562. Australian Academy of Sciences, Canberra.
- Christian GD. 1986. Spectrophotometric determination of iron. Analytical Chemistry, Fourth Edition(GD Christian ed.), pp 599, John Wiley & Sons. NY.
- Derry R Garrett KH LeRoux NW and Smith SE. 1976. Bacterially assisted plant process for leaching uranium ores. Geology, Mining and Extractive Processing of Uranium. (ML Jones ed.),pp 56-62, Institution of Mining and Metallurgy, London.
- DiSpirito AA and Tuovinen OH. 1982. Kinetics of iron uranous ion and ferrous iron oxidation by *Thiobacillus ferrooxidans*. Archive of Microbiology 133: 33-37.
- Duncan DW and Bruynesteyn A. 1971. Microbial leaching of uranium. New Mexico State Bureau of Mines and Mineral Resources, Circular No 118: 55-61.
- Elevatroski, EA 1977. Uranium ores and minerals. MINOBRAS, Dana Point, Calif.
- Ferroni GD Leduc LG and Todd M. 1986. Isolation and temperature characteristics of psychrotrophic strains of *Thiobacillus ferrooxidans* from the environment of a uranium ore. Journal of General Microbiology 32: 169-175.
- Fisher JR. 1966. Bacterial leaching of Elliot lake uranium ore. Trans. Can. Inst. Min. Metall. 69: 167-171.
- Fletcher JR. 1970. Metal winning from low-grade by bacterial leaching. Trans. Can. Inst. Min. Metall. 79: C247-C252.
- Guay R and Silver M. 1975. *Thiobacillus acidophilus* sp. nov., isolation and some physiological characteristics. Canadian Journal of Microbiology 21: 281-288.

- Guay R Silver M and Torma AE 1977. Ferrous iron oxidation and uranium extraction by *Thiobacillus ferrooxidans*. *Biotechnology and Bioengineering* 19: 727-740.
- Gow WA, McCreehy HH, Ritcey GM, McNamara VM, Harrison FA and Lucas BH. 1971. Bacterial-based processes for the treatment of Low-grade uranium ore. pp. 195-211. IAEA, Vienna, Austria.
- Gow WA. 1985. Recent advances in uranium ore processing. In: *Advances in uranium ore processing and recovery from non-conventional resources*. Proceedings of a Technical Committee Vienna, 26-29 September 1983. (IAEA-TC-491/1).
- Guay R, Silver M and Torma AE. 1976. Microbiological leaching of low-grade uranium ore by *Thiobacillus ferrooxidans*. *European Journal of Applied Microbiology* 3: 157-167.
- Harrison VF Gow WA and Ivarson KC. 1966. Leaching of uranium from Elliot Lake ore in the presence of bacteria. *Canadian Journal of Mining* 87(5):64-67.
- Kelly DP. 1976. Extraction of metals from ores by bacterial leaching: Present status and future prospects. *Microbial Energy Conversion* (HG Schlegel and J Barnea eds.), pp 329-338. Firma E. Goltre KG Gottingen. W. Germany.
- Lundgren DG and Silver M. 1980.. Ore leaching by bacteria. *Annual Review of Microbiology* 34: 263-283.
- MacGregor RA. 1966. Recovery of U₃O₈ by underground leaching. *CIM Bulletin* 59 (649): 583-587.
- Marchbank A. 1987. Update on uranium leaching at Denison mines. Proceedings of the fourth Annual General Meeting of BIOMINET. (R.G.L. McCreehy ed.), CANMET Special Publication SP 87-10. pp. 3-18. Canadian Government Publication Centre, Ottawa, Canada.
- Moghal MY. (1974). Uranium in Siwalik Sandstones, Sulaiman Range, Pakistan. Formation of uranium ore deposits. IAEA Technical Series No. IAEA-SM 183/41, 383-403.
- Ralph BJ. 1975. *Biotechnology Applied to Raw materials processing*. Comprehensive Biotechnology, The principles, Application and Regulation of Biotechnology in Industry, Agriculture and Medicine. (Murracy Moo Young ed.), Volume IV. pp. 201- 234. Pergamon Press, Oxford. U.K.
- Sasson A. 1975. The Earth's invisible garbage men. *Unesco Courier*, July, 1975: 20-29.
- Silverman MP and Lundgren DG. 1959. Studies on the chemoautotrophic iron bacterium *Ferrobacillus ferrooxidans*. 1. An improved medium and a harvesting procedure to secure high cell yields. *Journal of Bacteriology* 77: 642-647.
- Stevens CJ and Tuovinen OH. 1986. Ferrous iron oxidation, nitrogen fixation (acetylene reduction) and nitrate reductase activity by *Thiobacillus ferrooxidans*. Proceedings of the Second Annual General Meeting of BIOMINET (RGL McCreehy ed). CANMET Special Publication, SP 85-6. Canadian Government Publication Centre, Ottawa, Canada.
- Tomizuka N and Takahara Y. 1972. Bacterial leaching of uranium from Ningyo-Toge ores. Proceedings IV IFS: *Fermentation Technology Today* (Terui G ed.), pp 513-520, Society for Fermentation Technology, Osaka, Japan.
- Torma AE. 1986. Biohydrometallurgy as an Emerging Technology. *Biotechnology and Bioengineering Symposium* No. 16: 49-63.

Tuovinen OH. 1986. Acid leaching of uranium ore material with microbial catalysis. *Biotechnology and Bioengineering Symposium No. 16*: 65-72.

Tuovinen OH and Kelly DP. 1972. Biology of *Thiobacillus ferrooxidans* in relation to the microbiological leaching of sulfide ores. *Z. Alg. Mikrobiol.* 12: 311-346.