Hair Selenium Levels and Children's Classroom Behavior

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Abstract

The study investigated the relationship between children's hair-selenium levels and children's behavioral performance in school. Hair-selenium levels of 120 children drawn from a general school population were correlated with teachers' ratings of the children on the Walker Problem Behavior Identification Checklist (WPBIC). Parents were interviewed to control for confounding variables that may effect behavioral development. Increasing hair-selenium values correlated significantly with increased scores on the WPBIC scales measuring acting out, withdrawal, and total scale score. A continuing reexamination of selenium exposure in the young is needed in order to determine the margin of safety between potentially toxic and nutritionally required levels of selenium.

The importance of slight amounts of selenium in human and animal nutrition is well known (Ganther, 1974; Scott, 1981). At concentrations higher than needed for nutrition the element is toxic (Wilber, 1980), and individuals exposed to high doses of selenium are subject to CNS impairments resulting in behavioral disorders. Yang et al. (1983) described the central nervous system effects of selenium intoxication in inhabitants of a village in China. CNS abnormalities included peripheral anesthesia, acroparesthesia, numbness, convulsions, paralysis, motor disturbance, and hemiplegia. Smith and coworkers (Smith and Westfall, 1936; Smith et al., 1936) in their surveys of rural populations in seleniferous areas of Wyoming, South Dakota, and Nebraska reported behavioral correlates of selenosis which included irritability, depression, nervousness, emotional instability, and extreme lassitude.

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Although it has been assumed that the margin of safety is adequate between potentially toxic and nutritionally required levels of selenium, there is no literature which has examined selenium levels of a general population in relationship to sensitive measures of behavioral performance. The purpose of this study was to examine children's selenium levels in relationship to teachers' ratings of their classroom behavior.

In this study selenium levels were determined via hair samples and atomic absorption spectroscopy. Although blood values are the most widely used indicator of selenium status, there is increased interest in the use of hair as a diagnostic tool for the assessment of trace element status (Bowen, 1972; Passwater and Cranton, 1983; Petering et al., 1971). Hair collection is a less invasive procedure, and can be stored without loss of nutrients, and is a potential reflection of long term accumulation of a given substance. If proper procedures are used to minimize sampling error and environmental contaminants, highly sensitive and accurate measurements of hair trace element concentrations can be made (Maugh, 1978; Laker, 1982).

Methods

The 120 subjects in this study were randomly drawn from grades one through six at two elementary schools in Wyoming. The mean age of the subjects was 9.09 years with a standard deviation of 3.64. Sixty-three subjects were female.

Confounding Variables

Parents of subjects were interviewed via questionnaire or telephone in order to identify and control for the following confounding variables that affect behavioral development: mother and father's age at subject's birth, length of hospitalization at birth, birth order, birthweight, number of hospital admissions since birth, history of immunizations against childhood diseases, presence of father in the home, and mother and father's occupation and level of educational attainment. In addition, each child's school record was reviewed to determine whether there was a known or highly probable medical reason for a behavioral deficit, e.g., neurological disturbance. None of the children's school records contained a "probable cause."

Classification of Selenium Levels

After obtaining parental permission, children were asked to submit a small sample of hair (about 400 mg) for trace mineral analysis. None of the subjects reported using selenium containing shampoos (e.g., Selsun Blue, Iosel). Hair samples were collected from the nape of the child's neck, as close to the scalp as possible, by the senior researcher using stainless steel scissors. The hair samples were then submitted to Doctor's Data, Inc., a state and Center for Disease Control licensed laboratory in West Chicago, where they were analyzed with the atomic absorption spec-troprotometer, the graphite furnace, and the induction coupled plasma torch to determine the children's selenium levels.

Precise laboratory standards are used by Doctor's Data, Inc., to assure reliability of results and to meet reproducibility requirements. These include:

1. A blind sample is run from the initial steps through the entire procedure to assure reproducibility of methods.
2. At least one of every three tests is a standard. Working standards are made to assure proper values.
3. The in-house pool is completely remade and analyzed monthly to eliminate the possibility of precipitating elements and to assure reproducibility.
4. Temperature and humidity are controlled to assure reliability and consistency of the testing instruments.
5. The hair samples are weighted to the thousandths of a gram (.001g is equal to approximately four hairs), 1 inch (.0254m long); and only volumetric flasks, the most accurate available are used for diluting the ashed sample.
6. Lot-number control sheets for all reagents are used to assure uniformity. Records are kept and available for inspection.
7. All glassware is acid washed in-house before used and between each use, including acid prewashed disposable test tubes.
8. The water used at Doctor's Data, Inc. is virtually mineral free, rated at 18 + MEG.
9. Upon receipt the hair sample is washed thoroughly with deionized water, a non-ionic detergent, and an organic solvent to remove topical contaminants.

Walker Problem Behavior Identification Checklist (WPBIC)

The Walker Problem Behavior Identification Checklist (Walker, 1976) is a screening device designed for elementary teachers in selecting children with behavior problems who may need referral for further psychological evaluation, referral, and treatment. The WPBIC consists of 50 observable operational statements of classroom behavior that might limit a child's adjustment in school. Differential score weights are assigned to each statement based on their influence in handicapping a child's adjustment. Factoring the 50 items, there are 14 items relating to acting-out (aggressive and disruptive behaviors), 5 items relating to withdrawal (socially avoidant and passive behaviors), 11 items relating to distractibility (poor attentiveness and restlessness), 10 items relating to disturbed peer relations, and 10 items relating to immaturity.

Standardized on 534 elementary age children, the mean raw total score was 7.76 with a standard deviation of 10.53. A total scale score of 21 or greater separates disturbed behavior from non-disturbed behavior, and Walker (1976) reported the split half reliability of the scale at .98 and the difference between the means of disturbed and non-disturbed children on the scale significant beyond the .001 level.

In the present study, classroom teachers were instructed on how to fill out the scale by the senior researcher. All teacher ratings were based on observations of the child's classroom behavior for the past two months prior to hair collection.

Results

The subjects' mean hair-selenium value was 0.34 parts per million (ppm) with a standard deviation of 0.15 ppm and a range of 0.18 ppm to 0.72 ppm. According to Doctor's Data, Inc. (Doctor's Data, Inc., 1985), the accepted reference range (±1 SD) for hair-selenium is 0.16 ppm to 0.88 ppm. Thus, the subjects' hair-selenium values were well within the range of accepted laboratory norms.

The means and standard deviations for the subjects' scores on the WPBIC are presented in Table 1. The groups' WPBIC total score of 10.45 was well within the range of normal behavior (<21).

Table 1
Group WPBIC Scores (N = 64)

Scale	**Mean**	SD
Total Scale	10.45 '	14.06
Acting-Out	3.88	6.10
Withdrawal	1.32	2.92
Distractibility	2.84	3.24
Disturbed Peer Relations' '	1.34	2.93
Immaturity	1.09	2.10

Pearson Product correlations were run between confounding variables and the WPBIC scales as well as hair-selenium values. None of the confounding variables were correlated with the WPBIC scales or hair-selenium values.

A hierarchical multiple regression analysis using sets of predictor variables (Cohen and Cohen, 1975) was performed on each of the Walker scales. The incremental increase in explained variance attributable to selenium over and above age and sex was tested for significance. The order of entry was (1) age and sex and (2) selenium. The correlations involving selenium and subscale scores are in Table 2.

Table 2
Correlations of Selenium and Walker Scales

	Acting-Out	**Withdrawal**	**Distractibility**	**disturbed Peer Relations**	**Immaturity**	**Total Scale**
Selenium	**.32***	**.29***	**.28***	**.23***	**.26***	**.34***

*p<.01

As a check on some of the assumptions underlying the use of multiple regression, scatterplots were constructed between each of the Walker scale distributions and selenium. No patterns of curvilinearity were detected.

Residual plots were generated to test the equality of variance assumption and to identify outliers. The variability of standardized residuals was uniform over all cases for scale distribution. An outlier was defined as a case falling at least two standard deviations from the mean of the residual distribution. Less than *5%* of the cases were identified as outliers, and the cases were not constant across scales.

Although a moderately positive skew was detected in some of the scale distributions, the F test is robust to violations of normality, and correlations will not be deflated appreciably unless the skewness of one variable is opposite the skewness of the other variables. None of the predictor variables revealed a positive skew.

For the acting-out scale selenium contributed an additional *8%* to the explained variance, beyond age and sex, (F (1,116) = 5.42, p < .05). For the withdrawal scale selenium accounted for *8%* of the explained variance in addition to the cov-ariates(F = (1,116) = 5.58, p < .05). Finally, for the Walker total scale score selenium accounted for almost 9% of the variance, over and above age and sex, ($F (1,116) = 6.39$, $p < .05$); all other scale scores were nonsignificantly related to hair-selenium values.

Sex accounted for *8%* of the total scale score variance $F (1,118) = 6.39$, p. $< .01$. The correlation of sex (male = 0, female = 1) with selenium was significant, -.23 ($p < .05$). Age did not correlate significantly with selenium.

Discussion

The value of this study is that it presents baseline data on the relationship between hair-selenium values in children and sensitive measures of their behavioral performance in school. There is an absence of comparative data, as previous studies have focused on selenium levels in relationship to clinical symptoms, rather than validated measures of behavior.

Regression data indicated that selenium was significantly and positively related to increased scores on the acting-out and distractibility scales and the total scale score. Controlling for age and sex, it was found that 9% of the total scale score was accounted for by hair-selenium values. Behavioral performance is, of course, multifactorally determined, as there are a large number of genetic and environmental determinants, and for low selenium levels to account for 9% of the overall variance is noteworthy.

The potential adverse effects of selenium exposure on developmental processes may be of additional importance for several reasons.. First, the effects may signal the early stage of an ongoing toxic process that becomes more disabling with age. Second, it is known that there is considerable individual variation to metals. A metal that produces subtle behavioral alterations in many children may produce a severe behavior disorder in those who are especially susceptible because of genetic or other factors, e.g., malnutrition. Third, for the child's caretakers, some selenium induced behavioral effects (e.g., hyper-irritability) may make a child difficult to parent or instruct, leading to adult-child tensions that become amplified over time.

The significant behavioral correlates that we observed resemble those reported by Smith and coworkers (Smith and West-fall, 1939;.Smith et al., 1939) in their earlier surveys of rural populations in seleniferous regions of the western U.S.A. Those authors could not establish a direct relationship between the signs they observed in humans, e.g., irritability, nervousness,. emotional instability, because they could not show a definite association between clinical signs of individuals and the level of selenium in the urine. In our opinion, the variability of urine-selenium values and the lack of sensitive behavioral measures employed would make such correlations difficult to establish.

Few other reports deal with the chronic subclinical effects of environmental selenium on man. Kilness and Hochberg (1977) described four cases of amyothrophic lateral sclerosis, a neurological disease, over a 10-year period in South Dakota farmers unrelated but living within a radius of about seven miles of one another. The area is rural, sparsely settled, and is known for chronic selenium poisoning of farm animals. The authors suggested that a casual relationship existed between the high environmental selenium in the area and the observed amyotrophic lateral sclerosis in human beings residing there.

Ely et al. (1981) in a study of the trace metal content of 400 children in the Akron, Ohio, public schools reported significantly higher hair-selenium levels in learning disabled children when compared to a control group. Learning disabled children in special classes had significantly higher hair-selenium values than learning disabled children who remained in the regular classroom.

As previously noted, the hair-selenium values of the sample population were well within the accepted laboratory norms of Doctor's Data, Inc. Yang et al. (1983) in their study of endemic selenium intoxication in China postulated that the maximum tolerable level of hair-selenium is 3.75 ppm. Previous work by these authors (Keshan Disease Research Group, 1979a; 1979b) indicated that the minimum safe hair levels characteristic of populations protected against selenium deficiency (Keshan disease) was 0.16 ppm. Thus, the ratio of maximum selenium level tolerated versus minimum selenium level required would be 23, according to these authors. The data presented here suggest that such a margin of safety may be grossly overestimated if selenium uptake is examined in terms of behavioral performance, rather than clinical signs of morbidity.

Confidence in the associations reported here between WPBIC measures and hair-selenium values depends on the validity of hair as a marker of exposure. The classification of selenium to hair-selenium levels has been validated in a number of other studies. Hair-selenium values have been found to correlate with selenium dietary intake (Gallagher et al., 1984), with exposure to selenium in well water (Valentine et al., 1978), with endemic distribution of Keshan disease (Keshan Disease Research Group, 1979a; 1979b), with endemic distribution of selenosis (Yang et al., 1983), and with glutathione peroxidase levels (Bland, 1980).

The biological and developmental significance of our findings is not clear. While warranting replication, the increased WPBIC scores may be functional evidence of low level selenium induced neuronal damage. Selenium studies in monkeys have demonstrated behavioral effects with typical signs of brain lesion and lathyrism (Rudra, 1952), and Rosenberg et al. (1966) found that selenium changed the conduction properties of the neural signal. The subtle subclinical effects of selenium are little understood, however, due to the absence of studies relating sensitive measures of behavior to selenium exposure. The developing central nervous system is especially vulnerable to metal toxicity, and future studies should examine the specificities of selenium exposure in the young in order to determine the margin of safety between potentially toxic and nutritionally required levels of selenium.

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