Bull. Environ. Contam. Toxicol. (2003) 71:840–847
© 2003 Springer-Verlag New York Inc.
DOI: 10.1007/s00128-003-0212-0

Environmental
Contamination
and Toxicology

Immobilization of Chromium(VI) with Debris of Aquatic Plants

M. J. Hu,[1] Y. L. Wei,[1] Y. W. Yang,[2] J. F. Lee[2]

[1] Department of Environmental Science, Tunghai University, Taichung City, 407 Taiwan, Republic of China
[2] National Synchrotron Radiation Research Center, Hsinchu City, 300 Taiwan, Republic of China

Received: 10 September 2002/Accepted: 18 June 2003

Cr compounds have two common oxidation states: trivalent and hexavalent. Cr(VI) has been found to be carcinogenic and mutagenic (Maria, 1995). On the other hand, Cr(III) is recognized as a micronutrient necessary for human health. In the environment, Cr(VI) can be reduced to Cr(III) by Fe(II), MnO, humic substance, etc; while Cr(III) is oxidized to Cr(VI) by MnO_2. Cr(VI)-containing waste water from the electroplating industry will contaminate soil and underground water if it is not adequately treated. The conventional method for treating Cr(VI)-containing waste water is to reduce Cr(VI) to Cr(III) with reductants such as sulfur-containing compounds that are in reduced form. Cr(III) is precipitated by adjusting the pH of the solution to alkalinic condition (pH~10). Such a Cr(VI)-removing process requires various chemicals. Therefore the alternative method of removing Cr(VI) by using naturally-occurring organic substances (e.g., debris of plants) has great potential for practical application. The plants used to remove heavy metals from waste water should at least have two important characteristics: (i) a considerable sorption capacity, and (ii) a rapid growth rate. Various plants growing in a water system have been used to remove heavy metals from water. Water hyacinth was used to remove Cr(VI) from waste water (Lytle et al., 1998). Cr(VI) was reduced to Cr(III) at the roots and transferred to other parts of water hyacinth (Lytle et al., 1998). In leaves, Cr(III) was considered to be complexed with oxalate ligands (Lytle et al., 1998). Wan and Sun (1999) raised water hyacinth, pistia, duck weed, and floating fern in Cd-containing water and found that water hyacinth could better tolerate Cd toxicity than the others. Cr(III) was found to be more effectively removed by duck weed than Cu(II) (Wahaab, 1995). *Pistia stratiotes* (L.) demonstrated a rapid growth rate, and it removed Cd from 1—6 mg/L aqueous solution (Maine, 2001). X-ray absorption spectroscopy (XAS) technology was used to study the Zn sorbed by *Thlaspi caerulescens* (J. & C. Presl.) and Zn was determined to be complexed with histidine in the root and shoot; while Zn existed as a hydrated form in xylem (Salt et al., 1999). Eu(II)-containing water was remedied with *Eichhornia crassipes* (Mart. Solms.), and Eu(II) was found by SEM technique to be associated with the root surface through the carboxylic group and hydration (Kelley and Mielke, 1999). Lee et al., studied the sorption of heavy metals onto 27 living plants, and concluded that *Najas graminea* was a better candidate for sorbing heavy metals due to its rapid growth rate and high sorption capacity (Lee et al., 1998).

Correspondence to: Y. L. Wei

Some factors need to be carefully considered for using living plants to remove heavy metals from water. These include toxicity of heavy metals to plants, pH and temperature conditions, and supply of nutrients. Therefore dead biomass seems to have more advantages in the removal of heavy metals (Moffat, 1995). The objective of this study is to use the debris of *Najas graminea* and *Pistia stratiotes* to remove Cr(VI) from water.

MATERIALS AND METHODS

Najas graminea and *Pistia stratiotes* were collected from southern Taiwan. They were rinsed with de-ionized water, dried at 105±5℃ in an oven, and finally ground to a size smaller than 50 mesh. Experiments of Cr(VI) sorption onto three plant debris types (whole plant of *Najas graminea*, leaves of *Pistia stratiotes*, and roots of *Pistia stratiotes*) were carried out as follows. Each 100 mL Cr(VI)-containing solution (500, 1000, 2000, or 4000 mg/L) was end-to-end mixed with 5 g debris in a 100 mL PE bottle at a rotating speed of 30 rpm. The mixing time was 0.5, 1, 2, 4, 12, 36, or 48 hr. After the specified time, the mixture was solid-liquid separated using a N_2-pressurized filtration process. Cr concentration in the liquid phase was determined with a flame atomic absorbency spectrometer (FAAS) (Z-6100, Hitachi, Japan) that used C_2H_2/air flame for the atomizer, and the solid phase (Cr-sorbed debris) was dried at 30±5℃ until the weight remained constant. The change of morphology of the dried biomass debris prior to and after Cr(VI) sorption was studied with scanning electron microscopy (SEM) (JXA-840, JEOL, Japan) and XAS (Wiggler BL 17C, Synchrotron Radiation Research Facility, Taiwan, which had a storage energy of 1.5 GeV and a beam current of 120-200 mA). An XAS spectrum can be divided into three regions: (i) the pre-edge region, (ii) the X-ray absorption near edge structure (XANES), and (iii) the extended X-ray absorption fine structure (EXAFS). The XANES region contains information such as electronic state and intensity of d-orbital occupation. The EXAFS region can be analyzed for information on the molecular environment of the selected target element (e.g. Cr in this study). Coordination number, interatomic distance, and degree of array order of each coordinated shell can generally be derived through the analysis of the EXAFS region. Spectra of both XANES and EXAFS regions are collected in an XAS measurement. The collected XAS spectra were analyzed using WinXAS 2.0 software (Ressler, 1998) to determine Cr speciation in samples.

The quality control (QC) approach for the measurement of Cr in the liquid phase was as follows. Cr(III) stock solution (1000 mg Cr^{3+}/L, Merck) was diluted into five different concentrations (0—4.0 mg/L), followed by the measurement of Cr with the FAAS to produce a calibration curve. Triplicate measurements for each concentration were made in this study. The correlation coefficient (r^2) for any calibration curve was required to be greater than 0.995. A QC standard of certified Cr concentration was measured with the FAAS to check the validity of the calibration curve for each 20 measurements of the unknown samples. If the recovery yield of the measured QC standard was within 80—120%, the calibration curve was acceptable; otherwise, a new one was re-constructed and the last 20 unknown samples were re-measured for their Cr concentrations. The method detection limit (MDL) was determined as follows. An unknown sample was diluted to a level where the FAAS signal-to-noise ratio was just greater than three, and then the diluted unknown sample was measured by the FAAS in 12 replicates. Three times the standard deviation of the 12 replicate measurements was considered as the absorbency for MDL. The MDL for Cr was determined to be 32 μg Cr/L.

RESULTS AND DISCUSSION

Figure 1 shows the Cr sorption quantity with increasing sorption time from water containing 500—4000 mg/L chromium by *N. graminea*, and leaves and roots of *P. stratiotes*. From a quick glance at this figure, it is clear that the Cr sorption capacity was in the following decreasing order: roots of *P. stratiotes* > leaves of *P. stratiotes* > *N. graminea*. It appears that after 24-hr sorption, the Cr sorption by each debris type reached a solid-liquid equilibration, with a few exceptions. As shown in Table 1, these sorption data have been modeled with a Langmuir isotherm, and the correlation coefficient (r^2) was 0.943—0.970. However, the Langumir fit did not give a reasonable maximum sorption capacity. In contrast, the fitting of sorption data with the Freundlich equation resulted in reasonable results. The Freundlich equation is:

$$q_e = KC_e^{1/n}$$

where q_e : amount of sorbate adsorbed per unit weight of adsorbent (mg/kg)
K : the adsorption capacity (L/kg)
C_e: the equilibrium concentration of sorbate in solution (mg/L)
n : the adsorption intensity

The fitting results are indicated in Table 2. Both "K" and "n" values are in the following decreasing order: *Pistia* roots > *Pistia* leaves > *N. graminea*. This suggests that *Pistia* root has the strongest sorption capability, with *N. graminea* the weakest and the *Pistia* leaves in between.

Table 1. Fitting results based on the Langmuir isotherm

sample	correlation coefficient (r^2)	Q (mg/kg)
N. graminea	0.968	-
leaves of *P. stratiotes*	0.970	-
roots of *P. stratiotes*	0.943	-

-: cannot be reasonably calculated (negative values)

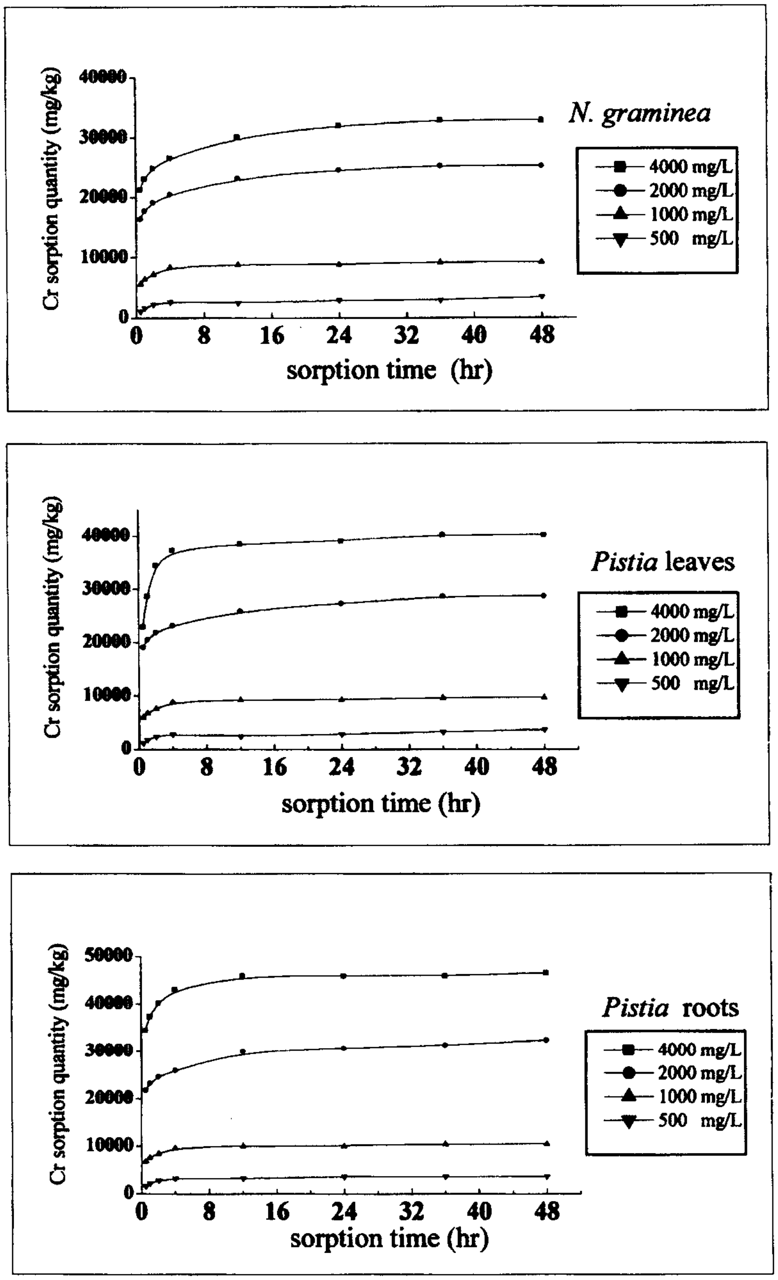


Figure1. Effect of sorption time on Cr sorption quantity by debris of *N. gramines*, Pistia leaves, and Pistia roots from 500—4000 mg Cr/L aqueous solution.

Table 2. Fitting results based on the Freundlich isotherm

sample	correlation coefficient (r^2)	K	n
N. graminea	0.916	0.84×10^{-5}	0.64
leaves of *P. stratiotes*	0.927	1.36×10^{-5}	0.95
roots of *P. stratiotes*	0.924	1.79×10^{-5}	1.01

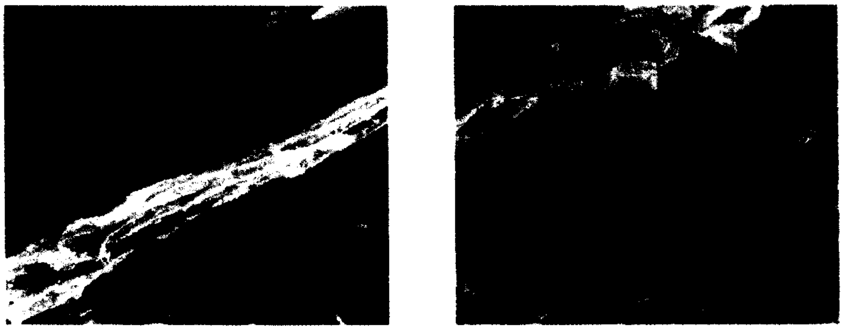
Figure 2 presents the morphology change of *N. graminea*, leaves of *P. stratiotes*, and roots of *P. stratiotes* prior to and after Cr(VI) sorption for 48 hr. It shows that the morphology of leaves of *P. stratiotes* was considerably changed after the Cr(VI) sorption. This was probably due to the oxidation capability of Cr(VI). In contrast, the morphologies of *N. graminea* and roots of *P. stratiotes* were not significantly altered after Cr(VI) sorption.

Figure 3 shows the XANES fitting results (left column) and their corresponding EXAFS (right column) simulation for the *N. graminea*, leaves of *P. stratiotes*, and roots of *P. stratiotes* sorbing Cr(VI) in the 2000 mg/L solution for 48 hr. The XANES spectra indicate an absence of the pre-edge peak that is a characteristic of Cr(VI) XANES specta (Bajt et al, 1993). During the 48-hr sorption, Cr(VI) onto all debris was reduced to Cr(III). To determine Cr speciation in these samples, three reference compounds were used: CrO_3, $Cr(OH)_3$, and $Cr_3(OH)_2(OOCCCH_3)_7$. CrO_3 was selected as a reference compound because it was used to prepare the Cr(VI) solution in this study. $Cr(OH)_3$ was selected because previous literature (Jardine et al, 1999) reported its existence in research related to the interaction between Cr(VI) and organic substance. The selection of $Cr_3(OH)_2(OOCCCH_3)_7$ was a difficult decision. In the early stage of this study, we found that XANES fitting was always unsuccessful by using various combinations of CrO_3, $Cr(OH)_3$,and Cr_2O_3, thus an effort was made to seek for various kinds of organic Cr(III) compounds. $Cr_3(OH)_2(OOCCCH_3)_7$ turned out to be the most suitable one in the aspects of the performance of XANES fitting and of the functional groups it contains. Previous literature has recognized that carboxylic and alcoholic groups were the most common functional groups in plants or humic substance that sorb heavy metal ions. Although there is minor deviation of the simulated EXAFS from the experimental one (see Figure 3), we suggest that $Cr(OH)_3$ precipitated onto these three debris types generally accounts for less than 50% of total Cr, while organic Cr(III) represents about 50—70% of the total sorbed Cr. We suggest that the organic Cr(III) bonds to the carboxylic and hydroxyl functional groups of the debris through an ion-exchange mechanism following its formation from Cr(VI) reduction by the debris. It should be noted that the minor deviation of EXAFS simulation might have been due to a greater noise signal at higher *k* (see Figure 3).

In conclusion, the debris of *N. graminea*, leaves of *P. stratiotes*, and root of *P. stratiotes* effectively sorbed Cr(VI) from Cr(VI) solution. Nearly all sorbed Cr(VI) was then reduced to Cr(III) by the debris. We suggest that about 53-67% of the Cr(III) was in an organic form, while the rest was in $Cr(OH)_3$ that was precipitated on the debris

(a) neat *N. graminea* (*a*) Cr(VI)-sorbed *N. graminea*

(b) neat *Pistia* leaves (*b*) Cr(VI)-sorbed *Pistia* leaves



(c) neat *Pistia* roots (*c*) Cr(VI)-sorbed *Pistia* roots

Figure 2. Morphologies of debris of *N. graminea*, *Pistia* leaves, and *Pistia* roots prior to (a-c) and after (*a-c*) Cr(VI) sorption.

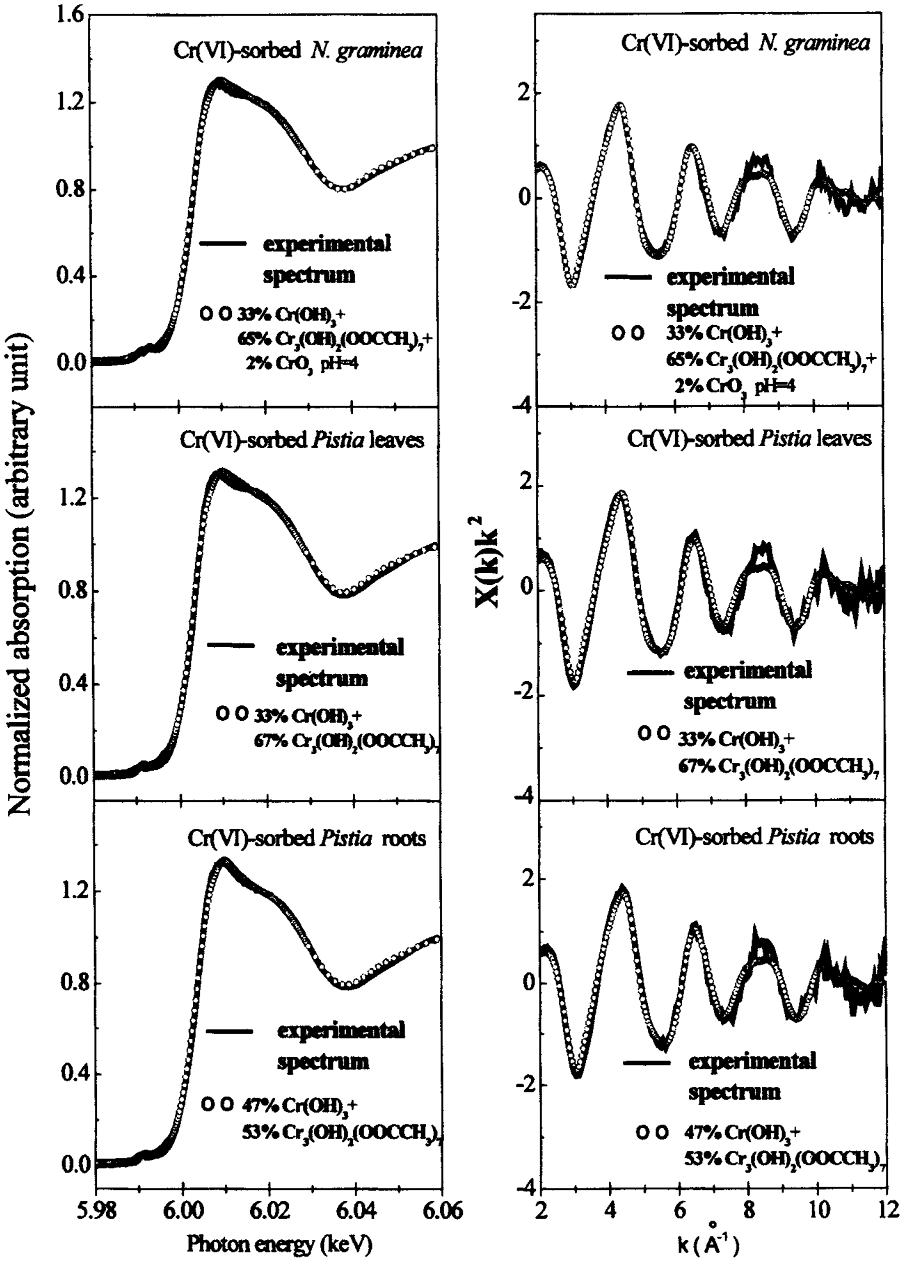


Figure 3. Fitting of XANES spectra and simulation of EXAFS spectra for the debris of *N. graminea*, *Pistia* leaves, and *Pistia* roots sorbed with Cr(VI).

Acknowledgments. We thank the NSRRC staff for their XAS assistance and the National Science Council of the Republic of China (grant number: NSC 90-2211-E-024-003). Presented at the first International Conference on Pollution Eco-Chemistry & Ecological Processes, Shenyang, P. R. China August 26-31, 2002.

REFFERENCES

Bajt S, Clark SB, Sutton SR, Rivers ML, Smith JV (1993) Synchrotron X-ray Microprobe Determination of Chromate Content Using X-ray Absorption Near-edge Structure. Anal Chem 65:1800-1804

Jardine PM, Fendorf SE, Mayes MA, Larsen IL, Brooks SC, Bailet WB (1999) Fate and Transport of Hexavalent Chromium in Undisturbed Heterogeneous Soil. Environ Sci Technol 33:2939-2944

Kelley C, Mielke RE (1999) Adsorption of Eu(II) onto Roots of Water Hyacinth. Environ Sci Technol 33:1439-1443

Lee CL, Tsen CW, Hsu CH, Chiau AA (1998) Heavy Metals Sorption by Aquatic Plant in Taiwan. Bull Environ Contam Toxicol 61:497-504

Lytle CM, Lytle FW, Yang N, Qian JH, Hansen D, Zayed A, Terry N (1998) Reduction of Cr(III) to Cr(VI) by Wetland Plants: Potential for In Situ Heavy Metal Detoxification. Environ Sci Technol 32:3087-3093

Maine MA, Duarte MV, Sune NL (2001) Cadmium Uptake by Floating Macrophytrs. Wat Res 35:2629-2634

Maria CG (1995) Toxic and Mutagenic Effects of Chromium(VI). Polyhedron 15:3667-3689

Moffat AS (1995) Plants Proving Their Worth in Toxic Metal Clean up. Environ Biol 269: 302-303

Ressler T (1998) WinXAS: A Program for X-ray Absorption Spectroscopy Data Analysis under MS-Windows. J Synchrotron Radiat 5:118-122

Salt DE, Prince RC, Baker AJM, Raskin I, Pickering IJ (1999) Zinc Ligands in the Metal Hyperaccumulator *Thlaspi caerulescens* as Determined Using X-ray Absorption Spectroscopy. Environ Sci Technol 33:713-717

Wahaab RA, Lubberding HJ, Alaerts GJ (1995) Copper and Chromium(3) Uptake by Duckweed. Wat Sci Technol 32:105-110

Wan KC, Sun EJ (1999) Response and Absorption of Cadmium by Four Water Plant. J Environ Protect Soc Republic of China 22:11-20