

Effects of Selenium Supplementation on Four Agricultural Crops

KATHLEEN M. CARVALHO,* MARIA T. GALLARDO-WILLIAMS,
ROBERT F. BENSON, AND DEAN F. MARTIN

Institute for Environmental Studies, Department of Chemistry, University of South Florida,
4202 East Fowler Avenue, Tampa, Florida 33620

Agricultural crops can be used either to remediate selenium-contaminated soils or to increase the daily selenium intake of consumers after soil supplementation using inorganic or organic selenium sources. In this study, four agricultural crops were examined for potential selenium enhancement. Soils containing tomato, strawberry, radish, and lettuce plants were supplemented with either an inorganic or an organic form of selenium. Two different soils, i.e., low Se and high Se containing, were also used. Statistically significant differences in appearance, fruit production, and fresh weights of the fruit produced were studied. Next, the amount of selenium retained in the edible fruits, nonedible plant, and soil for each was analyzed by acid digestion followed by hydride generation atomic absorption analysis. Finally, inhibition effects on the seeds of the agricultural plants were studied. The results show that supplementation with an inorganic form of selenium led to higher retention in the plants, with a maximum of 97.5% retained in the edible portion of lettuce plants.

KEYWORDS: Selenium; supplementation; crops

INTRODUCTION

For many years, selenium has been recognized as an essential trace element necessary for both human and livestock nutrition (1–3). With regard to human nutrition, there have been numerous attempts to associate low or suboptimal levels of selenium intake with a wide variety of human diseases, such as heart disease, cystic fibrosis, and cancer (4). It was reported that after selenium supplementation, an infant diagnosed with cystic fibrosis had a reversed positive sweat test (5). Selenium has been recognized to play an important role in several selenoproteins (6). The recent interest in selenium has resulted from the promise it has shown in the treatment of lung, prostate, and colon types of cancer (7). The work of Clark and co-workers led to the study of selenium compounds as a relevant group of cancer chemopreventive agents (8). At doses substantially higher than the physiological requirements, inorganic selenium protects laboratory animals against cancer of the mammary gland, colon, lung, pancreas, liver, and skin. Organic and some selenium-containing amino acids were also effective and have the same side effects (9). The National Research Council has established a Recommended Dietary Allowance (RDA) of selenium for humans, 55 and 70 $\mu\text{g}/\text{day}$ for men and women, respectively (10). However, most of the selenium humans take in is from foods, primarily meat and wheat, which are usually low in selenium content (11). Nutritional supplements have also been recommended to increase daily selenium; however, recent studies have shown that the amount of selenium in over-the-counter supplements can be much lower than advertised (12).

Therefore, one focus seems to be on increasing selenium intake through natural sources (13), such as garlic (14) and broccoli (15). It has been shown that the most effective way to increase the amount of selenium in cultivated crops is to add selenite or selenate to fertilizers, spray the crops with selenium salts, or treat the seeds with aqueous selenium (16–19).

Attempts have also been made to increase the amount of selenium available in the soil for animal fodder and human food. A range of 0.05–0.1 mg/kg dry matter is typically used; however, after chronic exposure to fodder that exceeds 1 mg/kg dry matter, toxic effects can be expected (20). Keeping this limit in mind, soils in different areas of the world may be characterized as selenium-deficient, selenium-adequate, and selenium-toxic. In Finland, selenium has been added to fertilizers since 1984, in the form of selenate, to increase the selenium in the soils (21). Vegetables rich in selenium have been shown to contribute as much as 28–32% of the daily intake in Northern Mexico (22).

The purpose of this study was to evaluate the feasibility of selenium supplementation of some commercial agricultural crops. This includes the form of selenium—both organic (selenomethionine) and inorganic (selenium dioxide)—sources to supplement, as well as the type of crop (four different species of plants) in a controlled environment. After the agricultural crops were analyzed and selenium was found to remain in the soil, the effects of selenium supplementation on germination and plant growth were studied. Selenium has not been demonstrated as an essential nutrient for plant growth; therefore, the effect of selenium concentration was also studied.

MATERIALS AND METHODS

Agricultural Crops. Tomato seeds (*Lycopersicon esculentum* var. Homestead, NK Lawn & Garden, Chattanooga, TN), lettuce seeds (*Lactuca sativa* var. Black Seeded Simpson, Fredonia Seeds, Minneapolis, MN), and radish seeds (*Raphanus sativum* var. Early Scarlet Globe, NK Lawn & Garden) were obtained and grown. Sweet Charlie strawberry plants were obtained from Armenia Nursery, Tampa, FL.

Growth Conditions. Plants were grown in a controlled environment phytotron room (Environmental Growth Chambers, Chagrin Falls, OH) with a photoperiod of 12 h light/12 h dark, a relative humidity of 80%, a constant temperature of 26 °C, and light intensity of 190 $\mu\text{einsteins m}^{-2} \text{ s}^{-1}$. Plants were grown in sand/soil pots and subjected to the following selenium supplementation conditions. The plants for each crop examined were divided into five treatment groups, each with four replicates: control sets, no selenium supplementation; test 1, each plant received a total of 1.5 mg of an inorganic form of selenium per kg of soil; test 2, each plant received a total of 1.5 mg of an organic form of selenium per kg of soil; test 3, each plant received a total of 40 mg of an inorganic form of selenium per kg of soil; and test 4, each plant received a total of 40 mg of an organic form of selenium per kg of soil. Tests 1 and 2 were considered to have received an "optimum" level of selenium and tests 3 and 4 to receive a "supraoptimal" or "high" amount of selenium. We chose the supplementation concentration of 1.5 mg of Se/kg of soil on the basis of previous studies (23, 24), and 40 mg of Se/kg of soil was used as a much higher amount to test toxicity effects.

Chemical Analysis. The amount of selenium in the fruit and vegetables, if any, was quantified by using hydride generation atomic absorption spectroscopy. The entire plant (leaves, stem, and roots) was then analyzed by atomic absorption spectroscopy to determine if any accumulation of selenium was present and if the plant absorbed the selenium but retained it in the green portions of plants rather than the fruit and vegetables. Last, the amount of selenium retained in the soil was determined by atomic absorption spectroscopy and energy dispersion X-ray fluorescence (XRF).

The fruit and vegetable samples were digested in the following manner, keeping in mind the volatile nature of selenium (25). Each individual fruit or vegetable was first weighed and then blended for 2 min. A 1-g fresh weight aliquot was weighed and then placed in a 150-mL Teflon beaker. Next, 5 mL of nitric acid (16 M) and 5 mL of deionized water were added. The sample was covered with a watch glass and heated for 10–15 min without boiling (90–95 °C). The sample was then allowed to cool, another 5 mL of nitric acid (16 M) was added, and the sample was heated (90–95 °C) for an additional 30 min. This step was repeated, and then 2 mL of deionized water and 3 mL of 30% hydrogen peroxide were added slowly, and the mixture was heated until effervescence ceased. Finally, 5 mL of hydrochloric acid (12 M) was added, and the mixture was refluxed for 10–15 min. The sample was cooled to room temperature and then diluted to 100 mL with 6% (v/v) HCl. Next, the sample was vacuum filtered in an all-glass filtration apparatus using a 0.45- μm Millipore membrane filter. Finally, the aliquot was diluted with 6% (v/v) HCl to 100 mL using a volumetric flask and then analyzed using atomic absorption.

The plant samples were digested in the following manner. The entire plant (leaves, stems, and roots) was weighed for a total fresh weight. Next, the plant was allowed to dry in the phytotron for 1 week. A dry weight was taken, and a 1-g portion was weighed and then placed in a 150-mL Teflon beaker. The sample was then treated with nitric acid, as with fruit and vegetable samples, and analyzed in the same way as before.

The soil samples were digested in the following manner. A soil sample weighing approximately 1 g was weighed and then placed in a 150-mL Teflon beaker. The samples were treated with nitric acid and analyzed using the same procedure as that used for fruit and vegetable samples.

The soil samples were also analyzed by energy dispersion XRF in order to determine whether any significant differences were observed in elements other than selenium. A 1-g sample was weighed and then ground into a powder using a mortar and pestle. Samples prepared in this manner were analyzed by energy dispersion XRF in a JEOL

Table 1. Data Summary for Comparison of Tomato and Strawberry Fruit Produced^a

	control	high organic	high inorganic	low organic	low inorganic
tomato	1.3 ± 0.2	0.0 ± 0.0 ^b	0.5 ± 0.3	2.3 ± 0.3	1.3 ± 0.2
strawberry	3.3 ± 2.1	2.0 ± 0.8	3.0 ± 0.8	2.0 ± 1.2	1.8 ± 1.0

^a $n = 4$. Scale, number of fruit produced. ^b Statistically significant difference at 95% confidence level.

scanning electron microscope (model JFM-840) in order to determine the amount of selenium and other significant elements for each soil sample. For each species studied, the elemental composition of the control was compared to that of the selenium-supplemented samples. We also obtained a soil sample from the Gulf Coast Research and Education Center, University of Florida Institute of Food (IFAS) and Agricultural Sciences, Dover, FL, as well as a sample from an anonymous strawberry farm in Plant City, obtained from IFAS, and analyzed these samples using XRF. One of the major observations we wanted to examine was the change in the amount of sulfur between the controls and the treatments. It has been shown that selenium will substitute for sulfur in plants (2); therefore, this was examined.

Seed Bioassays. Seeds from lettuce, tomatoes, and radishes were evaluated for inhibition effects from selenium supplementation. Seeds were washed with soap and water, and then with 5% bleach, rinsed with deionized water, and gravity filtered using Whatman No. 1 filter paper. Seeds were evaluated using a randomized design with each concentration in triplicate. Concentrations of 0, 10, 20, 50, 100, and 200 ppm were used in the bioassays. Using trays with individual wells, 10 seeds were counted and distributed to each well, which was lined with Whatman No. 1 filter paper. The trays were then covered with plastic wrap and a glass plate and placed under a 40-W fluorescent light with a photoperiod of 12 h light/12 h dark for 7 days. Inhibition parameters compared included fresh weight, dry weight, and percent germination of the seeds.

RESULTS AND DISCUSSION

After supplementation of the agricultural crops of interest was completed, several effects from the supplementation were examined, including differences in appearance, amount of selenium retained, and surrounding environmental changes.

Gross Morphological Changes. Examining differences in morphology focused on differences in the number of fruits produced, weight of fruits and vegetables, and any physical appearances of fruit and vegetables. The total numbers of fruits produced from the selenium-supplemented tomato and strawberry plants were counted and compared with the control plants. The results showed (Table 1) that for the tomato plants there was a significant difference between the control plants and high-organic treatment plants ($P = 0.02$, $n = 5$) with respect to the amount of fruit produced, with no fruit produced from the test plants; otherwise, no statistically significant differences between test and control plants were observed. For strawberry plants, there were no significant differences observed in fruit production (Table 1). The edible portions of the selenium-supplemented fruit and vegetable agricultural crops examined were individually weighed and compared with the controls (Table 2). For both the tomato and strawberry plants, no statistically significant differences were observed when the fresh weights of the fruits were examined. Statistically significant differences were observed for fresh weights of the edible portions of the radish plants, comparing the control with the low inorganic treatment ($P = 0.03$, $n = 5$), and of the lettuce plants, comparing the control with the high organic treatment ($P = 0.04$, $n = 5$).

Selenium Retention. After all the physical observations were made, the amount of selenium retained in the fruits or

Table 2. Data Summary for Fresh Weights (in Grams) of Tomato, Strawberry, Lettuce, and Radish^a

	control	high organic	high inorganic	low organic	low inorganic
tomato	35.0 ± 15.9	0.0 ± 0.0	28.5 ± 13.7	12.5 ± 8.1	34.5 ± 13.8
strawberry	7.1 ± 3.2	5.1 ± 1.2	8.7 ± 2.4	5.9 ± 3.2	9.5 ± 6.0
radish	5.3 ± 1.3	10.8 ± 0.9 ^b	9.0 ± 3.7	6.6 ± 0.9	9.8 ± 1.9 ^b
lettuce	0.65 ± 0.07	0.92 ± 0.10	0.93 ± 0.35	1.30 ± 0.87 ^b	1.03 ± 0.25

^a *n* = 4–10. Scale, grams. ^b Statistically significant difference at 95% confidence.

Table 3. Comparison of the Amount of Selenium Retained after Supplementation with Selenium^a

	control	high organic	low organic	high inorganic	low inorganic
tomato					
edible	0.13	0.00	0.30	0.27	0.42
nonedible	10.03	121.62	135.65	120.81	138.34
soil	0.25	2.30	0.96	1.82	1.09
strawberry					
edible	0.06	0.16	0.12	0.20	0.14
nonedible	9.27	110.11	122.51	106.57	120.83
soil	0.27	2.42	1.21	1.66	0.61
radish					
edible	0.03	0.42	0.14	0.28	0.16
nonedible	2.49	17.34	6.75	20.73	7.67
soil	0.35	2.70	1.27	2.00	0.73
lettuce					
edible	0.92	2.85	1.40	3.64	1.34
soil	0.08	0.09	0.18	0.09	0.05

^a *n* = 4. Scale, milligrams of Se per gram.

vegetables, plants, and soil was quantified using atomic absorption spectroscopy (**Table 3**). The amount of selenium in the edible portion of the fruit or vegetable crop was first determined for each crop studied. For all crops studied, the amount of selenium absorbed and retained in the edible portions was statistically significantly higher in the inorganic treatments when compared with organic treatments. This was a positive result, because supplementation with inorganic selenium would not only be more economical, but would also result in better retention of selenium than supplementation with organic selenium. Next, the amount of selenium retained by the inedible parts of the agricultural crop (i.e., leaves, stems, and roots) was collectively quantified, again using atomic absorption spectroscopy. For all of the crops studied, the amount of selenium absorbed and retained in the inedible portions was statistically significantly higher in the inorganic treatments than in the organic treatments. The amount of selenium retained in the soil for each agricultural crop was then quantified using atomic absorption spectroscopy. For the tomato and lettuce crops, there were no statistically significant differences observed for any of the treatments, another positive result favoring inorganic selenium supplementation. Although some selenium remains in the soil after treatment, using inorganic or organic selenium sources did not make a statistically significant difference, and inorganic selenium sources were less expensive than organic sources. For the strawberry and radish plants, the amounts of selenium remaining in the soil after the high inorganic treatments were statistically significantly lower than the amounts of selenium remaining in the soil after the high organic treatments. Again, this is another positive result favoring inorganic selenium supplementation because, although some selenium remains in the soil after treatment, using inorganic selenium would leave less selenium to accumulate in the soil.

Although selenium has not been demonstrated to be an essential element for plants, studied crops can assimilate it and respond in one of two ways: as a selenium source for humans or as a selenium scavenger for Se-contaminated soils, depending upon whether the edible or nonedible portions are to be used.

Our results show that selenium supplementation of these selected agricultural crops is feasible, with a wide range (between 0.1 and >97.5%) of selenium retained in the edible portion. The lettuce plants seemed to retain the most selenium (ca. 3–4%) in the edible portion of the plant compared to the other crops studied (**Table 3**).

Some plants (tomatoes and strawberries) managed to remove all of the added selenium in nonedible portions, which indicates that these plants could be used as selenium scavengers in Se-contaminated soils. The conditions for the scavenging procedure deserve consideration. Using an inorganic selenium source, such as selenium dioxide, proved to be better for increasing selenium content than using an organic selenium source, such as selenomethionine, for the following reasons. First, the edible portions of the plants examined retained statistically significant greater amounts of the inorganic selenium source used than the organic selenium source. Second, the inorganic selenium source was more cost-effective when compared with the organic selenium source. Finally, the amount of selenium left in the soil when the inorganic selenium source was used was less than that when the organic source was used. All of these factors have proven that using inorganic selenium as a source for selenium supplementation would be ideal, but the use of tomatoes as a Se scavenger could be even more promising.

Rayman has presented an interesting dichotomy: arguments for increasing selenium intake (26) versus the observation that diets have become steadily poorer in selenium (27). This seems especially true of European diets, owing to the reduction of imported wheat from North America (mainly Canada), and the same is likely true of American diets as well, owing to the shift toward fast foods. Thus, supplementation of food sources remains an attractive potential solution for American sources. In addition, selenium-deficient disorders have been recognized, apart from Keshan disease, and one may point to the conversion of harmless viruses to virulent ones in Se-deficient hosts (28). Se-deficient organisms can also develop more severe and prolonged lung inflammation when exposed to influenza virus, owing to harmful mutations in the virus. In addition, levels of selenium can be strong predictors of the outcome of HIV infection (29). There also seems to be cogent evidence supporting the benefits of supranutritional levels of selenium, i.e., that such levels have marked immunostimulant effects (30). Substantial evidence indicates that selenium may alter cancer at several sites and by multiple mechanisms (31).

Supplementing a Se-deficient diet can be managed in several ways. Rayman (26) suggests enhancements with Brazil nuts, but their fat content may limit the usefulness of these delicious nuts. Fish, crabs, and other shellfish are moderately good sources of selenium, but improvements in the concentration of selenium in blood and tissues are variable (27). On the other hand, the crops studied here have an attractive feature: they are nutritious, and tomatoes and lettuce, commonly used with many fast foods, could cater to contemporary tastes while providing a Se supplement benefit. One of the problems, however, is the effect of soil selenium on seeds, and this issue was addressed.

Surrounding Environmental Changes. Soil samples from the lettuce, tomato, strawberry, and radish plants were examined using XRF to determine weight percentages of significant elements (**Tables 4–7**). For the lettuce, tomato, and radish

Table 4. Weight Percentages of Elements Identified by XRF in Lettuce Soil Samples^a

	control	high organic	high inorganic	optimum inorganic	optimum organic
Na	0.24	0.00	0.00	0.00	0.00
Zn	0.00	0.00	0.00	0.00	0.32
Mg	0.40	0.00	0.00	0.00	0.00
Al	2.08	1.82	2.64	2.76	20.51
Se	0.00	0.00	0.00	0.00	0.00
Si	89.02	92.14	86.50	87.59	72.08
P	1.01	0.73	2.31	0.78	0.74
S	1.92	1.53	1.60	0.61	0.67
Ca	2.43	1.61	4.12	2.97	1.17
Fe	2.61	3.17	2.64	3.02	1.87
Cu	0.91	0.00	0.00	0.77	2.65
Ti	0.18	0.00	0.21	0.50	0.00
Cl	0.00	0.00	0.00	0.00	0.00
K	0.00	0.00	0.00	0.00	0.00

^a n = 4. Scale, weight percent.**Table 5.** Weight Percentages of Elements Identified by XRF in Tomato Soil Samples^a

	control	high inorganic	high organic	optimum inorganic	optimum organic
Na	1.58	1.50	0.00	1.92	1.00
Zn	0.00	0.00	0.00	0.00	0.00
Mg	1.93	1.80	0.74	2.62	1.50
Al	8.44	9.96	4.98	10.97	5.23
Se	0.00	0.00	0.00	0.00	0.00
Si	48.90	38.87	67.03	40.28	60.81
P	1.99	3.16	1.64	1.90	3.73
S	15.42	12.60	9.51	10.85	6.78
Ca	18.77	25.80	11.68	20.78	14.06
Fe	4.88	6.32	3.28	5.47	3.96
Cu	2.10	0.00	1.13	2.20	2.93
Ti	0.00	0.00	0.00	0.00	0.00
Cl	0.00	0.00	0.00	0.00	0.00
K	0.00	0.00	0.00	0.00	0.00

^a n = 4. Scale, weight percent.

samples, the amount of sulfur in the treatment soil samples was less than the initial amount of sulfur in the control. Also, with the high treatments, all of these plants were shown to absorb more selenium than the optimum treatment plants, and the amount of sulfur remaining in the soil for the optimum treatment plants was less than the amount of sulfur remaining in the soil for the high treatment plants. This suggests that the selenium is substituting for the sulfur in these plants, which has been previously demonstrated (2).

The strawberry soil samples for the control, all treatments, a sample obtained from IFAS, and an anonymous sample were examined for weight percentages of significant elements. It should be noted that the strawberry plants were obtained from a local nursery and that the soil used for this crop was not prepared in the laboratory. First, we compared elements observed in the IFAS and anonymous samples to elements in the control and treatment strawberry soil samples. It was observed that the weight percentages of silicon were higher in the IFAS and anonymous samples than in the control or treatment soil samples from our study. This suggests that the soil samples from IFAS and the anonymous samples contained more sand than the strawberry soil samples which were obtained from the local nursery. Other elements that had higher weight percentages in the control and treatment soil samples, when compared to the IFAS and anonymous soil samples, were phosphorus, calcium, and chlorine. The next comparison

Table 6. Weight Percentages of Elements Identified by XRF in Strawberry Soil Samples^a

	IFAS	anonymous	control	high organic	high inorganic	optimum organic	optimum inorganic
Na	0.00	0.00	0.00	2.01	1.57	0.00	0.00
Zn	0.00	0.00	0.00	0.00	0.00	2.68	0.00
Mg	0.72	0.50	1.82	2.18	1.95	1.66	0.00
Al	7.65	3.89	9.47	11.10	9.74	8.72	8.17
Se	0.00	0.00	0.00	2.21	0.00	0.00	0.00
Si	78.77	88.51	35.84	34.93	34.45	33.44	56.85
P	4.51	2.77	10.95	10.09	13.15	14.28	6.18
S	0.22	0.00	12.73	6.63	4.87	3.54	4.57
Ca	2.33	1.26	15.82	16.96	14.56	15.75	11.90
Fe	3.08	1.47	3.97	4.43	9.38	12.51	5.70
Cu	0.55	0.59	2.89	3.75	5.29	2.71	3.23
Ti	1.77	1.01	1.86	0.91	1.34	1.99	2.10
Cl	0.00	0.00	1.64	1.79	1.10	0.18	1.31
K	0.41	0.00	3.01	3.01	2.60	2.54	0.00

^a n = 4. Scale, weight percent.**Table 7.** Weight Percentages of Elements Identified by XRF in Radish Soil Samples^a

	control	high inorganic	high organic	optimum inorganic	optimum organic
Na	1.48	1.32	1.42	1.39	1.53
Zn	0.00	0.00	0.00	0.00	0.00
Mg	2.03	1.93	1.84	2.19	1.71
Al	7.34	8.29	6.93	8.30	6.72
Se	0.00	0.00	0.00	0.00	0.00
Si	48.66	42.99	50.39	47.67	53.66
P	1.24	2.26	2.88	2.83	2.99
S	15.05	14.18	11.42	9.92	8.22
Ca	17.83	20.43	18.24	19.83	17.46
Fe	4.55	5.92	5.06	5.19	4.98
Cu	1.82	2.68	1.82	2.68	2.73
Ti	0.00	0.00	0.00	0.00	0.00
Cl	0.00	0.00	0.00	0.00	0.00
K	0.00	0.00	0.00	0.00	0.00

^a n = 4. Scale, weight percent.

involved examining the amount of sulfur in the control soil sample and comparing it to the amount in the treatment soil samples. It was observed that the amount of sulfur in the treatment samples was lower than the amount of sulfur in the control plants, indicating that sulfur was being absorbed. Also, the amount of sulfur in the high treatment soil samples was higher than the amount of sulfur in the optimum treatment samples, which again suggests that selenium is substituting for sulfur in the plants that absorbed more selenium, and more sulfur is left in the soil of the plants that did not absorb as much selenium.

Inhibition on Seed Germination. For the lettuce seeds, **Figure 1** shows that the fresh weights decreased linearly with increasing selenium concentration. However, the dry weights remained constant over the range of selenium concentration. The percent germination showed a statistically significant linear decrease with an increase in selenium concentration. An EC₅₀ was determined by interpolation to be 36 ppm, at which 50% of the seeds showed inhibition of germination.

For the tomato seeds, **Figure 2** shows that the fresh weights and dry weights remained constant over the range of selenium concentration. The percent germination showed a statistically significant linear relationship: an increase in selenium concentration caused a decrease in percent germination. An EC₅₀ was determined by interpolation to be 29 ppm, at which 50% of the seeds showed inhibition of germination. Although the percent

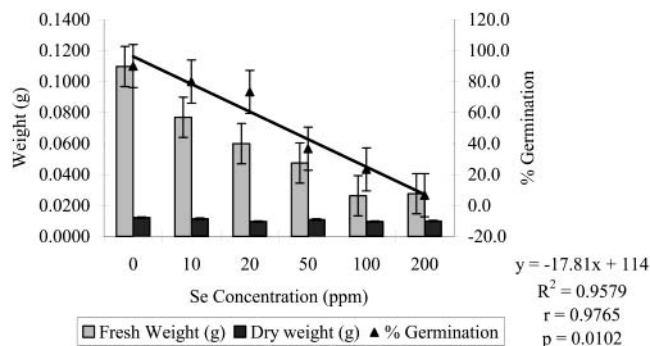


Figure 1. Effect of selenium on lettuce seed germination.

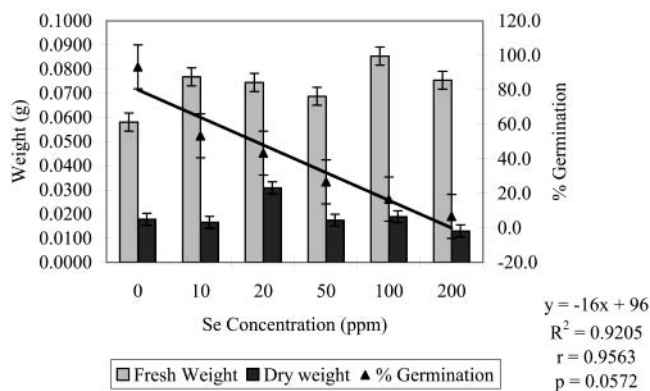


Figure 2. Effect of selenium on tomato seed germination.

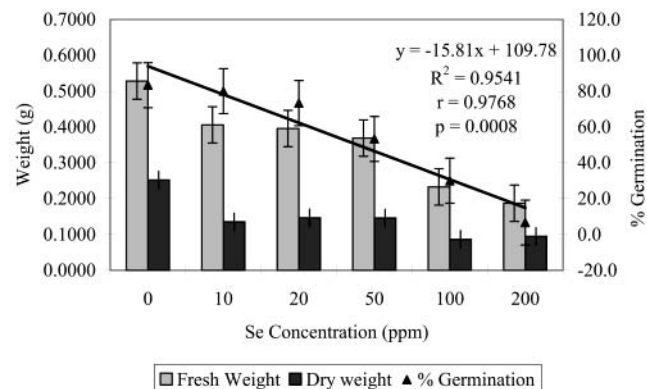


Figure 3. Effect of selenium on radish seed germination.

germination decreased at a lower concentration than for the lettuce seeds (29 ppm versus 38 ppm for 50% inhibition of germination), of those seeds which did germinate, the growth was not severely inhibited until a selenium concentration of 100 ppm.

For the radish seeds, **Figure 3** shows that the fresh weights showed a linear decrease with increasing selenium concentration; however, the dry weights remained constant. The percent germination showed a statistically significant linear decrease with an increase in selenium concentration. The EC_{50} was interpolated to give a concentration of 38 ppm, at which 50% of the seeds showed inhibition of germination. The percent germination was similar to the results from the lettuce seed study (38 ppm for 50% inhibition of germination).

The results from these seed bioassays indicate that several important factors need to be considered for agricultural selenium supplementation. First, although selenium is not an essential nutrient for plants, at high enough concentrations, the growth and germination of seeds can be inhibited. This has been previously demonstrated with selenate and selenite for cabbage,

lettuce, radish, sorgrass, turnip, and wheat (32). Finally, the most important conclusion which can be drawn from the seed bioassays is that, in all the agricultural crops studied, a concentration higher than 29 ppm, for the most sensitive seeds studied, is required to inhibit germination of the seeds. This is very important, because the highest concentration of selenium left in the soil from our agricultural crop studies was 1.9 ppm (high organic selenium treatment of tomatoes). The amount of selenium left in the soil after the first round of supplementation would not be a high enough concentration to inhibit the germination of the seeds of any of the agricultural crops that were studied. Therefore, the concentration of selenium left in the soil would not be toxic when new seeds were planted, and several crops could be grown and supplemented in the same soil.

LITERATURE CITED

- (1) Frankenberger, W. T., Jr.; Engberg, R. A. *Environmental Chemistry of Selenium*; Marcel Dekker: New York, 1998.
- (2) Anderson, J. W.; Scarf A. R. Selenium and plant metabolism. In *Metals and Micronutrients: Uptake and Utilization by Plants*; Robb, D. A., Pierpoint, W. S., Eds.; Academic Press: New York, 1983; pp 241–275.
- (3) Combs, G. F., Jr.; Combs, S. B. *The role of selenium in nutrition*; Academic Press: Orlando, FL, 1986.
- (4) National Research Council. *Selenium in Nutrition*, revised edition; National Academy Press: Washington, DC, 1983.
- (5) Wallach, J. D.; Garmaise, B. Cystic Fibrosis: A perinatal manifestation of selenium deficiency. In *Trace Substances In Environmental Health*; Hemphill, D. D., Ed.; University of Missouri Press: Columbia, MO, 1979; pp 469–476.
- (6) Burk, R. F.; Hill, K. E. Regulation of Selenoproteins. In *Annual Review of Nutrition*, 13th ed.; Olson, R., Bier, D., McCormick, D., Eds.; Annuals Reviews, Inc.: Palo Alto, CA, 1993; pp 65–81.
- (7) Clark, L. C.; Combs, G. F., Jr.; Turnbull, B. W.; Slate, E. H.; Chalker, D. K.; Chow, J.; Davis, L. S.; Glover, R. A.; Graham, G. F.; Gross, E. G.; Krongrad, A.; Leshner, J. L.; Park, H. K.; Sanders, B. B., Jr.; Smith, C. L.; Taylor, J. R. Effects of selenium supplementation for cancer prevention in patients with carcinoma of the skin a randomized controlled trial. *JAMA, J. Am. Med. Assoc.* **1996**, *276*, 1957–1963.
- (8) Ip, C. Lessons from basic research in selenium and cancer prevention. *J. Nutr.* **1998**, *128*, 1845–1854.
- (9) El-Bayoumi, K. The role of selenium in cancer prevention. In *Cancer: Principles and Practice of Oncology*, 4th ed.; DeVita, V. T., Jr., Hellman, S., Rosemberg, S. A., Eds.; Lippincott: Philadelphia, 1991; pp 1–15.
- (10) National Research Council. *Recommended dietary allowances*, 10th ed.; National Academy Press: Washington, DC, 1989.
- (11) Levander, O. A. A global view of human selenium nutrition. *Annu. Rev. Nutr.* **1987**, *7*, 227–250.
- (12) Carvalho, K. M.; Benson, R. F.; Booth, F. A.; Collier, M. J.; Martin, D. F. Analysis of commercial selenium-supplement tablets. *Florida Scient.* **2000**, *63*, 8–12.
- (13) Ip, C.; Lisk, D. J. Characterization of Tissue Selenium Profiles and Anticarcinogenic Responses in Rats Fed Natural Sources of Selenium-rich Products. *Carcinogenesis* **1994**, *15*, 573–576.
- (14) Ip, C.; Lisk, D. J. Efficacy of Cancer Prevention by High-selenium Garlic is Primarily Dependant on the Action of Selenium. *Carcinogenesis* **1995**, *16*, 2649–2652.
- (15) Finley, J.; Matthys, L.; Shuler, T.; Korynta, E. Selenium Content in Foods Purchased in North Dakota. *Nutr. Res.* **1996**, *723–728*.
- (16) Bisberg, B.; Gissel-Nielsen, G. The Uptake of Applied Selenium by Agricultural Plants. I. The Influence of Soil Type and Plant Species. *Plant Soil* **1969**, *31*, 287–298.

- (17) Gissel-Nielsen, G.; Bisberg, B. The Uptake of Applied Selenium by Agricultural Plants. 2. The Utilization of Various Selenium Compounds. *Plant Soil* **1970**, *32*, 382–396.
- (18) Gissel-Nielsen, G. Control of Selenium in Plants. *Risoe Rep.* **1977**, No. 370, 13 app.
- (19) Ylärinta, T. Increasing the Selenium Content of Cereals and Grass Crops in Finland. Dissertation, University of Helsinki, Finland, 1985.
- (20) Gissel-Nielsen, G. Effects of selenium supplementation of field crops. In *Environmental Chemistry of Selenium*; Frankenberger, W. T., Jr., Engberg, R. A., Eds.; Marcel Dekker: New York, 1998; pp 99–112.
- (21) Aro, A.; Ekholm, P.; Alfthan, G.; Varo, P. Effects of Selenium Supplementation of Fertilizers on Human Nutrition and Selenium Status. In *Environmental Chemistry of Selenium*; Frankenberger, W. T., Jr., Engberg, R. A., Eds.; Marcel Dekker: New York, 1998; pp 81–97.
- (22) Wyatt, C. J.; Meléndez, J. M.; Acuna, N.; Rascon, A. Selenium (Se) in Foods in Northern Mexico, Their Contribution to the Daily Se Intake and the Relationship of Se Plasma Levels and Glutathione Peroxidase Activity. *Nutr. Res.* **1996**, *16*, 949–960.
- (23) Cary, E. E.; Allaway, W. H. Selenium Content of Field Crops Grown on Selenite-treated Soils. *Agron. J.* **1973**, *65*, 922–925.
- (24) Eurola, M.; Ekholm, P.; Ylinen, M.; Koivistoinen, P.; Varo, P. Effects of Selenium Fertilization on the Selenium Content of Selected Finnish Fruits and Vegetables. *Acta Agric. Scand.* **1989**, *39*, 345–350.
- (25) Inhat, M. Selenium. In *Hazardous Metals in the Environment Techniques and Instrumentation in Analytical Chemistry*; Stoeppler, M., Ed.; Elsevier: Amsterdam, 1992; pp 457–515.
- (26) Rayman, M. P. The argument for increasing selenium intake. *Proc. Nutr. Soc.* **2002**, *61* (2), 203–215.
- (27) Rayman, M. Se brought to earth. *Chem. Br.* **2002**, *39* (10), 28–31.
- (28) Beck, M. A.; Nelson, H. K.; Shi, Q.; Van Dael, P.; Schiffrin, E. J.; Blum, S.; Barclay, D.; Levander, O. A. Selenium deficiency increases the pathology of an influenza virus infection. *FASEB J.* **2001**, *15*, 1481–1483.
- (29) Shor-Posner, G.; Miguez, M.-J.; Pineda, L.; Rodriguez, A.; Ruiz, P.; Castillo, G.; Burbano, X.; Lecusay, R.; Baum, M. Impact of selenium status on the pathogenesis of mycobacterial disease in HIV-1-infected drug users during the era of highly active antiretroviral therapy. *JAIDS, J. Acquired Immune Defic. Syndr.* **2002**, *29*, 69–173.
- (30) Kiremidjian-Schumacher, L.; Roy, M.; Wishe, H. I.; Cohen, M. W.; Stotzky, G. Supplementation with selenium augments the functions of natural killer and lymphokine-activated killer cells. *Biol. Trace Elem. Res.* **1996**, *52*, 227–239.
- (31) Kim, Y. S.; Milner, J. Molecular targets for selenium in cancer prevention. *Nutr. Cancer* **2001**, *40*, 50–54.
- (32) Carlson, C. L.; Kaplan, D. I.; Adriano, D. C. Effects of Selenium on Germination and Radicle Elongation of Selected Agronomic Species. *Environ. Exp. Bot.* **1989**, *29*, 493–498.

Received for review July 28, 2002. Revised manuscript received November 13, 2002. Accepted November 18, 2002.

JF0258555