Bioremediation and Biodegradation

In Situ Reduction of Chromium(VI) in Heavily Contaminated Soils through Organic Carbon Amendment

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ABSTRACT

Chromium has become an important soil contaminant at many sites, and facilitating in situ reduction of toxic Cr(VI) to nontoxic Cr(III) is becoming an attractive remediation strategy. Acceleration of Cr(VI) reduction in soils by addition of organic carbon was tested in columns pretreated with solutions containing 1000 and 10 000 mg L\(^{-1}\) Cr(VI) to evaluate potential in situ remediation of highly contaminated soils. Solutions containing 0, 800, or 4000 mg L\(^{-1}\) organic carbon in the form of tryptophosphate broth or lactate were diffused into the Cr(VI)-contaminated soils. Changes in Cr oxidation state were monitored through periodic micro-XANES analyses of soil columns. Effective first-order reduction rate constants ranged from \(1.4 \times 10^{-4}\) to \(1.5 \times 10^{-3}\) s\(^{-1}\), with higher values obtained for lower levels of initial Cr(VI) and higher levels of organic carbon. Comparisons with sterile soils showed that microbial populations were largely responsible for Cr(VI) reduction, except in the soils initially exposed to 10 000 mg L\(^{-1}\) Cr(VI) solutions that receive little (800 mg L\(^{-1}\)) or no organic carbon. However, the microbial populations in the viable soils are probably too low for direct enzymatic Cr(VI) reduction to be important. Thus, synergistic effects sustained in whole soil systems may have accounted for most of the observed reduction. These results show that acceleration of in situ Cr(VI) reduction with addition of organic carbon is possible in even heavily contaminated soils and suggest that microbially dependent reduction pathways can be dominant.

The wide range of chromium applications has resulted in its occurrence as a common contaminant in soils (Palmer and Wittbrodt, 1991; Proctor et al., 1997). In soils, Cr occurs in two oxidation states having very different behavior (Rai et al., 1989). Hexavalent Cr is generally more soluble, mobile, and toxic. The reduced Cr(III) forms are generally much less mobile and less hazardous. The negative logarithm of the electron activity (pe) of Cr(VI)–Cr(III) transformations is in the range of 14 to 5 over the pH range of 4 to 9. Because of this high pe, availability of reductants, and typically faster rates of Cr(VI) reduction relative to Cr(III) reoxidation, most Cr occurs in trivalent forms in uncontaminated soils. In soils that have been contaminated with Cr(VI), rates of reduction to Cr(III) are of great interest because of the vastly differing toxicities of these two oxidation states.

One general approach to remediating Cr(VI)-contaminated soils involves accelerating in situ reduction to Cr(III) (Higgins et al., 1998). Enhanced in situ Cr(VI) reduction is attractive because excavation and waste disposal are very costly. Although Cr(VI) is reduced to Cr(III) to some extent without intervention, rates of natural attenuation can be unacceptably slow. Thus, supplying reductants into soils and ground water is becoming attractive for accelerating Cr(VI) reduction. Inorganic treatments include H\(_2\)S injection (Thornton and Amonette, 1999; Kim et al., 2001), aqueous Fe(II) injection (Seaman et al., 1999), and use of reduced Fe solids in permeable reactive barriers (Blowes et al., 1997). Proposed organic-based Cr(VI) reduction strategies have included application of various carbon sources such as manure, molasses, and organic acids (Losi et al., 1994; Henny et al., 2001; Higgins et al., 1998; Perlmutter et al., 2001).

The effectiveness of organic carbon (OC) in reducing Cr(VI) depends on the concentration of the contaminant, reactivity of OC, and microbial activity. Higher Cr(VI) concentrations depress microbial activity (Chen and Hao, 1998), thereby diminishing several Cr(VI) reduction processes, including enzymatic reduction (Lovel, 1993), reduction by ferrous iron and sulfide (Fend-orf et al., 2000) sustained primarily through the activity of Fe- and S-reducing bacteria (Chapelle, 1992), and reduction by intermediate organic degradation products (Deng and Stone, 1996). Despite the fact that some contaminated soils and ground water contain levels of Cr(VI) exceeding 10 000 mg L\(^{-1}\) (Palmer and Wittbrodt, 1991; Sturges et al., 1992; Makdisi, 1992), very few laboratory studies have been conducted at concentrations relevant to remediating heavily contaminated environments.

The extent of Cr(VI) reduction in contaminated sediments also depends on the amount of native and added OC. Many forms of OC directly reduce Cr(VI), including phenols (Elowitz and Fish, 1994), organic acids (Deng and Stone, 1996), and humic substances (Wittbrodt and Palmer, 1995, 1996, 1997; Nakayasu et al., 1999). Chromium(VI) reduction rate constants for these organic species are highly pH dependent, becoming slow under

Abbreviations: \(k^*\), Wittbrodt–Palmer rate constant; \(k_{oc}\), effective first-order rate constant, dependent on organic carbon; \(K_c\), Monod half-velocity constant; OC, organic carbon; \(\Delta OC\), organic carbon added per mass of soil; SHA, soil humic acid; TSB, tryptophosphate broth; \(X\), biomass concentration; \(X_o\), fraction of humic acid oxidized; XANES, X-ray absorption near-edge structure; \(\mu_{max}\), Monod maximum specific reduction rate constant; \(2Cr\), total chromium.


Published in J. Environ. Qual. 32:1641–1649 (2003). © ASA, CSSA, SSSA

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neutral to alkaline conditions. Chromate reduction by OC also occurs through microbiologically mediated pathways. Available OC may stimulate activity of some Cr-reducing organisms within the native microbial community. Soil microorganisms can also contribute to Cr(VI) reduction through release of ferrous iron, sulfides, and reactive organic intermediates. Soluble organic ligands can kinetically influence the fate of subsurface Cr through Fe(II) and Fe(III) complexes, as well as through complexing Cr(III) in soluble forms (Buerge and Hug, 1998). Some Fe(III) reduction occurs through microbiologically produced electron shuttles such as reduced humic acids and hydroquinones (Nevin and Lovley, 2000). Mineral surfaces also catalyze Cr(VI) reduction by OC (Deng and Stone, 1996). This brief survey indicates that Cr reduction in soils occurs through complex interactions of synergistic and competing processes.

To understand the dynamics of contamination and in situ remediation processes, it is essential to include transport influences. One approach to including these effects involves conducting experiments in columns or soil aggregates in which transport can become diffusion-dominated. This approach was used in our previous studies on Cr contamination (Tokunaga et al., 2001, 2003) and is used here to examine OC-based remediation of Cr(VI)-contaminated soils. Competing effects of the initial level of Cr(VI) contamination and the amount of OC added were tested in small columns representing transects into soil aggregates. Our primary goals are to determine (i) the overall effect of OC availability on Cr reduction in soils, (ii) the collective importance of microbiologically dependent Cr(VI) reduction pathways versus abiotic pathways in highly contaminated soils, and (iii) whether or not spatial heterogeneity of Cr develops during OC-stimulated Cr reduction.

MATERIALS AND METHODS

Small soil columns, similar to those described in Tokunaga et al. (2001), were used to represent transects into subsurface domains where transport is diffusion-limited (Fig. 1). The upper surface of each column represents the exterior of an aggregate, through which components [water, O₂, CO₂, Cr(VI), OC] are exchanged between the system and its environment. Soil from the C horizon of Altamont clay (fine, smectitic, thermic Aridic Haploxerert) was collected at Altamont Pass (Alameda County, CA). The calcareous (1–4% calcite, pH 8.3), clayey (42% clay, 52% silt, 6% sand, 1% organic carbon) soil was crushed, homogenized, and passed through a 250-μm sieve. The native Cr concentration of this soil is 60 mg kg⁻¹, with greater than 95% as Cr(III). The Cr oxidation state was determined by X-ray absorption spectroscopy, as described later. This soil was packed into columns to an equivalent dry bulk density of 1.26 Mg m⁻³. Soil columns were 11.9 mm in diameter, 30 mm long, with porosities and pore volumes of 0.52 and 1.74 mL, respectively. The gravimetric water content of the soil at this stage was 0.074 g g⁻¹. Columns were sequentially treated with three different solutions. First, deionized water was added to achieve a soil water content of 0.30 g g⁻¹ (equivalent to 70% saturation), by allowing water to infiltrate into columns while the outflow port was open. Using texture and density information, the soil matric potential was estimated to be about −0.3 MPa under these preexposure conditions (Campbell, 1985). Columns were incubated at this level of water saturation for 8 d at room temperature to partially reactivate the microbial community while maintaining aerobic conditions. This was followed by ponding 1.10 mL of solutions containing 0, 1000, or 10 000 mg L⁻¹ Cr(VI), added as K₂CrO₄. These solutions were ponded on top of the partially saturated soils while the outflow port was still open, such that inflow continued and drainage of soil water occurred until the chromate pool receded into the soil surface. The outflow port was then sealed for the remainder of the experiment, such that further transport was by diffusion only. At the end of this contamination phase, soils contained either 0, 4.93, or 49.3 μmol of added Cr(VI) per g soil, and the water content was in the range of 0.40 ± 0.02 g g⁻¹ (95 ± 5% saturation). Five days after the initial exposure to Cr(VI) solutions, columns were ponded with a third solution containing various concentrations of organic carbon. Columns received 0.60 mL (0.345 pore volume) of either trypic soy broth or sodium lactate solutions as the OC remediation agent. Infiltration and slow evaporation diminished the initial ponding depths (4–5 mm) of these solutions, such that distilled water was periodically added to maintain about a 1-mm pool for the calomel reference electrode. Solutions with 0 (deionized water control), 800, or 4000 mg L⁻¹ OC were ponded on each column. This amounted to adding 0, 9.3, or 47 μmol of OC per g soil (0, 112, or 560 μg g⁻¹). Tryptic soy broth (TSB) was used as a carbon source representing decomposing plant tissue. Lactate was used in several columns since it is an easily metabolized carbon source, with related compounds now being tested for use in in situ bioremediation (Lütze et al., 2001; Evans and Kueniggerberg, 2001). The ponded surface of each column was capped to minimize evaporation, but vented through a segment of a hypodermic needle to maintain this boundary in equilibrium with atmospheric oxygen. Periodic uncapping for redox potential and pH measurements also helped to maintain this aerated boundary condition.

Redox potential profiles were periodically measured through a series of Pt electrodes embedded along columns. Electrodes were set at 0, 2, 4, 6, 10, 15, 20, and 25 mm, relative to the soil surface. For these measurements, a calomel reference electrode was dipped into the surface pool. The reference electrode was rinsed with alcohol and distilled water after removal from each column. Redox measurements presented later are relative to the standard hydrogen electrode, obtained by adding 245 mV to the raw calomel-referenced data. Measurements of pH were also obtained within the surface pool of each column.

Profiles of Cr concentrations and oxidation states in soil columns were obtained at various times after exposure to Cr(VI) solutions, using micro-X-ray absorption near edge structure (micro-XANES) spectroscopy. Micro-XANES measurements (Sutton and Rivers, 1999; Bertsch and Hunter,
RESULTS AND DISCUSSION

pH and Redox Potential Measurements

Measurements of pH in water ponded at the surface ranged from 6.9 to 8.2, with typical values ranging from 7.2 to 7.9. Redox potential measurements in the Cr(VI)-contaminated columns remained relatively high throughout the study, showing little if any spatial trends, and showed no significant response to OC additions (Fig. 3). Values were typically in the range of +250 to +400 mV (standard H electrode referenced). For comparison, soils that were not exposed to Cr(VI) supported wider ranges in redox potentials, and a significant shift to lower potentials with addition of OC (Fig. 3, left panel). Redox potential data for soils not exposed to Cr(VI) come from a +0 OC control column monitored during this study, and from a +800 mg L⁻¹ OC column [before Cr(VI) addition] tested in an earlier study (Tokunaga et al., 2001). The uncontaminated soils did show significant redox gradients, being more oxic at the surface and more reducing at greater depths. The high redox potentials in the Cr(VI)-contaminated soils provided an indirect indication that microbial activity was low in each of these systems, since higher respiration rates combined with diffusion-limited oxygen supply would result in lower Eh with depth. Such sustained high redox potentials and inferred low microbial activity suggest that the initial exposure of soils to very high (1000 and 10 000 mg L⁻¹) Cr(VI) concentrations resulted in long-term suppression of microbial communities. These high redox...
values also indicate that concentrations of two potentially important inorganic reductants, ferrous iron and sulfide, remained very low throughout the experiment.

**Microbial**

Analyses of microbial populations on Day 130 showed low cell counts, ranging from $9.5 \times 10^4$ to $2.1 \times 10^5$ per g soil. Most of these soils were exposed to 1000 mg L$^{-1}$ Cr(VI) solutions, with varying levels of added OC. Based on their very low rates of Cr reduction, even lower cell counts are expected in the soils initially exposed to 10,000 mg L$^{-1}$ Cr(VI) solutions, with varying levels of added OC. However, since these latter systems are being maintained for long-term studies, they have not yet been analyzed for their microbial characteristics. The highest cell count of $2.1 \times 10^5$ per g soil was obtained from a control [with neither Cr(VI) or OC added]. These low populations indicate that microbial community exposure to Cr(VI) solutions did not grow significantly in response to later addition of OC. The densities and activity were significantly higher in the bottom half of the columns vs. the top half of the column in all of the columns exposed to OC, although these differences were not large enough to generate gradients in Cr profiles (following section). The control soils [no Cr(VI) added] and the soils with 1000 mg L$^{-1}$ Cr(VI) and no OC did not show significant differences in density or activity between the top and the bottom of the columns. No colonies were observed on any of the plates of sterile soils, indicating less than $10^5$ culturable organisms per g sterile soil.

**Total Chromium and Chromium(VI) Profiles in Soil Columns**

Total chromium ($\Sigma$Cr) concentration profiles were relatively uniform within individual columns. Temporal trends in the distribution of $\Sigma$Cr between Cr(VI) and Cr(III) showed net reduction over time. Representative micro-XANES maps from the soil column exposed to 1000 mg L$^{-1}$ Cr(VI), then treated with +4000 mg L$^{-1}$ OC, are presented in Fig. 4. Results from other soil columns are shown in later graphs in terms of average values of Cr(VI) concentrations relative to initial values, since intracolumn gradients in Cr concentrations and oxidation states were not significant. Reduction of Cr(VI) occurred more rapidly with higher amounts of OC additions for both levels of initial Cr(VI), although the more highly contaminated soils [10 000 mg L$^{-1}$ initial Cr(VI) solutions] generally required longer times to achieve low Cr(VI) to $\Sigma$Cr ratios. The relatively uniform distribution of Cr(VI) at any given time within individual systems indicated spatially uniform reduction rates. If significantly higher Cr(VI) reduction occurred within certain regions, these locations would exhibit locally lower Cr(VI) to $\Sigma$Cr ratios, as well as locally higher $\Sigma$Cr. The latter enrichment would occur through Cr(VI) diffusion into the region supporting a higher reduction rate. The reaction-limited condition observed in these columns is in strong contrast to the diffusion-limited cases examined in our previous experiments (Tokunaga et al., 2001). The previous study involved Cr(VI) diffusion into microbially active aggregates with previously established reducing conditions. Such conditions favored very localized Cr(VI) reduction and Cr(III) precipitation. In the present work, Cr(VI) solutions were rapidly infiltrated into aerobic, unsaturated soil columns having relatively low microbial activity. Low microbial activities were probably maintained throughout this study because of Cr(VI) toxicity.

**Chromium(VI) Reduction Rates**

Time trends in ratios of Cr(VI) concentrations relative to initially added Cr(VI) concentrations were fit to first-order kinetic equations to permit comparisons among the different soil columns and with other studies (Fig. 5). Each data set was fit to:

$$ [\text{Cr(VI)}] = [\text{Cr(VI)}]_i \exp(-k_{oc} t) \quad [1] $$

where $[\text{Cr(VI)}]_i$ is the initial Cr(VI) concentration (mg kg$^{-1}$), the effective first-order rate constant $k_{oc}$ (s$^{-1}$) is assumed to depend linearly on the amount of OC added, and $t$ is time (s). Since Cr(VI) reduction in soils occurs via numerous interrelated processes, parameters from such expressions only approximately describe net rates. Individual Cr(VI) reduction pathways may better be described by other rate laws, as noted later for reactions with humic substances. A unique time zero for applying the above expression does not exist since the organic carbon solutions were diffused into the soils starting 5 d after infusion of Cr(VI) solutions. For simplicity, the time at which OC solutions were added was taken as time zero for fitting. For each of the two initial Cr(VI) concentrations, the three sets of Cr(VI) versus time data were fit simultaneously using Eq. [1] and:

$$ k_{oc} = a + b(\Delta\text{OC}) \quad [2] $$

$$ \Sigma \text{Cr}, \text{mg (kg soil)}^{-1} $$
Comparisons between measured reduction rates in these soils and rate constants for specific reactions can help evaluate the importance of various pathways. Here, two relevant pathways are considered: (i) direct reduction by the indigenous microbial community and (ii) reduction by aqueous Fe(II). Rates of Cr(VI) reduction in mixed cultures obtained from soils and sediments were characterized at stationary (no growth) phase by Schmieman et al. (1998). They used a Monod approach to characterize microbial Cr(VI) reduction:

$$\frac{d}{dt}[\text{Cr(VI)}] = \frac{\mu_{\text{MAX}} [X]}{K + [\text{Cr(VI)}]}$$

where $\mu_{\text{MAX}}$ is the maximum specific rate constant $[\text{mg Cr(VI)} (\text{g dry cells})^{-1} \text{h}^{-1}]$, $K$ is the half-velocity constant (mg L$^{-1}$, equal to the substrate concentration at which the reaction rate is half that of its maximum), and $[X]$ is the biomass concentration expressed as (g dry cells L$^{-1}$). The $\mu_{\text{MAX}}$ values they obtained were in a fairly narrow range, from 1 to 3.3 mg Cr(VI) (g dry cells h)$^{-1}$. Since our experiments were done with initial Cr(VI) concentrations of 1000 and 10 000 mg L$^{-1}$, while Schmieman et al. (1998) obtained $K$ values less than 2 mg L$^{-1}$, effectively zero-order microbial reduction of Cr(VI) is predicted over most of the experiment with Eq. [4]:

$$\frac{d}{dt}[\text{Cr(VI)}] = \mu_{\text{MAX}} [X]$$

Although our trends are closer to first-order than zero-order (Fig. 5), we continue with Eq. [4] to determine whether or not direct reduction of Cr(VI) by the microbial community is likely to be significant. For this purpose, we will use the highest value of $\mu_{\text{MAX}}$ reported by Schmieman et al. (1998) of 3.3 mg Cr(VI) (g dry cells h)$^{-1}$, combined with the highest microbial cell counts of 2.1 $\times$ 10$^9$ cells (g soil)$^{-1}$ obtained in our soil columns. Since the soil to water ratio in the columns is 2.55 g mL$^{-1}$, this population is equivalent to about 5.4 $\times$ 10$^9$ cells (mL pore water)$^{-1}$ = approximately 5.4 $\times$ 10$^9$ cells L$^{-1}$. Assuming an average live cell volume of 5 $\mu$m$^3$ (a high estimate), the total live cell volume is 2.7 $\times$ 10$^9$ $\mu$m$^3$ L$^{-1}$ = 2.7 $\times$ 10$^{-3}$ cm$^3$ L$^{-1}$. Assuming a wet cell density of 1.1 g cm$^{-3}$ and a solids content of 0.4 g g$^{-1}$ (Paul and Clark, 1996), $[X] = 1.2 \times 10^{-3}$ (g dry cells) L$^{-1}$. Finally, combining these $\mu_{\text{MAX}}$ and $[X]$ values yields a constant Cr(VI) reduction rate of $-1.1 \times 10^{-6}$ mg L$^{-1}$ s$^{-1}$ = $-0.094$ mg L$^{-1}$ d$^{-1}$. Such a zero-order rate could account for only 0.4

Table 1. Summary of Cr(VI) reduction rate constants.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Rate constant units</th>
<th>Initial Cr(VI)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>1000 mg L$^{-1}$</td>
</tr>
<tr>
<td>Nonsterile, +0 mg L$^{-1}$ OC†</td>
<td>s$^{-1}$</td>
<td>6.4 $\times$ 10$^{-8}$</td>
</tr>
<tr>
<td>Nonsterile, +800 mg L$^{-1}$ OC</td>
<td>s$^{-1}$</td>
<td>8.2 $\times$ 10$^{-8}$</td>
</tr>
<tr>
<td>Nonsterile, +4000 mg L$^{-1}$ OC</td>
<td>s$^{-1}$</td>
<td>1.5 $\times$ 10$^{-7}$</td>
</tr>
<tr>
<td>Sterile, +0 mg L$^{-1}$ OC</td>
<td>M$^{65}$ s$^{-1}$</td>
<td>1.0 $\times$ 10$^{-7}$</td>
</tr>
<tr>
<td>Sterile, +4000 mg L$^{-1}$ OC</td>
<td>M$^{65}$ s$^{-1}$</td>
<td>1.2 $\times$ 10$^{-7}$</td>
</tr>
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† Organic carbon.
to 3% of the measured Cr(VI) reduction by Day 200 (Fig. 5).

Aqueous Fe(II) is another potentially important reductant, especially under reducing conditions where its concentrations become appreciable. Chromate reduction at much lower concentrations of aqueous Fe(II) expected under the suboxic to oxic conditions of these soils was estimated through calculating Fe(II) concentrations in equilibrium with soluble iron hydroxides and oxyhydroxides, then applying Fe(II)-dependent rate expressions from the literature. Aqueous Fe(II) concentrations in equilibrium with ferric hydroxide were calculated over a range of suboxic and oxic $p_e$, for pH 7 to 8, using the equilibrium relation at unit water activity (Sposito, 1994):

$$\log(\text{Fe}^{2+}) = 16.4 - p_e - 3pH \quad [5]$$

Solubility products for relatively soluble Fe oxyhydroxides, ferrihydrite, amorphous ferric hydroxide, and soil ferric hydroxide from several other sources (Lindsay, 1979; Macalady et al., 1990; National Institute of Standards and Technology, 1997) yield Fe(II) concentrations that range within 1.5 log units of Eq. [5]. These Fe(II) concentrations were then used to calculate effective first-order Cr(VI) reduction rate constants, $k$, based on the results of Sedlak and Chan (1997) and Pettine et al. (1998). The calculated Fe(II) concentrations are plotted in Fig. 6A. Calculated Fe(II)-based $k$ values are shown in Fig. 6B, along with ranges of Pt electrode-based $p_e$ (from Fig. 3) and $k_{oc}$ (from Fig. 5) representative of the nonsterile soils. Redox measurements obtained with Pt electrodes have been reported to characterize the Fe(II)–Fe(III) couple reasonable well (Macalady et al., 1990; Matia et al., 1991). However, it should be noted that in our case the Pt electrode-based $p_e$ values assigned to the soil columns are very approximate since they come from measurements on mixed, nonequilibrium systems (Lindberg and Runnells, 1984). Furthermore, the relatively high redox potentials are indicative of Fe(II) concentrations that are too low to permit quantitative interpretation (Macalady et al., 1990; Stumm and Morgan, 1996). Nevertheless, the calculations summarized in Fig. 6B indicate that redox-predicted rate constants for Cr(VI) reduction by Fe(II) are consistent with much of the observed Cr(VI) reduction (Fig. 6B).

Several other factors are likely to contribute to the observed Cr(VI) reduction rates. These include redox reactions with soil organic matter and catalytic influences of oxide surfaces (Deng and Stone, 1996; Wittbrodt and Palmer, 1996; Buerge and Hug, 1999) and organo–Fe(II) complexes (Buerge and Hug, 1998). However, detailed comparisons with these previous studies were precluded by lack of information on concentrations of reactive organic functional groups and catalytic surface areas of specific minerals in our soils. Nevertheless, the strong influences of OC concentration and microbial activity on Cr(VI) reduction were demonstrated through comparisons with sterile soils, as discussed next.

**Reduction of Chromium(VI) in Sterile versus Nonsterile Soils**

The extent to which various microbially dependent pathways collectively contribute to Cr(VI) reduction in these soils was estimated through comparisons with reduction in sterile Altimont soils (Fig. 7). These comparisons are presented in terms of the unReduced Cr(VI) fraction remaining at Day 84 since the sterile soils were analyzed at that time. Values for the nonsterile soils were obtained from evaluating first-order fits at Day 84. Comparisons between the various sterile soils appeared to show slightly greater Cr(VI) reduction in response to OC addition [significant at $\alpha = 0.20$ and 0.07 for initial Cr(VI) concentrations of 1000 and 10 000 mg L$^{-1}$, respectively]. Nonsterile soils exposed to 1000 mg L$^{-1}$ Cr(VI) had about three to four times greater reduction than their sterile counterparts, indicating that microbially dependent pathways were dominant. In the systems exposed to 10 000 mg L$^{-1}$ Cr(VI) without additional OC, differences between sterile and nonsterile soils were not significant, indicating negligible microbial influences at these extremely high Cr concentrations. However, these extremely Cr(VI)-contaminated sys-
more acidic conditions tested by Wittbrodt and Palmer (1995, 1996, 1997). Since our study was done in soil columns, the native humic substances are largely associated with the solid phase, not the more accessible solution phase. On the other hand, the soil matrix facilitates Cr(VI)–OC redox reactions through catalytic influences of Fe(III) and other oxides (Deng and Stone, 1996) that were not nearly as abundant in the batch suspensions analyzed by Wittbrodt and Palmer (1995, 1996, 1997). Thus, magnitudes of $k''$ values in our sterile soils are anticipated to differ from those of acidic suspensions of humic substances.

An integral function of Eq. [6] that is linear with respect to time (Wittbrodt and Palmer, 1995, 1996, 1997) was used to obtain values of $[SHA]_0$ and $k''/H_{11630}$ in the following manner. This function has the form:

$$z/H_{11005}/H_{11002}[SHA]_0 k''/H_{11630}t$$

where:

$$z = -[SHA]_0 k''t$$

and the subscript zeroes denote initial concentrations. The variable $z$ is the product of $[SHA]_0$ times the variable $y$ of Wittbrodt and Palmer (1996). Thus $z/t$ gives $[SHA]_0 k''$.

In the sterile soils, $[SHA]_0$ is assumed to represent the sum of added OC and the available portion of native soil OC having an equivalent reactivity as TSB. Thus, a plot of the dependence of $z/t$ on added OC has an $x$ axis intercept at the negative of the equivalent native soil OC concentration. This procedure is illustrated in Fig. 8A, using the data from both levels of $[Cr(VI)]_0$ and with the organic carbon added expressed per unit soil mass. Although the intercept values for the two $[Cr(VI)]_0$ lines should be identical, they are $\pm 26\%$ relative to their average value. Using the average intercept, we estimate the concentration of OC in the original soil having the equivalent reactivity as TSB to be $340 \pm 90 \, \text{mg kg}^{-1}$. This amounts to only $3.4\%$ of the total soil OC concentration, reflecting the likelihood that dissolved TSB is much more available and reactive than most of the native soil OC. The Wittbrodt–Palmer rate constants for the sterile cases were then obtained from their associated $z/t$ and $[SHA]_0$ values (Eq. [7]). These rate constants are presented in Fig. 5 and Table 1.

An alternative estimate of the native soil OC fraction having similar reactivity as TSB can be obtained based on fitting of first-order rate expressions for Cr(VI) reduction in the nonsterile soils. Plots of the dependence of $k''$ on added OC for all nonsterile systems are shown in Fig. 8B. The linearity of each relation simply reflects the use of Eq. [2]. Note the convergence of each line on the $x$ axis (at $k'' = 0$) to a fairly common OC value. These intercept values of $-332 \pm 55 \, \mu\text{g OC}$ per g soil are similar to those obtained based on the Wittbrodt–Palmer model applied to the sterile soils. Thus, using two very different approaches, the portion...
Under the slightly alkaline-oxidizing conditions of these experiments, microbially dependent pathways dominated over abiotic reduction in all of the 1000 mg L\(^{-1}\) initial Cr(VI) systems and in the OC-amended 10,000 mg L\(^{-1}\) initial Cr(VI) system. However, microbial populations in our soils were so low that direct microbial (enzymatic) reactions were not likely to have contributed significantly to the overall Cr(VI) reduction. Aqueous-phase Cr(VI)–Fe(II) redox reactions may be significant if Fe\(^{2+}\) concentrations are in equilibrium with relatively soluble, ferric hydroxide–like phases. Overall, synergistic interactions between microbial activity, OC degradation, Fe\(^{2+}\), and mineral surfaces determine net rates of Cr(VI) reduction in these soils. The demonstrated accelerated reduction of very high levels of Cr(VI) by addition of OC indicate that this approach to in situ remediation may be promising. However, similar studies on different soil and sediment types are needed. Finally, information on long-term stability of artificially reduced Cr(VI) is needed.

### ACKNOWLEDGMENTS

We thank Tracy Letain, Dominique Joyner, and Andrew Mei of Lawrence Berkeley National Laboratory, Matt Newville of the University of Chicago, Bill Rao of the University of Georgia, and the GeoSoilEnviroCARS staff for assistance. We very much appreciate the help of Zuoping Zheng (LBNL), three anonymous reviewers, and the associate editors Dr. Ashok Alva and Dr. Dennis Corwin for their thorough internal and final review comments. Funding was provided through the Basic Energy Sciences, Geosciences Research Program, and the Natural and Accelerated Bioremediation Research (NABIR) program, Biological and Environmental Research, and U.S. Department of Energy, under Contract no. DE-AC03-76SF00098. Use of the Advanced Photon Source was supported by the U.S. Department of Energy, Basic Energy Sciences, Office of Science, under Contract no. W-31-109-Eng-38. Research carried out (in part) at the National Synchrotron Light Source, Brookhaven National Laboratory, which is supported by the U.S. Dep. of Energy, Division of Materials Sciences and Division of Chemical Sciences.

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### CONCLUSIONS

Addition of organic carbon accelerates chromate reduction in soils through abiotic and microbially dependent processes. Ferrous iron, native humic substances, and added OC support abiotic Cr(VI) reduction, which occurs at initial Cr(VI) concentrations as high as 10,000 mg L\(^{-1}\). Under the slightly alkaline-oxidizing conditions of these experiments, microbially dependent pathways dominated over abiotic reduction in all of the 1000 mg L\(^{-1}\) initial Cr(VI) systems and in the OC-amended 10,000 mg L\(^{-1}\) initial Cr(VI) system. However, microbial populations in our soils were so low that direct microbial (enzymatic) reactions were not likely to have contributed significantly to the overall Cr(VI) reduction. Aqueous-phase Cr(VI)–Fe(II) redox reactions may be significant if Fe\(^{2+}\) concentrations are in equilibrium with relatively soluble, ferric hydroxide–like phases. Overall, synergistic interactions between microbial activity, OC degradation, Fe\(^{2+}\), and mineral surfaces determine net rates of Cr(VI) reduction in these soils. The demonstrated accelerated reduction of very high levels of Cr(VI) by addition of OC indicate that this approach to in situ remediation may be promising. However, similar studies on different soil and sediment types are needed. Finally, information on long-term stability of artificially reduced Cr(VI) is needed.


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