Bull. Environ. Contam. Toxicol. (1990) 44:681–685

Bulletin of Environmental Contamination and Toxicology

Selenium in Tissues of Rats Fed Rutabagas Grown on Soil Covering a Coal Fly Ash Landfill

Gilbert S. Stoewsand,[1] Judy L. Anderson,[1] Leonard H. Weinstein,[2] Joseph F. Osmeloski,[2] Walter H. Gutenmann,[3] and Donald J. Lisk[3]

[1]Department of Food Science and Technology, New York State Agricultural Experiment Station, Geneva, New York 14456; [2]Boyce Thompson Institute for Plant Research, Cornell University, Ithaca, New York 14853; and [3]Toxic Chemicals Laboratory, New York State College of Agriculture and Life Sciences, Cornell University, Ithaca, New York 14853-7401, USA

Approximately 80 million tons of fly ash are produced by coal-burning power plants in the United States each year. Most is disposed in landfills which are capped with a layer of soil from about 0.5 to 1 meter thick. There has been concern that crops planted or voluntarily growing over such areas may absorb toxic elements which could be ingested by foraging animals or humans. Indeed, selenium has been shown to be taken up by vegetables,and field crops grown on soils to which fly ash was deliberately added in field and greenhouse studies (Furr et al. 1976, 1977, 1979; Gutenmann et al. 1979). Deep-rooted crops growing over capped landfills might expectedly absorb elements directly from the fly ash below. Shallow-rooted crops might also by contacting fly ash moved upward into the soil cap by earthworm activity or by absorption of such elements as dissolved ions moved upwards by capillarity. In the work reported here, rutabagas grown on soil covering a coal fly ash landfill were fed to rats and the movement of selenium from soil to plant to animal was determined.

MATERIALS AND METHODS

Plant growth was conducted at a closed fly ash landfill site about 32 km north of Ithaca near the eastern shore of Cayuga Lake in Lansing, New York. The fly ash derives from Milliken Station, a coal-burning electric power generating plant located there. Construction of the landfill began in 1976 and was closed in 1978 after capping the fly ash with 45-60 cm. of soil. After closing, a grass-legume crop mixture was seeded over it to prevent erosion. In the fall of 1987, a 3 m wide by 100 m long area was mowed and treated with "Roundup" herbicide to kill existing vegetation. The area was then plowed to a depth of 15-20 cm. In the spring of 1988, three plots, 2 m wide by 6.7 m long, were staked out at random in the plowed strip and were cultivated and "American Purple Top" rutabagas (Brassica napus) planted. Three plots of the same dimensions were prepared similarly and planted on soil adjacent to the fly ash landfill to serve as controls. The soil types in the cap and control areas included Ilion silty clay loam (fine-loamy, mixed, mesic mollic ochraqualfs) and Kendaia silt

Send reprint requests to Donald J. Lisk at the above address.

loam (fine-loamy, mixed, nonacid, mesic aeric haplaquepts). The pH of the soil in the cap and control areas was, respectively, 7.3 and 7.6. Normal cultural and pest control treatments were used during crop growth. In the fall, the plants were harvested, tops removed and discarded and the roots washed, brushed and peeled to assure the absence of soil or fly ash particles. The root material was mechanically chopped, freeze-dried, milled to a fine powdery consistency and mixed.

Weanling, 22-day-old, male Sprague-Dawley rats (Charles River, Kingston, NY) were housed in pairs in hanging polycarbonate solid bottom cages with pressed chipped-hardwood pads used as bedding. Initial body weights were 57.9 ± 1.8 g. Diets and water were provided ad libitum. The room was maintained at 23°C on a 12-h light/dark cycle. There were three dietary groups of rats with 10 rats assigned to each group, i.e., purified basal diet, and two rutabaga diets grown either on soil or on the fly ash landfill site. Rutabagas were peeled, freeze-dried, and ground to a powdery consistancy and incorporated at 25% into a complete diet as shown in Table 1. The rats were fed for 13 weeks.

Table 1. Experimental diets

Constituent	Basal %	Rutabaga %
Rutabaga[1]	-----	25.00
Casein	20.00	17.00
Cornstarch	15.00	15.00
Sucrose	25.00	15.00
Glucose	25.00	15.50
Non-nutritive fiber	5.00	3.00
Mineral Mix[2,3]	3.50	3.50
Corn Oil[4]	5.00	5.00
Vitamin Mix[2]	1.00	1.00
DL-Methionine	0.30	0.30
Choline bitartrate	0.20	0.20
Santoquin	0.0125	0.0125

[1]Freeze-dried. Grown on soil or fly ash.

[2]Report of the American Institute of Nutrition ad hoc committee on standards of nutritional studies (AIN 1977; 1980).

[3]Mineral mix further modified as per Mars et al. (1988).

[4]Amended with vitamin K_1 at a rate of 2 mg/kg diet.

At the end of the feeding period the rats were sacrificed by CO_2 inhalation and kidney, liver, muscle and spleen were removed and freeze-dried prior to the determination of selenium. Selenium was determined in the animal tissues and diets by wet ashing the samples with nitric and perchloric acids followed by analysis using the fluorescence procedure of Olson (1969).

RESULTS AND DISCUSSION

The mean weight gains and food intake of each dietary group of rats are presented in Table 2. There was a trend for the rats fed rutabagas grown on fly ash to exhibit a larger weight gain with increased food intake, as compared to the rats fed the soil-grown rutabagas. This may reflect the increased amount of protein in the fly ash-grown rutabagas as seen in the proximate analysis (Table 3).

Table 2. Weight gain and food intake of rats fed 25% rutabagas grown on soil or fly ash (FA)

Dietary treatment	Weight gain (g)	Food intake (g)
Basal	467.8 ± 13.8[a1]	1998.1 ± 48.5[a]
S Rutabagas	367.8 ± 11.1[b]	1552.4 ± 12.1[b]
FA Rutabagas	397.8 ± 11.8[b]	1669.8 ± 21.1[c]

[1]Mean ± SEM (n = 10); dissimilar letter superscripts indicate significant ($p \leq 0.05$) differences between respective treatment means.

Table 3. Proximate analysis of freeze-dried rutabagas

Sample	% Protein	% Fat	% Fiber	% Moisture	% Ash
Control	10.7	3.06	8.3	3.99	4.90
Fly ash	12.8	2.2	7.9	2.55	4.51

The selenium content of the control and fly ash-grown rutabagas was, respectively, 0.17 and 4.4 ppm, dry weight. This indicates that the roots of fly ash-grown plants absorbed selenium by several possible mechanisms: (1) the plant roots contacted the bed of fly ash below or fly ash particles moved into the soil cap above by earthworm activity, (2) roots absorbed selenium moved upwards into the soil layer by capillarity and (3) available selenium would also have been moved into the soil cap from the bed of fly ash especially by the deep-rooted legumes which were initially sown nine years earlier and allowed to grow and continuously decay on the soil surface.

The mean concentrations of selenium found in rat tissues are given in Table 4. The selenium content of each of the tissues from the rats fed the fly ash-grown rutabagas was significantly higher than that of the respective tissues representing the control or basal dietary treatments. The animals exhibited no external signs of toxicity during the feeding study. However, as seen in Table 5 relative liver weights of the rutabaga-fed rats significantly increased in size. This may indicate an adaptive response accompanied by microsomal enzyme induction to the ingestion of

Table 4. Selenium deposition in rat tissues

Dietary treatment	Selenium ppm dry weight in[1] Kidney	Liver	Muscle	Spleen
Basal	3.8 ± 0.1^{x}	1.9 ± 0.1^{x}	0.32 ± 0.01^{x}	1.7 ± 0.0^{x}
Control rutabaga	4.1 ± 0.1^{x}	2.2 ± 0.1^{y}	0.35 ± 0.01^{x}	1.7 ± 0.0^{x}
Fly ash rutabaga	6.4 ± 0.2^{y}	3.3 ± 0.0^{z}	0.89 ± 0.03^{y}	2.8 ± 0.1^{y}

[1]Mean ± standard error (n = 10); dissimilar letter superscripts indicate significant differences between respective treatment means ($p < 0.05$).

Table 5. Relative organ weights of rats fed 25% rutabagas grown on soil or fly ash (FA)

Dietary treatment	Liver % body wt	Kidney % body wt	Spleen % body wt
Basal	4.09 ± 0.10^{b}[1]	0.72 ± 0.01^{b}	0.16 ± 0.01^{a}
S Rutabagas	4.40 ± 0.09^{b}	0.82 ± 0.03^{a}	0.17 ± 0.01^{a}
FA Rutabagas	4.42 ± 0.09^{a}	0.81 ± 0.03^{a}	0.17 ± 0.01^{a}

[1]Mean ± standard error (n = 10); dissimilar letter superscripts indicate significant differences between respective treatment means ($p < 0.05$).

natural compounds present in both soil and fly ash-grown rutabagas (Crampton et al. 1977).

The possible environmental implications of the results reported here are speculative. A concentration of 4.4 ppm of selenium as found in the fly ash-grown rutabagas in this study is considered a near toxic dietary level for monogastric animals. However, ruminants can apparently tolerate far higher concentrations (Furr et al. 1978). It is more probable that grasses and legumes would be planted or voluntarily grow over soil-capped fly ash landfills than vegetables. It has been shown that sweet clover voluntarily growing on an uncapped fly ash landfill absorbed an extremely high concentration of selenium. When the latter crop was fed to goats and sheep high concentrations of selenium were deposited in animal tissues and excreted in milk (Furr et al. 1978). Legumes have also been shown to absorb elevated concentrations of selenium when grown on soil-covered fly ash landfills (Weinstein et al. 1989). The extent of selenium uptake by plants will depend, however, on a number of factors. These include the plant species, the concentration of selenium in the ash, the pH of the soil and fly ash, the thickness of the soil cap, rainfall and many others. The extent to which selenium from fly ash enters the human food chain will depend on any increases in numbers of fly ash landfills and the types of agriculture that may prevail there. At present, the concern may be for wildlife inhabiting such areas.

Acknowledgments. The authors thank Audrey V. Gardner for the proximate analyses of the rutabagas.

REFERENCES

AIN (1977) Report of the American Institute of Nutrition ad hoc Committee on Standards for Nutritional Studies. J Nutr 107:1340-1348

AIN (1980) Second Report of the ad hoc Committee on Standards for Nutritional Studies. J Nutr 110:1726

Crampton RF, Gray TJB, Grasso P, Parke DV (1977) Long-term studies of chemically induced liver enlargement in the rat. I. Sustained induction of microsomal enzymes with absence of liver damage on feeding phenobarbitone or butylated hydroxytoluene. Toxicology 7:289-306

Furr AK, Kelly WC, Bache CA, Gutenmann WH, Lisk DJ (1976) Multielement uptake by vegetables and millet grown in pots on fly ash amended soil. J Agric Food Chem 24:885-888

Furr AK, Parkinson TF, Hinrichs RA, Van Campen DR, Bache CA, Gutenmann WH, St. John LE, Pakkala IS, Lisk DJ (1977) National survey of elements and radioactivity in fly ashes: Absorption of elements by cabbage grown in fly ash-soil mixtures. Environ Sci Technol 11:1194-1201

Furr AK, Parkinson TF, Heffron CL, Reid JT, Haschek WM, Gutenmann WH, Bache CA, St. John LE, Lisk DJ (1978) Elemental content of tissues and excreta of lambs, goats, and kids fed white sweet clover growing on fly ash. J Agric Food Chem 26:847-851

Furr AK, Parkinson TF, Elfving DC, Gutenmann WH, Pakkala IS, Lisk DJ (1979) Elemental content of apple, millet, and vegetables grown in pots of neutral soil amended with fly ash. J Agric Food Chem 27:135-138

Gutenmann WH, Pakkala IS, Churey DJ, Kelly WC, Lisk DJ (1979) Arsenic, boron, molybdenum, and selenium in successive cuttings of forage crops field grown on fly ash amended soil. J Agric Food Chem 27:1393-1395

Mars YWHM, Lemmens AG, Beynen AC (1988) Dietary phosphorus and nephrocalcinosis in female rats. Nutr Rep Int 38:249-258

Olson OE (1969) Fluorometric analysis of selenium in plants. J Assoc Offic Anal Chem 52:627-634

Weinstein LH, Osmeloski JF, Rutzke M, Beers AO, McCahan JB, Bache CA, Lisk DJ (1989) Elemental analysis of grasses and legumes growing on soil covering coal fly ash landfill sites. J Food Safety 9:291-300

Received August 1, 1989; accepted November 20, 1989.