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EFFECTS OF FeS ON CHROMIUM OXIDATION MEDIATED BY MANGANESE OXIDIZERS

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ABSTRACT

Reductive immobilization of Cr(VI) has been widely explored as a cost-effective approach for Cr-contaminated site remediation. The long-term stability of the immobilized Cr(III), however, is a concern. Cr(III) is known to be oxidized by Mn oxides chemically and Mn-oxides could be produced through microbially mediated Mn(II) oxidation. This study examined the effect of FeS on Cr(III) oxidation mediated by *Pseudomonas putida*. The results showed that commercial granular FeS did not affect Cr(III) oxidation in the culture of P. *putida* with Mn(II), but freshly precipitated FeS slurry inhibited Cr(III) oxidation. A 10 mg/l of FeS did not inhibit the microbial growth, but delayed the production of Mn oxides, thus postponing potential Cr(III) oxidation. In the presence of excessive FeS slurry, both Cr(VI) and Mn oxides were reduced rapidly. The reduced Cr(III) could not be re-oxidized as long as freshly formed FeS was present, even in the presence of the manganese oxidizers.

INTRODUCTION

Cr(VI) is a known epithelial irritant and human carcinogen (IARC 1990) and is also toxic to many plants, aquatic animals and bacteria (USEPA 1985). Chromium contamination has been found in many industrial and federal sites in the United States, due to accidental leakages and improper disposals associated with its widespread usage in electroplating, cooling and other industries (Nieboer and Nriagu 1988; Ellis, Johnson *et al*. 2002). Many efforts have been devoted to developing technologies for efficient remediation of Cr pollution. One important approach is to detoxify Cr(VI) in soils and

water through its reduction by H_2S, since the reduced Cr species, *i.e.*, Cr(III) compounds, are less mobile and less toxic. However, potential re-oxidation of Cr(III) in the environment needs to be properly assessed. Mn oxides are primary oxidants for Cr(III) oxidation in the soil environment. Even in the systems without oxidative Mn-oxides, the reduced manganese(II) species could potentially be oxidized through microbial processes, because microbes may accelerate the oxidation rate by several orders of magnitude compared to the abiotic Mn(II) oxidation (Nealson, Tebo *et al*. 1988).

Our previous study showed that Cr(III) could be oxidized in water under aerobic conditions in the presence of Mn(II) and a manganese oxidizer, *Pseudomonas putida* (ATCC #23483). However, reductive immobilization of chromium could produce some reducing products. For example, FeS has been found to be the common product in contaminated soils treated with H_2S. It is not well understood how the reducing products affect Cr(III) stability in the presence of Mn(II) and manganese oxidizers. The objective of this study is to investigate the effect of FeS on Cr(III) oxidation mediated by *P. putida*.

MATERIALS AND METHOD

P. putida (ATCC #23483) was grown on a modified LEP medium in HEPES buffer at pH 7.5 (Boogerd and de Vrind 1987; Caspi, Tebo *et al*. 1998). Granular ferrous sulfide was obtained from Merck (100 mesh). A freshly-synthesized ferrous sulfide slurry was prepared following a procedure Butler and Hayes (2001). Microbial growth was monitored spectrophotometrically by measuring the absorbance at 600 nm and calibrated with plate count. The concentration of manganese oxides was measured by the Leuco Berbelin blue assay (Boogerd and de Vrind 1987). Chromate was determined using diphenylcarbazide colorimetric method (Deng and Stone 1996). Total soluble chromium, sulfur, iron and soluble Mn(II) were measured with inductively-coupled plasma spectroscopy (ICP).

RESULTS AND DISCUSSION

Effects of granular FeS: Figure 1 shows the changes in biomass, as well as Mn oxides and Cr(VI) concentrations, a function of time in the culture of *P. putida* grown on the modified LEP medium (Boogerd and de Vrind 1987; Caspi, Tebo *et al*. 1998) without (A) and with (B) granular FeS. The culture contained necessary micro nutrients, Mn(II) (100 µM), and Cr(III) (10 µM). The pH was controlled at 7 with HEPES buffer. The cell counts and concentration profiles were all similar for the two experimental systems. Biomass peaked at the 24th hr and then gradually decreased with time. Cr(VI) was detected following Mn oxides production at the 24th hr, indicating Cr(III) oxidation occurred after the formation of Mn oxides. When an additional 0.20 mM of Cr(III) was added at 240th hr, much faster Cr(VI) production was observed, with corresponding drop of Mn oxides concentration. ICP analysis (data not shown) showed no apparent changes of soluble sulfur and iron during the experiments, indicating that not much FeS was oxidized in the system. This type of FeS, therefore, did not significantly affect Cr(III) oxidation in the *P. putida culture.*

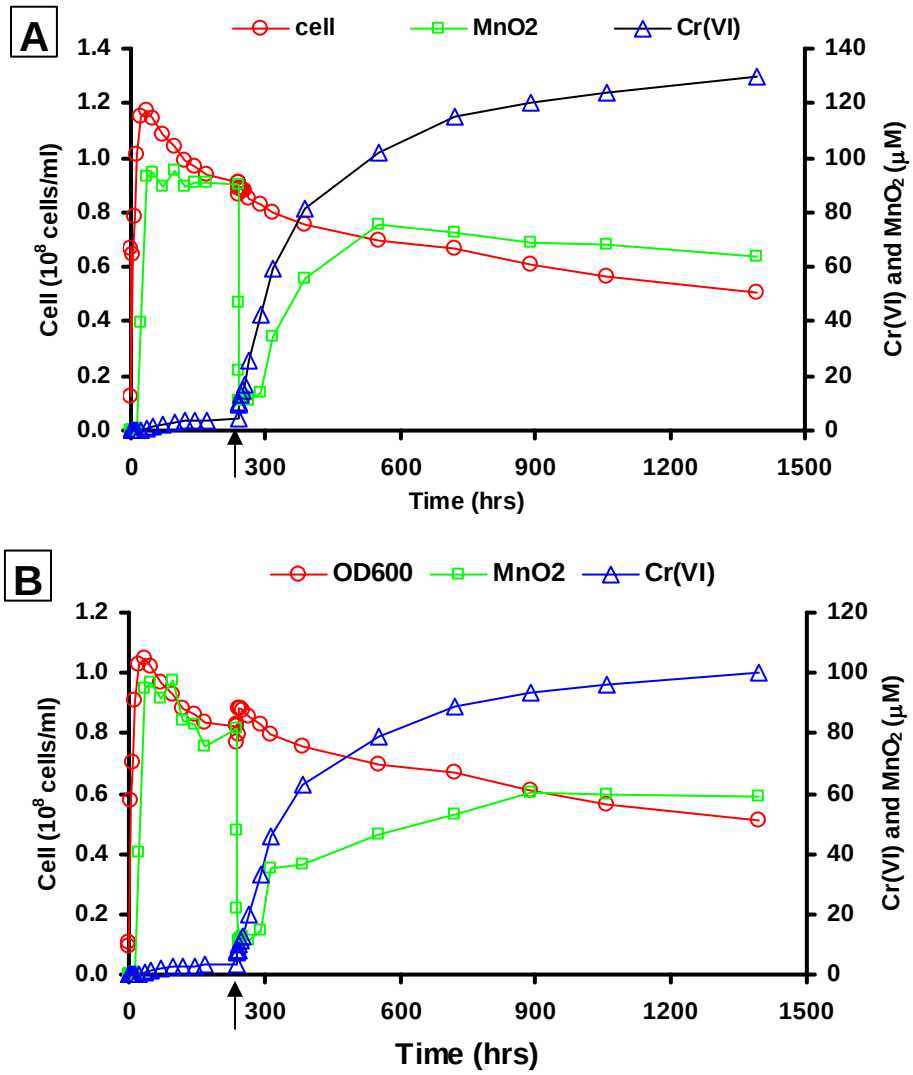

Figure 1. Effects of commercial granular FeS. A, no FeS; B, FeS (120 mg/l) present in the culture of *P. putida.* Arrows indicate time when an additional 0.20 mM of Cr(III) was added.

Effects of freshly-prepared FeS: Figure 2 shows the changes of biomass, Mn, Cr, Fe and sulfur in the culture of *P. putida* grown on the modified LEP medium in the presence of freshly precipitated FeS. The FeS slurry was added into the culture at a final concentration of 10 mg/l after the microorganism had been grown for 5 hrs. After adding the FeS slurry, the biomass continued to increase and reached the maximum of 1.3 x 10^8 cells/ml, suggesting that the microbial growth was not inhibited by the freshly-prepared FeS slurry. However, Mn oxidation production was delayed when compared to the systems with the granular FeS or with no FeS (Fig. 1). No Cr(V) was detected at all during the 600-hrs experiment. ICP results (Fig. 2B) showed that soluble Fe was released and soluble Cr concentration was decreased after addition of fresh FeS. It

appeared that soluble Cr(III) was precipitated through reactions with fresh FeS, decreasing its activity. A yellowish floating product, likely elemental sulfur, was found in the culture. It appeared that the freshly-prepared FeS was oxidized under such an aerobic condition. First step of FeS oxidation led to sulfide oxidation and Fe(II) release and Fe(II) oxidation took place as a second step. Oxidation of FeS in this form could inhibit Mn oxidation by P. *pudia*, therefore protecting Cr(III) from oxidation by Mn oxides.

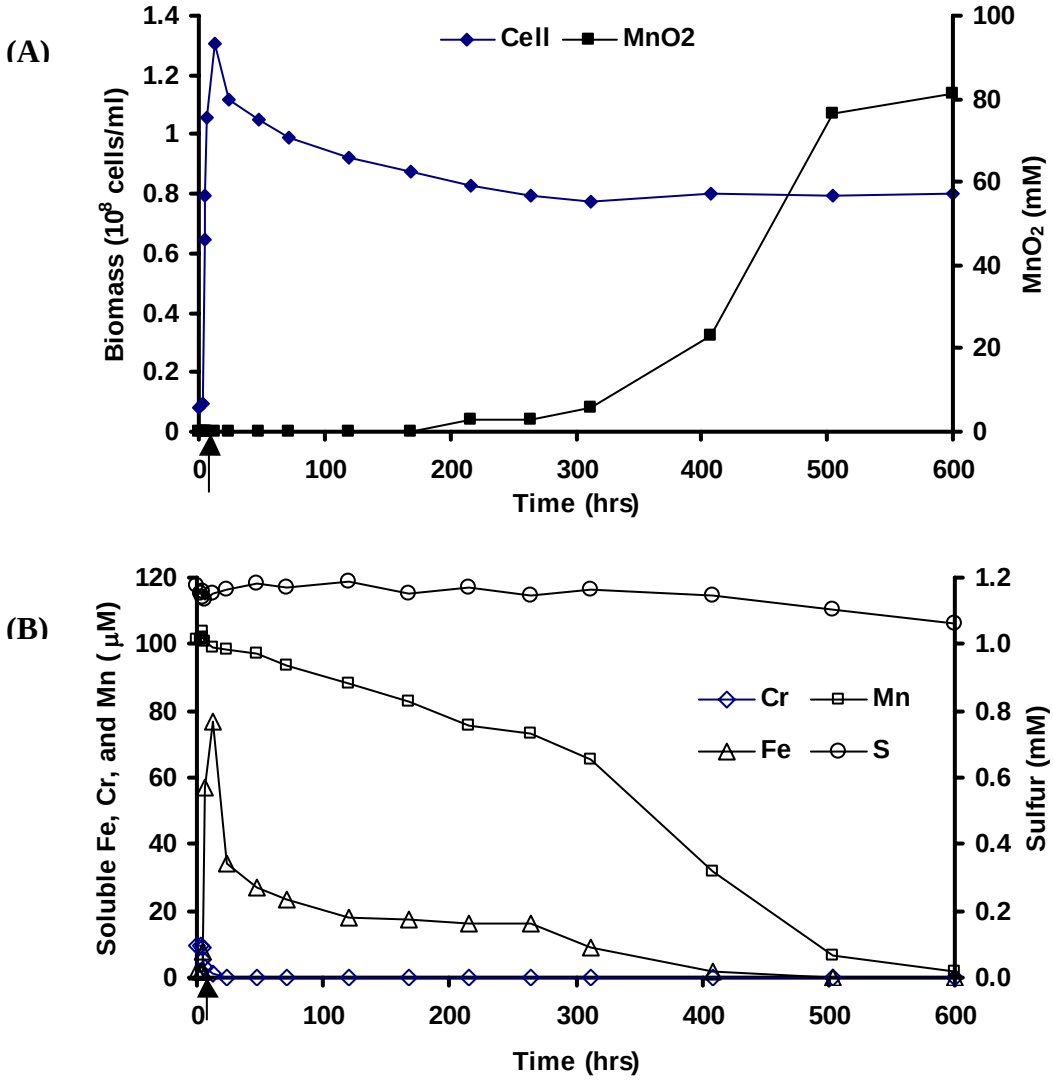

Figure 2. Effects of freshly prepared FeS slurry on Cr(III) oxidation. Arrows indicate addition of freshly precipitated FeS at the 5th hour.

When the fresh FeS was added at a final concentration of 20 mg/l into the culture of *P. putida*, previously formed Mn oxides were reduced immediately to a lower level, while Cr(VI) was rapidly reduced to zero (Fig. 3). No Cr(III) oxidation occurred afterwards

although Mn oxides were present in the experimental system. It suggests that the reduced Cr(III) was unlikely to be oxidized by Mn oxides in the system treated by FeS.

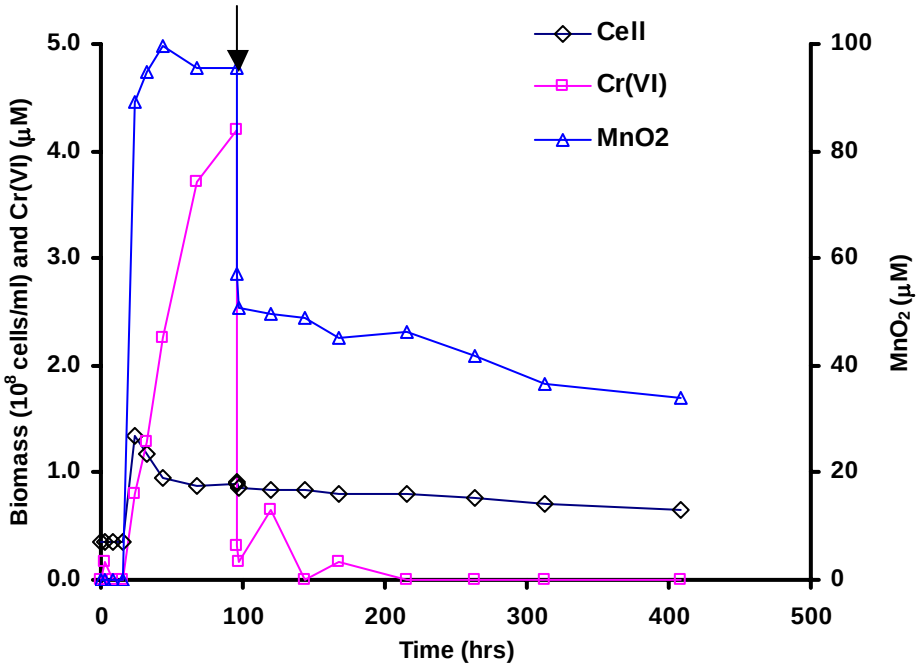

Figure 3. Effects of fresh FeS addition after formation of Mn oxides and Cr(VI) in the culture of *P. putida*. The arrow indicates addition of FeS (20 mg/l) at the 96th hour.

In summary, the presence of freshly precipitated FeS can inhibit Mn(II) oxidation that was mediated by manganese oxidizers, thus protecting Cr(III) from oxidation. Cr(III) reduced from Cr(VI) reduction by sulfide was unlikely to be re-oxidized by Mn oxides, even in the presence of manganese oxidizers.

ACKNOWLEDGEMENTS

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REFERENCES

Boogerd, F. C. and J. P. de Vrind (1987). "Manganese oxidation by Leptothrix discophora." *Journal of Bacteriology* **169**(2): 489-94. Field Reference Number: FIELD Journal Code:2985120R Field Call Number.

Butler, E. C. and K. F. Hayes (2001). "Factors Influencing Rates and Products in the Transformation of Trichloroethylene by Iron Sulfide and Iron Metal." *Environmental Science & Technology* **35**(19): 3884-3891.

Caspi, R., B. M. Tebo, *et al*. (1998). "c-Type cytochromes and manganese oxidation in Pseudomonas putida MnB1." *Applied and Environmental Microbiology* **64**(10): 3549-3555.

Deng, B. and A. T. Stone (1996). "Surface-Catalyzed Chromium(VI) Reduction: Reactivity Comparisons of Different Organic Reductants and Different Oxide Surfaces." *Environ. Sci. Technol.* **30**(8): 2484-2494.

Ellis, A. S., T. M. Johnson, *et al.* (2002). "Chromium isotopes and the fate of hexavalent chromium in the environment." *Science* **295**(5562): 2060-2062.

IARC (1990). Chromium, Nickel and Welding, Monogr. on the Evaluation of Carcinogenic Risks to Humans. Lyons, France, International Agency for Research on Cancer. 49.

Nealson, K. H., B. M. Tebo, et al. (1988). "Occurrence and mechanisms of microbial oxidation of manganese." *Adv. Appl. Microbiol.* **33**: 279-318.

Nieboer, E. and J. O. Nriagu (1988). Chromium in the Natural and Human Environments. J. O. Nriagu. New York, Wiley-Interscience. 20.

USEPA (1985). Ambient Water Quality Criteria for Chromium - 1984. Washington, D.C., Office of Regulations and Standards, Criteria and Standards Division, U.S. Environmental Protection Agency.