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Remediation of uranium contaminated soils with bicarbonate extraction and microbial U(VI) reduction

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SUMMARY

A process for concentrating uranium from contaminated soils in which the uranium is first extracted with bicarbonate and then the extracted uranium is precipitated with U(VI)-reducing microorganisms was evaluated for a variety of uranium-contaminated soils. Bicarbonate (100 mM) extracted 20–94% of the uranium that was extracted with nitric acid. The U(VI)-reducing microorganism, *Desulfovibrio desulfuricans* reduced the U(VI) to U(IV) in the bicarbonate extracts. In some instances unidentified dissolved extracted components, presumably organics, gave the extract a yellow color and inhibited U(VI) reduction and/or the precipitation of U(IV). Removal of the dissolved yellow material with the addition of hydrogen peroxide alleviated this inhibition. These results demonstrate that bicarbonate extraction of uranium from soil followed by microbial U(VI) reduction might be an effective mechanism for concentrating uranium from some contaminated soils.

INTRODUCTION

Methods are needed for removing radionuclides and heavy metals from contaminated soils. The ideal process for treating contaminated sites would selectively remove the contaminants of concern in a readily recoverable form, without excessive soil destruction. Sodium bicarbonate is known to effectively leach uranium from rocks and soils [2,13,14]. The uranium is held in solution in the form of U(VI)-carbonate complexes. Compared to other potential extractants such as strong acids which may have a number of deleterious effects on soil structure and chemistry, bicarbonate is relatively environmentally benign.

It has recently been demonstrated that U(VI)-carbonate complexes can be effectively removed from solution through the activity of U(VI)-reducing microorganisms [4,8]. The microorganisms reduce U(VI) to U(IV) which then precipitates as uraninite. Microbial U(VI) reduction may be more effective in removing U(VI) from bicarbonate solutions than are other removal techniques such as ion exchange resins and biosorption [8]. Furthermore, the uraninite precipitate from microbial U(VI) reduction is relatively pure and compact and more easily handled than the uranium that is adsorbed onto resins or biomass [8]. Microbial U(VI) reduction can be used to remove uranium from a variety of uranium-contaminated waters [8].

From these previous studies it was readily apparent that a potential mechanism to concentrate uranium from contaminated soils was to first extract the uranium with bicarbonate and then precipitate the uranium with U(VI)-reducing microorganisms. The uranium could then be recovered in a concentrated form for disposal or possible reuse. The bicarbonate extractant could be recycled to extract more uranium. The results from bench-scale studies reported here suggest that such a treatment strategy could be feasible.

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MATERIALS AND METHODS

Soil types

In this paper, both true soils containing uranium-bearing materials and other crushed earth materials, such as uranium ore and uranium mill tailings were studied (Table 1). For ease of description, all are referred to as soils. Mined, unprocessed uranium ore, subgrade ore, and uranium mill tailings can be found at uranium mining and milling sites. These materials themselves, and admixtures of them with ambient soils, are potential materials requiring remediation. The Denver Radium Superfund Site soil contained waste from the radium processing industry (active 1914–1926), for which processing and disposal procedures are very poorly documented. Typical waste from the radium industry included residues and precipitates, soils contaminated by contact with waste solutions, and crude or partially processed uranium ores [6]. Soil from the Aberdeen Proving Ground Artillery Range was contaminated with uranium as the result of test firing artillery shells constructed with ^{235}U-depleted uranium [3].

Extraction of uranium

Uranium was extracted with sodium bicarbonate (100 mM, pH 8.4) or nitric acid (1 N). Soils (10 g) were suspended in the extractant (50 ml) in polypropylene centrifuge bottles on a wrist action shaker. For the alkaline and mixed tailings, the nitric acid concentration was adjusted after determining the

TABLE 1

Extraction of uranium from soils and precipitation of uranium from bicarbonate extracts

Sample	Reference	Description	Sample preparation	Extractable uranium (μmol g^{-1}) Nitric acid[a]	Bicarbonate[b]	% of extracted uranium precipitated[c]
Ore		Stockpiled ore at abandoned uranium mine, South Dakota	Air dried, crushed to less than 2 mm	4.8	4.1	99
Acid tailings	[5]	Acid-leached uranium mill tailings, Texas	Dried at 110 °C, crushed to less than 0.6 mm	0.40	0.36	100
Mixed tailings	[7]	Mixed acid- and alkaline-leached uranium mill tailings, Utah	Dried to 100 °C, crushed to less than 2 mm	0.81	0.59	94
Alkaline tailings	[7]	Alkaline-leached uranium mill tailings, Utah	Dried to 100 °C, crushed to less than 2.0 mm, wet sieved to less than 44 μm	1.8	0.61	22 (75)[d]
Superfund site	[6]	Soil contaminated by the radium industry, Denver, Colorado	Air dried, crushed to less than 2 mm	2.5	0.49	19 (97)
Artillery range		Artillery Range soil contaminated with uranium artillery shells, Aberdeen Proving Grounds, Maryland	Air dried, crushed to less than 2 mm	0.31	0.29	22 (96)

[a]1 N nitric acid, 4 h.
[b]100 mM bicarbonate from Fig. 1.
[c]Uranium not passing through a 0.2-μm pore diameter filter after incubation of bicarbonate extract for 24 h with *D. desulfuricans* and H_2.
[d]Number in () indicates percent removal when extract was pretreated with peroxide to remove organics.

consumption of acid by carbonates in the sample using the acid neutralization method [1]. The extractions were at room temperature (ca. 20 °C). Preliminary studies indicated that extraction of the soils for 4 h with nitric acid yielded estimates for total uranium that were within 84–100% of the total uranium in the soils as determined by delayed neutron analysis.

The kinetics of U(VI) extraction with bicarbonate extracts were determined by subsampling (0.1 ml) over time and analyzing for soluble U(VI) as outlined below. Where noted, the solids were collected with centrifugation and all of the bicarbonate extract was removed and replaced with fresh bicarbonate solution. At the end of the extractions the extracts were passed through a cellulose nitrate filter (0.2-μm pore diameter, Nalgene, Rochester, NY, USA) and stored at 4 °C. The extracts were diluted (1:2, v/v) with water prior to the precipitation studies.

Microbial precipitation of uranium

Aliquots (10 ml) of the diluted soil extract or of bicarbonate buffer amended with uranyl acetate were added to 20-ml serum bottles, bubbled with N_2-CO_2 (80:20) for 5 min, and the bottles were sealed with thick butyl rubber stoppers. *Desulfovibrio desulfuricans* was used as the U(VI)-reducing microorganism as it was in previous studies on the bioremediation of uranium-contaminated waters [8]. As previously described [9], *D. desulfuricans* was grown in medium with lactate as the electron donor and sulfate as the electron acceptor. Washed cell suspensions were prepared under anoxic conditions in bicarbonate buffer [9] and added to the bicarbonate soil extracts in order to provide 2.5 mg of cell protein per bottle. H_2 (10 ml) was added to each bottle as the electron donor for U(VI) reduction and the extracts were incubated at 35 °C. Subsamples (0.1 ml) were taken over time with a syringe and needle and, in an anoxic chamber, were first diluted 1000 times and then filtered (0.2-μm pore diameter Gelman polysulfone filter, Gelman Sciences, Ann Arbor, MI, USA). U(VI) and total uranium in the filtrates were determined as outlined below.

As discussed below, the bicarbonate extracts from some soils were yellow and rates of U(VI) reduction and/or U(IV) precipitation were inhibited in the yellow extracts. Analysis of the yellow extracts with a Shimadzu TOC Total Organic Carbon Analyzer (Shimadzu Scientific Instruments, Columbia, MD, USA) indicated that they contained relatively high (>20 mg L^{-1}) concentrations of dissolved organic carbon. Therefore, when noted, these yellow extracts were diluted with a hydrogen peroxide solution (30%) rather than water. The extracts treated with hydrogen peroxide were incubated for 18

days at room temperature to allow the yellow color to clear and for the hydrogen peroxide to dissipate. The ability of *D. desulfuricans* to reduce U(VI) in the extracts was then evaluated as described above.

Analytical techniques

U(VI) in extracts and cell suspensions was measured with a kinetic phosphorescence analyzer (KPA-10; Chemchek Instruments, Richland, WA, USA) as previously described [10]. Concentrations of other materials present in bicarbonate and nitric acid extracts were determined using a directly coupled plasma spectrometer.

RESULTS AND DISCUSSION

Extraction of uranium

The effectiveness of the bicarbonate extraction ranged from 20 to 94% of the nitric acid-extractable uranium (Table 1). The kinetics of uranium extraction differed among the materials (Fig. 1). With ore and acid tailings prolonged or repeated extraction increased the final yield, while other materials did not greatly improve with additional treatment.

In the ore sample, 63% of the total uranium was solubilized in the first extraction, and 85% was removed after three successive extractions (Fig. 1). Most of the uranium was extracted within the first 24 h of the first extraction. Uranium recovery from the mill tailings varied, with best recovery in the acid tailings, and poorest recovery in the alkaline tailings (Fig. 1). As with the ore sample, most of the extractable uranium was recovered early in the first extraction. The reason for the poor recovery from the alkaline mill tailings may be that these are the residues of sodium carbonate/bicarbonate-leached uranium ore. It is expected that much of the bicarbonate-extractable uranium would have already been removed during the leaching of the ore and that most of the uranium left in the tailings would be resistant to further bicarbonate extraction.

In contrast with the ore and tailing samples, the Superfund Site soil required an extended extraction period and only a fifth of the nitric acid-extractable uranium was removed with repeated extractions (Fig. 1). In order to investigate whether the low recovery of uranium from this soil was due to non-extractable uranium phases or if soil components might be interfering with the extraction, 1 g of the highly extractable uranium ore was added to 9 g of the Superfund Site soil. All of the uranium added with the ore was recovered in a bicarbonate extraction of the mixed materials. With the Artillery Range soil, the bicarbonate extracted a quantity of uranium that was essentially equivalent to the nitric acid-extractable uranium (Table 1).

Although bicarbonate did not completely extract all of the uranium from some of the soils, the uranium that bicarbonate does not extract is probably highly immobile in most soils. Thus, in situations where the primary concern is potential contamination of water supplies as a result of uranium leaching from the soils, uranium that is not leached with bicarbonate may be of little concern. Furthermore, studies on bioavailability of uranium in soils have demonstrated that bicarbonate removes a much higher percentage of soil uranium than is bioavailable [14].

As expected, the bicarbonate extraction was far less destructive to the soils than 1 N nitric acid, releasing much less of major soil components like silica, aluminum, magnesium, calcium, and iron (Fig. 2). Whereas the nitric acid extracts often had high concentrations of copper (for example 162 and 270 μM in the extracts of the Superfund Site and Artillery Range soils, respectively), there was no detectable copper in the bicarbonate extracts. This is significant because copper can inhibit microbial U(VI) reduction [8].

Precipitation of uranium from soil extracts

When uranyl acetate was added to bicarbonate buffer as a pure uranium form, rates of uranium reduction by *D. desulfuricans* were much slower (Fig. 3) in 100 mM bicarbonate than in the 30 mM bicarbonate buffer that had been evaluated in previous studies [8]. Therefore, the 100-mM bicarbonate soil

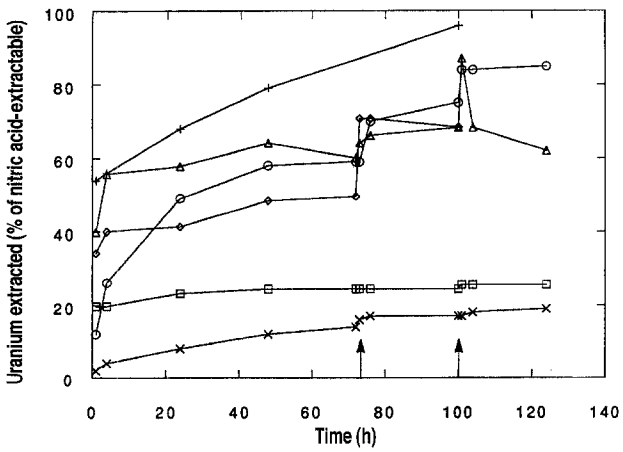

Fig. 1. Bicarbonate (100 mM) extraction of uranium from various soils. Arrows indicate replacement of the extract with fresh bicarbonate solution. Values were corrected for the entrained solution. –○–, ore; –△–, acid tailings; –◇–, mixed tailings; –□–, alkaline tailings; –×–, Superfund site; –+–, Artillery range.

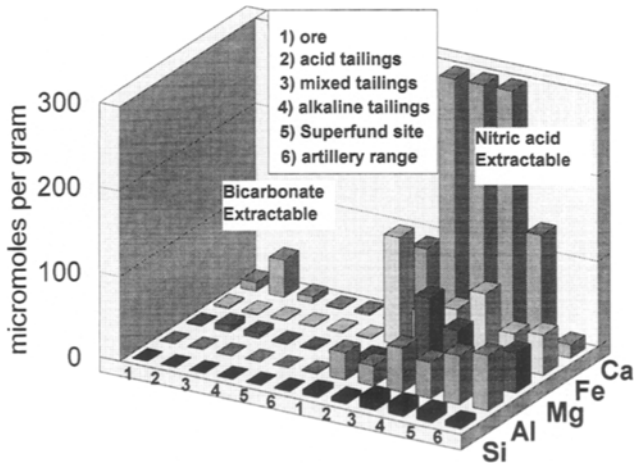

Fig. 2. Major soil components extracted by 100 mM sodium bicarbonate (72 h) and by 1 N nitric acid (4 h) from uranium-contaminated soils.

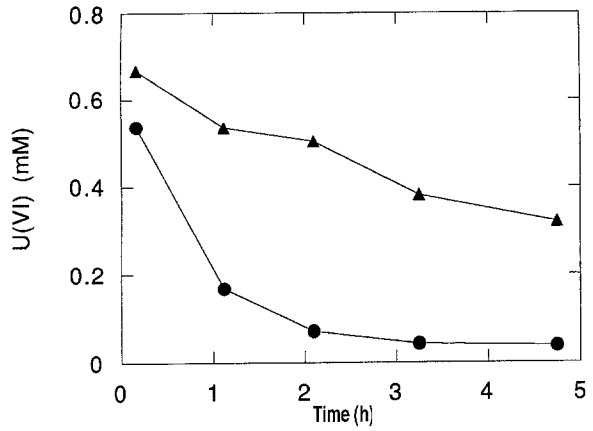

Fig. 3. Comparison of U(VI) reduction in solutions of uranyl acetate in 100 mM (–▲–) and 30 mM (–●–) sodium bicarbonate.

extracts were diluted with water to 33 mM before evaluating the potential for microbial U(VI) reduction to precipitate uranium from the extracts. However, in subsequent studies it was found that 30 mM bicarbonate extracted uranium from the soils at a rate and extent comparable to 100 mM bicarbonate. This means that in the application of this method, a dilution step would not be necessary as 30 mM bicarbonate could be used to leach the uranium rather than 100 mM.

Reduction of U(VI) to U(IV) readily proceeded in the bicarbonate extracts from the ore, tailings, and the Superfund Site soil (Fig. 4). Previous studies demonstrated that the loss of U(VI) over time could be attributed to U(VI) reduction as there is no U(VI) reduction under these conditions in the absence of a U(VI)-reducing microorganism and *D. desulfuricans* does not adsorb U(VI) [8,9,10]. U(VI) reduction was markedly slower in the Artillery Range soil but, even in this sample, over 80% of the U(VI) was reduced with 24 h.

The U(IV) that was produced in the extracts from the ore and the acid and mixed tailings was readily precipitated as evidenced by the fact that most of the uranium would not pass through a filter with a pore diameter of 0.2 μm (Table 1). However, most of the U(IV) in the extracts from the alkaline

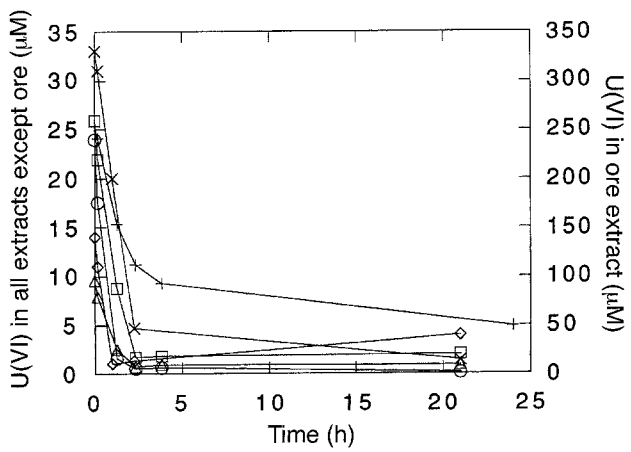

Fig. 4. *Desulfovibrio desulfuricans* reduction of U(VI) in bicarbonate soil extracts. –○–, ore; –△–, acid tailings; –◇–, mixed tailings; –□–, alkaline tailings; –×–, Superfund site; –+–, Artillery range.

tailings as well as the Superfund Site and Artillery Range soil passed through the 0.2-μm filter (Table 1). The alkaline tailings, Superfund Site, and Artillery Range soil extracts had a notable yellow color, presumably due to the presence of dissolved organic matter. When the extracts were diluted in a hydrogen peroxide solution (30%) rather than water and were allowed to digest for 18 days, the yellow color disappeared. When these treated extracts were then subjected to microbial U(VI) reduction the removal of uranium from solution was greatly increased (Table 1).

From these results, it seems likely that the poor precipitation in the yellow extracts resulted from the formation of U(IV) complexes with dissolved organic carbon which inhibited the formation of uraninite particles large enough to be trapped by the 0.2-μm filter. Further evidence supporting this conclusion is the finding that in the presence of chelators such as nitrilotriacetic acid and ethylenediaminetetraacetic acid, *D. desulfuricans* reduces U(VI) to U(IV) but the U(IV) does not precipitate (D.R. Lovley and J.C. Woodward, unpublished results).

Application

These results suggest that it may be possible to concentrate uranium from some contaminated soils by extracting the uranium with bicarbonate and then precipitating the extracted uranium with microbial U(VI) reduction. The bicarbonate, once freed of uranium, could be recycled for further soil extraction. The uranium would be precipitated out in a highly concentrated and pure form which would have low-volume disposal requirements or possible economic value. Depending upon the situation, the bicarbonate leaching could potentially be carried out in one of a variety of manners including in situ, in vats, or by heap leaching. In addition to *D. desulfuricans* a number of other known U(VI)-reducing microorganisms [10,11] or their enzymes [12] could be employed in the reductive-precipitation step.

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