CADMIUM, LEAD, COPPER AND ZINC IN FOOD, FECES AND ORGANS OF HUMANS

—Interrelationships in food and feces and
interactions in the liver and the renal cortex—

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ABSTRACT

The concentrations of Cd, Pb, Cu and Zn in food, feces and organs were analyzed in order to study the interrelationships among these metals in humans. Correlation coefficients and the ratio of all 4 metals in these media were calculated, and the following results were obtained: the ratios of Cd to Cu and Cd to Zn in the renal cortex showed 100 times higher and those ratios in the liver showed 10 times higher than those in food and feces. However, Pb/Cu and Pb/Zn ratios did not vary in these media. Intimate correlations between Cd and Zn were found in the organs in spite of the fact that they showed no correlation in food and feces. Cu and Zn indicated high correlations in food, feces and renal cortex, but not in the liver. Thus this study indicates that the proportions of the metals ingested via food and excreted in feces are almost the same, i.e., the absorption rates are not greatly different. However, once absorbed, the relationships among the metals are markedly altered, suggesting different metabolism by individual metal and/or possibly metal-metal interactions in the human organs, particularly in the renal cortex and the liver.

INTRODUCTION

There are many studies on the distribution of heavy metals in organs such as renal cortex and liver.^{1,2,3,4} Tsuchiya *et al.*⁵ and Friberg *et al.*⁶ reported, for example, that one half to two thirds of the total body burden of cadmium is distributed in the renal cortex and the liver. It is also known that more than 90 per cent inorganic lead is stored in the bone.⁷

However, effects of a heavy metal such as cadmium (Cd), lead (Pb), copper

(Cu) and zinc (Zn) on humans depend not only on the concentration of the metal in a critical organ but on the interaction with other metals and elements. There are a great many animal experiments, for instance, on antagonistic effects between Cd and Zn or methylmercury and selenium.^{8,9} There may also be synergism or additive effects between two or more metals. Such effects on humans are generally considered to be interactions of heavy metals.

As for the interaction of heavy metals in human organs, there are few reports, since it is difficult to conduct human experiments for observing the metabolism, interactions and effects of heavy metals. This study will not cover every aspect of metal-metal interaction, but will be restricted to the relationships and interrelationships among 4 metals: Cd, Pb, Cu and Zn concentrations in food, feces and human organs. From this study the author aimed to obtain basic information on the physiological interrelations among those metals.

MATERIALS AND METHODS

Feces study

Nineteen male medical students (21–24 years) and 17 female nursing students (20–22 years) from Keio University were selected for analysis of the metals in their feces. They all lived in Tokyo and had no illness during the period of observation and no known high exposure to the metals. Feces were collected daily for 5 days for each person (a total of 172 samples). Information on each subject was obtained, at the same time, including smoking and drinking habits, habitual medications, place of residence and food items ingested the day prior to sampling. However, this information was not used in this study because findings which may affect the result of the study were not obtained.

Each sample in a plastic bag was weighed as wet weight and stored immediately in a freezer. It was then put into a freeze-dryer for about 7–10 days to be lyophilized. The dry weight was recorded after lyophilization and then stored in a plastic container. Two grams of dry samples was put into an electric oven at 400°C for 36 hours. After ashing, 0.5N nitric acid (HNO₃) was added to the sample to a total volume of 20 ml. Zinc was determined by flame atomic absorption spectrophotometry (AAS) by taking 1 ml of the solution without extraction. For copper, lead and cadmium analyses, extraction by methylisobutylketone (MIBK) with chelation of sodium diethyldithiocarbamate (DDTC) was performed. One ml, 3 ml and 15 ml solutions were used for Cd, Pb and Cu analyses, respectively, and these solutions were analyzed by flame AAS.

Food study

The meals were donated by the administrative office of Keio University Hospital. Twenty days or 60 meals of food samples were collected and the three meals of each day were mixed together to make 20 samples for analysis. These were regular hospital meals prepared for the patients not requiring special diets. Beverages such as tea and milk were included in the daily food. The average daily calorie intake was 2298 ± 131 kcal, which was almost equal to the standard recommended by the Ministry of Health and Welfare.

The collected samples were mixed by electric mixer and lyophilized in a freeze-dryer. Ten grams of dried materials were prepared for analysis. Analytical methods were the same as those used in the feces study.

Organ study

Among the human organs and tissues, renal cortex and liver were represented since the former is the critical organ for cadmium poisoning and the latter is important for studying metal toxicology in general. Cadmium, lead, copper and zinc concentrations in the renal cortex and the liver were determined using 85 cadavers of both males and females, which had been autopsied at Keio University, Department of Legal Medicine. These were sudden death cases without known disease or chemical poisoning.

Five grams of liver and 1 gram of renal cortex were ashed by acids in Kjeldahl flasks. After digestion, the liquid was filled with 0.5N-HNO₃ up to 20 ml. The concentrations of four metals were determined by the same methods as performed in the feces study.

Since the method used for ashing organ samples was different from that used for feces, Cd concentrations in 10 samples of liver were measured using both dry and wet digestion in order to cross-check the methods. No significant differences in the results were found; the correlation coefficient was $r=0.90$ and the regression line was $Y_{\text{DRY}}=1.07X_{\text{WET}}-0.02$.

RESULTS

Feces study

As shown in Table 1, daily amounts of feces of the male students varied from 31.3 g to 250.7 g wet weight (7.4 g to 47.7 g dry weight). Average amounts were $116.2 \text{ g} \pm 79.5 \text{ g}$ calculated wet weight, or $27.2 \text{ g} \pm 17.3 \text{ g}$ dry weight. Female students showed less excretion of feces than males, i.e., from 29.1 g-wet weight (9.4 g-dry) to 141.0 g-wet weight (37.8 g-dry), the average being $84.9 \pm 55.4 \text{ g}$ wet weight ($19.9 \pm 11.4 \text{ g}$ -dry). Thus daily excretion of feces varied

Table 1 Means and standard deviations of daily excretion in feces during 5 days

	DRY Wt (g)	WET Wt (g)	D/W (%)	Cd (μ g/g)	Pb (μ g/g)	Pb (μ g/day)	Cu (μ g/g)	Cu (mg/day)	Zn (μ g/g)	Zn (mg/day)
MALE										
1	30.0 $\pm$ 15.6	150.5 $\pm$ 59.3	19.9 $\pm$ 2.8	0.78 $\pm$ 0.54	1.2 $\pm$ 0.4	36 $\pm$ 11	37 $\pm$ 9	1.12 $\pm$ 0.51	275 $\pm$ 78	8.2 $\pm$ 3.6
2	47.7 $\pm$ 19.0	250.7 $\pm$ 48.8	19.0 $\pm$ 0.6	1.06 $\pm$ 0.33	0.9 $\pm$ 0.1	43 $\pm$ 21	38 $\pm$ 6	1.80 $\pm$ 0.85	229 $\pm$ 49	10.9 $\pm$ 2.4
3	18.6 $\pm$ 6.8	65.2 $\pm$ 37.4	28.8 $\pm$ 15.3	1.02 $\pm$ 0.43	19.2 $\pm$ 12.3	37 $\pm$ 18	75 $\pm$ 4	0.85 $\pm$ 0.32	322 $\pm$ 79	6.0 $\pm$ 2.2
4	10.6 $\pm$ 6.3	39.7 $\pm$ 32.4	26.6 $\pm$ 7.7	2.52 $\pm$ 0.37	26.7 $\pm$ 12.6	20 $\pm$ 16	48 $\pm$ 4	0.83 $\pm$ 0.57	487 $\pm$ 66	5.1 $\pm$ 3.2
5	25.9 $\pm$ 10.0	140.9 $\pm$ 72.6	18.4 $\pm$ 5.6	1.04 $\pm$ 0.14	26.9 $\pm$ 12.0	42 $\pm$ 16	52 $\pm$ 17	1.34 $\pm$ 0.43	294 $\pm$ 118	7.6 $\pm$ 3.7
6	7.4 $\pm$ 3.6	31.3 $\pm$ 14.1	23.5 $\pm$ 6.7	1.47 $\pm$ 0.41	10.9 $\pm$ 6.6	11 $\pm$ 6	45 $\pm$ 8	0.84 $\pm$ 0.21	435 $\pm$ 66	3.2 $\pm$ 1.7
7	22.9 $\pm$ 13.7	87.2 $\pm$ 21.3	26.3 $\pm$ 2.1	0.80 $\pm$ 0.28	18.4 $\pm$ 13.2	36 $\pm$ 34	56 $\pm$ 19	1.29 $\pm$ 1.35	349 $\pm$ 89	8.0 $\pm$ 6.0
8	41.5 $\pm$ 23.6	190.1 $\pm$ 69.4	21.8 $\pm$ 7.5	1.20 $\pm$ 0.26	49.8 $\pm$ 24.3	59 $\pm$ 10	42 $\pm$ 5	1.75 $\pm$ 0.76	500 $\pm$ 106	20.7 $\pm$ 8.8
9	30.1 $\pm$ 12.1	131.1 $\pm$ 67.6	23.0 $\pm$ 3.5	1.39 $\pm$ 0.21	41.9 $\pm$ 12.7	54 $\pm$ 32	45 $\pm$ 15	0.84 $\pm$ 0.76	338 $\pm$ 61	10.8 $\pm$ 4.5
10	18.4 $\pm$ 13.1	98.4 $\pm$ 105.5	18.7 $\pm$ 7.6	0.88 $\pm$ 0.37	16.2 $\pm$ 19.0	33 $\pm$ 17	43 $\pm$ 20	0.80 $\pm$ 0.64	481 $\pm$ 917	8.9 $\pm$ 7.0
11	28.7 $\pm$ 11.1	83.1 $\pm$ 30.8	33.3 $\pm$ 4.4	1.48 $\pm$ 0.70	48.3 $\pm$ 12.1	76 $\pm$ 33	76 $\pm$ 31	2.19 $\pm$ 1.77	565 $\pm$ 138	16.2 $\pm$ 7.7
12	47.6 $\pm$ 22.1	190.8 $\pm$ 99.1	25.0 $\pm$ 4.3	1.51 $\pm$ 0.30	71.9 $\pm$ 44.9	81 $\pm$ 24	46 $\pm$ 9	2.19 $\pm$ 1.06	305 $\pm$ 46	14.5 $\pm$ 8.0
13	20.5 $\pm$ 2.9	66.5 $\pm$ 10.3	30.9 $\pm$ 1.4	1.22 $\pm$ 0.37	25.2 $\pm$ 8.6	28 $\pm$ 2	43 $\pm$ 13	0.84 $\pm$ 0.27	479 $\pm$ 81	9.9 $\pm$ 1.7
14	19.2 $\pm$ 9.0	59.7 $\pm$ 27.7	32.1 $\pm$ 1.4	1.79 $\pm$ 0.19	34.3 $\pm$ 17.5	25 $\pm$ 9	51 $\pm$ 10	0.97 $\pm$ 0.31	423 $\pm$ 106	8.1 $\pm$ 3.1
15	22.8 $\pm$ 16.4	113.7 $\pm$ 70.4	20.0 $\pm$ 3.8	1.05 $\pm$ 0.19	23.9 $\pm$ 18.3	73 $\pm$ 42	44 $\pm$ 8	1.01 $\pm$ 0.67	250 $\pm$ 102	5.7 $\pm$ 3.0
16	37.6 $\pm$ 27.5	126.4 $\pm$ 122.0	29.7 $\pm$ 2.6	1.73 $\pm$ 0.65	65.0 $\pm$ 85.0	42 $\pm$ 5	56 $\pm$ 14	2.12 $\pm$ 1.35	408 $\pm$ 87	15.3 $\pm$ 9.0
17	39.6 $\pm$ 7.5	192.5 $\pm$ 53.4	20.6 $\pm$ 2.6	0.95 $\pm$ 0.28	37.6 $\pm$ 16.1	42 $\pm$ 5	35 $\pm$ 5	1.37 $\pm$ 0.40	432 $\pm$ 117	17.1 $\pm$ 4.6
18	33.1 $\pm$ 3.7	157.5 $\pm$ 40.9	21.0 $\pm$ 5.7	1.40 $\pm$ 0.56	46.3 $\pm$ 21.4	40 $\pm$ 10	49 $\pm$ 7	1.63 $\pm$ 0.29	326 $\pm$ 104	10.8 $\pm$ 2.9
19	13.7 $\pm$ 12.6	64.7 $\pm$ 72.9	21.2 $\pm$ 7.2	2.07 $\pm$ 0.61	28.4 $\pm$ 33.4	29 $\pm$ 15	46 $\pm$ 11	0.63 $\pm$ 0.60	431 $\pm$ 149	5.9 $\pm$ 2.8
average	27.2 $\pm$ 17.3	116.2 $\pm$ 79.5	23.4 $\pm$ 7.3	1.33 $\pm$ 0.48	36.2 $\pm$ 31.2	45 $\pm$ 26	49 $\pm$ 16	1.33 $\pm$ 0.91	384 $\pm$ 142	10.4 $\pm$ 6.5
FEMALE										
1	19.1 $\pm$ 10.2	84.9 $\pm$ 56.8	22.5 $\pm$ 4.2	1.13 $\pm$ 0.25	22.6 $\pm$ 9.7	22 $\pm$ 11	43 $\pm$ 16	0.82 $\pm$ 0.46	274 $\pm$ 85	5.2 $\pm$ 3.6
2	21.8 $\pm$ 14.1	138.4 $\pm$ 82.9	15.8 $\pm$ 5.9	1.24 $\pm$ 0.36	27.1 $\pm$ 22.1	32 $\pm$ 30	31 $\pm$ 17	0.67 $\pm$ 0.47	239 $\pm$ 126	5.2 $\pm$ 3.7
3	19.3 $\pm$ 10.1	72.2 $\pm$ 35.3	26.7 $\pm$ 5.7	0.87 $\pm$ 0.18	16.8 $\pm$ 8.5	20 $\pm$ 9	36 $\pm$ 5	0.70 $\pm$ 0.27	272 $\pm$ 43	5.6 $\pm$ 3.7
4	14.8 $\pm$ 6.4	68.1 $\pm$ 33.5	21.7 $\pm$ 4.6	1.09 $\pm$ 0.57	16.1 $\pm$ 13.7	19 $\pm$ 8	41 $\pm$ 8	0.61 $\pm$ 0.26	379 $\pm$ 95	5.6 $\pm$ 1.9
5	21.1 $\pm$ 11.4	75.4 $\pm$ 44.0	28.1 $\pm$ 2.6	1.45 $\pm$ 0.27	30.7 $\pm$ 20.0	30 $\pm$ 22	65 $\pm$ 9	1.38 $\pm$ 0.68	416 $\pm$ 79	8.8 $\pm$ 6.0
6	16.7 $\pm$ 3.9	77.7 $\pm$ 8.4	21.5 $\pm$ 6.5	1.33 $\pm$ 0.82	23.1 $\pm$ 14.6	31 $\pm$ 14	55 $\pm$ 32	0.92 $\pm$ 0.71	557 $\pm$ 481	9.3 $\pm$ 10.0
7	20.4 $\pm$ 12.1	64.8 $\pm$ 45.5	31.6 $\pm$ 5.5	1.33 $\pm$ 0.22	27.2 $\pm$ 12.8	38 $\pm$ 25	61 $\pm$ 8	1.23 $\pm$ 0.71	426 $\pm$ 101	8.7 $\pm$ 5.4
8	19.1 $\pm$ 13.8	61.4 $\pm$ 47.1	31.2 $\pm$ 5.1	1.22 $\pm$ 0.34	23.5 $\pm$ 20.1	33 $\pm$ 27	63 $\pm$ 16	2.00 $\pm$ 0.70	398 $\pm$ 51	7.6 $\pm$ 5.5
9	23.4 $\pm$ 9.1	74.6 $\pm$ 31.7	31.4 $\pm$ 3.6	1.70 $\pm$ 0.76	39.9 $\pm$ 38.8	39 $\pm$ 12	64 $\pm$ 14	1.51 $\pm$ 0.91	860 $\pm$ 244	20.1 $\pm$ 7.3
10	13.8 $\pm$ 6.7	61.8 $\pm$ 32.1	22.4 $\pm$ 6.5	1.17 $\pm$ 0.35	16.2 $\pm$ 8.2	16 $\pm$ 9	39 $\pm$ 9	0.54 $\pm$ 0.25	365 $\pm$ 164	5.0 $\pm$ 1.3
11	20.3 $\pm$ 10.7	103.8 $\pm$ 74.5	19.5 $\pm$ 6.9	0.99 $\pm$ 0.14	20.1 $\pm$ 8.9	30 $\pm$ 10	38 $\pm$ 12	0.76 $\pm$ 0.27	297 $\pm$ 80	6.0 $\pm$ 2.4
12	18.2 $\pm$ 10.0	74.4 $\pm$ 60.2	24.5 $\pm$ 13.4	0.73 $\pm$ 0.35	13.3 $\pm$ 14.2	29 $\pm$ 15	43 $\pm$ 5	0.79 $\pm$ 0.47	374 $\pm$ 73	6.8 $\pm$ 3.3
13	9.4 $\pm$ 2.6	29.1 $\pm$ 7.5	32.5 $\pm$ 2.7	1.46 $\pm$ 0.54	13.8 $\pm$ 8.0	14 $\pm$ 7	57 $\pm$ 4	0.53 $\pm$ 0.14	355 $\pm$ 81	3.4 $\pm$ 1.1
14	16.2 $\pm$ 11.7	75.9 $\pm$ 46.5	21.3 $\pm$ 2.9	0.71 $\pm$ 0.13	11.5 $\pm$ 10.8	20 $\pm$ 20	46 $\pm$ 7	0.74 $\pm$ 0.63	287 $\pm$ 29	4.6 $\pm$ 3.1
15	37.8 $\pm$ 14.2	141.0 $\pm$ 59.5	26.8 $\pm$ 4.2	1.33 $\pm$ 0.45	50.3 $\pm$ 24.9	61 $\pm$ 39	44 $\pm$ 10	1.65 $\pm$ 0.65	413 $\pm$ 287	15.6 $\pm$ 14.2
16	20.3 $\pm$ 17.1	101.0 $\pm$ 73.7	20.1 $\pm$ 2.5	1.05 $\pm$ 0.10	21.4 $\pm$ 15.0	17 $\pm$ 22	30 $\pm$ 4	0.61 $\pm$ 0.54	221 $\pm$ 42	4.5 $\pm$ 4.8
17	23.0 $\pm$ 10.5	137.0 $\pm$ 66.2	19.0 $\pm$ 4.5	1.65 $\pm$ 0.13	42.9 $\pm$ 14.7	35 $\pm$ 22	45 $\pm$ 5	1.18 $\pm$ 0.48	351 $\pm$ 149	9.1 $\pm$ 3.0
average	19.9 $\pm$ 11.4	84.9 $\pm$ 55.4	23.4 $\pm$ 7.4	1.20 $\pm$ 0.45	23.9 $\pm$ 18.4	29 $\pm$ 22	47 $\pm$ 16	0.93 $\pm$ 0.59	376 $\pm$ 207	7.5 $\pm$ 6.4

greatly according to sex and individual. But average ratio of dry weight to wet weight indicated the same value in males and females. This means that water contents of feces by sex did not differ and were three-quarters or four-fifths of the feces.

As shown in Table 1, average daily fecal excretions of Cd, Pb, Cu and Zn for 5 days in male students were $36.2 \pm 31.2 \mu\text{g}$, $45 \pm 26 \mu\text{g}$, $1.33 \pm 0.91 \text{ mg}$ and $10.4 \pm 6.5 \text{ mg}$, respectively. Those in female students were $23.9 \pm 18.4 \mu\text{g}$, $29 \pm 22 \mu\text{g}$, $0.93 \pm 0.59 \text{ mg}$ and $7.5 \pm 6.4 \text{ mg}$, respectively. Since the concentrations of the 4 metals in both sexes showed similar levels, the differences in the fecal excretion of the metals between the male and the female students are considered to depend on the different amounts of feces.

Food study

Table 2 shows the results of food analyses. The average intake of Cd was $34.8 \pm 15.6 \mu\text{g}$, $60 \pm 15 \mu\text{g}$ for Pb, $0.91 \pm 0.42 \text{ mg}$ for Cu and $10.2 \pm 4.7 \text{ mg}$ for Zn per day. The data of the feces study are also presented in Table 2. It is interesting to note that each metal in daily food indicated amounts almost equal to that in fecal output, in spite of the fact that the food was collected from the hospital and feces from medical or nursing student who did not eat hospital diets.

Table 2
Average dietary intake and fecal excretion of 4 metals

	FOOD	FECES	
	(n=20)	MALE (n=91)	FEMALE (n=81)
DRY WEIGHT (g)	527.5 ± 130.4	27.2 ± 17.3	19.9 ± 11.4
WET WEIGHT (g)	2538.0 ± 100.1	116.2 ± 79.5	84.9 ± 55.4
D/W RATIO (%)	20.8 ± 5.1	23.4 ± 7.3	23.4 ± 7.4
CADMIUM (ppm)	0.07 ± 0.03	1.33 ± 0.58	1.20 ± 0.45
($\mu\text{g}/\text{day}$)	34.8 ± 15.6	36.2 ± 31.2	23.9 ± 18.4
LEAD (ppm)	0.13 ± 0.05	1.6 ± 0.6	1.4 ± 0.5
($\mu\text{g}/\text{day}$)	60 ± 15	45 ± 26	29 ± 22
COPPER (ppm)	1.7 ± 0.7	49 ± 16	47 ± 16
(mg/day)	0.91 ± 0.42	1.33 ± 0.91	0.93 ± 0.59
ZINC (ppm)	19 ± 7	384 ± 142	376 ± 207
(mg/day)	10.2 ± 4.7	10.4 ± 6.5	7.5 ± 6.4

Organ study

Figure 1 shows the concentrations of 4 metals in the renal cortex of males and females by age. Cadmium concentration increases sharply during 0–19 years, and gradually rises to maximum concentration between 40 and 50 years of age. Afterwards the concentration of Cd decreases by age. In the age group 0–9, Zn in the renal cortex shows much higher concentrations than any of the other metals, but Cd concentrations approach similar levels in the thirties. Both copper and lead maintain almost steady levels through all ages, although copper levels are much higher than lead.

Figure 2 shows the trends of the 4 metals in the liver. Copper indicates a different pattern from that of other metals, i.e., it decreases slightly according to age, whereas the other metals increase. The concentration of copper in the liver is two or three times higher than that in the renal cortex. Cadmium concentration in the renal cortex, on the other hand, indicates about ten times higher

Fig. 1 Concentrations of Cd, Pb, Cu and Zn in the renal cortex.

Fig. 2 Concentrations of Cd, Pb, Cu and Zn in the Liver.

than that in liver. Zinc and lead in the liver indicate the same levels as those in the renal cortex.

Figure 3 shows the Cd/Cu ratio in food, feces, liver and renal cortex of both males and females in their twenties. Although the results of these comparative studies are shown in Figures 3-6, it is important to remember that the materials were taken from different individuals. Nevertheless, the Cd/Cu ratio in food is almost equal to that in feces, the ratio in the liver is ten times higher and that in the renal cortex is about 100 times higher than that in food or feces.

Figure 4 also indicates the Cd/Zn ratio in the three media studied. The ratios in food and feces are equal to each other, and this level is 10 times lower than in the liver and 100 times lower than in the renal cortex. In comparing the Cd/Zn ratio with the Cd/Cu ratio, the former in all three media is 10 times lower than the latter. Figures 5 and 6 show the Pb/Cu and Pb/Zn ratio in the three media. The levels of Pb/Cu ratios in all three media, are 10 times higher than those of Pb/Zn ratios.

Fig. 3 Cd/Cu ratio in food, feces and organs.

Table 3
Correlations in food (n=20)

	Cd	Pb	Cu	Zn
Cd	—	.12	.29	-.03
Pb	—	—	.21	.02
Cu	—	—	—	.34
Zn	—	—	—	—

Table 4
Correlations in feces (n=172)

	Cd	Pd	Cn	Zn
Cd	—	.15*	.23**	.13
Pd	—	—	.36**	.36**
Cu	—	—	—	.46**
Zn	—	—	—	—

*: $p \leq 0.05$, **: $p \leq 0.01$

Table 5
Correlations in the liver (n=85)

	Cd	Pd	Cu	Zn
Cd		.17	-.14	.33**
Pb	—		.10	.21**
Cu	—	—		.09
Zn	—	—	—	

**: $p \leq 0.01$

Table 6
Correlations in the renal cortex (n=85)

	Cd	Pb	Cu	Zn
Cd		.08	.52**	.37**
Pb	—		.12	.14
Cu	—	—		.38**
Zn	—	—	—	

**: $p \leq 0.01$

Fig. 4 Cd/Zn ratio in food, feces and organs.

Fig. 5 Pb/Cu ratio in food, feces and organs.

Tables 3-6 indicate the correlation matrices of metal pairs in feces, food and organs. The unit of correlation used was ppm-dry weight for the reasons stated below. Despite of the fact that a low correlation between Cd and Zn was found in feces ($r=0.13$), and that no correlation was found in food ($r=-0.03$), there were close correlations in the liver ($r=0.33$) and renal cortex ($r=0.37$). Significant correlations between Cu and Zn were found in food, feces and renal cortex, but not in the liver.

Analysis of variance in feces

Analysis of variance in feces was made in order to compare the variation among individuals and the variation within individuals. Table 7 shows the F-values of the fecal amounts and the concentrations of the four metals for both

Table 7
Analysis of variance of 4 metals feces

	MALE					FEMALE				
	TERM [#]	DF	SS	MS	F	TERM [#]	DF	SS	MS	F
AMOUNT of FECES	A	18	6589.55	366.09	1.346	A	16	931.05	58.19	0.388
	B	72	20645.84	286.75		B	64	9575.71	149.93	
	T	90	27235.39			T	80	10526.76		
CONC. of CADMIUM	A	18	21.923	1.218	3.567**	A	16	2.779	0.174	0.816
	B	72	24.586	0.342		B	64	13.623	0.213	
	T	90	46.509			T	80	16.402		
CONC. of LEAD	A	18	11.94	0.663	2.294*	A	16	3.79	0.237	0.921
	B	72	20.82	0.289		B	64	16.46	0.257	
	T	90	32.76			T	80	20.25		
CONC. of COPPER	A	18	7.21	0.400	1.791*	A	16	7.82	0.489	2.420**
	B	72	16.09	0.223		B	64	12.92	0.202	
	T	90	23.30			T	80	20.73		
CONC. of ZINC	A	18	1480.32	82.244	5.532**	A	16	1083.75	67.73	1.816*
	B	72	1306.55	18.15		B	64	2387.02	37.30	
	T	90	2786.87			T	80	3470.77		

#: A: variation within individuals, B: variation among individuals, T: total variation, DF: degree of freedom, SS: sum of square, MS: mean square, F: F-value

*: $P \leq 0.05$, **: $P \leq 0.01$

male and female students. Concerning the fecal amounts for each sex, no significant differences of F-value were shown, indicating that variation within one individual over a five-day period does not differ from variation among individuals.

In the analysis of variance for the concentrations of all four metals among male students, F-value was significant. This indicates the existence of variation among individuals rather than variation within each individual male, i.e., the concentrations of the metals in feces vary more from individual to individual than within any single individual.

Female students showed significant inter-individual variation in Cu and Zn concentrations, but not in Pb and Cd concentrations, i.e., the variation of Pb and Cd concentrations within an individual does not differ so much from inter-individual variation. To explain this, except for two students, the female students were living in a dormitory at Keio University. They ate at the hospital cafeteria

or prepared and ate meals together in their rooms. This may partly explain the non-existence of significance in the variation among the females. In addition, the size of the samples may have been too small to show a significance.

Correlation matrix of four metals in feces

Table 8 shows the correlation matrix of pairs of the 4 metals in feces. Units of comparison for the metals were expressed as $\mu\text{g/g}$ -dry weight (ppm-dry). Tables 9 and 10 show the same matrix as Table 8 with the units expressed in $\mu\text{g/g}$ -wet weight (ppm-wet) and $\mu\text{g/day}$, respectively. It is interesting to note that the correlation coefficients expressed as ppm-wet are higher than those expressed as ppm-dry, and moreover that the highest correlations are found in the matrix described in daily amounts. For example, the correlation between Cd and Cu indicates $r=0.23$, $r=0.57$ in ppm-wet and $r=0.72$ in daily amount. With regard to the correlations which change the degree of coefficients according to the expression of three units, the following discussion can be made: a higher

Table 8
Correlations in feces (ppm-dry)

	Cd	Pb	Cu	Zn
Cd		.15*	.23**	.13
Pb	—		.36**	.36**
Cu	—	—		.46**
Zn	—	—	—	

*: $p \leq 0.05$

** : $p \leq 0.01$

Table 9
Correlations in feces (ppm-wet)

	Cd	Pb	Cu	Zn
Cd		.47**	.57**	.47**
Pb	—		.66**	.52**
Cu	—	—		.71**
Zn	—	—	—	

** : $p \leq 0.01$

Table 10
Correlations in feces (daily amount)

	Cd	Pb	Cu	Zn
Cd		.72**	.72**	.63**
Pb	—		.83**	.75**
Cu	—	—		.76**
Zn	—	—	—	

** : $p \leq 0.01$

Fig. 6 Pb/Zn ratio in food, feces and organs.

correlation expressed as "wet weight" rather than as "dry weight" is most likely influenced by water contents of feces. The highest correlation observed for "total daily intake" may possibly be influenced by both water content and amount of feces. Thus, in order to exclude these two factors and possibly other unknown factors, the author believes that "dry weight" is more appropriate for the calculation of the correlation coefficient between two metal concentrations in feces. However, it is important to conclude that the interrelationships among Cd, Pb, Cu and Zn are very close in feces as well as food.

DISCUSSION

Daily intake of the metals

Estimation of average total daily intakes of metals from food and beverages by human adults have been recently reported. As for cadmium excretion in

feces, Tati and co-workers,¹⁰ reported a range of 41.1 to 79.4 $\mu\text{g/day}$ for male students. Suzuki *et al.*,¹¹ found it to be $35.92 \pm 10.02 \mu\text{g/day}$ and $35.95 \pm 6.68 \mu\text{g/day}$ from two adult subjects. Fukushima¹² obtained Cd values from food analyses of 44.8–84.2 $\mu\text{g/day}$. From these results, the estimated average in Japan lies between 30–60 $\mu\text{g/day}$ of cadmium. There are many reports from other countries. Friberg *et al.*⁶ suggested that 50 μg of Cd may be ingested in most countries, with a probable range of 25 to 75 $\mu\text{g/day}$. Schroeder and Tipton⁷ estimated the average total lead in food in the United States to be 280 $\mu\text{g/day}$. Horiuchi¹³ gave a range of 239–318 $\mu\text{g Pb/day}$ for Japanese adults. A range of 100–500 $\mu\text{g/day}$ is generally accepted in the world as the daily intake of lead. As for daily Cu intake, Underwood¹⁴ noted that most western-style diets supply adults with 2–4 mg/day, while in Japan, Yamagata¹⁵ estimated 0.78–2.54 mg Cu/day. As for zinc, Sandstead *et al.*¹⁶ and Schroeder¹⁷ reported that adult diets in U.S. supply 12–15 mg/day. Osis *et al.*,¹⁸ from a comparative study of two types of hospital diets, noted a range of 7.0–16.3 mg Zn/day. They also stated that daily intake was greatly influenced by both the choice and source of food.

In this study, somewhat lower daily excretions of Cd, Pb, Cu and Zn have been observed than have been reported previously in Japan. These low intakes are consistent with the levels of these metals in hospital diets, although the feces samples were obtained from students who had not eaten hospital diets.

Interaction of the metals

Concerning interaction of the metals in organs or tissues, many reports on animal experiments have been presented. For instance, Petering and co-workers¹⁹ found in rats that Zn-insufficient food and water with low concentration of Cd caused a decrease in the Zn concentration in testes, but such was not found in the case of Zn-rich diets with low Cd. Schroeder *et al.*²⁰ reported that exposure to Cd increased Zn concentration in the liver and kidney. It has also been observed that Zn has a protective effect against Cd because of the rapid accumulation of Cd in the liver and kidney as a metal-binding protein called metallothionein.¹⁴

In human beings, Schroeder *et al.*¹⁷ found the Cd/Zn ratio in the renal cortex to be 0.6–0.7 for those 35–55 years of age, and Piscator and Lind² found it to be 0.73 for those up to 50 years of age. In this study, as shown in Figure 4, it was 0.49 among those 20–29 years of age. The trend of the Cd/Zn ratio in the renal cortex according to age is discussed elsewhere.²¹ The ratios of Cd/Zn as well as Cd/Cu in food and feces were almost the same, which suggests that the absorption rate in humans is constant. However, the ratios of these metals in

the liver and the renal cortex indicate different distribution.

As shown in Tables 3-6, Cd-Zn relationship becomes significant in the liver and renal cortex, but not in food or feces. On the other hand, there is close Cd-Cu relationship in food, feces and the renal cortex but not in the liver. This discrepancy can not be explained simply by the distribution of a single metal in an organ even though Cu seems to have special affinity to liver cells.¹⁴ The affinity of a metal to an organ is subject to change when another metal is distributed to that organ. In a cadmium polluted area, Zn as well as Cd in food and feces showed higher concentrations than in control area.²¹ When both Zn and Cd levels in food ingested are higher than normal values, the critical concentration of Cd in the renal cortex may be altered since both Cd and Zn have equal ability to produce metallothionein in the liver, resulting in interaction between the two metals. Finally, further research of metal interactions in humans are required for a better understanding of the toxico-kinetics of heavy metals.

CONCLUSION

1. The daily excretion in feces Cd, Pb, Cu and Zn in 19 medical students and 17 nursing students was slightly lower than those reported in other Japanese studies. The daily intake of the 4 metals estimated in regular hospital diets agreed fairly well with the daily excretion, especially among the medical students (all male).
2. In male students the concentrations of the metals (ppm-dry weight) in feces varied more among individuals than within individuals.
3. The heavy metals seem to have almost the same absorption rate, since the proportion of each metal in feces did not differ greatly from that in the hospital food. However, once absorbed the relationship among metals and/or distribution in organs change markedly. This phenomenon is caused not only by the affinity of a single metal for a specific organ, but also by interactions among metals.
4. More studies on living humans and post mortem studies are required in the future to observe these relationships among heavy metals in various media, especially in areas polluted by more than one metal.

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REFERENCES

1. Tipton, I. H.: The distribution of trace metals in the human body. In: Metal-binding in medicine. Seven, M. J. and Johnson, L. A., eds., Lippincott, Phil., 27-34, 1960
2. Piscator, M. and Lind, B.: Cadmium, zinc, copper and lead in human renal cortex. *Arch. Environ. Hlth.* 24: 426-431, 1972
3. Hammer, D. I., Calocci, A. V., Hasselblad, V., Williams, M. E. and Pinkerson, C.: Cadmium and lead in autopsy tissues. *J. Occup. Med.* 15: 956-963, 1973
4. Sumino, K., Hayakawa, K., Shibata, T. and Kitamura, S.: Heavy metals in normal human Japanese tissues. *Arch. Environ. Hlth.* 30: 487-494, 1975
5. Tsuchiya, K., Seki, Y. and Sugita, M.: Cadmium concentrations in the organs and tissues of cadavers from accidental death. *Keio J. Med.* 25: 83-90, 1976
6. Friberg, L., Piscator, M., Nordberg, G. and Kjellström, T.: Cadmium in the Environment. 2nd ed., CRC Press, Cleveland, 1974
7. Schroeder, H. A. and Tipton, L.: The human body burden of lead. *Arch. Environ. Hlth.* 17: 965-978, 1968
8. Magos, L.: The role of synergism and antagonism in the toxicology of metals. In: Effects and dose-response relationships of toxic metals. Nordberg, G. F. ed., Elsevier, Amsterdam, 1976, 491-497
9. Parizek, J.: Interrelationships among trace elements. In: Effects and dose-response relationships of toxic metals. Nordberg, G. F. ed., Elsevier, Amsterdam, 1976, 498-510
10. Tati, M., Katagiri, Y. and Kawai, M.: Urinary and fecal excretion of cadmium in normal Japanese: An approach to non-toxic levels of cadmium. In: Effects and dose-response relationships of toxic metals. Nordberg, G. F. ed., Elsevier, Amsterdam, 1976, 331-342
11. Suzuki, S. and Lu, C.: A balance study of cadmium, An estimation of daily input, output and retained amount in two subjects. *Ind. Hlth.* 14: 53-65, 1976
12. Fukushima, M.: Cadmium content in various foodstuffs. *Environmental Health Report (Kankyo Hoken Report)* 11: 22-25, 1972 (in Japanese)
13. Horiuchi, K.: Lead in the environment and its effects on man in Japan. *Osaka City Med. J.* 16: 1-28, 1970
14. Underwood, E. J.: Trace elements in human and animal nutrition. 4th ed., Academic Press, New York, 1977
15. Yamagata, N.: Copper concentration in food and daily intake in man. *Biryo Genso, Sangyo Tosho*, Tokyo, 1977, 188-189 (in Japanese)
16. Sandstead, H. H., Prasad, A. S., Schlert, A. R., Farid, Z., Miale, A. Jr., Bassily, S. and Darby, W. J.: Human zinc deficiency, endocrine manifestations and response to treatment. *Am. J. Clin. Nutr.* 20: 422-442, 1967
17. Schroeder, H. A., Nason, A. P., Tipton, I. H. and Balassa, J. J.: Essential trace elements in Man; zinc: Relation to environmental cadmium. *J. Chron. Dis.* 20: 179-210, 1967
18. Osis, D., Kramer, L., Wiatrowski, E. and Spencer, H.: Daily zinc intake in man. *Am. J. Clin. Nutr.* 25: 582-588, 1972
19. Petering, H. G., Johnson, M. A. and Stemmer, K. L.: Studies of zinc metabolism in the rat. I, dose-response effects of cadmium. *Arch. Environ. Hlth.* 23: 93-101, 1971
20. Schroeder, H. A. and Nason, A. P.: Interactions of trace metals in rat tissues, cadmium and nickel with zinc, chromium, copper and manganese. *J. Nutr.* 104: 167-178, 1974
21. Tsuchiya, K. and Iwao, S.: Interrelationships among zinc, copper, lead and cadmium in food, feces and organs of humans. *Environ. Health Perspect.* (to be published)