

Pilot-Scale in Situ Bioremediation of Uranium in a Highly Contaminated Aquifer. 1. Conditioning of a Treatment Zone

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To evaluate the potential for in situ bioremediation of U(VI) to sparingly soluble U(IV), we constructed a pilot test facility at Area 3 of the U.S. Department of Energy Natural and Accelerated Bioremediation Research (NABIR) Field Research Center (FRC) in Oak Ridge, TN. The facility is adjacent to the former S-3 Ponds which received trillions of liters of acidic plating wastes. High levels of uranium are present, with up to 800 mg kg⁻¹ in the soil and 84–210 μM in the groundwater. Ambient groundwater has a highly buffered pH of ~3.4 and high levels of aluminum (12–13 mM), calcium (22–25 mM), and nitrate (80–160 mM). Adjusting the pH of groundwater to ~5 within the aquifer would deposit extensive aluminum hydroxide precipitate. Calcium is present in the groundwater at levels that inhibit U(VI) reduction, but its removal by injection of a high pH solution would generate clogging precipitate. Nitrate also inhibits U(VI) reduction and is present at such high concentrations that its removal by in situ denitrification would generate large amounts of N₂ gas and biomass. To establish and maintain hydraulic control, we installed a four well recirculation system parallel to geologic strike, with an inner loop nested within an outer loop. For monitoring, we drilled three boreholes perpendicular to strike across the inner loop and installed multilevel sampling tubes within them. A tracer pulse with clean water established travel times and connectivity between wells and enabled the assessment of contaminant release from the soil matrix. Subsequently, a highly conductive region of the subsurface was prepared for biostimulation

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by removing clogging agents and inhibitors and increasing pH. For 2 months, groundwater was pumped from the hydraulically conductive zone; treated to remove aluminum, calcium, and nitrate, and supplemented with tap water; adjusted to pH 4.3–4.5; then returned to the hydraulically conductive zone. This protocol removed most of the aqueous aluminum and calcium. The pH of the injected treated water was then increased to 6.0–6.3. With additional flushing, the pH of the extracted water gradually increased to 5.5–6.0, and nitrate concentrations fell to 0.5–1.0 mM. These conditions were judged suitable for biostimulation. In a companion paper (Wu et al., *Environ. Sci. Technol.* 2006, 40, 3978–3987), we describe the effects of ethanol addition on in situ denitrification and U(VI) reduction and immobilization.

Introduction

Uranium is a major groundwater contaminant at U.S. Department of Energy (DOE) sites, and poses risks for liver damage and cancer. For 31 years, trillions of liters of acidic plating wastes containing high levels of uranium and nitric acid were generated at the Y-12 Facility, Oak Ridge, TN and were discarded in the unlined S-3 Ponds. In 1983, attempts were made to neutralize and denitrify the waste, and the area was capped and converted to a parking lot. Despite these measures, contamination continued to migrate from the source along geologic strike and dip to greater depths and to surface discharge points. To assess the potential for immobilization of uranium and other radionuclides and metals, the DOE established a Field Research Center (FRC) at the Y-12 Facility as part of the DOE Natural and Accelerated Bioremediation Research (NABIR) program. Since 2001, we have performed experiments in FRC Area 3, an area immediately adjacent to and downgradient from the S-3 Ponds along strike. To enable hydraulically controlled experiments, we developed a recirculation system and facilities for above-ground removal of clogging agents and inhibitors and used a stepwise procedure to condition the site prior to biostimulation (Figure 1).

As shown in Table 1, ambient groundwater is acidic (pH 3.4–3.5) and buffered by high levels of aluminum (up to 20 mM). It also contains high levels of nitrate, originally introduced as nitric acid (up to ~160 mM), sulfate introduced as sulfuric acid (up to ~10 mM), calcium (25 mM), metals, and organics (2, 3). Solid-phase analyses of a core sample from well FW104 gave total carbonate extractable U, NO₃⁻, SO₄²⁻, and PO₄³⁻ of 450, 780, 900, and 940 mg kg⁻¹, respectively (4). Zones with the highest U concentrations were dominated by clay- and silt-sized particles consisting primarily of illite. Fe(III) and Al(III) (hydr)oxides (>30 000 mg kg⁻¹), are abundant, typically as coatings on clay minerals (2). Soluble uranium is present at highly toxic levels of 84–210 μM, exceeding the U.S. Environmental Protection Agency drinking water standard (0.126 μM or 0.03 mg L⁻¹) by one thousand times. Most of this uranium migrates along strike within a narrow region of preferential flow—most likely a fracture zone ~1 m thick at a depth of 10.9–12 m where there is a high specific discharge of 0.1–1.0 m d⁻¹. Analyses of soil within this zone revealed high levels of sorbed or precipitated U(VI) (ranging up to 800 mg kg⁻¹) and phosphate (~1000 mg kg⁻¹) in close association with iron oxides. Thus, even though sorption or precipitation of U(VI) provides a natural mechanism of U(VI) immobilization, levels of U(VI) in the water

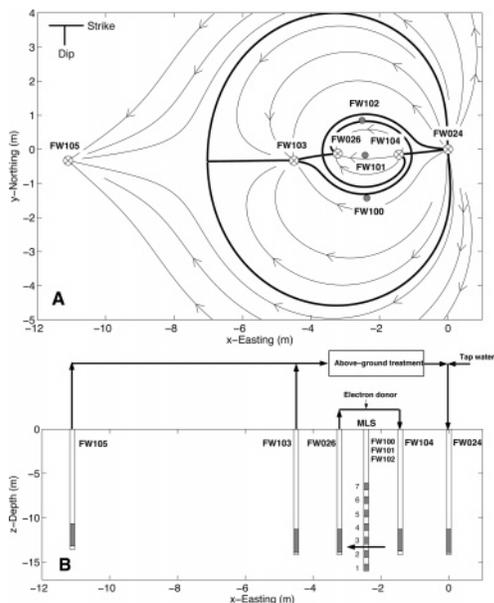


FIGURE 1. Layout of the below-ground system. (A) Plan view of the well system. The solid lines are streamlines with arrows indicating the flow direction, and the dark lines are the separation streamlines delineating the outer and inner flow cells. (B) Schematic diagram illustrating the overall strategy for in situ reduction of U(VI).

TABLE 1. Typical Chemical Properties of Contaminated Groundwater from FRC Area 3 Wells FW026 and FW024 before Bioremediation (on May 30, 2001)^a

nitrate	133 mM	U-233	<0.004 μM
sulfate	9.9 mM	U-234	0.004 μM
chloride	7.3 mM	U-235	0.43 μM
Al ³⁺	18.2 mM	U-236	0.043 μM
Ca ²⁺	25.1 mM	U-238	212 μM
Co ²⁺	0.02 mM	Tc-99	34 nCi/L
Fe ³⁺	0.17 mM	TOC	73 mg L ⁻¹
K ⁺	2.4 mM	COD	200 mg L ⁻¹
Na ⁺	32.9 mM	acetone	440 $\mu\text{g L}^{-1}$
Mg ²⁺	6.8 mM	2-butanone	76 $\mu\text{g L}^{-1}$
Mn ²⁺	2.4 mM	ethanol	200 $\mu\text{g L}^{-1}$
Zn ²⁺	0.035 mM	chloroform	34 $\mu\text{g L}^{-1}$
Ni ²⁺	0.19 mM	dichloromethane	39–42 $\mu\text{g L}^{-1}$
Sr ²⁺	0.018 mM	<i>cis</i> -1,2-DCE	723 $\mu\text{g L}^{-1}$
Si	0.81 mM	PCE	2051 $\mu\text{g L}^{-1}$
pH	3.5	TCE	111 $\mu\text{g L}^{-1}$
temperature	17.4 °C		

^a TOC = Total organic carbon; COD = chemical oxygen demand. All data were from FW026 except for organics which were from FW024. Other constituents of FW024 were similar to those in FW026.

remain unacceptably high, underscoring the need for a more effective means of immobilization. A promising option is microbial reduction of U(VI) to sparingly soluble U(IV), but biostimulation adjacent to or within the source zone poses several challenges.

(1) The low pH (~3.4) is not favorable for the growth of most organisms.

(2) Increasing the pH in situ is not desirable because aluminum hydroxide precipitates form at approximate pH > 4.5 and calcium solids precipitate at near-neutral pH. Because of their high concentration at the site, these solids will clog the aquifer, well screens, and installations, and create new surfaces for U(VI) sorption, confounding efforts to determine the mechanisms of soluble U(VI) removal.

(3) Nitrate is present at levels that are inhibitory to U(VI) reduction, and should therefore be decreased to a low level.

(4) In situ denitrification of bulk nitrate should be avoided because nitrate levels are very high (up to 80–160 mM),

increasing the probability of incomplete denitrification. Any nitrite so formed could oxidize U(IV) back to the more soluble U(VI) (5). Also, complete denitrification of as little as 1.35 mM nitrate generates N₂ in excess of its solubility limit (assuming 0.5 mole N₂/mole NO₃⁻ and N₂ solubility of 0.675 mM at 20 °C and 1.0 atm). The presence of a gas phase can alter flow direction and decrease solid surface area accessible for sorption/desorption and redox reactions.

(5) Calcium concentrations are above 2.0 mM at neutral pH conditions and above inhibit U(VI) reduction because of the formation of stable Ca–U–CO₃ complexes (6) and could be a factor for bioreduced U(IV) reoxidation (7).

(6) Other substances in the groundwater, such as chloroform or Ni, could potentially affect U(VI) reduction.

(7) The aquifer consists of highly fractured weathered shale with a soil matrix characterized by regions of low permeability and high porosity, so the rate of removal of contaminants from the aqueous phase will depend on the rates of mass transfer of contaminants from the low permeability matrix blocks to higher permeability regions within fractures.

The above issues were addressed in the following five stages.

(1) **Selection of a Treatment Zone.** Flow meter measurements and point dilution tests were used to identify a region of high flow (8, 9). Analyses of both groundwater and core samples ensured that the targeted region had high levels of U.

(2) **Installation of a Nested Recirculation System.** A four-well recirculation system was installed parallel to strike, creating a protective outer recirculation loop and nested inner recirculation loop. Three multilevel sampling wells were installed perpendicular to strike between the inner loop injection and extraction wells (as shown in Figure 1). A clean water tracer test confirmed well-to-well connectivity and enabled estimation of the rates of mass transfer of contaminants from the soil matrix.

(3) **Removal of Inhibitors and Clogging Agents.** Water extracted from the treatment zone was treated above ground to remove bulk aqueous Al, Ca, and nitrate, adjusted to pH 4.3–4.5, and injected back into the treatment zone.

(4) **Increase of Subsurface pH to a Value Better Suited for Microbial Activity.** Contaminant-free water was adjusted to pH 6.0–6.3 and injected, gradually increasing the pH of the subsurface by addition of a K₂CO₃ solution at low level via recycle line to a level favorable for robust microbial activity.

(5) **Biostimulation of in Situ Denitrification and U(VI) Reduction.** Ethanol was selected and intermittently added to stimulate in situ denitrification of residual nitrate and reduction of U(VI) (1).

In this article, we describe steps 1 through 4; in a companion article (1), we describe step 5.

Materials and Methods

Treatment Zone Selection. Two criteria were used for selection of a treatment zone: hydraulic conductivity and uranium concentrations. High hydraulic conductivity was needed for hydraulic control and to enable chemical delivery. High uranium concentration was needed to ensure appreciable changes in soluble uranium concentration in response to chemical and biological manipulations and provided an opportunity for testing of source control strategies.

The aquifer underlying the Y-12 Facility contains weathered saprolite overlying shale bedrock with limestone interbeds. Groundwater flow occurs primarily in thin zones of interconnected fracture networks with high hydraulic conductivity (8). From borehole flowmeter measurements and point dilution tests, a region of high hydraulic conductivity was identified at 10.9–12 m depth, a region where saprolite

transitions to bedrock limestone and shale (9–10). This region was also the region of highest uranium concentrations (11) and was therefore selected as the target zone for conditioning and remediation. Pumping wells were screened over the 11.5–13.7 m depth interval.

Below-Ground System Design. A recirculating well system was designed to provide hydraulic control for chemical delivery, to ensure adequate mixing, and to create a treatment zone protected from the inflow of ambient water. Paired wells have been used effectively for delivery of chemicals, mixing of nutrients, and extraction of contaminants (12–14), but complete recirculation can only be achieved when wells are aligned perfectly parallel to regional flow (15). Uncertainties and variations in regional flow direction make this impractical (16). Even if regional flow is unidirectional and wells are aligned to that direction, drilling variability can still result in borehole misalignment (10). Such deviations were in fact observed for the wells installed at our site. As a result, some screened intervals were offset from their intended locations. Despite this misalignment, the four-well nested cell design used in this research enabled hydraulic control despite deviations from the ideal. The modeling used as the basis for this design is described elsewhere (15).

As shown in Figure 1, the below-ground system consisted of an outer loop injection well (FW024), an inner loop injection well (FW104), an inner loop extraction well (FW026), an outer loop extraction well (FW103), and three boreholes (FW100, FW101, and FW102) containing multilevel sampling (MLS) wells located between the inner loop injection and extraction wells (15). In routine operations, the outer well pair defined an outer recirculation loop, and the inner pair defined an inner recirculation loop, nested within the outer loop. A third extraction well (FW 105) served as a source well for water entering the above-ground treatment system and gave additional control for the outer loop. The outer loop was expected to respond to changes in regional flow, while the shape and recirculation characteristics of the inner cell were expected to remain stable. The functions of the outer cell were to protect the inner cell from ambient groundwater high in nitrate and acidity, to maintain recirculation zone shape as discussed above, and to capture water escaping the inner cell due to imperfect recirculation. Detailed information about the well-system design and flow field manipulation is provided elsewhere (15). Each MLS borehole contained seven separate sampling tubes (diameter 1.9 cm) at the following depths below ground surface: 6.10 m (level 7), 7.62 m (level 6), 9.14 m (level 5), 10.67 m (level 4), 12.19 m (level 3), 13.7 m (level 2), and 15.24 m (level 1). Specific MLS wells were designated by borehole number 100, 101, or 102, and depth 1–7. For example, FW 101-2 specifies borehole 101, level 2. The above-ground treatment system is described in the Supporting Information. A fluidized bed reactor (FBR) was used for biological denitrification (17).

Forced Gradient Tracer Study and Measurement of Mass Transfer Rates from the Soil Matrix. To assess communication between the injection and extraction wells and to identify regions of preferential flow, a tracer study was performed on November 3–5, 2002, i.e., ten months before conditioning of the treatment zone. The flow rates used are indicated in Supporting Information Table S1. Y-12 Plant tap water adjusted to pH 3.8 with HCl was injected to FW024 at a flow rate of 2.36 L min⁻¹ and into FW104 at a rate of 4.0 L min⁻¹. Water was extracted from FW026 at 3.0 L min⁻¹ and from FW103 at 1.4 L min⁻¹. These rates were maintained for 96 h. The outer loop injection solution contained 5 mM CaCl₂ to maintain the ionic strength of the groundwater. Initially, water injected at the inner loop injection well FW104 also contained 5 mM CaCl₂, but after 18.1 h, the salt solution added was changed to an ~5 mM MgBr₂ solution, giving an influent concentration of 9.36 mM of Br⁻. After 15.7 h, the CaCl₂

solution (5mM) was again injected. Extracted water was discharged into a wastewater tanker. Samples were periodically withdrawn from all MLS well ports (FW100, FW101, and FW102) and from the extraction wells (FW026 and FW103) for analysis of bromide and other ions. All injection and extraction of water stopped after 96 h, but water samples were taken periodically for many months thereafter. The rebound of ion concentrations was used to estimate rates of matrix pore diffusion/desorption.

Conditioning of the Treatment Zone. Conditioning of the treatment zone was accomplished by flushing first with clean water at pH 4.3–4.5 to remove nitrate, aluminum, and calcium, then with contaminant-free water at pH 6.0–6.3 to increase subsurface pH from day 9 to 136. Both steps involved use of the nested well recirculation system (Figure 1) and above-ground treatment (Figure S1). Contaminated groundwater was pumped from the subsurface to the above-ground treatment system to be treated and stored as clean water. More clean water was injected than extracted and treated. This was achieved by supplementing treated water extracted from down-gradient well FW105 with Y-12 Plant tap water (Figure 1A and B). The resulting flow rate for injection at FW024 was three times the extraction flow rate at FW103. The tap water had a pH of 8.0 and the following major ion concentrations (mM): chloride, 2.82–3.38; nitrate, 0.04–0.048; sulfate, 0.24–0.26; Ca, 0.68–0.75; Al, <0.007.

Injection and extraction flow rates for this period are summarized in Table S1. This operational phase involved two major steps, detailed below. From days 9 to 68, treated water plus tap water was adjusted to pH 4–4.5 before injection. The purpose of this step was to remove Al and Ca without risk of chemical clogging. Flushing the treatment zone with contaminant-free water at low pH (4.3–4.5) enabled displacement, extraction, and above-ground removal of Al without below-ground precipitation of aluminum hydroxide, which forms above pH 4.5. This choice of pH also minimized U escape from the treatment zone: increasing pH from 3.4 to 4.5 increases sorption of uranium on site sediment (18) and decreases desorption (1). During the pH 4.3–4.5 flush, extracted water was treated to remove Al, Ca, and nitrate, mixed with tap water, adjusted to pH 4.3–4.5 in the above-ground system, and injected into wells FW024 and FW104. Initially there was considerable variability in injection and extraction flow rates because of varying field conditions (Table S1). Beginning on day 58, however, stable flow rates were obtained at the following values: 0.45 L min⁻¹ for extraction from FW103, 0.45 L min⁻¹ for extraction from FW026; 0.45 L min⁻¹ for injection at FW104; and 1.35 L min⁻¹ for injection at FW024. These flow rates were used for the remainder of the conditioning phase and for biostimulation.

From days 69 to 137, the clean water was injected at a pH of 6–6.3. The purpose of this step was to increase pH to a range suitable for robust microbial activity. A pH range of pH 6.0–6.3 was selected because sorption of U(VI) to the soil is maximum within this range, and flushing operations would only remove small amounts of U(VI) from the soil matrix. This pH was also less than optimum for growth of methanogens, which compete for electron donor, do not reduce U(VI), and produce methane—another potential aquifer clogging agent. Groundwater extracted from wells FW103 and FW026 was pumped to the storage tank in the above-ground treatment system then treated as described above. The treated water was mixed with tap water, adjusted to pH 6.0–7.0, then injected into FW024 and FW104. An exception was days 89–107 when water was injected directly into FW104 without above-ground treatment.

Analytical Methods. Uranium concentrations in all water samples, which were taken in the field and acidified with HNO₃ (final concentration 0.4 N), were determined by kinetic phosphorescence analysis using a KPA-11 analyzer (Chem-

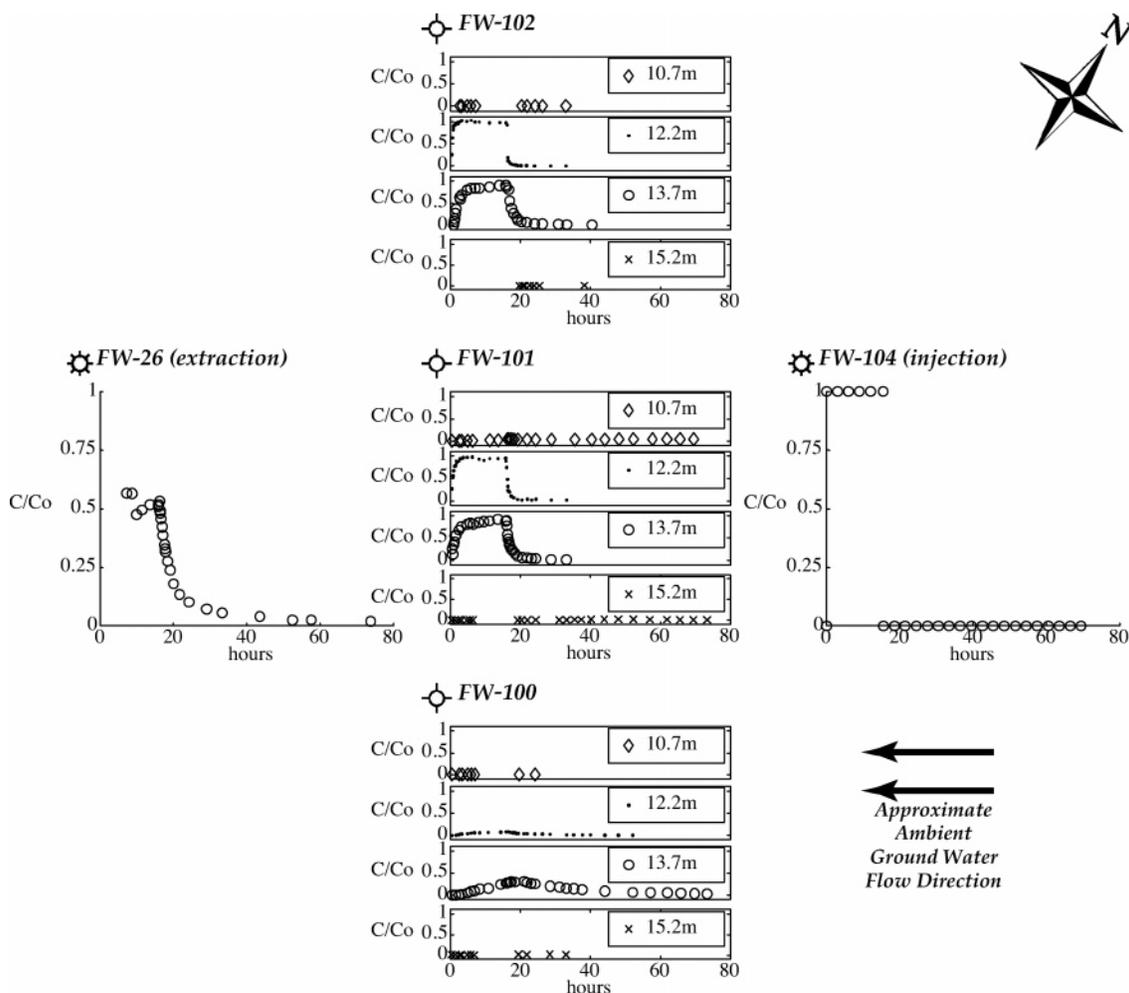


FIGURE 2. Schematic plan view showing wells at approximate locations and bromide breakthrough curves during the tracer study (November 4–8, 2002).

Chek Instruments, Richland, WA). Chemical oxygen demand (COD) was used as an overall indicator of the consumption of electron donors (ethanol and its metabolic intermediate acetate). COD, sulfide, and turbidity were measured using a Hach DR 2000 spectrophotometer (Hach Chemical, Loveland, CO). Anions (including NO_3^- , Br^- , Cl^- , SO_4^{2-} , and PO_4^{3-}) were analyzed with an ion chromatograph equipped with an IonPac AS-14 analytical column and an AG-14 guard column (Dionex DX-120, Sunnyvale, CA), and cations (Al, Ca, Fe, Mn, Mg, U, and K, etc.) were determined using an inductively coupled plasma mass spectrometer (ICPMS) (Perkin-Elmer ELAN 6100) as described elsewhere (2).

Results and Discussion

Inorganic Solids Production due to Increased pH. In titration studies with site groundwater, precipitate formed when pH was increased from 3.50 to 8.75 with K_2CO_3 (Figure S2). The quantity increased as pH increased. White aluminum hydroxide flocs formed at $\text{pH} > 4.5$. Dense calcium carbonate precipitates formed at $\text{pH} > 7.0$ with the most dramatic precipitation forming at $\text{pH} > 8.0$. The amount of dry solids produced at $\text{pH} 6.0$ was 1.6 g L^{-1} , and the amount produced at $\text{pH} 7$ was 2.2 g L^{-1} . Equilibrium calculations indicated a dissolved Al concentration of $\sim 0.5 \mu\text{M}$ at $\text{pH} 6.0$ and $\sim 0.4 \mu\text{M}$ at $\text{pH} 7.0$, with $\text{Al}(\text{OH})_3$ constituting 66 and 87% of the total Al. Site groundwater contained 18 mM Al, which could theoretically generate $\sim 1.5 \text{ g L}^{-1} \text{ Al}(\text{OH})_3$ —a quantity consistent with titration results. These data indicated that in situ adjustment of pH would lead to excessive accumulation

of solids and subsequent loss of hydraulic control. They would also generate surfaces for U sorption, confounding identification of soluble U(VI) removal mechanisms. Al and Ca were therefore removed above ground, prior to increasing subsurface pH.

Flow Pattern Characterization and Rebound of Solute Concentrations. A forced gradient tracer test established travel times and connectivity between wells and enabled estimates of rates of mass transfer from the soil matrix. Figure 2 illustrates bromide concentrations measured at MLS wells and extraction well FW026 during the tracer test. A detailed analysis with modeling is provided elsewhere (15). Tracer breakthrough at a depth of 12–14 m was more pronounced than at other depths indicating a zone of fast flow, consistent with flow meter measurements showing elevated hydraulic conductivity over this depth interval (9–10). Less bromide was detected at FW100 than at FW101 and FW102 as this well was located down dip from the source and tracer moved preferentially along strike. Faster response was observed at FW101 and FW102 at a depth of 12–14 m; i.e., FW101-2 (13.7 m depth), FW101-3 (12.2 m depth), FW102-2 (13.7 m depth), and FW102-3 (12.2 m depth), which are down-gradient strike-parallel wells. The results indicated connectivity between wells within a high-conductivity region suitable for controlled experiments.

The use of acidified clean water enabled an assessment of contaminant release from the soil matrix. Several ions were monitored during and after the forced gradient tracer study. Figure 3 describes the change of the ions in FW101-3.

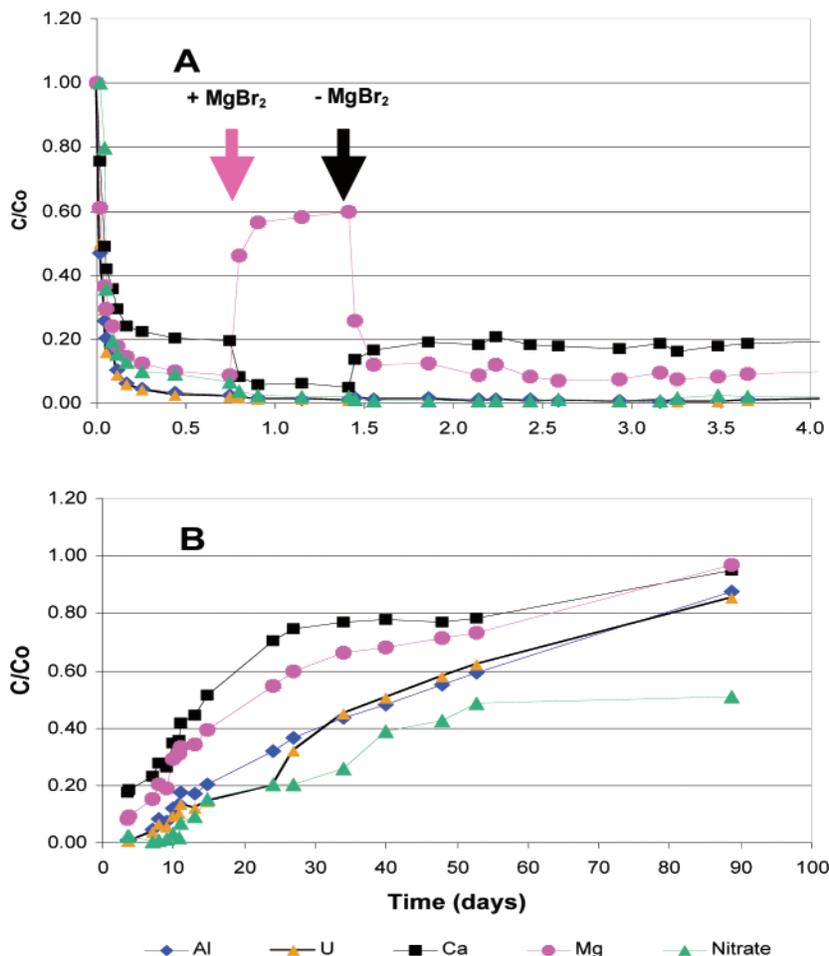


FIGURE 3. (A) Decrease in Al, Ca, Mg, and U concentrations during forced-gradient tracer study in well FW101-3. MgBr₂ was injected as tracer at $t = 0.75$ d and stopped at $t = 1.4$ d. CaCl₂ was added to injection water except for tracer injection period. (B) Rebound of Al, Ca, and U concentrations at FW102-3 after the forced-gradient tracer study. After 8 months, nitrate concentration increased to 80% of its initial level. Solute concentrations before acid water flush were as follows: 19.8 mM Al; 300 μ M uranium; 34.5 mM Ca; 92 mM Mg; and 154 mM nitrate.

All solutes in other MLS wells followed a similar pattern, with fast displacement during the forced gradient flush followed by a slow rebound under natural gradient conditions. The rebound was largely attributable to mass transfer from stagnant micropores (immobile zones) within the soil matrix to the fractured zones (mobile zones). These observations were consistent with prior data indicating domains of vastly different permeability, where most of the flow occurs within interconnected fractured zones on a millimeter scale but these zones contribute little to the overall void-space (19–23). Storage of groundwater and its dissolved constituents occurs predominantly in immobile zones (8)—large regions characterized by high porosity, small pore size, and large surface area for sorption of ions, such as U(VI). Rates of release of nitrate from these storage regions were quantified using a modified mobile–immobile model (24). A slow mass-transfer rate was obtained, indicating that the immobile domain would be a long-term source for compounds stored within it and would control the bioavailability of U(VI) in recirculation schemes. The tracer test also indicated that acidic flushing to remove nitrate, Al, and Ca would be a long-term task. Model projections indicated that nitrate in the immobile region would be depleted to low levels (<0.1 mM) by flushing over a period of about one year. Subsequent observations confirmed this prediction (1). The model and the breakthrough data also indicated that the mobile domain would respond rapidly to flushing, suggesting that low concentrations of nitrate, Al, and Ca could be maintained by

vigorous recirculation coupled with removal of substances diffusing in from the mobile zone. These predictions were also confirmed.

Operation of Above-Ground System. The performance of the above-ground treatment system and the resulting composition of the water used for the injection to subsurface are summarized in Tables 2 and 3. FBR influent nitrate concentrations decreased from 80 mM to around 1.0–2.0 mM (Figure S3), alleviating the potential for clogging by gas and biomass upon electron donor addition. A more detailed description of FBR operations is given elsewhere (17). The quality of the treated water (<1.0 mM nitrate and 1–3 FTU) satisfied objectives for re-injection to the subsurface.

Effects of Flushing on Removal of Al, Ca, and U. Flushing with acidified clean water (days 9–68) removed bulk aluminum and other metals and avoided in situ precipitation. Al levels fell from 12.6 to <0.03 mM in FW103 and to <0.3 mM in FW026. Ca levels fell from 25 to < 2.5 mM in both wells (Figure S4). The Al concentration in FW026 was near the solubility of solid-phase Al at pH 4.4, suggesting a source of Al near the extraction well. However, at these low levels with 98% of the Al and 90% of the Ca removed, there was less risk of clogging due to the formation of Ca and Al precipitate.

Al and Ca concentrations decreased to <0.1 and 1.0 mM, respectively, during the pH adjustment period (days 69–137) (Figure S4A and B). Figure S4C illustrates the accompanying change in pH at the inner loop recirculation wells as pH increased at injection well FW104. The pH of

TABLE 2. Performance of Above-Ground Treatment System and the Quality of Water Injected to the Subsurface during the Clean Water Flush (Days 1–137)^a

period: item	flush at pH 4.2–4.5 (days 1–68)			flush at pH 6.0–6.3 (days 69–137)		
	influent	FBR eff.	inject.	influent	FBR eff.	inject.
nitrate (mM)	80 to 10	0.2 to 1.0	0.2 to 0.5	4.6 to 1.5	0.05 to 0.5	0.05 to 0.5
sulfate (mM)	1.2 to 1.0	1.0 to 1.2	0.85 to 1.0	0.8 to 1.0	0.8 to 1.0	0.5 to 0.6
chloride (mM)	3.0 to 4.0	3.0 to 4.0	3.0 to 4.0	3.0 to 4.0	3.0 to 4.0	3.0 to 4.0
Al ³⁺ (mM)	1.85 to 0.07	0.03	<0.03	0.07 to 0.03	<0.03	<0.03
Ca ²⁺ (mM)	7.1 to 2.5	1.9 to 2.1	1.0 to 1.25	2.2 to 1.8	1.5 to 2	1.0 to 1.3
uranium (μM)	210 to 6.3	2.0 to 2.5	0.17 to 0.4	2.7 to 3.4	2.4 to 2.7	2.4 to 2.7
K ⁺ (mM)	5.1 to 2.6	7.2 to 8.0	5.1 to 6.4	2.5 to 5.9	5.2 to 7.7	2.6 to 5.5
Na ⁺ (mM)	2.6 to 1.3	1.3 to 2.6	1.3 to 2.0	1.2 to 2.2	1.3 to 2.6	0.7 to 1.7
Mg ²⁺ (mM)	0.82 to 0.49	0.49 to 0.8	0.49 to 0.8	0.5 to 0.82	0.5 to 0.82	0.45 to 0.8
Mn ²⁺ (mM)	0.2 to 0.10	0.09 to 0.1	0.03	0.06 to 0.1	0.04 to 0.09	0.45 to 0.8
COD (mg/l)	5 to 2	10 to 30	2 to 10	1 to 2	10 to 30	0 to 5
FTU	0 to 1	1 to 3	0 to 2	0 to 1	1 to 3	0 to 2
pH	4.0 to 4.5	8.0 to 8.5	4.2 to 4.5	4.0 to 4.5	6.8 to 8.0	6.0 to 6.3

^a Influent = treatment system influent. FBR eff. = effluent of biological fluidized bed reactor. Inject. = water injected to subsurface. Injected water was a mixture of treated water and tap water. COD = Chemical oxygen demand. FTU = Formazine turbidity unit. Turbidity of the FBR treated water was measured after filtration. Before filtration, the turbidity varied from 8 to 40 FTU.

TABLE 3. Changes in Water Quality Before and After the Clean Water Flushes (Day 69 Was the Last day of the pH 4.3–4.5 Flush and Day 137 Was the Last day of the pH 6.0–6.3 Flush)^a

item	date	Well						
		FW026	FW104	FW103	FW101-2	FW101-3	FW102-2	FW102-3
Ca ²⁺ (mM)	pre-flush	25.15	59.1	54.2	57.4	36.7	57.9	28.4
	day 69	1.73	1.21	2.18	1.98	1.42	1.68	1.23
	day 137	1.32	1.35	2.73	1.51	1.04	1.53	1.36
Al ³⁺ (mM)	pre-flush	18.4	13.6	10.6	16.7	9.55	7.53	12.1
	day 69	0.39	0.043	0.05	0.07	0.23	0.054	0.045
	day 137	0.12	0.001	0.03	0.007	0.014	0.023	0.003
uranium (μM)	pre-flush	158.4	139.5	94.1	134.0	134	47.5	150
	day 69	6.8	1.43	2.2	1.94	3.97	1.94	3.22
	day 137	5.1	2.8	2.7	2.39	3.04	3.29	3.5
nitrate (mM)	pre-flush	132	208	198	183	114	271	137
	day 69	1.73	0.18	1.20	0.28	0.48	0.51	0.68
	day 137	0.78	0.10	0.43	0.11	0.12	0.12	0.11
pH	pre-flush	3.22	3.83	3.80	3.78	3.95	4.00	3.72
	day 69	4.50	4.55	4.56	nd	nd	nd	nd
	day 137	5.16	6.26	6.27	6.23	5.89	6.33	6.26

^a nd = Not determined. Throughout the flushing periods, the composition of water in injection well FW104 was the same as that of the water injected.

extraction well FW026 dropped below 5.0 during a period when groundwater was recirculated directly without base addition (days 89–107), but increased thereafter from 4.5 to 5.2. As the solid-phase equilibrated, the pH values at MLS wells FW101-2, FW101-3, FW102-2, and FW102-3 increased to 6.1–6.2. The slower increase in FW026 was likely due to residual acidity from Al in the soil matrix and the fact that the extraction well captured water over a broad region.

At the pH levels used for flushing (5.8–6.2) the sediments have higher U(VI) adsorption partition coefficients and lower desorption partition coefficients (1, 18) compared to values for the ambient groundwater–sediment system at pH 3.6. Thus, flushing had minimal impact on solid-phase concentrations. The maximum uranium content in sediments from cores recovered during installation of the inner loop injection well was 800 mg kg⁻¹. In the subsequent biostimulation phase, the concentrations of U on all sediment samples recovered from recirculation and monitoring wells were equal to or greater than 800 mg kg⁻¹ (1). However, it should be noted that the U concentrations measured in core samples varied depending on location and could be significantly different from levels in sediments around the wells. Nevertheless, as

noted above, the pH was maintained within a range that would maximize adsorption and minimize desorption. It was thus anticipated that the vast majority of the uranium would remain in situ after flushing, and subsequent observations were consistent with that expectation.

Removal of Nitrate by Flushing. Figure 4 illustrates the change in nitrate concentrations of FW026, FW103, and three MLS wells for both flushing periods. During the pH 4.3–4.5 flush (days 9–68), nitrate concentration decreased rapidly from >100 to around 1.0–2.0 mM in both inner loop and outer loop extraction wells. In all of the MLS wells, nitrate concentrations rapidly decreased at depths 12.2 and 13.7 m (FW100-2, 100-3, 101-2, 101-3, 102-2, and 102-3) but only slowly decreased at depths 10.7 and 15.2 m. These results are consistent with tracer test results, showing more pronounced tracer response at 12–14 m compared to other depths, due to preferred flow in fractures. FW100 exhibited a somewhat slower and less pronounced response compared to the other MLS wells. This was also consistent with the tracer study, where the response was not as fast as at other MLS wells. Nitrate concentrations occasionally jumped to high values—probably due to occasional intrusion of contaminants from

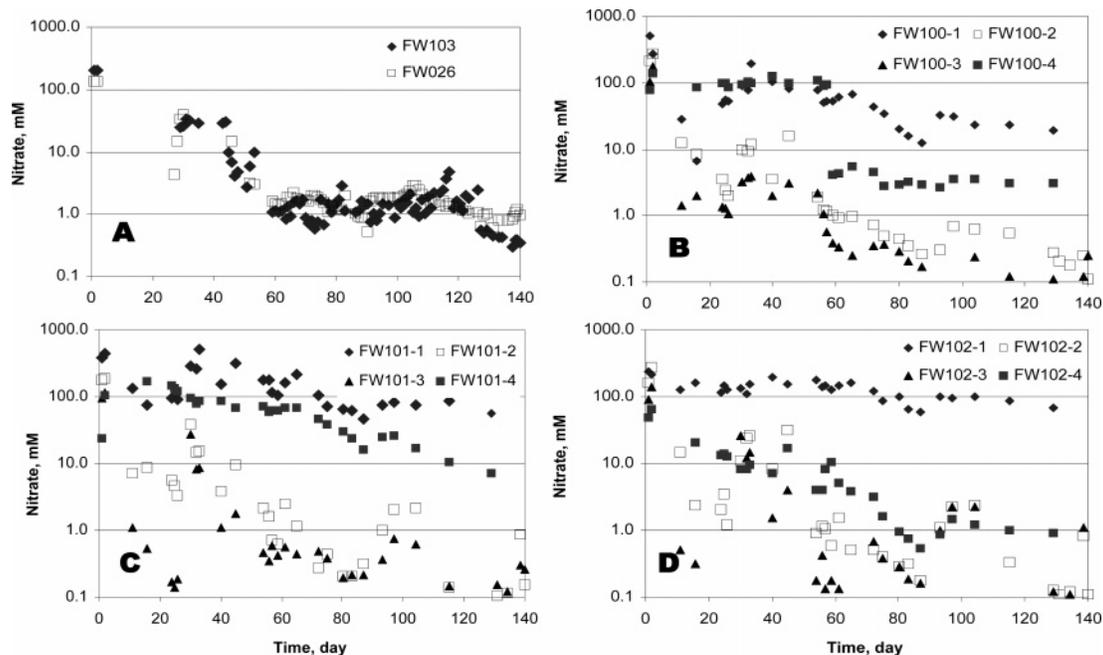


FIGURE 4. Changes of nitrate profiles in inner loop and outer loop extraction wells and MLS wells during the clean water flush period (day 0–140). Note that nitrate concentrations are displayed on a logarithmic scale.

the outer recirculation loop or release of trapped nitrate from the soil matrix.

By day 137, the system was deemed suitable for biostimulation based on the pH of the MLS wells (1). A comment on chemical fouling is merited here. Shortly after initiation of biostimulation, white precipitate formed on the pump head of extraction well FW026, clogging the screen. The precipitate was soluble in a solution of HCl (4%). SEM analysis indicated that it was $\text{Al}(\text{OH})_3$ with small fractions of uranium, sulfur, silicon, and phosphorus (Figure S5). Only two such clogging events occurred. Given that ambient levels of aluminum were over 100 times higher than the residual that caused these fouling events, it is likely that above-ground removal of aluminum prevented unmanageable fouling.

Implications. This work demonstrates that a nested-well recirculation system can be used to achieve hydraulic control for remediation, even in a complex, highly contaminated aquifer. The approach used should have general value for pilot-scale testing at sites where ambient contamination would otherwise prevent a controlled assessment of the potential for remediation.

Staged conditioning of an aquifer is a generally useful strategy, but the sequence and nature of the conditioning steps required will depend on site conditions and the selection pressures needed to support the desired microbial ecology. At a large field site in Michigan, conditioning of an aquifer was accomplished by adjustment of pH to 8 (25). In that case, the pH adjustment step created a niche for introduction of a non-native bacterium. At the FRC Area 3, additional conditioning was necessary for removal of inhibitors and clogging agents, and conditioning was accomplished in two steps because of the nature of the contaminants present. A staged strategy with recirculation of contaminant-free water decreased concentrations of clogging agents and inhibitors in the bulk solution, and allowed controlled biostimulation of U(VI) reduction, as described in the companion article (1). Addition of contaminant-free water also provided a means of estimating rates of contaminant mass transfer. These strategies should also have general value at highly contaminated sites.

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Supporting Information Available

Injection and extraction flow rates for tracer test and subsurface flush; above-ground treatment system and operation; production of inorganic solids due to an increase in pH; metals removal during conditioning; SEM photo of precipitates. This material is available free of charge via the Internet at <http://pubs.acs.org>.

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