

**CARACTERIZACIÓN DE LA ABSORCIÓN DE CU, NI Y ZN POR EL MÉDANO (*S. kali*) PARA PROPÓSITOS DE FITORREMEDIACIÓN****CHARACTERIZATION OF CU, NI AND ZN UPTAKE IN TUMBLEWEED (*S. kali*) FOR PHYTOREMEDIATION PURPOSES**

G. de la Rosa<sup>1,2\*</sup>, J. R. Peralta-Videa<sup>3</sup>, J. G. Parsons<sup>3</sup>, G. Cruz-Jimenez<sup>1</sup>,  
I. Cano-Aguilera<sup>1</sup> y J. L. Gardea-Torresdey<sup>2,3</sup>

<sup>1</sup>Facultad de Química, Universidad de Guanajuato, Col. N. Alta s/n, C.P. 36050, Guanajuato, Gto., México

<sup>2</sup>Environmental Science and Engineering, Ph.D. Program

<sup>3</sup>Chemistry Department, The University of Texas at El Paso, El Paso, TX 79968, U.S.A.

Recibido 1 de Julio 2005; Aceptado 4 de Diciembre 2005

**Resumen**

Estudios previos han demostrado que *Salsola kali*, una planta desértica, es capaz de absorber cantidades significativas de Pb y Cr. Más aún, se ha propuesto como una posible planta hiperacumuladora de Cd. Ya que ésta especie ha mostrado ser una posible fitorremediadora, el objetivo de esta investigación fue obtener información más completa acerca de la tolerancia hacia diferentes metales pesados y los posibles mecanismos bioquímicos. En ésta investigación las semillas se germinaron y las plantas se crecieron por 15 días en un medio de agar conteniendo 0-40 mg L<sup>-1</sup> Cu(II), Ni(II) or Zn(II). Los resultados indicaron que Zn benefició el crecimiento mientras que Ni fue extremadamente tóxico. El único metal que se acumuló en cantidades considerables fue el Cu (1300 mg kg<sup>-1</sup> en parte aérea). Los estudios de rayos X sugieren que la traslocación de Zn y Ni se lleva a cabo sin cambio en el estado de oxidación y ambiente de coordinación, lo que no sucede con Cu. Se encontró que en los tejidos de las plantas Zn se enlazó a nitrógeno y oxígeno, Cu a azufre y oxígeno y Ni a nitrógeno y oxígeno. Los resultados que se presentan en este papel indican que *S. kali* puede considerarse una opción interesante para la remediación de suelos conteniendo cantidades moderadas de Cu, Ni y Zn.

**Palabras clave:** *S. kali*, Cu, Ni, Zn, fitorremediación, estudios de XAS.

**Abstract**

Tumbleweed plants (*S. kali*) accumulate considerable amounts of Pb, Cd and Cr in their tissues. Moreover, this plant has been proposed as a potential Cd hyperaccumulator. In order to obtain a more complete profile for metal accumulation and tolerance, experiments were conducted in agar media individually contaminated with 0-40 mg L<sup>-1</sup> of Cu(II), Ni(II), and Zn(II). Zn was rather beneficial as it promoted root and shoot elongation as well as biomass accumulation. Plants exposed to Cu accumulated 1300 mg kg<sup>-1</sup> dry weight in the aerial plant part, indicating a potential hyperaccumulation. X-ray absorption spectroscopic (XAS) studies showed that tumbleweed plants absorb and move Zn and Ni from the roots to the leaves without changes in oxidation state and coordination environment; however, Cu is transported to the aerial part probably bound to different compounds in different plant tissues. Oxygen/nitrogen were identified as ligands for Zn; Cu was observed complexed to sulfur and oxygen, while Ni was bound to nitrogen/oxygen. The results obtained in this research indicate that *S. kali* can be considered as an option to cleanup polluted soils containing moderate amounts of Cu, Ni, and Zn.

**Keywords:** *S. kali*, Cu, Ni, Zn, phytoremediation, XAS studies.

**1. Introducción**

Heavy metal contaminated soils are usually remediated using leaching, solidification, chemical reduction, and electrokinetics, among other techniques (Barceló and Poschenrieder, 2003). However, the use of these techniques is sometimes limited due to their high cost and potential toxicity. In recent years phytoremediation -the use of living plants for the remediation of polluted soils and

waters- has been recognized as an excellent alternative (Brooks, 1998). Recent reports indicate that phytoremediation is already being used to clean up polluted soils in Europe and the U.S.A. (van der Lelie et al., 2001, Bañuelos et al., 2005).

A plant suitable for phytoremediation purposes must have a high biomass production and hyperaccumulate metals, among others (Baker and Brooks, 1989). Unfortunately, most hyperaccumulators identified up to date produce low

\*Autor para la correspondencia: E-mail: delarosa@quijote.ugto.mx  
Tel: (473)7320006 ext. 8139; Fax: (473)7320006 ext. 8108

biomass (Barceló and Poschenrieder, 2003). On the other hand, only a few of those are desert plant species (Gardea-Torresdey et al., 2004; Reeves and Brooks, 1983; Aldrich et al., 2003). Therefore, scientists are constantly working in the identification of potential phytoremediators in different areas of the world.

Copper (Cu), nickel (Ni), and zinc (Zn) are some plant micronutrients that form part of the structure of important biomolecules (Salt et al., 2002; Rengel, 1999). However, high concentrations of these elements in the growth medium may become toxic for non-tolerant plants. Nevertheless, tolerant plants may concentrate these elements far above from ordinary requirements, and they may be used for the reclamation of metal laden soils. On the other hand, the understanding of metal accumulation and tolerance mechanisms is of extreme importance for the successful development of a phytoremediation technology. In the study of such mechanisms, different techniques can be used. Among those, X-ray absorption spectroscopy (XAS) presents several advantages over others since no sample pretreatment is needed. XAS provides information about oxidation states and coordination environments of the elements analyzed (Gardea-Torresdey et al., 2005a).

Previous studies have demonstrated that tumbleweed (*Salsola kali*), a drought resistant shrub, is a potential hyperaccumulator of Cd and Cr (de la Rosa et al., 2004; Gardea-Torresdey et al., 2005b). In addition, it has been found that Cd in plant tissues is present as Cd-S and Cd-O (de la Rosa et al., 2004). Also, it has also been demonstrated that arsenate is biotransformed by this plant to produce the less toxic As(III)-sulfur complex (De la Rosa et al., 2006). The purpose of the present research was to determine the uptake ability of tumbleweed plants germinated and grown in a solid media containing individual concentrations of Cu(II), Ni(II), and Zn(II). XAS experiments were also performed in order to obtain information about the possible biotransformation of these metals by tumbleweed plants. The information presented herein provides a more complete profile for heavy metal accumulation, tolerance, and uptake mechanisms in *S. kali* plants.

## 2. Materials and methods

### 2.1 Media preparation and seed planting

Tumbleweed seeds were obtained from wild plants growing in El Paso, Texas. Growth media was prepared using a modified Hoagland nutrient solution as previously described by Peralta et al. (2001). Metals were supplied individually as  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ ,  $\text{Ni}(\text{NO}_3)_2$ , and  $\text{Zn}(\text{NO}_3)_2$ . Agar ( $5\text{g L}^{-1}$ ) was added and subsequently each medium was poured into Mason jars for sterilization (30 min,  $120^\circ\text{C}$ ,  $1\text{ kg cm}^{-2}$ ) using a Market Forge Sterilmatic Autoclave. After sterilization, the media were

allowed to solidify and  $35 \pm 5$  tumbleweed seeds disinfected with captan were planted under a sterile environment. Plants were allowed to grow for 15 days in a growth chamber ( $24^\circ\text{C}$ , 12 h dark/light cycle at  $19\text{ }\mu\text{mol m}^{-2}\text{ s}^{-1}$ ). Treatments consisted of 3 replicates for statistical purposes (Data was analyzed using SPSS Version 11.0).

### 2.2 Effect of Cu(II), Ni(II), and Zn(II) on tumbleweed growth

Ten plants per replicate per treatment were randomly selected and measured. Plants were rinsed using 0.01 M  $\text{HNO}_3$  and washed with DI water (deionized water) to remove any metal adsorbed to the roots (Aldrich et al., 2003). Subsequently, roots, stems and leaves were cut to separately evaluate biomass and metal uptake. Samples were oven dried at  $70^\circ\text{C}$  for 72 h, weighed and data analyzed.

### 2.3 Elemental analysis in plant tissue

Dried samples were digested with 6 mL trace pure  $\text{HNO}_3$  using a CEM microwave oven following the USEPA 3051 method (Kingston and Jassie, 1988). After digestion, the sample volume was adjusted to 10 mL using double deionized water. Elemental analysis was performed using an Inductively Coupled Plasma Optical Emission Spectrometer (ICP/OES) Optima 4300 DV. Spiked standards were used for QA/QC (Quality assurance/Quality control). The blank and four calibration standards were used to acquire the calibration curve ( $r^2 \geq 0.999$ ).

### 2.4 X-ray absorption spectroscopy experiments (XAS)

Treated plants were randomly selected, washed, and cut as previously described. The samples were then submerged in liquid nitrogen for 45 min and lyophilized for 2 days. Afterwards, samples were ground, loaded into 1-mm thick aluminum sample holders and sealed using Kapton® tape. XAS experiments were performed at Stanford Synchrotron Radiation Laboratory (Stanford University, Menlo Park, CA, USA) on Beam Line 7-3. Fluorescence spectra were taken using a 13-Ge detector, a 3 GeV beam energy, a 60-100 mA beam current, and a Si(220) double crystal monochromator. The beam line was detuned by 50%, and 2-4 scans were averaged to improve the signal to noise ratio. The XAS data analysis was performed using the WinXAS software as described by De la Rosa et al. (2004).

### 3. Results and discussion

#### 3.1 Evaluation of plant growth

Root and shoot elongation of plants treated with 0-40 mg L<sup>-1</sup> of either Cu(II), Ni(II), or Zn(II) was evaluated at the end of the experimental period (Figs. 1 a and b). As observed, the data indicated that tumbleweed plants tolerated well the Zn concentration of 40 mg L<sup>-1</sup>. In addition, it was observed that all Zn treatments significantly promoted root and shoot elongation as compared to the Zn-free plants. These data may imply that Zn is rather beneficial for tumbleweed plants at the concentrations used in this study. On the other hand, plants were unable to survive in Ni(II) and Cu(II) treatments above 10 and 20 mg L<sup>-1</sup>, respectively. A hormetic effect (Calabrese and Baldwin, 2003) in shoot elongation was observed in plants exposed to 5 mg L<sup>-1</sup> Cu(II) and Ni(II). A positive significant correlation for Ni(II) concentration in growth media and root size was obtained (0.820, Pearson, 2-tailed,  $P < 0.01$ ). However, after this concentrations plants died. In addition, 10, and 20 mg Cu(II) L<sup>-1</sup> significantly affected root and shoot plant growth. Peralta-Videa et al. (2002) also found that Zn(II) positively affected root and shoot elongation in alfalfa plants grown in soils individually contaminated with several metals including Ni(II), Cu(II), and Zn(II). In addition Peralta-Videa et al. (2001) reported that in alfalfa plants, the presence of Zn(II) at 40 mg L<sup>-1</sup> in the growth media significantly increased root elongation, while Ni(II) and Cu(II) produced a reduction in root size.

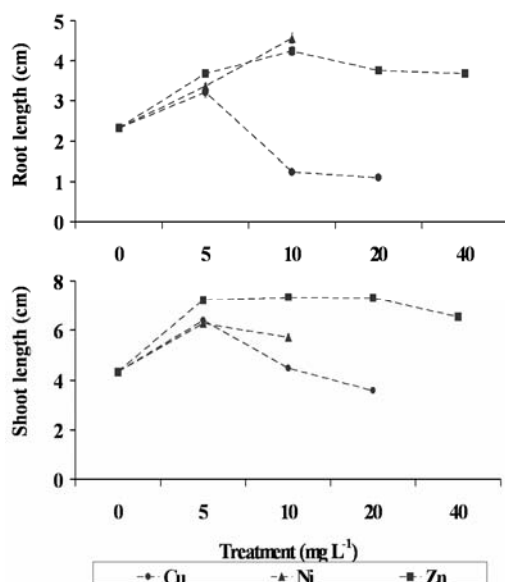


Fig. 1. (a) Roots and (b) Shoots elongation in tumbleweed plants germinated and grow in Cu(II), Ni(II) concentration of 0-40 mg L<sup>-1</sup>. Data are means of 30 plants  $\pm$  S.E.

As for biomass production, roots were the most sensitive in terms of metals treatments and concentrations, while biomass production in leaves was not significantly affected (Table 1). The presence of 40 mg Zn L<sup>-1</sup> produced a 33% increment in root biomass, as compared to controls. In addition, in the 10 and 20 mg L<sup>-1</sup> treatments, Zn was also beneficial for biomass production in stems, as compared to Ni and Cu. Table 1 and Fig. 1 (a) also show that in the 10 mg L<sup>-1</sup> treatment, Ni positively affected both, root biomass and root elongation.

Table 1. Biomass production in tumbleweed plants treated with different concentrations of Cu(II), Ni(II), and Zn(II). Data are means of 30  $\pm$  SE. Different letters indicate significant differences\*

Treatment (mg L <sup>-1</sup> )	Root weight (mg)	Stem weight (mg)	Leaf weight (mg)
0	3.6 $\pm$ 0.2 <sub>B</sub>	7 $\pm$ 0.5	5 $\pm$ 0.03
Zn 5	3.8 $\pm$ 0.3 <sub>B</sub>	6.5 $\pm$ 0.3	4.8 $\pm$ 0.3
Cu 5	3.3 $\pm$ 0.3	7 $\pm$ 0.5	6.3 $\pm$ 1
Ni 5	4.3 $\pm$ 0.4	5.7 $\pm$ 0.5	5 $\pm$ 0.1
Zn 10	3.3 $\pm$ 0.36 <sub>BB</sub>	7.4 $\pm$ 0.08 <sub>a</sub>	5 $\pm$ 0.5
Cu 10	3 $\pm$ 0.1 <sub>b</sub>	6 $\pm$ 0 <sub>b</sub>	4.3 $\pm$ 0.3
Ni 10	5 $\pm$ 0.3 <sub>a</sub>	5 $\pm$ 0.4 <sub>b</sub>	5.1 $\pm$ 0.3
Zn 20	3.8 $\pm$ 0.2 <sub>B</sub>	7.7 $\pm$ 0.1 <sub>a</sub>	5 $\pm$ 0.3
Cu 20	3.5 $\pm$ 0.03	5.5 $\pm$ 0.6 <sub>b</sub>	4.1 $\pm$ 0.4
Zn 40	5.4 $\pm$ 0.4 <sub>A</sub>	7.8 $\pm$ 0.4	5.8 $\pm$ 0.5

\*Uppercase letters indicate significant differences in biomass accumulation among same metal treatments. Lowercase letters indicate significant differences in biomass accumulation in plants treated with different metal at the same concentration.

#### 3.2 Evaluation of Zn, Cu, and Ni uptake by tumbleweed plants

##### Zn uptake

Fig. 2 displays the results of (a) Zn, (b) Cu, and (c) Ni uptake by tumbleweed plants. Similar to Cd uptake by this plant (de la Rosa et al., 2004), the content of Zn in root tissues increased as metal concentrations in the growth media increased. In every treatment, roots concentrated more Zn, followed by stems and leaves. As the Zn level increased in the agar, Zn concentration in roots increased proportionally. However, Zn transport to stems and leaves did not follow the same trend. These results may imply that a restriction mechanism for Zn translocation is present in *S. kali* or that this metal is being used by the plant at the root level. Similar results were found in alfalfa plants (Peralta et al., 2001). Other plant species have shown a similar Zn uptake ability. Matthews et al. (2004) reported the accumulation of 3 600, and 1 700 mg Zn kg<sup>-1</sup> in dry roots and leaves tissues, respectively, in the wetland grass *G. fluitans* exposed to 40 mg L<sup>-1</sup> Zn. According to Brooks (1998), a Zn hyperaccumulator

stores 1% or more of this metal in the aerial part, preferably the leaves. This means that in the 40 mg Zn L<sup>-1</sup> treatment, tumbleweed only accumulated one tenth the amount required for hyperaccumulation. However, since no phytotoxic effects were identified in Zn treated plants, it is possible that tumbleweed plants can be used in the phytostabilization of soils containing higher amounts of this metal (Barceló and Poschneider, 2003).

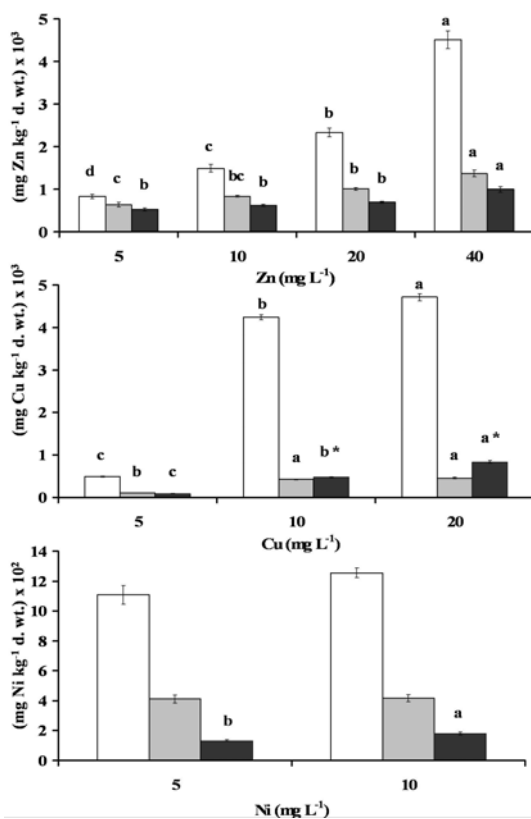


Fig. 2. (a) Zinc (b) Cu, and (c) Ni concentration in tumbleweed roots  $\square$ , stems  $\square$ , and leaves  $\blacksquare$ . Data are mean  $\pm$  S.E (n=3). Different letters indicate significant differences in metal content in the same plant tissues among different treatments ( $P < 0.05$ ).

#### Cu uptake

As previously shown, tumbleweed did not tolerate Cu concentrations in the growth media above 20 mg L<sup>-1</sup>. In Fig. 2(b), an asterisk indicates statistically significant differences in Cu content in stems and leaves. Surprisingly, at 10 and 20 mg Cu L<sup>-1</sup> levels, Cu was translocated to the leaves. The translocation is more easily observed in the 20 mg Cu L<sup>-1</sup> treatment. In this treatment, leaves tissues were able to store almost twice the amount found in the stems. The threshold for Cu hyperaccumulation is of 1000 mg kg<sup>-1</sup> d.wt. (Brooks, 1998). The results reported herein indicate that tumbleweed shoots (stems and leaves) accumulated about 1300 mg Cu kg<sup>-1</sup> d.wt. However, this amount is less than the one found in

shoots of alfalfa plants exposed to Cu concentrations similar to those used in the present study (4100 mg Cu kg<sup>-1</sup>) (Peralta-Videa et al., 2001). On the other hand, tumbleweed concentrated more Cu than *E. splendens*. This plant only accumulated 1751 and 15 mg Cu kg<sup>-1</sup> d.wt. in roots and shoots when grown in soils containing Cu extractable concentrations of 114 mg kg<sup>-1</sup>, which is about 5 times the highest Cu concentration used in these experiments (Jiang et al., 2004). According to the data obtained herein, tumbleweed could also potentially be used to clean up Cu polluted soils containing bioavailable concentrations below 20 mg L<sup>-1</sup>.

#### Ni uptake

No statistically significant differences ( $P < 0.05$ ) were found for Ni concentrations in roots and stems in the treatments where plants survived (Fig. 2(c)). On the other hand, Ni concentrations in leaves of plants exposed to 10 mg Ni L<sup>-1</sup> were significantly higher ( $P < 0.05$ ) than the concentration in leaves treated with 5 mg Ni L<sup>-1</sup>. However, the difference was not significant (an increase of 26% in Ni concentration was observed). Plants grown with 10 mg Ni L<sup>-1</sup> concentrations contained in shoots (stems plus leaves) less than 50% the amount of Ni found in roots. Peralta-Videa et al. (2001) reported that alfalfa plants grown in agar containing 10 mg Ni L<sup>-1</sup> accumulated 1913 and 713 mg Ni kg<sup>-1</sup> d.wt. in roots and shoots tissues, respectively. These amounts were 2 and 1.2 times the Ni concentrations accumulated in roots and shoots of tumbleweed plants.

#### 3.3 XAS experiments

##### XAS studies of tumbleweed plants exposed to Zn(II)

Fig. 3(a) displays the XANES spectra of tumbleweed roots, stems, and leaves tissues treated with 40 mg Zn(II) L<sup>-1</sup> as well as the agar-Zn media. Fig. 3(b) shows the spectra for the Zn model compounds used in this study. The results obtained in these experiments demonstrated that Zn(II) remained in the same oxidation state in agar, roots, stems, and leaves after the period of treatment (edge energy at approximately 9.67 keV). On the other hand, by comparing the spectra it can be seen that Zn in plant tissues is most probably coordinated in a similar environment to that in zinc phosphate, zinc 2,4-pentadionate, zinc citrate and zinc oxalate. Table 2 displays the FEFF fittings on Zn in plant tissues, Zn in agar, and Zn model compounds. In this table, the interatomic distances and coordination numbers are given. According to these data, Zn can be coordinated to oxygen/nitrogen atoms at different interatomic distances varying from 1.93 to 2.13. In addition, Zn was found to be present in coordination numbers of 2 and 4. No evidence of Zn-S binding

was found in any plant tissue (Fig. 3 and Table 2). Table 2 shows that the Zn-O in zinc pentadionate has a CN of 6.0. Since this coordination number was not identified in the FEFF fittings, it is possible that the geometry of such binding is different from that observed in zinc pentadionate. In addition, a Zn-N complex may be present. These results agree with those published by Küpper et al. (2004) who reported that in *T. caerulescens* (Ganges ecotype) Zn was present in the Zn-O form while absent as Zn-S. Salt et al. (1999) reported that in xylem sap of *T. caerulescens* plants, small amounts of Zn were bound to organic acids. On the other hand, Sarret et al. (2001) reported that in *Phaseolus vulgaris* plants, Zn was present as zinc phosphate.

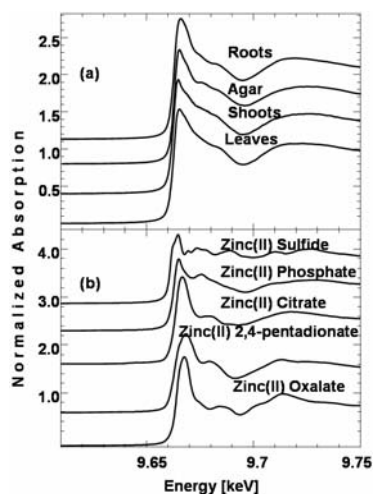


Fig. 3. (a) XANES of Zn uptaken by roots, shoots and leaves of tumbleweed grown in agar based media contaminated with 40 mg Zn(II) L<sup>-1</sup>. Also included is the XANES of Zn in the agar. (b) XANES spectra of Zn(II) model compounds.

#### XAS studies of tumbleweed plants exposed to Cu(II)

The results of the XANES experiments for tumbleweed plants treated with 20 mg Cu L<sup>-1</sup>, agar laden with Cu and the model compounds are presented in Figs. 4(a-c), and their corresponding FEFF fittings in Table 3. The edge energy of Cu in all plant tissues and agar is of 8.99 keV, which is the same as in the model compounds. Then, Cu in plant tissues is present as Cu(II). On the other hand, it seems that Cu in the agar media and root tissues is present in a coordination environment very similar to that in copper acetate or copper nitrate as the spectra are very alike. In addition, in the tumbleweed leaves and stems, a biotransformation may have occurred. Compounds having Cu-S entities may have formed as indicated by the similarity in copper sulfide and plant samples. The data is confirmed by the results presented in Table 3. In this table it can be seen that in roots, the ligand that best matches the Cu present in this tissue is Cu-O with an interatomic distance of 1.95 Å and a coordination number of 6.1. In stems

Cu is coordinated to oxygen and sulfur at interatomic distances of 2.03 and 2.26, respectively. Therefore, since in tumbleweed plants the Cu-S and Cu-O bounds were identified, it is very likely that in roots and stems of tumbleweed plants Cu may be bound to organic acids, to polysaccharides present in the cell wall, and sugars, while in leaves it seems that Cu is primarily bound to sulfur probably from phytochelatin. Similar results were obtained by Polette et al. (2000) in *L. tridentata* plants.

Table 2. FEFF fittings of Zn in agar media, Zn model compounds and Zn uptaken by tumbleweed roots, stems and leaves from agar contaminated with 40 mg Zn L<sup>-1</sup>

Sample	Interaction	CN	R(Å)	σ <sup>2</sup> (Å <sup>2</sup> )
Zinc in leaves	Zn-O/N	2.0	1.93	0.0010
	Zn-O/N	4.0	2.06	0.0066
Zinc in stems	Zn-O/N	2.0	1.96	0.0034
	Zn-O/N	2.0	2.01	0.0011
Zinc in roots	Zn-O/N	4.0	1.98	0.0055
	Zn-O/N	2.0	2.13	0.0031
Zinc in agar	Zn-O/N	4.0	1.96	0.0031
	Zn-O/N	2.0	2.12	0.0017
Zinc(II) 2,4-Pentadionate	Zn-O	6.0	2.02	0.0052
Zinc(II) Citrate	Zn-O	6.0	2.04	0.0069
Zinc(II) Phosphate	Zn-O	4.0	1.96	0.0049
Zinc(II) Oxalate	Zn-O	6.0	2.12	0.0054
Zinc(II) Sulfide	Zn-S	4.0	2.34	0.0028

Note: CN is the coordination number, R is the interatomic distance given in angstroms, and σ<sup>2</sup> is the Debye-Waller factor given in angstroms squared.

#### XAS studies of tumbleweed plants exposed to Ni(II)

Fig. 5(a) shows the XANES spectra of Ni in tumbleweed tissues and agar media. Fig. 5(b) displays the XANES spectra of Ni model compounds. The edge energy of Ni in plant and agar samples was of 8.34 keV. This energy was exactly the same as in model compounds indicating that Ni in plants tissues and agar media was present in the Ni(II) oxidation state. The XANES spectra of Ni in the agar and the three plant tissues are very similar, indicating that the coordination environment in all the samples is almost the same. In addition, the XANES of Ni in roots, stems, and leaves tissues of tumbleweed plants is very similar to the XANES for nickel acetate, however it is not exactly the same. Table 4 shows the FEFF fittings for Ni in agar and plant samples as well as the model compounds. From the results in Fig. 5 and Table 4, it can be concluded that in tumbleweed plants Ni is preferably bound to nitrogen/ oxygen moieties, probably from aminoacids.

By using XAS, Krämer et al. (2000) identified Ni binding in tissues of the non-Ni accumulator *T. arvense*. The results pointed out that most of the Ni

absorbed by this plant was bound to glycine and histidine (Ni-N) and only a small portion was bound to citrate. Since the Ni-N bound was identified in tumbleweed tissues, it is very likely that either histidine or another amino acid may be the Ni-coordinating compounds. Similar to the results obtained in the Zn studies, it seems that sulfur does not participate in Ni binding in tumbleweed tissues. However, oxygen and nitrogen moieties are involved in Ni uptake by this desert plant. The results reported in the present paper indicate that tumbleweed plants were low tolerant to nickel in the growth media. However, more research is needed in order to determine if the absence of Ni-S complexes is related to the low Ni tolerance in tumbleweed plants.

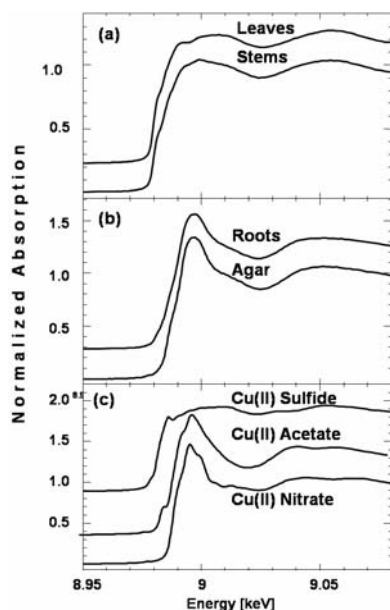


Fig. 4. (a) XANES of Cu uptaken into the leaves and stems of tumbleweed plants grown in agar contaminated with 20 mg Cu(II) L<sup>-1</sup>. (b) XANES of Cu uptaken into the roots in Tumbleweed plants and the XANES of the growth media contaminated with 20 mg Cu(II) L<sup>-1</sup>. (c) XANES of Cu model compounds.

Table 3. FEFF fittings of copper uptaken by tumbleweed roots, stems and leaves from agar contaminated with 20 mg Cu L<sup>-1</sup>, copper in contaminated agar, and Cu(II) model compounds.

Sample	Interaction	CN	R(Å)	$\sigma^2(\text{Å}^2)$
Copper in leaves	Cu-S	3.0	2.26	0.0059
	Cu-Cu	1.0	2.68	0.0087
Copper in stems	Cu-O	1.8	2.03	0.015
	Cu-S	2.0	2.26	0.0070
	Cu-Cu	1.0	2.73	0.010
Copper in roots	Cu-O	6.1	1.95	0.0041
Copper in agar	Cu-O	6.0	1.95	0.0040
Copper(II) sulfide	Cu-S	3.0	2.27	0.0061
Copper(II) nitrate	Cu-O	6.0	1.97	0.0010
Copper(II) acetate	Cu-O	5.0	1.96	0.0064

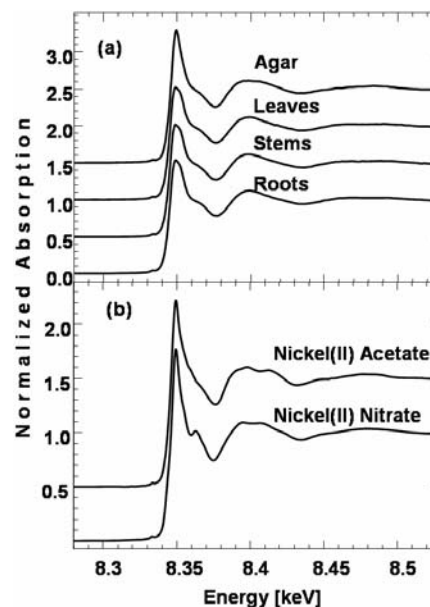


Fig. 5. (a) XANES of Ni uptaken into the leaves, stems, and roots of tumbleweed plants grown in agar contaminated with 10 mg Ni L<sup>-1</sup>, and the XANES of the growth media contaminated with 10 mg Ni L<sup>-1</sup>; (b) XANES of Ni model compounds.

Table 4. FEFF fittings of nickel uptaken by tumbleweed roots, stems and leaves from agar contaminated with 10 mg L<sup>-1</sup> nickel, nickel in contaminated agar, and nickel(II) model compounds (nickel(II) nitrate and nickel(II) acetate).

Sample	Interaction	CN	R(Å)	$\sigma^2(\text{Å}^2)$
Nickel in leaves	Ni-N	2.0	2.03	0.0020
	Ni-N	4.0	2.11	0.0035
	Ni-C	4.0	2.87	0.0074
Nickel in stems	Ni-N	2.0	1.99	0.0010
	Ni-N	4.0	2.11	0.0013
	Ni-C	4.0	2.86	0.0062
Nickel in roots	Ni-N	2.0	2.14	0.0010
	Ni-N	4.0	2.03	0.0016
	Ni-C	4.0	2.85	0.0068
Nickel in agar	Ni-O	6.0	2.05	0.0046
Nickel(II) nitrate	Ni-O	6.0	2.05	0.0038
Nickel(II) acetate	Ni-O	5.0	2.07	0.0039

Note: CN is the coordination number, R is the interatomic distance given in angstroms, and  $\sigma^2$  is the Debye-Waller factor give in angstroms squared.

## Conclusions

The results of the present research demonstrated that tumbleweed may hyperaccumulate Cu as it concentrated 1300 mg Cu kg<sup>-1</sup> d.wt. in the aerial part. In addition, Zn concentrations up to 40 mg L<sup>-1</sup> did not produce any phytotoxic effect, while 10 mg Ni L<sup>-1</sup> were lethal for the plants. The largest

amount of Zn was accumulated in the roots possibly indicating either the presence of a restriction mechanism or the use of this metal by roots. XAS studies demonstrated that organic acids, cell wall, and sugars may be involved in Zn and Cu binding in tumbleweed tissues. In addition, aminoacids may participate in Ni uptake and transport, while phytochelatins may also be responsible for Cu detoxification. Furthermore, it is very possible that phosphate groups participate in Zn coordination in *S. kali* plants. The results obtained in this research indicated that *S. kali* can be considered an option for the clean up of polluted soils containing moderate amounts of Cu, Ni, or Zn.

### Acknowledgments

The authors acknowledge financial support from the NIH (Grant S06GM8012-33), the SSRL/DOE funded Gateway Program and UTEP's Center for Environmental Resource Management (CERM), the HBCU/MI Environmental Technology Consortium (funded by the US DOE). Portions of this research were carried out at the Stanford Synchrotron Radiation Laboratory, a national user facility operated by Stanford University on behalf of the U.S. DOE, Office of Basic Energy Sciences. Guadalupe de la Rosa thanks the Universidad de Guanajuato.

### References

- Aldrich, M.V., Gardea-Torresdey, J.L., Peralta-Videa, J.R. and Parsons, J.G. (2003). Uptake and reduction of Cr(VI) to Cr(III) by Mesquite (*Prosopis* spp.): Chromate-plant interaction in hydroponics and solid media studied using XAS. *Environmental Science and Technology* 37, 1859-1864.
- Bañuelos, G., Terry, N., Leduc, D.L., Pilon-Smits, E.A. and Mackey, B. (2005) Field trial of transgenic Indian mustard plants shows enhanced phytoremediation of selenium-contaminated sediment. *Environmental Science and Technology* 39(6), 1771-1777.
- Barceló, J. and Poschenrieder, C. (2003). Phytoremediation: Principles and perspectives *Contributions to Science* 2, 333-344.
- Baker, A.J.M. and Brooks, R.R. (1989). Terrestrial higher plants which hyperaccumulate metallic elements—a review of their distribution, ecology and phytochemistry, *Biorecovery* 1, 81-126.
- Brooks, R.R. (1998). Plants that hyperaccumulate heavy metals: their role in phytoremediation, microbiology, archaeology, mineral exploration and phytomining, CAB International, Oxon, UK.
- Calabrese, E.J. and Baldwin, L.A. (2003). The hormetic dose-response model is more common than the threshold model in toxicology. *Toxicology Science*, 71, 246-250.
- De la Rosa, G., Peralta-Videa, J.R., Montes, M., Parsons, J.G., Cano-Aguilera, I. and Gardea-Torresdey, J.L. (2004). Cadmium uptake and translocation in tumbleweed (*Salsola kali*), a potential Cd-hyperaccumulator desert plant species: ICP/OES and XAS studies. *Chemosphere* 55, 1159-68.
- De la Rosa, Parsons, J.G., Martinez-Martinez, A., Peralta-Videa, J.R. and Gardea-Torresdey, J.L. (2006). Spectroscopic Study of the impact of arsenic speciation on arsenic/phosphorous uptake and plant growth in tumbleweed (*S. kali*). *Environmental Science and Technology*, 40, 1991-1996.
- Gardea-Torresdey, J.L., Peralta-Videa, J.R., Montes, M., De la Rosa, G. and Corral-Diaz, B. (2004). Bioaccumulation of cadmium, chromium and copper by *Convolvulus arvensis* L.: impact on plant growth and uptake of nutritional elements. *Bioresource Technology* 92, 229-235.
- Gardea-Torresdey, J.L., Peralta-Videa, J.R., de la Rosa, G. and Parsons, J.G. (2005a). Phytoremediation of heavy metals and study of the metal coordination by X-ray absorption spectroscopy, *Coordination Chemistry Reviews*, in press.
- Gardea-Torresdey, J.L., De la Rosa, G., Peralta-Videa, J.R., Montes, M., Cruz-Jimenez, G. and Cano-Aguilera, I. (2005b). A Study of the Differential Uptake and Transportation of Trivalent and Hexavalent Chromium by Tumbleweed (*Salsola kali*), *Archives of Environmental Contamination and Toxicology* 48, 225–232.
- Jiang, L.Y., Yang, X.E. and He, Z.L. (2004). Growth response and phytoextraction of copper at different levels in soils by *Elsholtzia splendens*, *Chemosphere* 55, 1179-1187.
- Kingston, H.M. and Jassie, L.B. (Eds.). (1988). ACS Professional Reference Book Series, Am. Chem. Soc., Washington, D.C.
- Krämer, U., Pickering, I.J., Prince, R.C., Raskin, I. and Salt, D.E. (2000). Subcellular localization and speciation of nickel in hyperaccumulator and non-accumulator *Thlaspi* species, *Plant Physiology* 122, 1343-1353.
- Küpper, H., Mijovilovich, A., Meyer-Klaucke, W. and Kroneck, P.M.H. (2004). Tissue- and Age-Dependent Differences in the Complexation of Cadmium and Zinc in the Cadmium/Zinc Hyperaccumulator *Thlaspi caerulescens* (Ganges Ecotype) Revealed by X-Ray Absorption Spectroscopy, *Plant Physiology* 134, 748–757.

- Matthews, D.J., Moran, B.M., McCabe, P.F. and Otte, M.L. (2004). Zinc tolerance, uptake, accumulation and distribution in plants and protoplasts of five European populations of the wetland grass *Glyceria fluitans*, *Aquatic Botany* 80, 39-52.
- Peralta-Videa, J.R., Gardea-Torresdey, J.L., Tiemann, K.J., Gomez, E., Arteaga, S., Rascon, E. and Parsons, J.G. (2001). Uptake and effect of five heavy metals on seed germination and plant growth in alfalfa (*Medicago sativa*), *Bulletin of Environmental Contamination and Toxicology* 66, 727-734.
- Peralta-Videa, J.R., Gardea-Torresdey, J.L., Gomez, E., Tiemann, K.J., Parsons, J.G., de la Rosa, G. and Carrillo, G. (2002). Potential of alfalfa Plant to Phytoremediate Individually Contaminated Montmorillonite-Soils with Cd(II), Cr(VI), Cu(II), Ni(II), and Zn(II). *Bulletin of Environmental Contamination and Toxicology* 69, 74-81.
- Polette, L.A., Gardea-Torresdey, J.L., Chianelli, R.R., George, G.N., Pickering, I.J. and Arenas, J. (2000). XAS and microscopy studies of the uptake and bio-transformation of copper in *Larrea tridentata*, *Microchemical Journal* 65, 227-236.
- Reeves, R.D., and Brooks, R.R. (1983). Hyperaccumulation of lead and zinc by two metallophytes from mining areas of central Europe. *Environmental Pollution* 31, 277-285.
- Rengel, Z. (1999). Heavy metals as essential nutrients. In: Prasad, M.N.V. (ed.) Heavy metal stress in plants: from biomolecules to ecosystems, 2<sup>nd</sup> Ed. Springer-Verlag, India.
- Sarret, G., Vangronsveld, J., Manceau, A., Musso, M., D'Haen, J., Menthonnex, J.J. and Hazemann, J.L. (2001). Accumulation forms of Zn and Pb in *Phaseolus vulgaris* in the presence and absence of EDTA, *Environmental Science and Technology* 35, 2854-2859.
- Salt, D.E., Prince, R.C., Baker, A.J.M., Raskin, I. and Pickering, I.J. (1999). Zinc ligands in the metal hyperaccumulator *Thlaspi caerulescens* as determined using X-ray absorption spectroscopy. *Environmental Science and Technology* 33, 712-717.
- Salt, D.E., Prince, R.C. and Pickering, J. (2002). Chemical speciation of accumulated metals in plants: evidence from X-ray absorption spectroscopy, *Microchemical Journal* 71, 255-259.
- Van der Lelie D., Schwitzguebel, J.P., Glass, D.J., Vangronsveld, J. and Baker, A. (2001) Assessing phytoremediation's progress in the United States and Europe, *Environmental Science and Technology* 35, 446A-452A.