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Trace Metal Content of Hair

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Trace Metal Content of Hair

II. Cadmium and Lead of Human Hair in Relation to Age and Sex

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Cadmium and lead content of hair of males and females was found to be age-dependent. Regression analysis showed two male groups with respect to cadmium, namely those under and those over 12 years of age, while the data revealed only one male group with respect to lead. Analysis of the data also indicated that hair cadmium distribution for females delineated one group below, and one above, 50 years. Females showed hair lead which fell into two groups, one below and the other above 30 years of age. Cadmium and lead in the hair of both sexes showed a high degree of association of the metals ($r=0.4052$ ($P=.001$) for males and $r=0.2563$ ($P=.02$) for females). The data indicate that comparisons of metallic content of hair in humans must be limited to a narrow age range and to one sex.

Previously we¹ have reported that the levels of the biologically essential metals, zinc and copper, in the hair of a relatively large sample of an urban population ranging in age from 2 to 88 years varied with age in the case of both sexes. In males there was an increase in hair content of both metals during childhood and then a decline after physiological maturity, the inflection point being at approximately 12 years of age. In the case of females, our data were available only for individuals older than 14 years of age. In these, the level of zinc declined while the copper content increased with age.

Samples of hair from the same individuals in the study mentioned above were analyzed for the toxic metals, cadmium and lead, in order to ascertain whether the hair content of these

metals also varied with age in a manner similar to that found for the essential metals. Such data are important in deciding upon the suitability of hair as a biopsy material to assess the exposure of human beings to, and their absorption of, toxic metals in the general or occupational environment.

Materials and Methods

Samples of hair from 95 males in the age range of 2 to 88 years and from 83 females in the age range of 14 to 84 years were taken from the predominantly white urban population of Cincinnati as previously reported.¹ These samples were identical with those used in the earlier report.

The hair samples were individually washed as described in the earlier report,¹ being treated successively with (1) organic solvents to remove oils, lacquers, and particulate matter; (2) a mild anionic detergent to remove adsorbed heavy metals; (3) deionized water to remove the detergent; and (4) acetone to dry the hair. Storage was in glassine envelopes.

The washed and dried hair samples were wet-ashed with a mixture of nitric and perchloric acids (5:1 v/v) and analyzed by the atomic absorption spectrophotometric method.¹ After zinc and copper had been determined in the 10% nitric acid solution in which the ash from each sample was dissolved, the remainder of the solution was measured and evaporated to about one fifth its volume. This solution was then analyzed for cadmium and lead. The concentration procedure was necessary because of the low levels of cadmium and the lower sensitivity of the lead determination as compared with the analysis of zinc, copper, and cadmium.

Statistical Analysis.—In accord with our previous experience, the variable influence of age on the contents of cadmium and lead in the hair of each sex was evaluated by applying conventional methods of covariance analysis to log transforms of both the age of the individual and the concentration of the respective metals found in the hair. Regressions were fitted to the log transforms by the method of least

squares. In some instances, regressions were fitted separately to two age periods corresponding to major points of inflection suggested by initial plots of the data. The Student *t* test and variance ratios (Fisher *F* test) were used to evaluate the statistical significance of the data. The extent of the correlation between the concentrations of any two of the four metals (zinc, copper, cadmium, and lead), without regard to age, was evaluated from the original data.

Results

Precision of Analytical Methods.—The same 29 aliquots of a standard mixture used to determine the sensitivity of the methods for zinc and copper¹ were also used to test the reliability of our methods for cadmium and lead. The mixture, in addition to zinc and copper, contained Cd^{+2} , $0.100\mu g/ml$ (as nitrate), and Pb^{+2} , $1.00\mu g/ml$ (as nitrate), and, as before, the aliquots were carried through the ashing procedure as well as the atomic absorption spectrophotometric determination procedure.

The results presented in Table 1 show that the degree of precision of the analyses for both cadmium and lead was lower than that found for zinc and copper,¹ but was satisfactory for the level of testing for which the methods were designed.

We have also tested the precision of our methods for cadmium and lead in a manner similar to that described before,¹ as shown by the data of Table 2. Here we give the means and statistical variations of ten replicates of one large hair sample. These data demonstrate that there was a high degree of consistency in the sample values for both cadmium and lead, and that the methods employed were sensitive enough to detect the effect of age and sex, which were the physiological variables under consideration.

Cadmium and Lead in Hair.—The data for average concentration of

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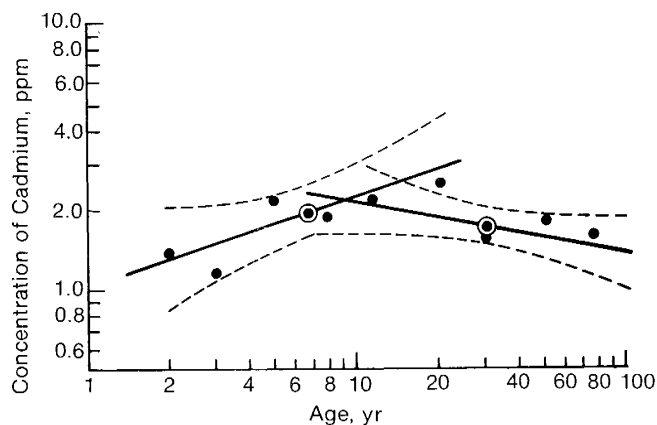
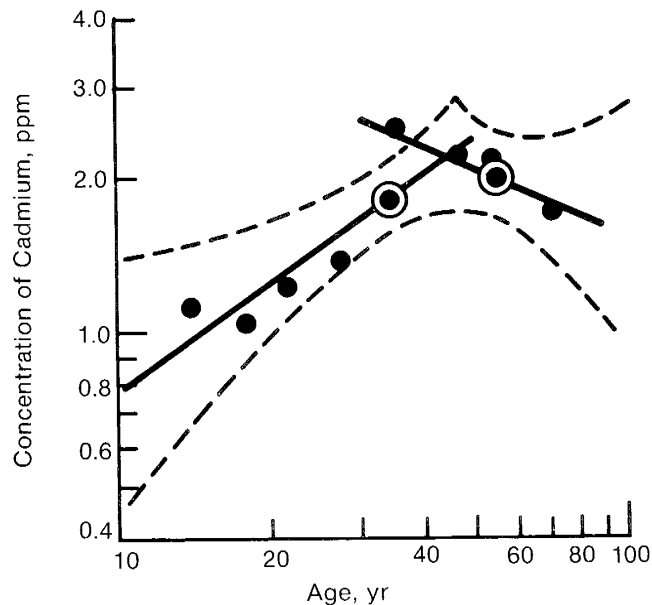


Fig 1.—Log-normal regression lines (with 95% fiducial limits) of hair cadmium of males in age ranges of 2 to 24 and 6 to 88 years. Individual points represent means of data grouped in intervals of 0.1 or 0.2 log units. Encircled points indicate means of age and metal content.

Fig 2.—Log-normal regression lines (with 95% fiducial limits) of hair cadmium of females in age ranges 14 to 56 and 30 to 84 years. (Details as indicated in Fig 1 legend.)



Element	Added, $\mu\text{g/ml}$	Recovery, $\mu\text{g/ml}$		
		Mean	SE	Range
Cadmium	0.100	0.130	0.0055	0.080-0.200
Lead	1.00	0.94	0.037	0.60-1.30

*Twenty-nine aliquots of standard solution of cadmium and lead as nitrates were carried through analytical procedure, including ashing, and analyzed by atomic absorption spectrophotometry.

Element	Means of Ten Replicate Analyses, $\mu\text{g/ml}$
Cadmium	0.55 ± 0.07 (SE)
Lead	3.96 ± 0.32 (SE)

cadmium in the hair of males and its variation with age are shown in Fig 1, and those data pertaining to females are shown in Fig 2. Although the axes are marked in conventional units, it should be noted that the regression lines are logarithmic. The encircled points represent the logarithmic means of age and cadmium around which the variance has been computed. The solid lines represent the predicted concentrations, and the dotted lines represent the fiducial limits of the predicted values. The points along each curve represent averages when the data are grouped in log-age intervals of 0.1 or 0.2; they show general conformity of the data handled in this manner to the estimated regression.

Figures 3 and 4 present values for lead of hair from males and females respectively. The Figures were formulated in a manner identical with the method mentioned above. The regressions shown are those which by analysis of variance showed correlations indicating the highest statistical significance we were able to find, ie, $P = .05$ or less. The statistical significance of each regression tested is given in Table 3.

The data shown in Fig 1 indicate that cadmium in hair of males increased with age to the 20th year ($P = .04$) and probably remained at an elevated level or decreased slightly thereafter ($P = .05$); the peak concentration was not clearly defined.

The concentration of cadmium in the hair of females (Fig 2) increased from the 14th year (earliest age of sample) to a peak between 40 and 50 years of age ($P = .001$). Thereafter the value remained high.

Lead in the hair of males in our population (Fig 3) appeared to behave differently from the pattern found for

cadmium. It gradually declined from a high value of 25 ppm at age 2 years to a low value of about 10 ppm at age 85 ($P = .01$).

In the case of the females (Fig 4), hair lead concentration rose sharply from a low of 4 ppm at 14 years to a peak value of about 40 ppm at 35 years of age ($P = .02$) and then decreased sharply to a low of 2 ppm at 84 years ($P < .01$). This pattern is similar to that found for cadmium in the hair of females, but is sharper in its rise and fall.

Statistical Correlations Between Metals.—In Table 4 we have summarized all of our data on the hair concentrations of zinc, copper, cadmium, and lead for the 95 male and 83 female subjects used for this study. These data show the means and standard errors for each metal in each sex, the correlation coefficients, the values for the Student t test, and the probabilities of values of t larger than those shown in random samples.

These compilations demonstrate the following relationships: In male hair, zinc is related to copper ($r = 0.3591$) and to lead ($r = 0.3218$), copper is associated with cadmium ($r = 0.2262$), and cadmium and lead show a high degree of correlation ($r = 0.4052$). In female hair, copper and cadmium show a correlation ($r = 0.2034$), and cadmium and lead show a high degree of association ($r =$

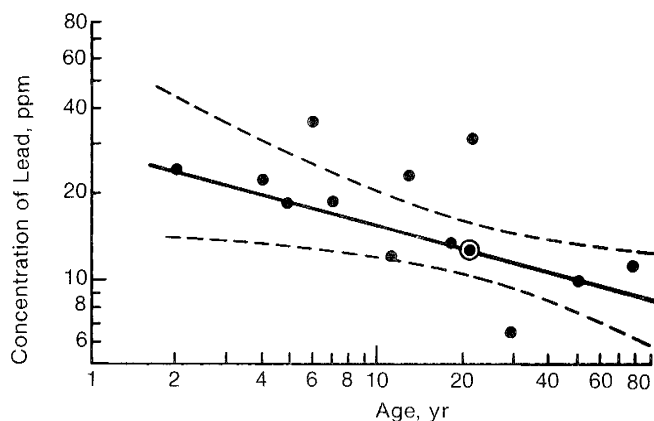


Fig 3.—Log-normal regression lines (with 95% fiducial limits) of hair lead of males in age ranges 2 to 88 years. (Details as indicated in Fig 1 legend.)

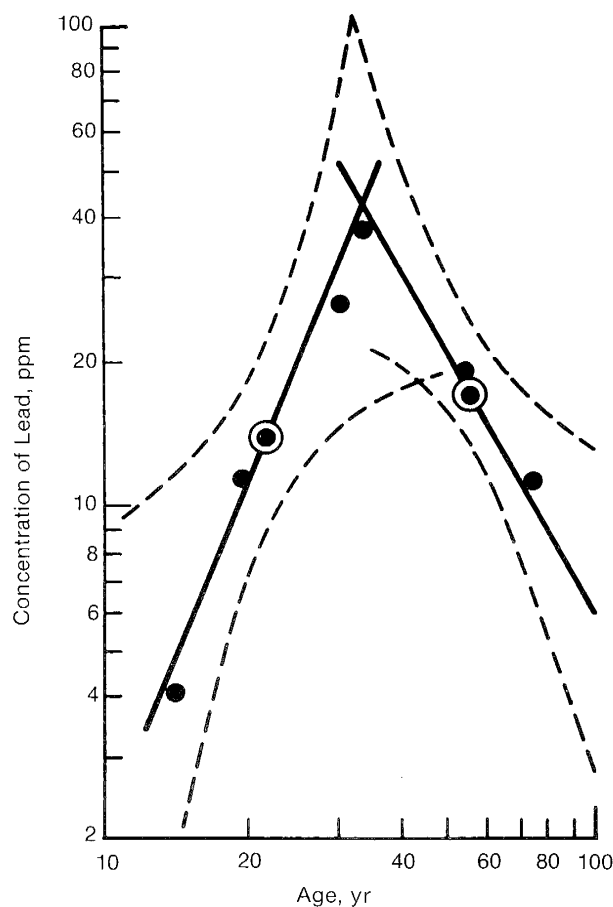


Fig 4.—Log-normal regression lines (with 95% fiducial limits) of hair lead of females in age ranges 14 to 30 and 30 to 84 years. (Details as indicated in Fig 1 legend.)

0.2563). These relationships are noteworthy in that they probably indicate differences between the sexes in their metabolism of these metals.

The lack of correlation between the concentrations in males of zinc and cadmium was surprising since their patterns of distribution with age were somewhat similar.

Comment

Lead in the hair of children has been used as a biopsy material by Kopito and co-workers^{2,3} to indicate the extent of exposure to lead and the presence of chronic plumbism, but the use of hair cadmium as an index for judging environmental and occupational exposures of human beings has only recently been considered. Thus in 1968, Petering and Yeager⁴ reported the preliminary findings of the age and sex relationships of cadmium and lead which are presented in detail

Metal	Subjects		Mean Log Concentration, $\mu\text{g/gm}$	Mean Log Age, yr	Regression Coefficient, Slope	r	t	df	P
	Sex	Age Span, yr							
Cadmium	M	2-24	0.2940	0.8186	0.3390	0.3449	2.17	35	0.04
Cadmium	M	6-88	0.2360	1.4870	-0.1924	0.1952	1.75	77	0.05
Cadmium	M	20-88†	0.2040	1.6464	0.0025	0.0013	0.01	59	0.5
Cadmium	F	14-56	0.2616	1.5323	0.6915	0.4088	3.32	55	0.001
Cadmium	F	30-84	0.3033	1.7455	-0.4420	0.1278	0.98	58	0.3
Lead	M	2-88	1.0972	1.3320	-0.2702	0.2629	2.63	93	0.01
Lead	M	2-20†	1.2618	0.8031	-0.1436	0.1316	0.77	34	0.4
Lead	M	18-88†	1.6474	0.9343	0.3402	0.1114	0.85	57	0.4
Lead	F	14-30	1.1225	1.3258	2.4931	0.5126	2.74	21	0.02
Lead	F	30-84	1.2367	1.7461	-1.8058	0.3432	2.81	59	0.01

*Herein, r represents correlation coefficient; t , Student t value; df, degrees of freedom; and P , probability of larger value of t in random samples.

†Regression not shown in the Figures.

here, and in 1969, Schroeder and Nason⁵ also indicated that there may be variations of cadmium and lead in the general population due to age and sex. Recently Nordberg and Nishiyama⁶ have shown that cadmium injected into mice quickly finds

its way to the hair follicles. The data presented here offer additional information on some of the factors which need to be weighed in consideration of the suitability of hair to assess exposure and absorption of cadmium and lead.

Table 4.—Correlations of Concentrations of Zinc, Copper, Cadmium, and Lead in Hair, Irrespective of Age*						
83 Females						
Metal		Zinc	Copper	Cadmium	Lead	
95 Males	Metal					
		Zinc	152 ± 6			
		Copper		29.6 ± 2.8		
		Cadmium			2.43 ± 0.26	
95 Males	Zinc	<i>r</i>	0.0932	0.0712	0.0603	
		<i>t</i>	0.84	0.64	0.54	
		<i>P</i>	145 ± 4	.4	>.5	>.5
95 Males	Copper	<i>r</i>	0.3590	0.2034	0.1463	
		<i>t</i>	3.71	1.87	1.33	
		<i>P</i>	<.001	34.7 ± 6.7	.05	>.1
95 Males	Cadmium	<i>r</i>	0.0438	0.2262	0.2563	
		<i>t</i>	0.42	2.24	2.39	
		<i>P</i>	>.6	.025	2.20 ± 0.2	.02
						24.4 ± 2.7
95 Males	Lead	<i>r</i>	0.3218	0.0947	0.4052	
		<i>t</i>	3.28	0.92	4.27	
		<i>P</i>	.001	>.3	.001	18.3 ± 1.8

*Diagonal axis represents concentrations (expressed as parts per million) ± SE; *r* represents correlation coefficient; *t*, Student *t* value; and *P*, probability of larger value of *t* in random samples.

The data clearly show that there are age variations in cadmium and lead of hair, and that these differ between the sexes. Therefore, it is important to limit comparisons to those values from one sex and from a fairly narrow age cohort, eg, a decade at most. These data also help to establish ranges or means which may be expected to be found in an urban population not exposed occupationally. Thus, our data suggest that cadmium in the general population not exposed occupationally will range between about 0.5 and 2.5 ppm, in both sexes, regardless of age above 2 years.

Similarly our data suggest that lead ranges between about 2 and 30 ppm. The value of 30 ppm as an upper limit for "normal," nontoxic, nonoccupational exposure was set by El-Dakhkhny and El-Sadik⁷ as a basis for using hair to assess toxic exposure of workers in lead industries. Their washing procedure was a very drastic one which would probably lead to low results. Therefore, limits on hair values should include specification of washing procedures until a standard method is accepted.

The criticism of using hair for determining occupational hazards in plants where cadmium is used as reported by Nishiyama and Nordberg⁸ is similar to that of Bate,⁹ and need not be of great concern since these authors used methods and conditions

which were unphysiological and extreme in the exposure of the hair to metal ions. Their results cannot appropriately be related to real-life situations of exposure. Under most occupational and environmental conditions, both lead and cadmium in the atmosphere are in particulate form, and come into contact with hair which has a protective film of oil or other organic matter. Proper washing of the hair samples will remove the adhered particles. If there is still any question about the significance of high results, pubic and axillary hair samples may be analyzed, ie, samples from areas not readily or ordinarily exposed to atmospheric particulate matter.

The high correlation of cadmium and lead in both males and females seems to us to be important in suggesting that a possible unknown factor in lead poisoning of children may in fact be an associated exposure to cadmium. Furthermore, blood is not a suitable material to analyze for cadmium, since the metal remains in blood only very briefly and, in consequence, the levels are always extremely low. Therefore, studies of cadmium and lead in blood would be inadequate to demonstrate associated exposure, while analysis of hair might lead to important findings.

The high correlation between cadmium and copper in males and fe-

males deserves additional study since it is known that cadmium can influence the metabolism of copper.¹⁰ Similarly, the demonstration of a correlation between zinc and lead in males merits attention by experimentalists since both metals reflect a dietary and an environmental exposure. The absence of correlation between zinc and cadmium in hair of both sexes is surprising in view of the known interactions found in experimental animals.

Finally our report of a high level of lead in the hair of children is of considerable interest since the children in our study were from families with middle class socioeconomic and educational backgrounds and represent a different environment from those who reside in poor neighborhoods. The observation suggests unsuspected exposures of children, perhaps even in utero, which need to be sought out.

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