

# Determination and Chemical Speciation of Selenium in Farmlands from Southeastern Spain: Relation to Levels Found in Sugar Cane

Juana P. Diaz-Alarcón,<sup>†</sup> Miguel Navarro-Alarcón,<sup>\*,†</sup> Herminia López-García de la Serrana,<sup>†</sup> Carlos Asensio-Drima,<sup>‡</sup> and Maria C. López-Martínez<sup>†</sup>

Department of Nutrition and Bromatology, Faculty of Pharmacy, University of Granada, E-18071 Granada, Spain, and Department of Edaphology, Faculty of Pharmacy, University of Granada, E-18071 Granada, Spain

The concentration and the oxidation state of selenium in farmland soils, sewage sludges, and sands was determined by using hydride generation atomic absorption spectrometry. Total Se concentrations ranged from 0.071 to 0.390  $\mu\text{g/g}$ , whereas  $\text{Se}^{\text{IV}}$  concentrations varied from not detectable to 0.050  $\mu\text{g/g}$ . Selenium is predominantly present as  $\text{Se}^{\text{VI}}$ ; a minor fraction was found as  $\text{Se}^{\text{IV}}$ . We also studied the influence of pH, its values varied between 7.03 and 8.33, on the oxidation state and the content in Se and its possible correlation with the selenium uptake and accumulation in the sugar cane plants. Significant differences were observed between the levels of total Se ( $p < 0.01$ ) and  $\text{Se}^{\text{IV}}$  ( $p < 0.05$ ) in farmland soils, whether they are located in the industrialized or nonindustrialized areas. Total Se is found in higher concentrations and mainly as  $\text{Se}^{\text{IV}}$  (selenite) in industrialized area. The content of total Se in the samples of sugar cane taken from the zone is independent of the concentration of total Se and  $\text{Se}^{\text{IV}}$  in farmland soils of the zone ( $p < 0.05$ ).

**Keywords:** *Se speciation; pH; farmlands and sewage sludges; uptake by sugar cane; influence of human and industrial activities*

## INTRODUCTION

Selenium is found in the earth's crust in concentrations that vary from 0.5 to 0.09  $\mu\text{g/g}$ ; it is frequently related with minerals that contain sulfur (National Research Council, 1983). The existence of Se in soil is related to several factors, such as geographic location, kind of rock, oxidation–reduction potential, pH, nature of drainage waters, and type of plant grown (Kabata-Pendias and Pendias, 1984; Elrashidi et al., 1987; Alexander et al., 1988; Banuelos and Meek, 1990; Simonoff and Simonoff, 1991; Paya-Pérez et al., 1993; Zhao et al., 1993; Wahl et al., 1994).

Normally, there is a direct correlation between soil Se content and the plants grown on such soil (Varo et al., 1988; Golubkina et al., 1992; Levander and Burk, 1994) so that high levels of Se may concentrate in some species of accumulator plants. However, the fact of finding high levels of Se in soils does not always imply that Se can be found in plants producing toxic effects as the Se uptake depends on different factors (Bureau et al., 1988) such as the species of plant, the way in which Se appears in soils, the presence of other ions in the solution of the soil,  $\text{SO}_4^{2-}$  (Bisbjerg and Gissel-Nielsen, 1969; Mikkelsen et al., 1988), the management of the crop, pruning, and harvest time.

The soil pH also plays an important role as, in those soils with an acid character, Se is found mainly in the form of selenite, not very soluble and assimilable, while in alkaline soils, Se ( $\text{SeO}_3^{2-}$ ) becomes oxidated and then selenate is produced; selenate is much more soluble and easily assimilable. Moreover, it can originate several selenium-containing organic compounds (Hartfiel and Bahnert, 1988; Banuelos and Meek, 1990; Gondi et al., 1992; Zhao et al., 1993).

This study describes techniques to widen Se speciation methods to assess total Se and  $\text{Se}^{\text{IV}}$  levels in

farmland soils, sewage sludges, and sands and its later direct determination by hydride generation atomic absorption spectrometry. Furthermore, pH values of the samples have been determined for their correlation with the oxidation state prevailing in Se in these samples. This way, we could have better knowledge of the factors that influence both bioavailability and, as a consequence, Se levels in the plants grown in this zone, which values were formerly determined (Diaz-Alarcón et al., 1994a).

Apart from this, we have also found it interesting to study the influence of human and industrial activities on Se levels in farmland soils of the zone and the influence on concentrations of this element in sugar cane plants (the most important crop of the zone) grown in these soils (Diaz-Alarcón et al., 1994a).

## MATERIALS AND METHODS

**Apparatus.** The samples were mineralized in an Investor sand bath. All atomic measurements were made with a Perkin-Elmer Model 1100B atomic absorption spectrometer equipped with a Perkin-Elmer MHS-10 hydride generator. A selenium hollow cathode lamp (Perkin-Elmer Corp.) was operated under the conditions recommended by the manufacturer. A spectral slit width of 2.0 nm was selected to isolate the 196.0-nm line. All analyses were performed in peak height mode to calculate absorbance values. The measurements of pH were carried out with a Radiometer Model 26 pHmeter.

**Reagents.** All solutions were prepared with ultrapure water with a specific resistivity of 18 M $\Omega$  cm obtained by filtering double-distilled water through a Milli-Q purifier (Millipore) immediately before use. A commercially available 1000  $\mu\text{g/mL}$  selenium standard solution (prepared from  $\text{SeO}_2$  in 0.5 mmol/mL  $\text{HNO}_3$ ) was used (Tritisol, Merck). The alternate standards for calibration (10 mg/L and 100  $\mu\text{g/L}$ ) were prepared by serial dilutions with 1.9% HCl solution.  $\text{HNO}_3$  (65%), HCl (37%),  $\text{HClO}_4$  (65%) (Carlo Erba),  $\text{NaBH}_4$ , and NaOH (Merck analytical grade) were also used.

**Samples.** Farmland soil samples were obtained from the fields of the study zone, according to the random model

<sup>†</sup> Department of Nutrition and Bromatology.

<sup>‡</sup> Department of Edaphology.

suggested by Bridges and Davidson (1982). The material (2 kg) was taken from the arable layer at 0–30 cm depth in 32 different sampling sites. Sewage sludge samples (500 g) were collected in four different places in a gully that crosses the study zone, the place where sewage of the zone is received. Sand samples (500 g) were collected in two different places in the gully's outlet to the sea. All samples were dried during 2 weeks in the laboratory at ambient temperature, homogenized, sieved through a 340  $\mu\text{m}$  sieve, and finally stored in polyethylene containers at  $-18^\circ\text{C}$  until the analysis was carried out.

**Procedure.** Lyophilized sample (300 mg) was placed in a 60-mL volumetric flask, and the sample was mineralized by addition of 4 mL of concentrated  $\text{HNO}_3$  and heating at  $80^\circ\text{C}$  for 1 h in a sand bath. Another 3 mL of  $\text{HNO}_3$  was added, and heating continued for an additional 3 h until the sample was completely mineralized (Díaz-Alarcón et al., 1994b). The digest obtained was cooled, diluted to 10 mL with deionized water, and then filtered through a Whatman filter paper to remove solid residues. Then,  $\text{Se}^{\text{VI}}$  was reduced to  $\text{Se}^{\text{IV}}$  by taking a 1-mL aliquot in a test tube and adding 1 mL of concentrated HCl and heating at  $100^\circ\text{C}$  for 10 min in a thermostated bath. When cool, this was diluted to volume with 2.0 (w/v) HCl solution. A 10-mL aliquot was then placed in the reaction vessel of the hydride generation system. Total Se determination was carried out using the HG-AAS technique. Finally, absorbance values were correlated with total Se concentrations by comparing with calibration plotting obtained after the application of the standard addition method to one soil sample.

For the determination of  $\text{Se}^{\text{IV}}$  in these samples, we used a method quite similar, although slightly modified, to the one used by Ericzon et al. (1989). Amounts of 300 mg of dried, homogenized sample were mineralized with 25 mL of 1 M  $\text{HNO}_3$  solution and heated at  $70^\circ\text{C}$  for 16 h in a sand bath. The digests were cooled, diluted to 25 mL with deionized water, and then filtered. A 9-mL aliquot was taken from the final filtrate and 1 mL of concentrated HCl was added to obtain a 1.2 M ClH solution. After 2 h in such conditions, the determination of Se was carried out by using the HG-AAS technique (Díaz-Alarcón et al., 1994b).

The Se in the remaining oxidation states ( $\text{Se}^0$ ,  $\text{Se}^{-\text{II}}$ ,  $\text{Se}^{\text{VI}}$ ) was calculated by the difference between values of total Se and  $\text{Se}^{\text{IV}}$ .

In order to determine pH, 20 g of a dried sample were weighed in a reaction vessel where 20 mL of deionized water was added (Ministerio de Agricultura, 1982). It was continuously stirred until total solubility of the soil in water was obtained. The measurement was carried out with the pHmeter. A Beckman buffer (Beckman Instruments Inc.), with a pH interval from 4.00 to 9.18 was used.

The determination of total Se in sugar cane samples was that previously used for the measurement of this element in crops of the zone (Díaz-Alarcón et al., 1994a).

**Reproducibility and Accuracy.** The reproducibility of the Se measurements was obtained following the procedures indicated by Díaz-Alarcón et al. (1994b). The reference standard consisted of river sediment [National Institute of Standards and Technology (NIST) standard reference material (SRM) 2704] with a noncertified Se content of  $1.10\ \mu\text{g/g}$  (not sufficiently homogeneous for certification).

**Method.** Because of its specificity and sensibility, hydride generation atomic absorption spectrometry of acid-digested samples is one of the most frequently recommended techniques for the determination of Se in soils, ashes, and sewage sludges (Ericzon et al., 1989; Banuelos and Meek, 1989, 1990).

During the last few years, there has been much concern about the determination of Se in soils. As a result of this, analytical techniques have been developed to determine the chemical speciation of the several forms in which Se is found in soils, ashes, and sediments by the prior separation and determination by HG-AAS (Ericzon et al., 1989; Velinsky and Cutter, 1990). This technique is the one we have used in our study.

**Data Analysis.** In order to specify the differences among the results obtained and definitely establish whether there are significant differences in Se concentration and pH in samples

**Table 1. Recovery of Total Selenium from Spiked Samples**

sample	Se present, mg	Se added, mg	Se found, mg	Se rec, %
farmland soil-32	0.004	0.000	0.004	100.00
	0.004	0.005	0.009	100.00
	0.004	0.010	0.013	92.86
	0.004	0.015	0.017	89.47
sewage sludge-1	0.006	0.000	0.006	100.00
	0.006	0.005	0.011	100.00
	0.006	0.010	0.016	100.00
	0.006	0.015	0.021	100.00

**Table 2. Precision Study**

sample	<i>n</i>	mean Se content, <sup>a</sup> $\mu\text{g/g}$	RSD %
farmland soil-5	7	$0.440 \pm 0.021$	4.77
farmland soil-6	7	$0.520 \pm 0.052$	9.99
NIST SRM 2704	10	$1.120 \pm 0.031$	2.73

<sup>a</sup> Mean Se content  $\pm$  standard deviation.

considered (farmland soils, sewage sludges, sands), and in total Se level in sugar cane samples, we made a statistical analysis of the variance. The comparison of means was performed either by a one-way ANOVA (level of significance was set at 0.05) or the Kruskal–Wallis test (level of significance was set at 0.01) using the Statgraphics 6.0 Software Package (STSC, Inc.). Analysis of variance was applied after checking the normal distribution and homogeneity of variances of sample populations using the Kolmogorov–Smirnov and Bartlett tests, respectively, as well as random and paired samples. Regression analysis between pH and  $\text{Se}^{\text{IV}}$  values was applied (level of significance was set at 0.05).

## RESULTS AND DISCUSSION

The analytical technique we propose is suitable for the determination of total Se and  $\text{Se}^{\text{IV}}$  in soils, sewage sludges, and sands. This technique can be used not only in quality control assays of soils but also in analysis of products that are used in agricultural treatments. Besides, our method is potentially useful in order to control the distribution of the different chemical forms in which Se can be found in soils and sediments.

The accuracy of the method was evaluated by recovery tests. Mean recovery for the added samples was 97.79% (Table 1). The total Se concentration determined in NIST SRM 2704 was  $1.12\ \mu\text{g/g}$  ( $n = 10$ ) for a noncertified value of  $1.10\ \mu\text{g/g}$ .

The technique was accurate and reproducible. The relative standard deviation was appropriately low (Table 2) and the analytical concentration determined was very close to the reference value for SRM 2704.

In Tables 3 and 4, we can observe the ranges and mean concentrations of total Se,  $\text{Se}^{\text{IV}}$ , and Se corresponding to the remaining oxidation states ( $\text{Se}^{-\text{II}}$ ,  $\text{Se}^0$ ,  $\text{Se}^{\text{VI}}$ ) in farmland soils, sewage sludges, and sands.

The application of a one-way ANOVA shows the existence of significant differences between total Se levels in farmland soils and sands ( $p < 0.05$ ). This result is correlated with the fact that farmland soils and sewage sludges belong to the group of calcareous and salicarcic fluvisols, respectively (Cabrera et al., 1993), a group (fluvisols) with an elevated content in organic matter. This characteristic is the reason for its large capacity for retaining Se. This result agrees with the opinions of the other authors about the influence of the geological characteristics of soil on the total content in this element (Ure et al., 1979; McKeague and Wolynetz, 1980; Kabatapendias and Pendias, 1984; Alexander et al., 1988; Erkamov, 1992; Zhao et al., 1993).

**Table 3. Concentrations of Total Selenium and Se<sup>IV</sup> (μg/g, Dry Weight) in Farmland Soils, Sewage Sludges, and Sands in the Province of Granada (Southeast Spain)**

sample	n	total Se		Se <sup>IV</sup>	
		mean Se <sup>a</sup>	range	mean Se <sup>a</sup>	range
farmland soils	32	0.212 ± 0.072	0.078–0.390	0.023 ± 0.007	0.017–0.050
sewage sludges	4	0.175 ± 0.036	0.121–0.196	0.017 ± 0.004	0.013–0.020
sands	2	0.080 ± 0.012	0.071 ± 0.088	nd <sup>b</sup>	nd <sup>b</sup>

<sup>a</sup> Mean selenium content ± standard deviation. <sup>b</sup> Not detectable.

**Table 4. Concentrations of Se Corresponding to the Remaining States of Oxidation (Se<sup>VI</sup>, Se<sup>0</sup>, Se<sup>-II</sup>) (μg/g, Dry Weight) in Farmland Soils, Sewage Sludges, and Sands in the Province of Granada (Southeast Spain)**

sample	n	Se <sup>VI</sup> , Se <sup>0</sup> , Se <sup>-II</sup>	
		mean Se <sup>a</sup>	range
farmland soils	32	0.189 ± 0.069	0.061–0.370
sewage sludges	4	0.158 ± 0.038	0.101–0.183
sands	2	0.080 ± 0.012	0.071–0.088

<sup>a</sup> Mean ± standard deviation.

**Table 5. pH Values in Farmland Samples, Sewage Sludges, and Sands in the Province of Granada (Southeast Spain)**

sample	n	pH value <sup>a</sup>	range	RSD, %
farmland soils	32	7.55 ± 0.27	7.11–7.98	3.56
sewage sludges	4	7.70 ± 0.33	7.38–8.14	4.24
sands	2	8.27 ± 0.08	8.22–8.33	0.94

<sup>a</sup> Mean ± standard deviation.

The existence of significant differences in Se<sup>IV</sup> levels in farmland soils and sewage sludges was determined and the differences were compared with those found in sands (included in the group of miscellany beach). It is probable, as mentioned above, that this result is related to soil edaphologic characteristics.

As we can see in Table 3, Se<sup>IV</sup> levels are quite low in comparison with total Se levels; for that reason, we have found Se, in most cases, in the remaining oxidation states (Se<sup>VI</sup>, Se<sup>-II</sup>, Se<sup>0</sup>). This result shows that Se is predominantly present in soils as selenates rather than selenites, due to selenite oxidation to selenates, a finding that is reinforced by the slightly alkaline character of the analyzed samples (Table 5), according to the reports by other investigators (Hartfiel and Bahnners, 1988; Banuelos and Meek, 1989, 1990; Gondi et al., 1992; Zhao et al., 1993).

It has been observed that significant differences exist ( $p < 0.05$ ) between the pH of soils and of sediments as

compared with that of sands. This fact establishes the correlation between pH and the type of farmland that has been considered, as other authors had determined (Ure et al., 1979; Gondi et al., 1992; Zhao et al., 1993).

In order to check the correlation of pH values with Se distribution in its various chemical forms a regression analysis study between pH and Se<sup>IV</sup> was performed. A linear correlation with the equation  $\text{Se}^{\text{IV}} (\mu\text{g/g}) = 0.1589 - 0.0181 \text{ pH}$  ( $r = -0.6532$ ,  $p < 0.05$ ) was observed. This result shows that when the alkalinity of the sample increases, Se<sup>IV</sup> (selenite) levels decrease owing to oxidation to Se<sup>VI</sup> (selenate), as it is easily assimilated and bioavailable for plants (Hartfiel and Bahnners, 1988; Banuelos and Meek, 1989, 1990; Gondi et al., 1992). These data are correlated with the low levels of Se<sup>IV</sup> found in the samples (Table 3) due to their slightly alkaline character.

The levels of total Se in soils (farming soils, pasture ground, industrial soils, sediments) that have been determined by other authors in different zones of the world is given in Table 6. Comparing them with that found in farmland soils in the present study, we deduce that our results are generally low. We must also take into account that our soil samples are located in an industrialized area, in which higher levels of Se are found, as Hartfiel and Bahnners (1988) determined in industrial soils in Germany (Table 6).

In sewage sludges, the average concentration of total Se found in our study, 0.175 μg/g, is much lower than those found in sediments of different regions of the United States, 0.728, 28.1, and 0.920 μg/g (Table 6).

In farmland soils of the zone of study, significant differences were found (Table 7) between the levels of Se<sup>IV</sup> ( $p < 0.05$ ) and the levels of total Se ( $p < 0.01$ ) after dividing the zone into two areas [area A, nonindustrialized; area B, industrialized (paper mill, town of Motril, fuel tanks, oil refinery, Port of Motril, and industrial complex)]. The highest levels of both Se<sup>IV</sup> and total Se corresponded to the industrialized area (Table 7). This

**Table 6. Concentrations of Total Se in Soils and Sediments As Determined in Other Countries by Other Authors (μg/g, Dry Weight)**

sample	country	mean	range	ref
soils	USA	1.500		Gutenmann et al., 1979
soils	Scotland	0.170		Ure et al., 1979
soils	Canada	0.300		McKeaque and Wolynetz, 1980
Darien soil	USA	4.000		Keith Furr et al., 1980
Teel soil	USA	4.800		Keith Furr et al., 1980
earth's crust			0.050–0.090	National Research Council, 1993
soils	Ireland	1200		Alexander et al., 1988
farmland soils	Germany	0.123		Hartfiel and Bahnners, 1988
grazing soils	Germany	0.158		Hartfiel and Bahnners, 1988
industr. soils	Germany	0.652		Hartfiel and Bahnners, 1988
sediments	USA	0.728	0.728 ± 0.058	Velinsky and Cutter, 1990
sediments	USA	28.1	28.1 ± 0.833	Velinsky and Cutter, 1990
sediments	USA	0.920	0.920 ± 0.043	Velinsky and Cutter, 1990
soil	USA	1.7		Fio and Fujii, 1990
soils	USA		1.00–3.00	Wu and Huang, 1991
soils	New Zeland	0.100		Simonoff and Simonoff, 1991
soils	Yugoslavia	0.200		Maksimovic et al., 1992
soil	China	0.130	0.075–0.180	Tiang et al., 1992
soil	China	0.323		Zhao et al., 1993

**Table 7. Analysis of Variance of the Selenium Concentrations (Total Se and Se<sup>IV</sup>) (μg/g, Dry Weight) and pH Values in Farmland Soil Samples, and of the Total Selenium in Sugar Cane Samples**

	p	area			
		industrialized		not industrialized	
		mean value	95% LSD interv for mean	mean value	95% LSD interv for mean
total Se <sup>a</sup>	0.0045 <sup>c</sup>				
Se <sup>IV</sup> <sup>a</sup>	0.0016 <sup>d</sup>	0.028	0.026–0.031	0.020	0.018–0.022
pH <sup>a</sup>	0.0286 <sup>d</sup>	7.42	7.31–7.52	7.63	7.55–7.71
total Se <sup>b</sup>	0.3676 <sup>d</sup>	1.881	1.081–2.681	1.268	0.739–1.797

<sup>a</sup> In farmland soils samples. <sup>b</sup> In sugar cane samples. <sup>c</sup> Kruskal–Wallis test ( $p < 0.01$ ); it denotes a statistically significant difference. <sup>d</sup> One-way ANOVA ( $p < 0.05$ ); it denotes a statistically significant difference.

result establishes the existence of the influence of human and industrial activities on Se levels in farmland soils of the zone, as previously established Hartfiel and Bahnert (1988) in a study made in Germany.

In view of these results, the levels of total Se in farmland soils were correlated with those ones found in the 24 samples of sugar cane cultivated in southeastern Spain (Díaz-Alarcón et al., 1994a). These samples were selected among all the crops that we analyzed because sugar cane is the predominant crop of the zone (Díaz-Alarcón et al., 1994a) and a considerable number of sugar cane samples were available for this study. After this correlation, no significant differences were discovered between the levels of total Se in the samples of sugar cane that are grown in the industrialized area in comparison with those of the nonindustrialized area ( $p < 0.05$ ) (Table 7). These data lead us to the conclusion that the absorption and bioavailability of Se in sugar cane is independent from its content in soil. However, the total Se levels in sugar cane in area B (industrialized) are slightly higher than those found in area A (nonindustrialized). This result is ratified by the fact that the pH of area A (with a mean value of 7.62) is significantly higher than that of area B (with a mean value of 7.41) ( $p < 0.05$ ) (Table 7). This difference shows the prevailing presence of Se as Se<sup>IV</sup> in the soil of area B, which is not easily bioavailable and absorbed by sugar cane.

Although, Se<sup>VI</sup> is more easily assimilated by plants (Bisbjerg and Gilssen-Nielsen, 1969; Banuelos and Meek, 1989, 1990; Gondi et al., 1992), the low levels of total Se and Se in the remaining oxidation states that we have found in farmland soils in this area, as well as the fact that the traditional crops usually have low tolerance to Se (Mikkelsen et al., 1988), as Se in the species of nonaccumulative plants generally interrupts protein synthesis, would imply low levels of Se absorbed and bioavailable for the plants grown in the zone (Díaz-Alarcón et al., 1994a) and, as a result, low levels of Se in such plants. This fact would lead to a low quantity of Se in the daily dietary intake of inhabitants who live in the zone and consume the crops grown there (Díaz-Alarcón et al., 1994a).

Despite all, future research is needed in that direction leading to better knowledge of Se and plant interrelationships.

#### LITERATURE CITED

Alexander, J.; Högberg, J.; Thomassen, Y.; Aaseth, J. Selenium. In *Handbook on Toxicity of Inorganic Compounds*; Seiler, H. G., Sigel, H., Eds.; Dekker: New York, 1988.

- Banuelos, G. S.; Meek, D. W. Selenium Accumulation in Selected Vegetables. *J. Plant. Nutr.* **1989**, *12*, 1255–1272.
- Banuelos, G. S.; Meek, D. W. Accumulation of Selenium in Plants Grown on Selenium-Treated Soil. *J. Environ. Qual.* **1990**, *19*, 772–777.
- Bisbjerg, B.; Gissel-Nielsen, G. Uptake of Applied Selenium by Agricultural Plants. *Plant Soil* **1969**, *31*, 278–298.
- Bridges, E. M.; Davison, D. A. *Principles and Applications of Soil Geography*; Longman Group Ltd.: New York, 1982.
- Bureau, R. G.; MacDonald, A.; Jacobson, A.; Mary, D.; Grattan, S.; Sheunau, C.; Swanton, B.; Scherer, D.; Abrams, M.; Epstein, X.; et al. Selenium in Tissues of Crops Sampled from the Westside of the San Joaquin Valley, California. In *Selenium Contents in Animal and Human Food Crops Grown in California*; Publication 3330; University of California-Davis: Davis, CA, 1988.
- Cabrera, C. Evaluation of the Environmental Contamination by Lead and Cadmium in the Coastal Zone of the Province of Granada. Doctoral Thesis, Service of Publications of the University of Granada; Granada, Spain, 1993.
- Díaz-Alarcón, J. P.; Navarro-Alarcón, M.; López-García de la Serrana, H.; López-Martínez, M. C. Determination of Selenium Levels in Vegetables and Fruits by Hydride Generation Atomic Absorption Spectrometry. *J. Agric. Food Chem.* **1994a**, *42*, 2848–2851.
- Díaz-Alarcón, J. P.; Navarro-Alarcón, M.; López-García de la Serrana, H.; López-Martínez, M. C. Determination of Selenium in Fresh Fish from Southeastern Spain for Calculation of Daily Dietary Intake. *J. Agric. Food Chem.* **1994b**, *42*, 334–337.
- Elrashidi, M. A.; Adriano, D. C.; Workman, S. M.; Lindsay, W. L. Chemical Equilibria of Selenium in Soils: a Theoretical Development. *Soil Sci.* **1987**, *144*, 141–152.
- Ericzon, C.; Pettersson, J.; Andersson, M.; Olin, A. Determination and Speciation of Selenium in End Products from a Garbage Incinerator. *Environ. Sci. Technol.* **1989**, *23*, 1524–1528.
- Erkamov, V. V. Biogeochemical Regioning Problems and the Biogeochemical Selenium Provinces in the Former USSR. *Biol. Trace Elem. Res.* **1992**, *33*, 171–85.
- Fio, J. L.; Fujii, R. Selenium Speciation Methods and Application to Soil Saturation Extracts from San Joaquin Valley, California. *Soil Sci. Soc. Am. J.* **1990**, *54*, 363–369.
- Golubkina, N. A.; Shagova, M. V.; Spirichev, V. B.; Khristenko, P. P.; Alftan, D.; Laaksonen, P.; Muratov, I. M.; Vachaeva, N. M. The Selenium Content of Food Products and the Blood of Inhabitants of Norilsk. *Vopr. Pitan.* **1992**, *4*, 43–45.
- Gondi, F.; Danto, G.; Feher, J.; Bogye, G.; Alftan, G. Selenium in Hungary. The Rock-Soil-Human System. *Biol. Trace Elem. Res.* **1992**, *35*, 299–306.
- Gutenmann, W. H.; Pakkala, I. S.; Chuney, D. J.; Kelly, W. C.; Lisk, D. J. Arsenic, Boron, Molybdenum, and Selenium in Successive Cuttings of Forage Crops Field Grown on Fly Ash Amended Soil. *J. Agric. Food Chem.* **1979**, *27*, 1393–1395.
- Hartfiel, W.; Bahnert, N. Selenium Deficiency in the Federal Republic of Germany. *Biol. Trace Elem. Res.* **1988**, *15*, 1–12.
- Kabata-Pendias, A.; Pendias, H. *Trace Elements in Soils and Plants*; CRC Press, Inc.: Boca Raton, FL, 1984.
- Keith Furr, A.; Parkinson, T. F.; Bache, C. A.; Gutenmann, W. H.; Pakkala, I. S.; Lisk, D. J. Multielement Absorption by Crops Grown on Soils Amended with Municipal Sludge Ashes. *J. Agric. Food Chem.* **1980**, *28*, 660–662.
- Levander, O. A.; Burk, R. F. Selenium. In *Modern Nutrition in Health Disease*, 8th ed.; Shils, M. E., Olson, J. A., Shike, M., Eds.; Lea and Feiger: London 1994.
- Maksimovic, Z. J.; Djuric, I.; Jovic, V.; Rsumovic, M. Selenium Deficiency in Yugoslavia. *Biol. Trace Elem. Res.* **1992**, *33*, 187–196.
- Mckeague, J. A.; Wolynetz, M. S. Background Levels of Minor Elements in Some Canadian Soils. *Geoderma* **1980**, *24*, 299–306.
- Mikkelsen, R. L.; Bingham, F. T.; Page, A. L. Factors Affecting Selenium Accumulation by Agricultural Crops. In *Selenium in Agriculture and the Environment*; Jacobs, L. W., Ed.; SSSA Spec. Publ. 23; ASA and SSSA: Madison, WI, 1988.

- Ministerio de Agricultura. *Official Methods of Soils and Waters Analysis*; Madrid, Spain, 1982.
- National Research Council (Subcommittee on Selenium; Committee on Animal Nutrition; Board of Agriculture). *Selenium in Nutrition*; National Academy Press; Washington, DC, 1983.
- Paya-Pérez, A. B.; Goetz, L.; Springer, A.; Nielsen, A. Sorption of Selenium(IV) in a Soil Profile: an Application of Radiochemical Techniques. *J. Trace Microprobe Tech.* **1993**, *11*, 143–158.
- Simonoff, M.; Simonoff, G. *Le Selenium et la Vie*; Masson: Paris, 1991.
- Tian, J.; Qian, Q.; Chai, Z.; Zhang, P.; Xu, Q.; Mao, D. Study on Low Selenium Environment of Exi Autonomous Prefecture, Hubei Province. *Chin. Sci. Bull.* **1992**, *37*, 1612–1617.
- Ure, A. M.; Bacon, M. L.; Berrow, M. L.; Watt, J. J. The Total Trace Element Content of Some Scottish Soils by Spark Source Mass Spectrometry. *Geoderma* **1979**, *22*, 1–23.
- Varo, P.; Alfthan, G.; Ekholm, P.; Aro, A.; Koivistoinen, P. Selenium Intake and Serum Selenium in Finland: Effects of Soil Fertilization with Selenium. *Am. J. Clin. Nutr.* **1988**, *48*, 324–329.
- Velinsky, D. J.; Cutter, G. A. Determination of Elemental Selenium and Pyrite-Selenium in Sediments. *Anal. Chim. Acta* **1990**, *235*, 419–425.
- Whal, C.; Benson, S.; Santolo, G. Temporal and Spatial Monitoring of Soil Selenium at Kesterson Reservoir, California. *Water Air Soil Pollut.* **1994**, *74*, 3–4.
- Wu, L.; Huang, Z. Z. Selenium Accumulation and Selenium Tolerance of Salt Grass from Soils with Elevated Concentrations of Se and Salinity. *Eccotoxicol. Environ. Saf.* **1991**, *22*, 267–282.
- Zhao, C.; Ren, J.; Xue, C. Selenium in Soils of Selenium-Rich Areas in Ziyang County. *Turang Xuebao* **1993**, *30*, 253–259.

Received for review July 11, 1995. Accepted May 7, 1996.®

JF950433Z

® Abstract published in *Advance ACS Abstracts*, June 15, 1996.