U.S. Food and Drug Administration, Code of Federal Regulations (21), Office of the Federal Register, Washington, D.C., 1974.
Van Campen, D. R., J. Nutr. 103, 139 (1973).
Van Campen, D., Gross, E., J. Nutr. 99, 68 (1969).

Van Campen, D., Mitchell, E. A., J. Nutr. 86, 120 (1965). Wheby, M. S., Crosby, W. H., Blood 22, 416 (1963). Wheby, M. S., Jones, L. G., Crosby, W. H., J. Clin. Invest. 43, 1433 (1964).

Received for review February 18, 1977. Accepted August 2, 1977. Supported in part by the Department of Agriculture Grant 12-14-100-9918(61).

# Selenium and Arsenic Levels in Soybeans from Different Production Regions of the United States

R. D. Wauchope

Whole soybeans [*Glycine max* (L.) Merr.] from Arkansas, Florida, Indiana, Iowa, Maryland, Mississippi, and North Carolina have been analyzed for total arsenic and selenium. Arsenic levels were 0.1 ppm (fresh weight) or less for all samples and were independent of selenium levels. Selenium levels in soybeans reflected levels in the soil in the region where the soybeans were grown, varying from 0.07 ppm or less in low-selenium areas (Maryland, Indiana, Florida, and North Carolina) to 0.16 ppm (Arkansas), 0.28 ppm (Iowa), and 0.90 ppm (Mississippi). The selenium was associated with the oil-free fraction of the soybean. Selenium levels in the soybeans from Stoneville, in the Mississippi "Delta" floodplain, were high in comparison with levels in soybeans from other areas considered to have adequate soil selenium levels, suggesting that soils in this area may unusually rich in available selenium. These results show that soybeans may be a significant source of selenium and possibly other trace nutrients in human and animal diets.

Ferretti and Levander (1976) recently reported selenium (Se) levels in soybean [Glycine max (L.) Merr.] food products and found quite variable levels in the range 0.02-0.7 ppm, with good correlation between Se levels and soy protein content. Since these products are being increasingly used as meat substitutes, there is concern that soy products with the lower ranges of Se will not adequately provide as much Se as the replaced meat. Ferretti and Levander listed three possible sources of variation in the soy product selenium levels: (1) differences in selenium content of the soybeans at harvest; (2) losses of Se in processing; and (3) addition of other ingredients to the soy products that may or may not contain substantial amounts of selenium. This report is concerned with source (1).

The Se content of forage and grain crops varies widely and is a function of the content of Se in the soil where they are grown, as well as other soil factors such as pH and texture. Wheat (Triticum aestivum L.) is the most studied example (Kubota et al., 1967). Few data are available on Se levels in fresh soybeans, however. Ferretti and Levander reported 0.07 ppm fresh weight; Olson (1969) reported a single value of 1.5 ppm. Klayman and Gunther (1973) list values of 0.07 ppm for soybeans from the eastern United States, 0.54 for Nebraska sovbeans (an area of highly seleniferous soils), and 0.1 ppm for midwestern soybeans, indicating considerable variation of levels with production area. At the time of appearance of the article by Ferretti and Levander (1976) the authors had been applying a new flameless atomic absorption technique to the measurement of arsenical herbicide residues in soybean grain grown at Stoneville (Wauchope and McWhorter, 1977). Since the technique is equally useful for selenium,

we tried it on the same soybean digests used for arsenic analysis and were surprised at the high levels found (in excess of 1 ppm for some samples). To verify these results we have analyzed a variety of Stoneville-grown soybeans, along with soybean samples from six other states for comparison. Although most of the important production areas of the U.S. (Dovring, 1974) are represented by these samples, the number of samples is too small for the results to be taken as definitive for each area. We also include results of arsenic (As) analyses, since they were simple to obtain from the same digested samples and are of interest both from a nutritional and toxicological standpoint.

#### MATERIALS AND METHODS

Soybeans were obtained through the ARS Soybean Production Research Unit at Stoneville, Miss. Fresh soybeans were ground in a Wiley mill and stored in a freezer until used. Two or more 4-g samples of the beans were digested with 80 mL of nitric-perchloric acids (3:1) using Bethge condensers to contain  $HClO_4 \cdot 2H_2O$  fumes (Griffen et al., 1974, 1975). We found that slow digestion was necessary and used round-bottom flasks and heating mantles, instead of hot plates and the Erlenmeyer flasks provided with the Bethge apparatus by the manufacturer. Rapid digestion with high-wattage hot plates caused an average loss of 30% of the Se content.

At the end of digestion the approximately 20 mL of 70% perchloric acid remaining in the apparatus was diluted to 50 mL with distilled water. Samples (1 mL) of the diluted digest were analyzed by sodium borohydride reduction of the Se or As to hydrogen selenide or arsine, followed by flameless atomic absorption assay (Wauchope, 1976). Optimization of carrier gas flow rates and the introduction of a very small flow (a few milliliter/min) of air into the carrier gas gave a working range of 5–200 ppb Se or As in the digest with this method, with a detection limit (3 × blank variance) of 2 ppb, equivalent to 0.03 ppm in the soybeans.

Southern Weed Science Laboratory, U.S. Department of Agriculture, Agriculture Research Service, in cooperation with the Mississippi Agricultural and Forestry Experiment Station, Stoneville, Mississippi 38776.

Table I. Selenium Content of Whole Soybeans from Ames, Iowa Area

0.24 ± 0.03
$0.17 \pm 0.04$
$0.48 \pm 0.04$
$0.30 \pm 0.08$
$0.39 \pm 0.07$
$0.08 \pm 0.02$

<sup>*a*</sup> Mean values for two or more samples  $\pm$  SD.

Recoveries of 0.25 ppm Se added to soybean meal were 80-95%. Arsenic recoveries were previously reported to be 80-88% (Wauchope, 1976). Values reported were not corrected for recovery and are listed in the tables on a fresh weight basis.

In order to determine if the Se was associated with the oil or protein fraction of the beans, two samples of "Davis" soybean meal from a loam soil at Stoneville were extracted for 1 h in a Soxhlet extractor with 100 mL of hexane. The meal and oil were then freed of hexane and analyzed as above.

## **RESULTS AND DISCUSSION**

Kubota et al. (1967) used extensive crop Se analytical data (forages, wheat, and feed grain) along with soil and geological information, to establish boundaries of four types of geographic areas classified by crop-Se ranges (see Figure 1). The classification was as follows: I, areas where more than 80% of the crops contained less than 0.05 ppm Se (dry weight); II, areas where more than 80% of the crops contained less than 0.10 ppm Se; III, areas with both low and adequate Se levels; IV, areas where more than 80% of the crops contained more than 0.10 ppm Se.

Kubota et al. stated "...A very limited number of comparisons indicated that corn and soybeans tended to be similar in Se content to the alfalfa in the same area...", but did not indicate whether these data were for whole plants, foliage, grain, etc. Our grain data confirm this suggestion without exception. The discussion below is based on the boundaries defined by Kubota et al., and the descriptions of soil origins are taken from their paper.

Arkansas. One sample of "Mack" soybeans from Stuttgart was analyzed. Arsenic content was below the detection limit and the average selenium content was 0.16  $\pm$  0.04 ppm for duplicate digests. These soybeans were grown on a Crowley silt loam, a wind-deposited soil from parent material in the Midwest that is relatively high in Se, and this may explain the similarity of the Arkansas



Figure 1. Shaded areas are soybean production areas of the United States (redrawn from Dovring, 1974). Triangles show locations where soybean samples were grown. Regional distributions of selenium, based on soil, alfalfa, and other forage analyses, are given by heavy lines (redrawn from Kubota et al., 1967): I, selenium deficient; II, low selenium; III, variable selenium; IV, adequate to high selenium.

soybean Se levels to those from Iowa (see below).

Florida and North Carolina. Samples of "Bragg", "Cobb", "Hood", "Hutton", "Mineira", and "Ransom" varieties from Gainesville, Fla., and "Ransom" from Clinton, N.C. were analyzed. These areas were classified by Kubota as Se deficient, and neither As nor Se was detected in any of these samples.

Indiana. "Woodworth" soybeans from Buffton, Evansville, Greenfield, Lafayette, and Sullivan were analyzed. Indiana is composed of old low-Se sedimentary rocks, and no Se was detected in any of the soybean samples. Low levels of arsenic  $(0.05-0.07 \text{ ppm}, \text{SD} \pm 0.04)$  were found in the Buffton, Lafayette, and Sullivan samples, and none was detected in the Evansville and Greenfield samples.

Iowa. Central Iowa is on the eastern side of a Se-adequate area (Figure 1). Soybeans from Ames gave Se levels equal to or higher than those of Stuttgart, Ark. (Table I). There were significant differences in Se levels between

Table II.	Arsenic and Seleniu	m Content of Whole	Soybeans and	Soybean Frac	ctions from Stone	eville, Mississippi
-----------	---------------------	--------------------	--------------	--------------	-------------------	---------------------

	Arsenic cor	ntent, ppm <sup>a</sup>	Selenium co	ntent, ppm <sup>a</sup>	<u>.</u>
Variety	Loam	Clay	Loam	Clay	
Bragg	n.d. <sup>b</sup>	n.d.	$1.29 \pm 0.06$	$0.47 \pm 0.07$	
Cajeme	$0.06 \pm 0.03$	$0.04 \pm 0.02$	$0.53 \pm 0.10$	$1.06 \pm 0.06$	
Davis	$0.05 \pm 0.04$	n.d.	$0.96 \pm 0.13$	$0.58 \pm 0.04$	
Forrest	$0.07 \pm 0.06$	n.d.	$0.96 \pm 0.09$	$1.09 \pm 0.13$	
Hill	$0.13 \pm 0.14$	n.d.	$1.20 \pm 0.11$	$0.39 \pm 0.09$	
Hood 75	$0.03 \pm 0.03$	n.d.	$0.98 \pm 0.05$	$0.24 \pm 0.03$	
Lee 68	$0.10 \pm 0.04$	n.d.	$1.08 \pm 0.13$	$0.31 \pm 0.01$	
Lee 74	0.11 <sup>c</sup>	$0.04^{c}$	$0.84 \pm 0.06$	$0.56 \pm 0.04$	
Mack	$0.06 \pm 0.04$	n.d.	$1.00 \pm 0.23$	$0.67 \pm 0.05$	
Pickett 71	$0.06 \pm 0.01$	$0.05 \pm 0.04$	$1.18 \pm 0.05$	$107 \pm 0.03$	
Ransom	0.13 <sup>c</sup>	$0.18^{c}$	d	$1.57 \pm 0.23$	
Tracy	n.d.	n.d.	$\overline{1.71} + 0.21$	$0.98 \pm 0.29$	
Average <sup>e</sup>	$0.07 \pm 0.04$	$0.03 \pm 0.05$	$1.1 \pm 0.3$	$0.7 \pm 0.4$	
Davis, meal			$0.80 \pm 0.01$		
Davis, oil			n.d.		

<sup>*a*</sup> Mean values for two or more digestions  $\pm$  SD. <sup>*b*</sup> n.d. = <0.03 ppm detection limit. <sup>*c*</sup> One sample. <sup>*d*</sup> No sample. <sup>*e*</sup> Average assuming 0.01 ppm for n.d. samples. samples of the different varieties. These varietal differences appear worthy of further investigation since the Iowa soil might be expected to be more homogeneous than the samples suggest if one assumed similar uptake by all varieties.

Mississippi. The Stoneville soybeans were obtained from Bosket fine sandy loam and Sharkey clay sites situated approximately one-quarter mile apart. The loam-grown soybeans were the only samples besides the Indiana samples which consistently had arsenic levels above the detection limit. This soil may be higher than usual in available As due to previous arsenical pesticide use in cotton production. The Se levels in soybeans from both soils were quite variable (Table II). The loam soil soybeans had higher Se levels, with some exceptions, suggesting that this alluvium is either higher in Se or the Se present is more available.

The consistently relatively high Se levels from the Mississippi Delta soybeans is somewhat surprising, particularly in comparison with the Arkansas and Iowa levels. All three areas were grouped by Kubota in the "selenium-adequate" central region of the United States. The most likely source of Se in the Stoneville soils may be deposition of sediments from the Arkansas River, which drains high-Se soils on the eastern side of the lower Rocky Mountains. Studies on soil Se levels in the Stoneville area will be a subject for future study.

The separately analyzed oil and oil-free meal from Stoneville "Davis" soybeans indicated that the Se was entirely associated with the oil-free meal (Table II).

## SUMMARY

Although the number of samples from some areas was small, it is obvious that order-of-magnitude variations in Se content in fresh soybeans occurs between different major production areas in the United States. Where Se is present in the soil, soybeans appear to be particularly efficient at concentrating it in the grain. This effect has been observed for other trace elements, specifically molybdenum (Harris, et al., 1965; Hawes et al., 1976), copper, zinc, and boron (Martens et al., 1974), and arsenic (Wauchope and McWhorter, 1977). It appears from these data that Mississippi and Iowa soybeans could be a significant resource of Se in human and animal diets.

### ACKNOWLEDGMENT

The author thanks T. C. Kilen, ARS Sovbean Production Research, Stoneville, Miss., for obtaining the soybean samples and for helpful discussions and R. J. Ferretti, formerly of the ARS Vitamin and Nutrition Laboratory, Beltsville, Md., for help in preparation of the manuscript.

#### LITERATURE CITED

- Dovring, F., Sci. Am. 230, 14 (1974).
- Ferretti, R. J., Levander, O. A., J. Agric. Food Chem. 24, 54 (1976).
- Griffen, H. G., Hocking, M. B., J. Chem. Educ. 51, A289 (1974).
- Griffen, H. G., Hocking, M. B., Lowery, D. G., Anal. Chem. 47,
- 229 (1975) Harris, H. B., Parker, M. B., Johnson, B. J., Argon. J. 57, 397 (1965).
- Hawes, R. L., Sims, J. L., Wells, K. L., Argon. J. 68, 217 (1976). Klayman, D. L., Gunther, W. H. H., "Organic Selenium Compounds: Their Chemistry and Biology", Wiley Interscience, New York, N.Y., 1973, pp 652-655.
- Kubota, J., Allaway, W. H., Carter, D. L., Cary, E. E., Lazar, V. A., J. Agric. Food Chem. 15, 448 (1967).
- Martens, D. C., Carter, M. T., Jones, G. D., Agron J. 66, 82 (1974).
- Olson, O. E., J. Assoc. Off. Anal. Chem. 52, 627 (1969).
- Wauchope, R. D., At. Absorp. Newsl. 15, 64 (1976).
- Wauchope, R. D., McWhorter, C. G., Bull. Environ. Contam. Toxicol. 17, 165 (1977).

Received for review May 31, 1977. Accepted October 10, 1977. Mention of a trademark or proprietary product does not constitute a guarantee or warranty of the product by the U.S. Department of Agriculture and does not imply its approval to the exclusion of other products that may also be suitable.