MASTER COPY

968

J. ENVIRON. QUAL., VOL. 27, SEPTEMBER-OCTOBER 1998

Selenium Uptake by Plants from Soils Amended with Inorganic and Organic Materials

H. A. Ajwa,* G. S. Bañuelos, and H. F. Mayland

ABSTRACT

Depending on its concentration and chemical form, Se functions as an essential element or potential toxicant to humans, livestock, and waterfowl. Application of seleniferous organic materials to soils may increase plant-available Se content and pose health hazards. This study assessed Se uptake by two successive plantings of canola (Brassica napus cv. Westar) and multiple clippings of tall fescue (Festuca arundinacea L. cv. Fawn) grown in soils [Hanford sandy loam (coarse-loamy, mixed, thermic Typic Durixeralfs) and Panoche clay loam (fine-loamy, mixed, superactive, thermic Typic Torriorthents)] amended with 1.5 mg Se kg⁻¹ soil as inorganic selenate (Se O₄²⁻) or seleniferous organic materials [alfalfa (Medicago sativa L.), Astragalus praelongus, or cattle (Bos tauris) manure] under growth chamber conditions. Tissues of canola and tall fescue accumulated much greater concentrations of Se from the inorganic SeO₄²⁻ treatment compared to the treatments with seleniferous organic materials. The addition of crop residue or animal manure to the SeO4--treated soils considerably reduced Se accumulation by both plant species. In soils amended with seleniferous organic materials, more than 80% of the Se remained in soils after two plantings of canola and all clippings of tall fescue. The slow release of plant-available Se in soils amended with seleniferous organic materials suggests the use of these materials to control the concentrations of Se in crops grown on nonseleniferous soils.

CELENIUM has received considerable attention as an Subscription essential micronutrient in the diet of animals and as a toxic element (Ohlendorf, 1989). Dietary Se levels required by animals ranges from 0.05 to 0.3 mg Se kg⁻¹ dry matter (DM) depending on animal species and level of vitamin E in the feed (Ohlendorf, 1989; Mayland, 1994; Oldfield et al., 1994). Animals obtain Se mainly from consumption of forage plants and soil Se content is the primary determinant of the Se content of animal feed and forage. Most soils contain no more than 0.1 mg Se kg⁻¹ soil, but soils derived from Cretaceous shales may contain 1 to 2 mg Se kg⁻¹ soil or higher (Mayland et al., 1989). In Se-deficient areas, several practices have been proposed to increase Se concentrations in pasture crops. Such practices including foliar application, inorganic fertilization, seed treatment, and soil incorporation of fly ash, municipal refuse incinerator ash, or sewage sludge (Logan et al., 1987; Mayland et al., 1989;

Published in J. Environ. Qual. 27:1218-1227 (1998).

Arthur et al., 1992). Another viable strategy to supplement animal diets is blending plant material containing high Se concentrations with low Se-containing forage (Bañuelos and Meek, 1990; Bañuelos et al., 1992). Recently, animal Se supplementation practices were questioned because of the possibility of an increase in the environmental burden of Se derived from animal manure (U.S. FDA, 1993) or seleniferous plant residues added to soils (Oldfield et al., 1994).

The solubility and bioavailability of mineralized Se will depend on the type of plant residue incorporated in the soil. For example, protein-bound selenomethionine and selenocystine are the two Se-containing amino acids most commonly found in non-accumulating plant tissues like alfalfa. In contrast, the Se in Se-accumulating plants, like some Astragalus spp., is mostly water-soluble and found in nonprotein forms such as Se-methylselenocysteine (Mayland, 1994). Based on the assumption that organic Se compounds (such as selenomethionine and selenocystine) present in some plant species are mineralized in a manner similar to their analogous organic S compounds (Shrift, 1973), the mineralization products of organic Se and S should be similar. Such Se products would be dominated by selenate (SeO_4^{2-}) , selenite (Se O_3^{2-}), elemental Se (Se⁰), and gaseous dimethylselenide [(CH₃)₂Se] and dimethyldiselenide [(CH₃)₂Se₂] (Mayland et al., 1989; Benson et al., 1992). The mineralization products in soils are, however, controlled by several factors including redox potential, precipitation-dissolution and adsorption-desorption reactions, the presence of other salts, and microbial immobilization (Mikkelsen et al., 1989; Parker et al., 1991; Blaylock and James, 1994; Zawislanski and Zavarin, 1996).

The exact forms of Se in seleniferous organic materials have not been well characterized. Martens and Suarez (1997a) investigated the assimilation of SeO_4^{2-} by alfalfa and mineralization of fresh alfalfa residue in soil. They found that 25% of the total Se assimilated was present as free Se-amino acids, Se-methylcysteine, selenomethionine, and selenocystine, but 30% of the assimi-

1218

H.A. Ajwa and G.S. Bañuelos, USDA-ARS, Water Management Res. Lab., 2021 S. Peach Ave., Fresno CA 93727; and H.F. Mayland, USDA-ARS, Northwest Irrigation and Soil Research Lab., Kimberly ID 83341. Received 16 June 1997. *Corresponding author (hajwa@ asrr.arsusda.gov).

Abbreviations: U.S. FDA, U.S. Food and Drug Administration; CEC. cation exchange capacity; DM, dry matter; GLM, general linear model; SAS, statistical analysis system; Seⁱⁿ, inorganic Se without organic material; Se^{ij}_{se}, inorganic Se plus succinate; Seⁱⁿ_{All}, inorganic Se plus nonseleniferous alfalfa tissues; Seⁱⁿ_{All}, seleniferous alfalfa tissues: Seⁱⁿ_{All}, seleniferous manure from cattle-fed alfalfa tissues; Seⁱⁿ_{All}, seleniferous manure from cattle-fed Astragalus praelongus tissues; Seⁱⁿ_{All}, seleniferous manure plus seleniferous manure from Astragalus praelongus tissues; Kⁱⁿ, control.

1219

lated Se was mineralized to water-soluble, non-amino acid, selenide-Se, and the remaining organic selenide-Se persisted in protein form. In another study, Martens and Suarez (1997b) found different mineralization and volatilization rates for the selenoamino acids, selenomethionine, and selenocystine in soils amended with animal manure. Their findings suggested that if selenomethionine is the dominant amino acid Se species in plant residue or animal manure, then most of the Se will be volatilized. But little volatilization is expected from selenocystine because it is very unstable in soil and will rapidly oxidize to Se^{2–}, and eventually to Se⁰ (Martens and Suarez, 1997b).

Although plant uptake of Se in inorganic forms $(SeO_4^{2-} and SeO_3^{2-})$ has been investigated extensively (Mayland et al., 1989; Bañuelos et al., 1991, 1993), plant uptake of organic Se has rarely been reported. Abrams et al. (1990) reported that selenomethionine was rapidly absorbed by wheat (Triticum aestivum L.). In a Hoagland's solution study, Williams and Mayland (1992) found that selenomethionine and selenocystine are absorbed by both a Se accumulator (two-grooved milkvetch, Astragalus bisulcatus) and a non-Se accumulator (western wheatgrass, Pascopyrum smithii) plant species, and that Se bioavailability follows the order: selenomethionine > selenocystine = SeO_4^{2-} . Sequential extraction techniques used by Zawislanski and Zavarin (1996) suggested that a large portion of total Se in soils can be in the organic fraction. Therefore, release of lowmolecular-weight Se compounds during organic matter decomposition may eventually be a source of Se for plant uptake. Little is known about plant availability of Se from different seleniferous plant residues and animal manures. Such information is needed to better understand the Se budget in the environment and to help determine strategies for lowering Se concentrations in soils and to supplement animal diets. Objectives of this study were to: (i) quantify the uptake of Se by canola and tall fescue grown in different soils amended with inorganic SeO₄²⁻, seleniferous alfalfa and seleniferous A. praelongus residues, and manures from cattle that consumed these two plant species, and (ii) assess changes in total and water-extractable Se concentrations in soils amended with seleniferous organic materials.

MATERIALS AND METHODS

Soils

Two nonseleniferous surface (0-20 cm) soils typical of central California were selected for this experiment: Hanford sandy loam soil (coarse-loamy, mixed, thermic Typic Durixeralfs), representative of the east side of the California Valley, and Panoche clay loam soil (fine-loamy, mixed, superactive, thermic Typic Torriorthents), representative of the west side. Field moist soil samples of the two soils were brought into the laboratory, mixed, and passed through a 5-mm sieve. At the time of sampling, Hanford and Panoche soils had the following properties: pH, 6.1 and 8.0; organic C, 2.6 and 5.6 g kg⁻¹; total N, 0.21 and 0.91 g kg⁻¹; sand, 610 and 320 g kg⁻¹; clay, 80 and 360 g kg⁻¹; and cation exchange capacity (CEC), 5.3 and 21 cmol_c kg⁻¹, respectively. The pH was determined by a glass combination electrode (soil/water ratio, 1:2.5), organic C by the method of Mebius (1960), CEC by neutral 1 Mammonium acetate method as described by Chapman (1965), and particle-size distribution by the pipette method of Kilmer and Alexander (1949). In preparation for planting, soil for each pot (3 kg of soil, on an oven-dry basis) was mixed on a plastic sheet with 100 mL of a nutrient solution containing 100 mg N as CO(NH₄)₂, 50 mg P as KH₂PO₄, 133 mg K as K₂SO₄, 14 mg Mg as MgSO₄, 11 mg Mn as MnSO₄, 12 mg Zn as ZnSO₄, 4 mg Cu as CuSO₄, 1.4 mg B as Na₂B₄O₇, 14 mg Fe as FeSO₄, and a total of 115 mg S as SO₄ (Allen et al., 1976). The soil was then treated with a SeO₄²⁻ solution with or without an organic material as described below.

Organic Materials

Organic materials selected as soil amendments were A. praelongus (no common name) tissues, alfalfa tissues, and manures from cattle fed these two plant tissues. Astragalus praelongus, a Se accumulator, was harvested as immature plants growing on seleniferous rangeland soils of Cretaceous geologic strata in southwestern Wyoming. Plants were clipped at 5 to 10 cm stubble height, air dried, and ground to pass a 2 mm sieve. The harvested A. praelongus residue contained 554 mg Se kg⁻¹ dry matter (DM). A portion of ground A. praelongus was mixed with nonseleniferous alfalfa and fed to ruminating adult cattle. Manure produced by cattle was collected, air dried, and ground to pass a 2 mm sieve. Selenium in this manure was derived entirely from the A. praelongus and averaged 10 mg Se kg⁻¹ DM.

Seleniferous alfalfa was produced on Nibley silt loam soil (fine, mixed, mesic Aquic Argiustoll) near Richmond in northeastern Utah. Alfalfa was in the vegetative growth stage when sprayed with aqueous solution of sodium selenite (Na₂SeO₃) and later sprinkler irrigated. Growth continued and seleniferous alfalfa was harvested at early bud stage, air dried, baled, subsampled, and ground to pass a 2 mm sieve. Selenium concentration in the seleniferous alfalfa residue was 112 mg Se kg⁻¹ DM. Seleniferous alfalfa was fed to adult cattle, and manure was collected, air dried, and ground as described above. Manure derived from cattle-fed seleniferous alfalfa contained 51 mg Se kg⁻¹ DM. Nonseleniferous alfalfa was grown without Se addition and processed like the seleniferous alfalfa.

Soil Treatments

Eight Se treatments were applied to the soils to obtain a final total Se concentration of 1.5 mg Se kg⁻¹ soil from inorganic or seleniferous organic sources (Table 1). In the inorganic SeO₄²⁻ treatment (Seⁱⁿ), SeO₄²⁻ was added to soils without any organic material. In the succinate-C treatment (Se_{Suc}^{in}), succinate-C was added to SeO²⁻-treated soils with irrigation water every 6 d at a rate of 0.5 g C kg⁻¹ soil d⁻¹ to achieve 30 g C kg⁻¹ soil in 60 d. The soluble C treatment was included to assess the effect of enhanced microbial activity caused by a readily available C source on Se uptake by plants. In the Se_{AM}^{in} and Se_{AM-M}^{in} treatments, SeO_4^{2-} -treated soils were amended with nonseleniferous alfalfa residue or with manure from cattle fed A. praelongus, respectively, at 30 g kg⁻¹ soil. At a 3% application rate, the Sein-M treatment contained only 0.3 mg Se kg⁻¹ soil as organic Se. Therefore, the Seⁱⁿ_{Au-M} treatment was complimented with a predetermined amount of Na2. SeO₄ solution to obtain the desired total Se concentration of 1.5 mg Se kg^{-1} soil.

In the organic Se treatments, a final total concentration of 1.5 mg Se kg⁻¹ soil was achieved by adding only seleniferous organic materials. These included alfalfa residue (Se^M_M), ma-

Table 1. Inorganic and organic Se treatments applied to Hanford sandy loam and Panoche clay loam soils.

т		Form of Se added	
l reatment symbol	Treatment	Inorganic†	Organic‡
		— mg Se kg	r' soil —
Sel	Inorganic Se without organic material	1.5	0
Set.	Inorganic Se plus succinate	1.5	0
Sein	Inorganic Se plus nonseleniferous alfalfa tissues	1.5	0
Se ⁱⁿ Asi-M	Inorganic Se plus seleniferous manure from cattle-fed Astragalus praelongus tissues	1.2	0.3
Sette	Seleniferous alfalfa tissues	0	1.5
Se ^M K-M	Seleniferous manure from cattle-fed alfalfa tissues	0	1.5
Ser.	Seleniferous A. praelongus tissues	0	1.5
Seam	Seleniferous manure from cattle-fed A. praelongus	0	1.5
K'	Control (soil only)§	0	0
K ⁸ All	Control (soil plus nonseleniferous alfalfa tissue)	0	0

† Selenium was added as selenate solution (Na₂SeO₄).

[‡] Total Se concentration in the seleniferous materials were: alfalfa tissue, 112 mg kg⁻¹; manure from alfalfa tissue, 51 mg kg⁻¹; A. praelongus tissue, 554 mg kg⁻¹; and manure from A. praelongus tissue, 10 mg kg⁻¹. § Hanford soil contained undetectable amount of Se. Panoche soil contained <0.04 mg Se kg⁻¹ soil.

nure from cattle-fed alfalfa (Se_{Alf-M}^{ot}), A. praelongus residue (Se_{As}^{ot}), and manure from cattle-fed A. praelongus (Se_{As-M}^{ot}). Except for the Se_{Asf-M}^{ot} treatment, the organic material was applied at 30 g kg⁻¹ soil. In the Se_{Asf-M}^{ot} treatment, the seleniferous A. praelongus manure was applied at a greater rate (150 g kg⁻¹ soil) to obtain a final Se concentration of 1.5 mg Se kg⁻¹ soil. Because the amounts of Se in seleniferous alfalfa or A. praelongus residues would exceed the desired 1.5 mg Se kg⁻¹ soil if applied at 30 mg kg⁻¹ soil, these residue were diluted by mixing with nonseleniferous alfalfa residue. The control treatments (K^0 and K_{Alf}^0) were included to assess effects of Se or plant residue on the DM yield.

After adding the respective treatment, the soil was mixed thoroughly, subsampled to determine total and water-extractable Se, and transferred into a 4 L plastic pot lined with double polyethylene bags. The moisture content of soil in each pot was adjusted with deionized water to a soil-matric potentials of -0.1 MPa and preincubated at room temperature for 14 d.

Growth Experiments

Four seeds of canola were planted 2 cm deep in each pot and thinned after 7d to two plants. Four freshly collected clones of tall fescue were planted in each pot. Clonal material was used to reduce the genetic variation. Certified "Fawn" tall fescue was maintained as distinct clones in an irrigated field nursery at Kimberly, ID. Seven days prior to initiation of this study, ramets were removed from one clone of "Fawn" and air freighted overnight to Fresno. Canola seeds and tall fescue ramets were grown in growth chambers (Environmental Growth Chambers, Chagrin Falls, OH) under light/dark temperatures of 22°C/18°C, with a light period of 16 h. The photosynthetic photon flux density in the growth chamber was measured once per week and ranged between 350 and 400 μ mol m⁻² s⁻¹. The experimental design was completely randomized with four replicates for each treatment, plant species, and soil type. Another four replicates of each treatment were left bare (without plants) to estimate Se losses from each soil induced by microbial activity. Throughout the experiment, water lost by evapotranspiration was replaced, thus maintaining soil-water potential of -0.05 to -0.1 MPa.

After 60 d, canola plants were harvested and separated into stems and leaves. The canola roots were removed from soil and washed with deionized water. During the 60 d, tall fescue shoots were clipped 2 cm above the soil in 20-d intervals, and the three clippings were combined and designated as first harvest. Tall fescue clippings 4, 5, and 6 were combined into one composite identified as the second harvest. Tall fescue roots were collected only after clipping 6.

After the first harvest, the soil planted with canola was mixed, subsampled, and analyzed for total and water-extractable Se. The nutrient solution described above was added and canola was again replanted and grown in the same pots for another 60 d under the same conditions. After the second growth period, both canola and tall fescue were harvested, and Se in the plant tissue and soil was determined.

Soil and Plant Tissue Analyses

The collected soil samples at preplant and each respective harvest were oven dried at 55°C for 7 d and ground to pass an 850 μ m sieve. At this temperature, Se volatilization losses were negligible. Water-soluble Se was determined on a saturated soil paste, and total Se was determined by atomic absorption with continuous hydride generation after wet acid digestion (HNO₃, H₂O₂, and HCl at 90°C for 24 h) as described by Bañuelos and Meek (1990).

All harvested plant parts were oven dried at 50°C in a forced draft oven for 7 d, weighed, and ground in a stainless steel Wiley mill equipped with an 850 μ m sieve. Plant tissue Se was determined by atomic absorption with continuous hydride generation after wet acid digestion as Bañuelos and Akohoue (1994) described. The NIST Standard Wheat Flour (SRM 1567; Se content of 1.1 ± 0.2 mg kg⁻¹) was used as an external quality control for Se analyses of plant samples. Selenium in plant tissues and soils was determined by a Thermo Jarrell Ash atomic absorption spectrometer (Smith-Hieftje 1000, Franklin, MA) equipped with an atomic vapor accessory hydride generator. Selenium measurements were made at the most sensitive resonance line (190.0 nm) using air-acetylene flame atomizer and hollow cathode lamp.

Statistical Analysis

The general linear model (GLM) procedure of the statistical analysis system (SAS) version 6.03 (SAS Institute, 1988) was used to compare DM yields between treatments, plant shoot Se concentrations between first and second harvests, and soil Se concentrations between the two soils for each respective harvest. No transformation of data was performed. Dry matter data were normally distributed, and specific contrasts in the GLM procedure were used to test for significant differences in shoot Se concentrations.

RESULTS

Soil Selenium

Bare Soils

Percentages of the total and water-extractable soil Se in the bare Hanford soil (without plants) at preincubation (initial), preplant, and each respective harvest are shown in Fig. 1. These percentages were calculated relative to the initial total Se concentration $(1.5 \pm 0.1 \text{ mg} \text{ kg}^{-1})$ measured immediately after adding the respective treatments (Table 1). Percentages of total Se measured in Panoche soil were similar to these shown for Hanford soil, and were not significantly different ($P \le 0.05$) between the soils for any of the treatments. Differences in





tive harvest of canola and tall fescue are shown in Fig. 2 and 3, respectively. These percentages were calculated relative to total Se concentrations measured in soils prior to planting. After the first harvest of canola, <20% of the added inorganic Se remained in the Seⁱⁿ or Seⁱⁿ_{Suc} treatments, but greater percentages of Se remained in soils with inorganic Se amended with nonseleniferous alfalfa or seleniferous organic materials. Statistical analysis for differences in total soil Se between Hanford and Panoche soils planted to canola showed that only the Seⁱⁿ_{Suc} and Se^X_{Ast} treatments of the first and second harvests of canola were significantly different ($P \le 0.05$). In comparison with results presented for Hanford soil (Fig.

2), the percentage of total Se concentrations in Panoche soil in the first and second harvests were 44 and 23% for the Se^m_{xet} treatment, respectively, and 86 and 71% for the Se^m_{Ast} treatment. There were no significant differences in total soil Se between Hanford and Panoche soils planted to tall fescue for any of the other treatments.

Plant Growth

Leaf mean DM yields of canola grown on Hanford soil ranged from 6.0 g for the Se^{α}_{All} treatment to 9.3 g for the Se^{α}_{Ast} treatment, and leaf DM yields of canola grown on Panoche soil ranged from 5.1 to 8.7 g for



Fig. 3. Total and water-extractable Se concentrations in Hanford soil at preplant and after second harvest (Harvest 2) of tall fescue. The superscripts "in" and "or" indicate inorganic and organic, respectively. For identification of treatments, see Table 1. Error bars represent the coefficient of variation (CV%) among replicates.

Se $_{\text{Mit}-M}^{\text{M}}$ and Se $_{\text{Mit}}^{\text{M}}$ treatments (data not shown), respectively. Although leaf DM yields of canola varied among the treatments within each soil type, differences in DM yields were significant ($P \le 0.05$) only between Seⁱⁿ or K^0 treatments and other treatments in the second harvest. Moreover, there were no significant differences ($P \le 0.05$) in DM yields between the two harvests of canola for each respective treatment, regardless of soil type. There were no significant differences ($P \le 0.05$) in canola stem DM yields among the various treatments or between the two harvests for each soil.

Total shoot mean DM yields of tall fescue grown in Hanford soil ranged from 11.7 g for the Seⁱⁿ treatment to 15.4 g for the Seⁱⁿ_{Ast-M} treatments, and yields of tall fescue grown in Panoche soil ranged from 10.8 to 17.6 g for the K^0 and Se $_{Ast}^{or}$ treatments (data not shown), respectively. In contrast to canola, shoot DM yields of tall fescue were significantly ($P \le 0.05$) greater for the second harvest than in the first harvest, regardless of treatment or soil type. Significant differences between the two harvests of tall fescue were attributed to better established root system and consequential new growth. For each harvest, there were no significant differences ($P \le 0.05$) among the various treatments.

Plant Selenium

Selenium accumulation in the various parts of canola and tall fescue grown in Hanford and Panoche soils are shown in Tables 2 and 3, respectively. Mean leaf tissue concentrations of canola grown in Hanford and Panoche

Treatment†	First harvest‡			Second harvest‡		
	Leaves	Stems	Roots	Leaves	Stems	Roots
· · · · · · · · · · · · · · · · · · ·	mg kg ⁻¹					
Hanford soil						
Se	283.8 (13.2)	54.8 (0.8)	87.5 (9.3)	5.7 (0.8)	0.6 (0.2)	0.8 (0.1)
Set.	213.8 (5.2)	50.1 (2.2)	55.1 (6.4)	7.7 (1.3)	1.2 (0.3)	2.3 (0.4)
Set.	41.0 (9.3)	18.5 (0.1)	14.8 (1.4)	3.6 (0.2)	0.8 (0.4)	1.4 (0.5)
Sein	29.2 (12.8)	14.8 (2.8)	14.3 (1.5)	1.1 (0.2)	0.6 (0.2)	2.2 (0.3)
Soft.	1.6 (0.2)	0.6(0.1)	0.6 (0.2)	1.5 (0.1)	0.7 (0.1)	1.6 (0.2)
Setting	1.6 (0.3)	0.9 (0.3)	1.8 (0.1)	2.2 (0.9)	0.7 (0.1)	2.3 (0.2)
Seam	6.7 (1.4)	2.8 (9.6)	2.5 (0.5)	0.8 (0.1)	0.3 (0.1)	0.9 (0.1)
Se ^r An	3.9 (0.4)	1.4 (0.5)	2.0 (0.1)	5.0 (0.9)	0.8 (0.1)	3.6 (0.3)
Panoche soil						
Sei	217.8 (5.4)	57.0 (3.5)	83.2 (5.8)	8.5 (1.2)	4.3 (0.5)	3.7 (1.2)
Set	128.1 (14.2)	37.3 (0.7)	62.2 (4.0)	33.1 (3.6)	5.6 (1.7)	5.8 (0.9)
Se ⁱⁿ .	82.8 (5.1)	18.8 (1.3)	18.6 (1.8)	5.6 (2.7)	3.2 (0.7)	3.0 (0.6)
Set	33.5 (8.7)	16.6 (3.1)	16.3 (3.0)	5.2 (0.3)	2.9 (0.7)	4.2 (0.1)
Set.	2.3 (0.4)	0.5 (0.1)	0.9 (0.2)	3.5 (0.2)	2.7 (1.3)	2.2 (0.2)
Set	1.6 (0.3)	0.9 (0.7)	1.6 (0.3)	6.3 (0.5)	2.1 (1.0)	2.8 (0.7)
Sof	6.2 (0.6)	1.7 (0.3)	1.9 (0.2)	1.6 (0.2)	0.8 (0.1)	1.0 (0.1)
Sean	5.0 (0.4)	1.5 (0.8)	3.7 (0.4)	9.8 (0.5)	4.9 (2.1)	4.0 (1.0)

Table 2. Accumulation of Se in canola grown in Hanford sandy loam and Panoche clay loam soils.

† See Table 1 for the corresponding treatments.

t Values are the means from four replications with the standard deviation in parentheses.

soils receiving only inorganic Se were 284 and 218 mg Se kg⁻¹ DM, respectively, and mean shoot concentrations of tall fescue grown in the respective soils were 75 and 52 mg Se kg⁻¹ DM. Selenium accumulation by canola tissues (Table 2) was consistently greater than by tall fescue tissues (Table 3) in all treatments of the first harvest, regardless of soil type.

All plant tissues from canola and tall fescue grown in the inorganic SeO₄²⁻ treatment that did not receive organic amendment (Seⁱⁿ) accumulated the greatest amounts of Se compared to all other treatments (Tables 2 and 3). Differences in leaf Se concentrations of canola were significant ($P \le 0.05$) between the two harvests of any inorganic Se treatment (Seⁱⁿ, Seⁱⁿ_{Ast-M}, and Seⁱⁿ_{Suc}). The addition of succinate-C (Seⁱⁿ_{Suc}), nonseleniferous alfalfa residue (Seⁱⁿ_{Att}) or *A. praelongus* manure

Table 3. Accumulation of Se in tall fescue grown in Hanford sandy loam and Panoche clay loam soils.

	First harvest‡	Second	harvest‡
Treatment†	Shoots§	Shoots§	Roots
		mg kg-1	
Hanford soil			
Sein	75.2 (5.0)	74.6 (4.8)	29.3 (1.7)
Set	33.6 (6.2)	20.4 (2.4)	17.0 (1.8)
Set	13.2 (3.1)	1.4 (0.3)	2.1 (0.2)
Set	6.2 (0.8)	5.5 (0.6)	5.5 (1.8)
Set	0.4 (0.1)	0.8 (0.1)	1.4 (0.1)
Settem	0.7 (0.2)	0.9 (0.2)	2.6 (0.3)
Ser.	0.9 (0.1)	0.8 (0.1)	1.8 (0.4)
Set	0.9 (0.2)	1.2 (0.2)	3.5 (0.3)
Panoche soil			
Sein	52.4 (3.2)	17.5 (2.4)	14.6 (0.3)
Set.	35.6 (4.5)	13.4 (1.5)	13.3 (0.5)
Sein	17.7 (2.4)	5.0 (0.4)	14.2 (1.9)
Sein	2.7 (0.4)	3.5 (0.8)	2.9 (0.7)
Set	1.2 (0.1)	1.7 (0.3)	2.8 (0.5)
Setun	1.4 (0.3)	2.3 (0.6)	3.0 (0.2)
Set	0.8 (0.3)	0.9 (0.4)	2.3 (0.8)
Seam	2.2 (0.3)	3.3 (0.5)	4.8 (0.2)

† See Table 1 for the corresponding treatments.

‡ Values are the means from four replications with the standard deviation in parentheses.

§ Includes stems and leaves.

 $(Se_{a_{si-M}}^{i_{a_{si-M}}})$ to soils containing soluble SeO_4^{2-} reduced Se accumulation in both canola and tall fescue plants.

The reduction in tissue Se in the Seⁱⁿ_{Suc} treatment of the first harvest was greater in Panoche clay loam soil than in Hanford sandy loam. Tissue Se concentrations of both canola and tall fescue grown in the Seⁱⁿ_{Suc} treatment were less than those grown in the Seⁱⁿ treatment of either soil. In the second harvest of Panoche soil, the converse was true; that is, canola tissues in the Seⁱⁿ_{Suc} treatment accumulated more Se than in the Seⁱⁿ treatment. In contrast to alfalfa residue or animal manure, the addition of succinate-C to Panoche soil maintained a large portion of added SeO²⁻₄ in the water soluble forms that were readily available for plant uptake in the second harvest.

In contrast to inorganic SeO₄²⁻ treatments, differences among the organic Se treatments or between the two harvests of canola (Table 2) were inconsistent and varied for each soil. For tall fescue (Table 3), there were no significant differences ($P \le 0.05$) in shoot Se concentrations of any of inorganic or organic Se treatments between the two harvests. Except for Se^{or}_{Asi} treatment, both canola and tall fescue accumulated significantly $(P \le 0.05)$ greater amounts of Se from the seleniferous organic treatments (Se $_{Alt}^{or}$, Se $_{Alt-M}^{or}$, and Se $_{Ast-M}^{or}$) in the second harvest than in the first harvest. There were no significant differences ($P \le 0.05$) in Se concentrations for first and second harvests of tall fescue or for the first harvest of canola shoots grown in both soils amended with organic seleniferous materials. For the second harvest of canola, differences in leaf Se concentrations between Seas and Seas and between Seaf and Se^{or}_{Ast-M} in both soils were significant. The effect of C substrate addition (Seⁱⁿ vs. Seⁱⁿ_{Suc} + Seⁱⁿ_{Alf} + Seⁱⁿ_{Ast-M}) and the effect of plant residue (Seⁱⁿ + Seⁱⁿ_{Suc} vs. Se^{or}_{Alf} + Se^{or}_{Ast}) on Se uptake were significantly different in both harvests of canola and tall fescue. Selenium uptake from plant residue vs. manure (Sear + Sear vs. Sear + Se^{or}_{Ast-M}) were only significantly different ($P \le 0.05$) in the second harvest of canola.

DISCUSSION

Soil Selenium

The percentage of Se loss from the bare soils that did not receive nutrient solution was <10% (data not shown). The addition of nutrient solution to the Seⁱⁿ treatment of bare soils (Fig. 1) enhanced Se losses up to 20% of the added SeO₄²⁻. Karlson and Frankenberger (1989) found that addition of some elements (N, Co, Zn, and Ni) to Se-laden soils resulted in greater volatilization losses of Se due to enhanced microbial activities. In our study, direct measurements of Se volatilization were not made. Because there were no leaching losses from pots, we assumed that losses in total Se were due to microbial volatilization.

A large portion of added SeO_4^{2-} in Se_{Alf}^{in} , Se_{Ast-M}^{in} , and Sein treatments was not recovered in the water extract after 120 d, possibly due to slow conversion of the added SeO_4^2 into SeO_4^2 that can be tightly adsorbed on soil surfaces. These results are consistent with other studies on effects of organic C addition on SeO₄²⁻ immobilization in soils (Calderone et al., 1990; Neal and Sposito, 1991). Neal and Sposito (1991) found that addition of dextrose resulted in transforming a large portion (64-90%) of added SeO $_4^{2-}$ into organically associated forms. They also noted wetting and drying of soil promoted rapid transformation of SeO²⁻ into other forms, such as SeO_3^{2-} and organic Se, that were adsorbed to soil surfaces. In our study, however, the immediate decrease (within 30 min) in water-soluble Se in Seⁱⁿ and Se^{in}_{Ast-M} treatments possibly was due to inability of the extraction technique to remove all of the added SeO_4^{2-} from the organic materials for Se analysis.

Less than 20% of total Se added as seleniferous organic materials to soils (Se^M_{All}, Se^M_{All}-M, Se^M_{Asl}, and Se^M_{Asl-M} treatments) was lost in 120 d. Selenium losses from Se^M_{All} and Se^M_{All-M} treatments were slower than losses in the Se^M_{Asl} treatment. These results could be due to the fact that Se in alfalfa is mainly present as protein bound selenomethionine and selenocystine that are less water soluble than the nonprotein forms of stored Se in *A. praelongus* tissues, like Se methylselenocysteine (Mayland, 1994). The slower release of Se from Se^M_{Asl-M} than from Se^M_{Asl} treatment may be due to absorption of soluble inorganic compounds and Se amino acids by the digestive tract of animals resulting in more refractory Se compounds in manure.

Plant Selenium

Selenium accumulation by canola tissues (Table 2) was consistently greater than by tall fescue tissues (Table 3) in all treatments of the first harvest, regardless of soil type. These results were expected because of canola's high affinity for S and its apparent inability to discriminate between absorbing Se and S species from the soil (Mayland et al., 1989), especially when Se is present in soils as soluble SeO_4^{2-} . Similar results on Se accumulation by tissues of canola and tall fescue have been reported in earlier studies (Bañuelos and Meek, 1990; Bañuelos et al., 1993, 1997). The ability of a plant

to absorb and accumulate Se, however, depends on Se forms and the presence of soluble ions (e.g., SO_4^{2-} and Cl^-) in soil solution (Parker et al., 1991; Ba-fuelos et al., 1996).

All plant tissue from canola and tall fescue grown in the inorganic SeO_4^{2-} treatment that did not receive organic amendment (Sein) accumulated the greatest amounts of Se compared to all other treatments (Tables 2 and 3). Addition of alfalfa residue or cattle manure to soils likely stimulated microbial assimilatory reduction of the added SeO₄²⁻, thus reducing its bioavailability. Our results support earlier studies that found a reduction in plant Se with increased organic C in soils (Bisbjerg and Gissel Nielsen, 1969; Levesque, 1974). Blaylock and James (1994) found that the addition of Mn oxides and organic acids (ascorbic and gallic acids) to some soils increased the amount of soluble Se and enhanced the oxidation of SeO₄ to SeO₄. In our study, the effect of succinate-C on Se accumulation by canola was less pronounced in Hanford soil than in Panoche soil. This possibly was due to greater plant absorption of Se from Hanford soil during the first harvest such that there was less soil Se available for plant uptake in the second harvest.

Leaf tissues of canola accumulated up to 10 mg Se kg^{-1} DM in the second harvest when grown in the organic-treated soils (Table 2), even though water-extractable Se concentrations were very small (Fig. 2). These results suggest that canola absorbed forms of Se other than the water-soluble forms (i.e., adsorbed, organic, etc.), or that canola's root activities may have increased Se availability by changing the redox potential in the rhizosphere (Blaylock and James, 1994; Bañuelos et al., 1997). The immobilized organic Se could have been released from dead microbial cells (Cook and Brilliant, 1987) that could become readily available for plant uptake in the second harvest.

With a few exceptions, both plants grown in soils amended with seleniferous organic materials accumulated Se concentrations at or below the toxic levels for animals (<3 mg Se kg⁻¹ DM (Mayland et al., 1989). Continuous application of seleniferous organic materials, however, may increase soil and plant Se concentrations. Cattle and sheep (*Ovis aries*) can consume slightly seleniferous forage plant tissues (4–5 mg kg⁻¹ DM) without suffering Se toxicity (Mayland et al., 1989). Animals consuming plant tissues containing 5 to 20 mg Se kg⁻¹ DM for a prolonged period are likely to suffer chronic or even acute Se poisoning (Girling, 1984). In our study, tissues of both canola and tall fescue grown in soils treated with inorganic SeO₄²⁻ contained Se concentrations that may cause acute toxicity to grazing animals.

Selenium Budget

The Se mass balance for various treatments of Hanford and Panoche soils is presented in Table 4. These calculations were based on total soil Se differences between preplant and corresponding harvests (Fig. 2 and 3) and absolute amounts of Se accumulated by whole plant (based on Se concentration and total dry mass of

Table 4. Percentage of Se accumulated in canola and tall fescue tissues and the "unaccounted for" Se losses relative to total soil Se losses during 120 d.

	Canola		Tall fescue	
Treatme nt †	Accumulated in tissues‡	Unaccounted for losses§	Accumulated in tissues‡	Unaccounted for losses§
		q	%	
Hanford soil				
Se	49	41	29	26
Set.	54	33	23	21
Seit	19	25	5	26
Set	29	9	24	4
Set.	3	23	2	17
Set	8	12	15	4
Set	10	19	4	15
Sen M	13	8	6	8
Panoche soil				
Sein	62	14	25	15
Set	42	35	28	6
Set	34	17	16	15
Set	26	9	10	4
Set	7	17	4	16
Set	9	6	20	7
Set.	5	30	4	22
Set	14	3	9	8

† See Table 1 for the corresponding treatments.

2 Calculations were based on total soil Se differences between preplant and final harvest corresponding harvest, and total Se amount accumulated by the plant.

§ The "unaccounted for" Se loss is the percentage of Se not recovered in plant tissues or in soil, (100 - % in plant tissues - % remaining in soil).

all plant parts harvested). Percentages of "unaccounted for" Se losses are those not recovered in plant tissue or soil. The percentage of Se accumulated by two harvests of canola and all clippings of tall fescue relative to total soil Se loss was the greatest in the Seⁱⁿ and Seⁱⁿ_{Suc} treatments. The addition of nonseleniferous alfalfa (Seⁱⁿ_{Alf} treatment) slightly affected "unaccounted for" Se losses from the SeO²⁺₄-treated soils, whereas addition of cattle manure (Seⁱⁿ_{Alf} treatment) greatly reduced these losses.

For seleniferous organic material treatments (Se_{All}^{a} , Se_{All-M}^{a} , Se_{Asl}^{a} , and Se_{Asl-M}^{a}), percentages of Se accumulated by plants relative to total soil Se losses varied widely between the two soils and among various treatments. The "unaccounted for" Se losses were, in general, less in cattle manure treatments (Se_{All-M}^{a} and Se_{Asl-M}^{a}) than in plant residues treatments (Se_{Asl-M}^{a} and Se_{Asl-M}^{a}) than in plant residues treatments (Se_{Asl-M}^{a} and Se_{Asl-M}^{a}). These results suggest that a large fraction of Se in seleniferous plant residues incorporated into soils would be volatilized, but only a small fraction of Se in seleniferous cattle manure would be volatilized, and therefore, Se may accumulate in soils.

Although direct measurements of volatilization were not made, Se volatilization can be estimated for each pot from the "unaccounted for" Se losses. For example, the largest amount of the "unaccounted for" Se loss was in the Seⁱⁿ treatment (41%). Therefore, 1845 µg Se pot⁻¹ (41% × 4500 µg Se pot⁻¹) was lost in 120 d, or 15.4 µg pot⁻¹ d⁻¹. The estimated volatilization rate of two canola plants in 0.04 m² surface area was ~384 µg Se m²⁻ d⁻¹. The smallest amount of the "unaccounted for" Se loss was for the Se^X_{Ast-M} treatment (3%), which has an estimated volatilization rate of ~28 µg Se m²⁻ d⁻¹. In comparison, the estimated volatilization rates in the Seⁱⁿ and Se $_{Asi-M}^{cr}$ treatments of the bare soils were 140 and 19 µg Se m²⁻ d⁻¹. Terry and Zayed (1994) reported that other *Brassica* spp. volatilize Se at rates between 280 and 340 µg Se m²⁻ d⁻¹. However, Se accumulation and volatilization rates by canola under controlled growth conditions are expected to be greater than those under field conditions.

CONCLUSIONS

Addition of plant residue or animal manure to soils treated with inorganic SeO_4^{2-} enhanced Se losses from bare soils. Accumulation of Se by both plant species was initially greater with inorganic SeO₄²⁻ than with seleniferous organic Se sources, and uptake was generally greater from Hanford sandy loam soil than Panoche clay loam soil. In soils treated with inorganic SeO_4^{2-} , tissue Se concentrations in successive plantings of canola greatly declined, whereas tissue Se concentrations in multiple clippings of tall fescue remained high. Addition of nonseleniferous plant residues or animal manures to soils containing soluble SeO₄²⁻ significantly decreased Se uptake by both canola and tall fescue. Both plant species grown in soils amended with seleniferous organic materials accumulated Se concentrations at or below the toxic levels for animals. Caution should be taken, however, when extrapolating results obtained under controlled conditions to the field condition or soils containing other forms of Se. Long-term field studies are needed to monitor Se transformation in soils amended with seleniferous crop residues and organic wastes.

ACKNOWLEDGMENTS

We are greatful to Dr. Dean Martens, USDA-ARS, Ames, 1A, for his helpful discussions. Thanks to Dr. K.E. Panter, USDA-ARS, Logan, UT, for the supply of seleniferous alfalfa and Astragalus praelongus.

REFERENCES

- Abrams, M.M., C. Shennan, R.J. Zasoski, and R.G. Burau. 1990. Selenomethionine uptake by wheat seedlings. Agron. J. 82: 1127-1130.
- Allen, S.E., G.L. Terman, and L.B. Clements. 1976. Greenhouse techniques for soil-plant-fertilizer research. National Fertilizer Development Center, Tennessee Valley Authority Bull. Y-104.
- Arthur, M.A., G. Rubin, and R.E. Schneider. 1992. Uptake and accumulation of selenium by terrestrial plants growing on a coal fly ash landfill. Part 2. Forge and root crops. Environ. Toxic. Chem. 11:1289-1299.
- Bañuelos, G.S., H.A. Ajwa, B. Mackey, L. Wu, C. Cook, S. Akohoue, and S. Zambrzuski. 1997. Evaluation of different plant species used for phytoremediation of high soil selenium. J. Environ. Qual. 26: 639-646.
- Bañuelos, G.S., and S. Akohoue. 1994. Comparison of wet digestion and microwave digestion on selenium and boron analysis in plant tissues. Comm. Soil Sci. Plant Anal. 25:1655-1670.
- Bañuelos, G.S., G.E. Cardon, C.J. Phene, L. Wu, S. Akohoue, and S. Zambrzuski. 1993. Soil boron and selenium removal by three plant species. Plant Soil 148:253-263.
- Banuelos, G.S., R. Mead, and S. Akohoue. 1991. Adding seleniumenriched plant tissue to soil causes the accumulation of selenium in alfalfa. J. Plant Nutr. 14:701-713.
- Bañuelos, G.S., R. Mead, L. Wu, P. Beuselinck, and S. Akohoue. 1992. Differential selenium accumulation among forage plant spe-

cies grown in soils amended with selenium-enriched plant tissue. J. Soil Water Conserv. 47:338-342.

- Bañuelos, G.S., and D.W. Meek. 1990. Accumulation of selenium in plants grown on selenium-treated soil. J. Environ. Qual. 19:772-777.
- Bañuelos, G.S., A. Zayed, N. Terry, L. Wu, S. Akohoue, and S. Zambrzuski. 1996. Accumulation of selenium by different plant species grown under increasing sodium and calcium chloride salinity. Plant Soil 183:49-59.
- Benson, S.M., T.K. Tokunaga, and P. Zawislanski. 1992. Anticipated soil selenium concentrations at Kesterson Reservoir. Rep. 33080. Lawrence Berkeley Lab., Univ. of California, Berkeley.
- Bisbjerg, B., and G. Gissel-Nielsen. 1969. The uptake of applied selenium by agricultural plants. I. The influence of soil type and plant species. Plant Soil 31:287-298.
- Blaylock, M.J., and B.R. James. 1994. Redox transformation and plant uptake of Se resulting from root-soil interactions. Plant Soil 158: 1-17.
- Calderone, S.J., W.T. Frankenberger, Jr., D.R. Parker, and V. Karlson. 1990. Influence of temperature and organic amendments on the mobilization of selenium in sediments. Soil Biol. Biochem. 22: 615-620.
- Chapman, H.D. 1965. Cation-exchange capacity. p. 891-901. In C.A. Black et al. (ed.) Methods of soil analysis. Part 2. Agron. Monogr. 9. ASA, Madison, WI.
- Cook, T.D., and K.W. Brilliant. 1987. Aquatic chemistry of selenium: Evidence of biomethylation. Environ. Sci. Technol. 21:1214-1219.
- Girling, C.A. 1984. Selenium in agriculture and the environment. Agric. Ecosys. Environ. 11:37-65.
- Karlson, U., and W.T. Frankenberger, Jr. 1989. Accelerated rates of selenium volatilization from California soils. Soil Sci. Soc. Am. J. 53:749-753.
- Kilmer, V.J., and L.T. Alexander. 1949. Methods of making mechanical analysis of soils. Soil Sci. 68:15-24.
- Levesque, M. 1974. Some aspects of selenium relationships in eastern Canadian soils and plants. Can. J. Soil Sci. 54:205-214.
- Logan, T.J., A.C. Chang, A.L. Page, and T.J. Ganje. 1987. Accumulation of selenium in crops grown on sludge-treated soil. J. Environ. Qual. 16:349-352.
- Martens, D.A, and D.L. Suarez. 1997a. Selenium in water management wetlands in the semi-arid west. Hortic. Sci. (in press)
- Martens, D.A., and D.L. Suarez. 1997b. Mineralization of seleniumcontaining amino acids in two California soils. Soil Sci. Soc. Am. J. 61:1685-1694.

- Mayland, H.F. 1994. Selenium in plant and animal nutrition. p. 29-45. In W.T. Frankenberger, Jr. and S. Benson (ed.) Selenium in the environment. Marcel Dekker, New York.
- Mayland, H.F., L.J. James, K.E. Panter, and J.L. Sonderegger. 1989. Selenium in seleniferous environments. p. 15-50. In L.W.G. Jacobs (ed.) Selenium in agriculture and the environment. SSSA Spec. Publ. 23. SSSA, Madison, WI.
- Mebius, L.J. 1960. A rapid method for the determination of organic carbon in soil. Anal. Chim. Acta 22:120-124.
- Mikkelsen, R.L., A.L. Page, and F.T. Bingham. 1989. Factors affecting selenium accumulation by agricultural crops. p. 65-94. In L.W. Jacobs (ed.) Selenium in agriculture and the environment. SSSA Spec. Publ. 23. SSSA, Madison, WI.
- Neal, R.H., and G. Sposito. 1991. Selenium mobility in irrigated soil columns as affected by organic carbon amendment. J. Environ. Qual. 20:808-814.
- Ohlendorf, H.M. 1989. Bioaccumulation and effects of selenium in wildlife. p. 133–177. *In* L.W. Jacobs (ed.) Selenium in agriculture and the environment. SSSA Spec. Publ. 23. ASA and SSSA. Madison, WI.
- Oldfield, J.E., R. Burau, G. Moller, H.M. Ohlendorf, and D. Ullrey. 1994. Risks and benefits of selenium in agriculture. Council Agric. Sci. Tech. Issue Paper Suppl. 3. Ames, IA.
- Parker, D.R., A.L. Page, and D.N. Thomas. 1991. Salinity and boron tolerances of candidate plants for the removal of selenium from soils. J. Environ. Qual. 20:157-164.
- SAS Institute. 1988. SAS user's guide: Statistics. 6th ed. SAS Inst., Cary, NC.
- Shrift, A. 1973. Metabolism of selenium by plants and microorganisms. p. 763-814. *In* D.L. Klayman and W.H.H. Gunther (ed.) Organic selenium compounds: Their chemistry and biology. John Wiley & Sons, New York.
- Terry, N., and A.M. Zayed. 1994. Selenium volatilization by plants. p. 343-368. In W.T. Frankenberger, Jr. and S. Benson (ed.) Selenium in the environment. Marcel Dekker, New York.
- U.S. Food and Drug Administration. 1993. Federal register. 58:47962-47973. U.S. Gov. Print. Office, Washington, DC.
- Williams, M.C., and H.F. Mayland. 1992. Selenium absorption by twogrooved milkvetch and western wheat grass from selenomethionine, selenocystine, and selenite. J. Range Manage. 45:374-378.
- Zawislanski, P.T., and M. Zavarin. 1996. Nature and rates of selenium transformations: A laboratory study of Kesterson Reservoir soils. Soil Sci. Soc. Am. J. 60:791-800.