

University of Norte Dame, Norte Dame, Indiana. May 11-13-1976

EFFECT OF SELENIUM INTAKE IN HUMANS AND IN RATS

Werner G. Jaffé*

The interest in the nutritional implications of selenium came from the identification of this element as the causative agent of the alkali disease in horses and somewhat later also of the blind staggers disease of cattle. The toxicological aspects were therefore investigated much earlier than the role of selenium as an essential element (1). It was only after factor 3 was identified as a selenium-containing compound (2) that this element became of general interest for nutritionists. As is so often the case, the pendulum swung completely to the other side, and the toxicological aspects were clouded by the possible nutritional implications, which nevertheless still remain obscure.

From studies with animals it was concluded that the human dietary requirement for selenium is likely to be in the range of 0.1 to 0.2 ppm of the wet weight of the diet. Total intake under normal conditions has been estimated as 50-150 mcg/day/person (3).

The effect on human nutrition is nearly unknown. It will depend on many factors. Plants differ widely in their capacity to take up selenium from the soil, because this element may be more or less available depending on edaphological factors and of the plants themselves (1). As selenium is metabolically related to sulphur, plants with a high sulphur content may be likely to take up selenium more readily than others.

My own interest in selenium came from a chance observation that some lots of defatted sesame seeds, when tested in rats for biological value turned out to be highly toxic, producing anemia, prolonged bleeding time after cutting the tip of the tail for blood sampling, and a high mortality from severe liver lesions (4). The explanation of these toxic effects could be traced to high selenium content. In order to localize the seleniferous areas of the country we obtained through the local public health services a total of 1055 urine samples of school children representing 59 different localities of all regions of Venezuela and also a total of 268 samples of 5 different kinds of foods (corn, rice, milk, beans and sesame). The results of the selenium analysis of these samples permitted us to define 4 geographic zones according to differences in the urine and food selenium levels (5). In the area with the highest urine selenium content the mean value was 0.235 mcg/ml and 39% of the food samples had over 3 ppm selenium. The highest individual urine value was 3.9 mcg/ml. The medium selenium urine secretion in the lowest area was 0.0985 mcg/ml and no food samples had a selenium content greater than 3 ppm. These observations prompted us to initiate several different studies on chronic selenium toxicity in rats and humans, the results of which will be presented briefly.

First we conducted experiments with rats in order to gain information to be used in a later investigation with humans living in the seleniferous area. Among other findings we observed that young rats consuming a diet in which the selenium level of 3 ppm was supplied by high selenium sesame press cake grew

*Department of Food Science, Instituto Nacional de Nutrición, Caracas 101, Apartado 2049, Venezuela.

significantly less than controls with 0.5 ppm of selenium. When the diets were supplemented with L-lysine, the limiting essential amino acid in sesame proteins, the growth difference between controls and animals on the high selenium diets was detectable only at 5 ppm of selenium. Moreover, growth was always better in the animals fed the lysine-supplemented diets even with dietary levels as high as 10 ppm of selenium (4). The effect of lysine on selenium toxicity is probably indirect; it improves the biological value of sesame proteins. We used for all further experiments a lysine-supplemented diet based on sesame press cake with less than 1 ppm of selenium as control, with 4.5 ppm of selenium as moderate level and with 10 ppm as high level seleniferous. In order to find biochemical signs of selenium toxicity which could be used in studies with humans, different parameters were investigated in the blood of selenium fed rats (6). Values of hemoglobin (Hb), prothrombin activity, fibrinogen, and packed blood cell volume (PCV) were lower in rats on the diets with selenium than in the controls (Table 1). Glutamic-pyruvic transaminase (GPT), glutamic-oxalo-acetic transaminase (GOT) and alkaline phosphatase serum levels were also significantly affected in rats by selenium intake (Table 2). From the data reported it can be concluded that the dietary protein level has an important influence on selenium toxicity. The animals on the high protein diet were less affected by the selenium content than those consuming the normal diet.

TABLE 1

PERFORMANCE OF RATS ON SELENIUM CONTAINING DIETS

Diet	Mortality	Hb	PCV	Fibrinogen mg %	Prothrombin activity	Animals with liver lesions	Spleen weight/ body weight	Liver Se mcg/g	Weight gain g in 6 weeks
18% protein 0.5 ppm Se	0/12	14.7	43.9	166.4	96.3	0/12	0.207	0.72	157
18% protein 10 ppm Se	4/12	12.3	40.8	65.5	71.3	10/12	0.544	7.3	61
26% protein 10 ppm Se	1/12	13.8	46.6	93.2	84.7	5/12	0.390	6.7	85

With these results in mind, a field study was undertaken in Villa Bruzual, a village with highest selenium excretion values within the seleniferous area on 111 school children and with 50 children from Caracas serving as controls (7). Selenium levels of whole blood were about 3 times higher in the first group than in the second, i.e. 0.82 and 0.35 ppm, urinary concentrations were 0.64 and 0.35 ppm respectively. The overall hemoglobin and hematocrit values were somewhat lower in Villa Bruzual than in Caracas but no correlation between high selenium blood or urine levels and low hemoglobin or hematocrit values were found. Dietary surveys showed that the children in Villa Bruzual consumed less milk and meat than those in Caracas. Therefore, the differences in hemoglobin were probably due to different nutritional conditions and not to different selenium intake.

TABLE 2

SERUM ENZYME LEVELS IN RATS FED FOR 6 WEEKS EXPERIMENTAL

SELENIFEROUS DIETS

Selenium content of diet ppm	Alkaline phosphatase μ /ml	GOP μ /ml	GPT μ /ml
0.5	6.3	58	20
4.5	8.8	68	26
10.0	18.9	77	43

Values for prothrombin activity, transaminases and alkaline phosphatase were normal in all children and no correlation with blood selenium levels was apparent. Symptoms of dermatitis, loose hair and pathological nails were more frequent among the children in the seleniferous area than in those living in Caracas (Table 3). It was concluded that the intake of selenium in amounts resulting in urinary excretion of levels as high as 0.6 ppm probably did not result in serious health hazards at least in children living under the conditions of this survey.

TABLE 3

BIOCHEMICAL AND CLINICAL OBSERVATIONS IN CHILDREN FROM A HIGH SELENIUM AREA (VILLA BRUZUAL) AND FROM CARACAS

Location	No. of cases	Hb g/100 g	PCV	Prothrombin activity %	Blood Se mcg/ml	Urine Se mcg/ml	Pathological symptoms %
Villa Bruzual	111	12.8	39	95	0.813	0.645	40
Caracas	50	14.8	42	95	0.355	0.224	14

Ingestion of significant amounts of selenium has been related to high incidence of dental caries (8). In the group of children living in the seleniferous area this incidence was slightly lower than that observed in Caracas. It must be pointed out, however, that the level of fluor in the drinking water of the former regions is much higher than that in Caracas.

In Venezuela, no correlation between the incidence of congenital malformations, as revealed by local public health statistics and selenium urine levels could be detected (9). (Congenital malformations in animals have been observed after high selenium intake (1). A note from Colombia dating back over 200 years mentions the frequent birth of monstrous babies to Indian women living in areas which are known to be highly seleniferous (1).

The highest selenium content detected by us in any crop product was a sample of defatted sesame seeds containing 48 ppm. We therefore obtained and analyzed 136 samples of sesame from 20 different countries and found levels of 3 ppm or more of selenium in samples from 14 different countries (10). As sesame is produced mostly in the seleniferous area of Venezuela and the press cake is widely used as an ingredient in animal feed, it is a likely source of selenium in milk, eggs and meat. Sesame flour has been used in feeding programs for undernourished children. Care should be taken about the selenium content in this case in view of the possible relation between protein nourishment and selenium toxicity. Wheat with 180 ppm of selenium has been described in Colombia (11). We have observed 14 ppm in corn and 18 ppm in rice samples from the high selenium area in Venezuela. Smith and Westfall in their classical studies (12) found levels of up to 18 ppm in cereals and vegetables from Wyoming, South Dakota and Northern Nebraska. These values are in the same order of magnitude as those found by us in the area of Villa Bruzual in Venezuela.

In a recent follow-up study we have analyzed samples from 42 different food products bought in Caracas. The data thus obtained were used to calculate the average selenium intake. The results were compared with those reported by Morris and Levander for Beltsville (13) and Thompson et al. (14) for some Canadian diets. It was concluded that the normal dietary selenium intake in Caracas is in the order of 325 mcg/day as compared to 70 mcg for Beltsville. Canadian diets are also lower than those from Caracas containing between 1/3 and 2/3 the amount of selenium. In 2 independent sets of urinary selenium determinations in 77 and 50 school children from Caracas, the mean levels were 0.162 and 0.355 ppm. If these children's daily urinary volume estimated as one 1/ day, the total excretion would be 0.16 and 0.35 mg/day. This is evidently considerably above the safe level recommended by Glover (15) who considers an excretion of 0.1 mg/l to be the maximum allowable concentration for persons exposed industrially to selenium or for a rural population. In a recent calculation, Sakurai and Tsuskiya (3) estimated the normal daily intake from foods to be about 100 mcg. In Japan about half of this comes from fish and shellfish, while in Caracas about 80% comes from eggs, milk products and meats. The availability of selenium from different foods may not be the same in all cases. In the aforementioned survey in Villa Bruzual, average selenium urine levels were three times those observed in the control group from Caracas. The subgroup of 17 children with the highest level excreted 1.021 mcg/ml of urine and had a serum level of 1.112 mcg/ml of selenium. One fourth of all children had serum selenium levels above 1 mcg/ml. Nearly all cases of pathological nails, dermatitis, and nausea were found in this group, but when compared with the subgroup of 11 children with the lowest serum level (0.330 mcg/ml and 0.266 mcg/ml), selenium in the urine, no significant differences in serum GOT, GPT, alkaline phosphatase, and of prothrombin activity could be detected.

Therefore, the only signs to correlate with serum and urine selenium levels were the somewhat poorly defined clinical findings about nausea and pathological nails. It would be highly desirable to count with some sensitive biochemical indices which correlate with selenium intake. It should also be pointed

out that all our studies have been done with school children of 7 to 14 years of age. It may be that in older persons living in the seleniferous area more clinical and biochemical signs could have been detected. The observations of Smith and Westfall which were done on a group of persons of all ages are similar to ours and also comprise a similar range of urinary selenium levels (12). A tentative conclusion would be that the serum selenium level in the subgroup with the highest selenium excretion, i.e. 1 mcg/ml (1 mg/l) is indicative of a toxic level of intake because of the higher incidence of clinical symptoms. This would mean that the maximum allowable concentration in the urine, as defined by Glover (15) has a safety range of about 1:10.

In order to gain more information on the possible consequence of chronic selenium intake in the borderline toxic range we kept rats for two generations on seleniferous diets. In litters born to mothers fed a diet containing 4.5 ppm selenium apportioned by seleniferous sesame press cake with L-lysine added and kept on this diet after weaning, the liver selenium levels decreased steadily; these levels increased in stock rats fed the same seleniferous diet after weaning. At 14 weeks the rats which were born on the 4.5 ppm selenium diet had liver levels significantly lower than those of the controls which had consumed the same diet from weaning, but at birth had only 30% of the liver selenium as compared with the experimental group. It could be shown that this adaptation depended on the length of time the dams had been fed the seleniferous diet during pregnancy and that the protection against accumulation of selenium was clearly detectable also with a diet containing 10 ppm of selenium (16).

The rats from dams fed the seleniferous diet during pregnancy were partly protected against the toxic effects of selenium after weaning. The incidence of mortality, liver lesions, and hyperplasia of the spleen were much lower in the adapted than in the non-adapted rats. Hemoglobin blood concentration, hematocrit, prothrombin activity, fibrinogen, glutamic-pyruvic and glutamic-oxaloacetic transaminase and alkaline phosphatase were much less affected in the rats from selenium fed dams than in the corresponding controls. The differences were larger in the animals killed 4 and 6 weeks after weaning and later tended to get smaller, an observation that may indicate that an adaptation mechanism to selenium intake exists not only in the embryonic phase of life but also thereafter (17).

We tried to find out whether a similar adaptive effect would be detectable in humans. The subgroup of children born in Villa Bruzual was compared with the group living in that area for less than two years. There was a small difference in serum and urine selenium, the children from the first subgroup showing the higher levels, but no significant difference could be found between these groups in any of the other parameters studied. A dietary survey among these children revealed that those living for a short time in the area consumed much less locally grown foods than those which were born there. Therefore, selenium intake may not have been the same in both subgroups.

There probably are areas with much higher selenium content in the soil than that studied by us in Venezuela. Benavides and Silva Mujica published detailed geochemical observations from Colombia indicating levels much higher than those found by us (11). It would be highly desirable to conduct epidemiological studies in these regions and to include investigations on persons of all age groups and on the incidence of liver lesions in autopsy material. This kind of work would be required in order to obtain definite information on the importance of seleniosis in humans.

REFERENCES

1. Rosenfeld, J. and Beath, O.A. Selenium, geobotany, biochemistry, toxicity and nutrition. Academic Press, New York, 1964.
2. Schwarz, K. and Foltz, C.M. Factor 3 activity of selenium compounds. J. Biol. Chem. 233: 245, 1958.
3. Sakurai, H. and Tsuchiya, K. A tentative recommendation for the maximum daily intake of selenium. Environ. Physiol. Biochem. 5: 107, 1975.
4. Chávez, J.F. and Jaffé, W.G. Nivel tóxico de selenio en dietas para ratas. Arch. Latinoamer. Nutr. 17: 69, 1967.
5. Mondragón, M.C. and Jaffé, W.G. Selenio en alimentos y en orina de escolares de diferentes zonas de Venezuela. Arch. Latinoamer. Nutr. 21: 185, 1971.
6. Jaffé, W.G., Mondragón, M.C., Layrisse, M. and Ojeda, A. Síntomas de toxicidad de selenio orgánico en ratas. Arch. Latinoamer. Nutr. 22: 467, 1972.
7. Jaffé, W. G., Ruphael, M., Mondragón, M.C. and Cuevas, M.A. Estudio clínico y bioquímico en niños escolares de una zona selenífera. Arch. Latinoamer. Nutr. 22: 595, 1972.
8. Hadjimarkos, D.M. Effect of trace elements on dental caries. Adv. Oral Biol. 3: 253, 1968.
9. Jaffé, W.G. and Vélez, F. Selenium intake and congenital malformations in humans. Arch. Latinoamer. Nutr. 23: 515, 1973.
10. Jaffé, W.G., Chávez, J.F. and Mondragón, M.C. Contenido de selenio en muestras de ajonjolí (Sesamum indicum) procedentes de varios países. Arch. Latinoamer. Nutr. 19: 299, 1969.
11. Benavides, S.T. and Silva Mujica, F. Seleniosis, Instituto Geográfico "Agustin Codazzi", Bogotá, 1965.
12. Smith, M.I. and Westfall, B.B. Further field studies on the Se problem in relation to public health. Publ. Health Rep. U.S. 52: 1375, 1973.
13. Morris, V.C. and Levander, O.A. Selenium content of foods. J. Nutr. 100: 1383, 1970.
14. Thompson, J.N., Erdoy, P. and Smith, D.C. Selenium content of food consumed by Canadians. J. Nutr. 105: 274, 1975.
15. Glover, J.R. Selenium in human urine: a tentative maximum allowable concentration for industrial and rural populations. Ann. Occup. Hyg. 19: 3, 1967.
16. Jaffé, W.G. and Mondragón, M.C. Adaptation of rats to selenium intake. J. Nutr. 97: 431, 1969.
17. Jaffé, W.G. and Mondragón, M.C. Effects of ingestion of organic selenium in adapted and non-adapted rats. Br. J. Nutr. 33: 387, 1975.