Cadmium: In vivo Measurement in Smokers and Nonsmokers

Abstract. *Absolute amounts of cadmium (in milligrams) in the left kidney and concentrations of cadmium (micrograms per gram) in the liver were measured in vivo in 20 healthy adult male volunteers. Organ cadmium levels of smokers were significantly elevated above those of nonsmokers. No relationship was evident between body stores of cadmium (liver and kidney) and cadmium or β_2-microglobulin in urine or blood. The average total body burden of cadmium in man at age 50 is estimated to be 19.3 milligrams for nonsmokers and 35.5 milligrams for smokers (38.7 pack-year smoking history). Biological half-time for the whole body was, on average, 15.7 years (10- to 33-year range). Dietary absorption was 2.7 micrograms per day. Cigarette smoking resulted in the absorption of 1.9 micrograms per pack.*

Short- and long-term effects of occupational exposure to cadmium have been observed for many years (*1*). The general population, however, is also exposed to cadmium. Combustion of coal and oil contributes to the level of cadmium in the environment, and it also appears that solar energy technology may add to the contamination. Trace amounts are present in air, water, food, and tobacco. In the nonindustrialized areas, dietary intake and smoking are the major routes of exposure.

Overt pulmonary and renal dysfunction has been observed in workers industrially exposed to cadmium (*1-3*). The potential consequences of increased lifetime exposures to low levels of contamination in the general population are not known. However, autopsy studies have revealed increased cadmium levels in individuals who had emphysema (*4*), hypertension (*5*), and Itai-Itai disease (*6*).

For a study of the long-term effects on humans of low levels of cadmium and of the possible role of cadmium in chronic diseases, it is essential that accurate data on existing levels and distributions of cadmium body burden be available. These data are necessary for the formulation of dose-response relationships at low levels of exposure, and also to serve as reference data for monitoring changes in the body burden of future populations.

Data on cadmium in the tissues in man have been derived primarily from autopsy studies; at age 50, an American has an estimated average body burden of 30 mg (*1, 7*). Cadmium distribution in the body is nonuniform, approximately half being located in the kidneys and the liver. The kidneys retain the largest absolute amount and also have the highest concentration. The high degree of localization within these organs has made possible direct in vivo measurements of cadmium in man (*8, 9*).

The partial body neutron activation (PBNA) technique is brief and noninvasive, and it carries minimal risk (localized dose of 0.067 rad). It is based on the specific nuclear properties of ^{113}Cd, a naturally occurring stable isotope (12.2 percent abundance). This isotope has a high probability (20,000 barns) of capturing thermal neutrons. In the capture process, excited ^{114}Cd is produced which, in turn, promptly decays ($<10^{-14}$ second) to the ground state. This de-excitation is accomplished by the emission of a cascade of gamma rays, which are detected externally to the body. As the gamma rays are emitted promptly after neutron capture, the subject must be irradiated and counted simultaneously (Fig. 1). Prior to the irradiation, the organs are accurately located by means of an ultrasonic scanner. Present detection limits are 2.5 mg for the kidney, and 1.5 μg/g for the liver (*8*).

The cadmium content of the liver and left kidney of 20 healthy male volunteers was measured by PBNA. Levels in the urine and blood plasma were measured by graphite furnace atomic absorption spectrophotometry preceded by wet ashing (*10*); β_2-microglobulin levels were measured by a radioimmunoassay technique (*11*). Analysis of variance (*12*) for correlations among organ content, blood and urine concentration, and β_2-microglobulin data showed a significant rela-

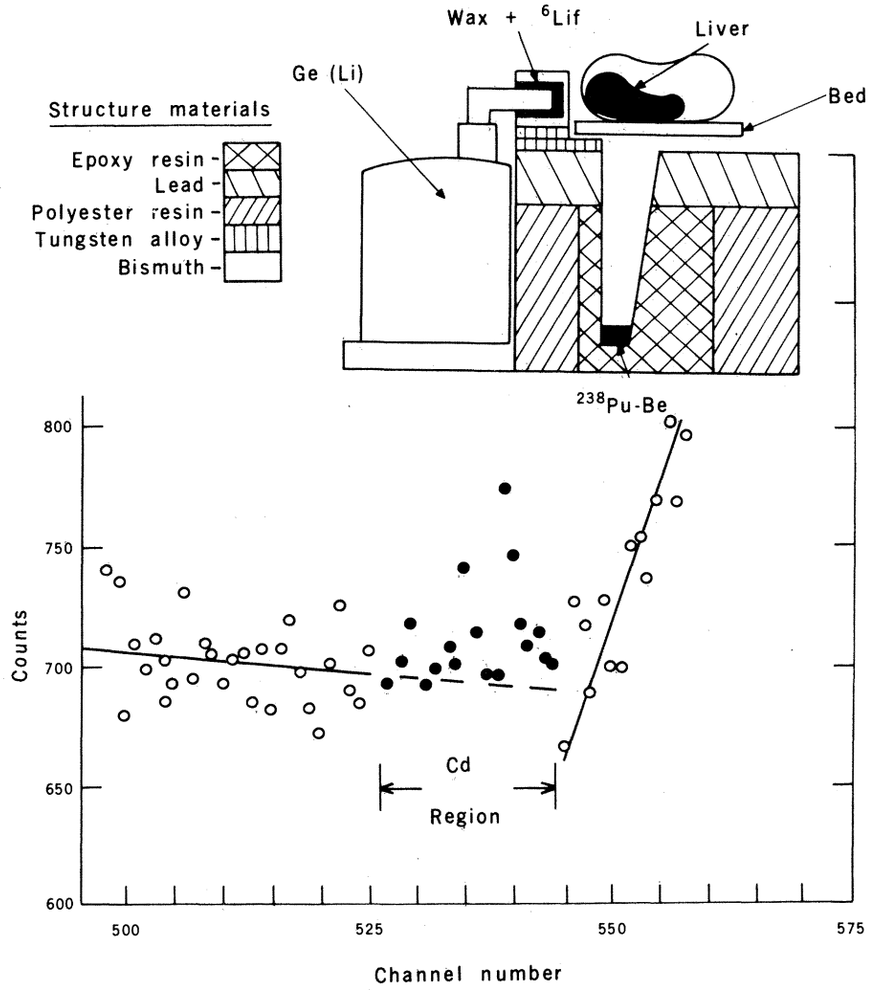

Fig. 1. At the top of the figure is a diagram of the experimental setup for in vivo measurement of liver cadmium in man. The subject is positioned over the collimated neutron beam (^{238}Pu-Be neutron source). Two large-volume Ge(Li) detectors are positioned to the side of the subject. Extensive shielding is provided to prevent the detectors from directly viewing the neutron source. The lower portion of the figure shows a gamma ray spectrum of cadmium in the left kidney of a smoker (age 52). The counts are for a single detector and represent approximately 4.1 mg of cadmium.

0036-8075/79/0720-0323$00.50/0

Table 1. Physical characteristics and cadmium data for 20 healthy male volunteers.

Parameter	All volunteers ($N = 20$)	Nonsmokers ($N = 8$)	Smokers ($N = 12$)	P
Age*	51 ± 9	52 ± 9	50 ± 9	N.S.†
Weight* (kg)	81.2 ± 16.5	83.5 ± 15.5	78.5 ± 17.0	N.S.
Height* (cm)	174 ± 9	175 ± 8	173 ± 10	N.S.
Plasma cadmium* (μg/liter)	2.2 ± 1.0	1.6 ± 0.8	2.5 ± 1.1	N.S.
Urinary cadmium* (μg/liter)	2.3 ± 1.2	1.7 ± 1.0	2.7 ± 1.3	N.S.
Plasma β_2-microglobulin* (mg/liter)	1.6 ± 0.5	1.7 ± 0.2	1.5 ± 0.6	N.S.
Urinary β_2-microglobulin* (μg/liter)	91 ± 86	50 ± 38	119 ± 127	N.S.
Kidney cadmium‡ (mg)	4.5 (1.9)	3.1 (2.0)	5.8 (1.7)	0.05
Liver cadmium‡ (μg/g)	2.9 (1.6)	2.3 (1.6)	4.1 (1.6)	0.05
Total body cadmium‡ (mg)	28.5 (1.6)	19.3 (1.5)	35.5 (1.6)	0.01
Smoking index* (pack-years)			38.7 ± 17	
Years of smoking*			27 ± 8	

*Arithmetic mean ± standard deviation. †N.S. = not significant ($P < .05$), t-test. ‡Geometric mean (standard deviation).

Table 2. Dietary and inhalation absorption rates based on kidney and liver cadmium data. The data for body burden are expressed as geometric mean (standard deviation); the numbers in parentheses under biological half-time represent the range; and the time of exposure is given as the arithmetic mean ± standard deviation.

Daily source of cadmium	Body burden (mg)	Biological half-time* (years)	Time of exposure (years)	Absorption rate
Diet	19.3 (1.6)	14.7 (10 to 37)	52 ± 9	2.7 g/day(< 4 percent)†
Cigarettes	16.2 (1.6)‡	16.5 (11 to 33)	27 ± 8§	1.9 g/pack(47 to 95 percent)‖

*$t_{1/2}$ = 14.7 years for nonsmokers, $t_{1/2}$ = 16.5 year for smokers. The range of values is also given. †Daily diet contains 75 to 100 μg of cadmium. ‡Additional body burden of smokers = 35.5 to 19.3 mg. §Average number of years of smoking cigarettes. ‖Inhaled cigarette smoke contains 2 to 4 μg per pack.

tion only between kidney and liver cadmium levels ($r = .6, P < .01$). Although the urinary excretion of cadmium was elevated when kidney content was high (on a group basis), the scatter of the data was too large to permit the kidney burden to be predicted for an individual. No relation was found between the level of cadmium in the blood and that in kidney or liver.

Since cigarettes contain approximately 2 μg of cadmium each (*13*), it was of interest to consider the body burden of smokers and nonsmokers separately (Table 1). The pack-year index, defined as number of packs of cigarettes smoked per day times the number of years of smoking, was used to estimate the cadmium exposure due to cigarette smoking (*14*). The smokers had a mean smoking index of 38.7 pack-years, with an average daily consumption of 1.4 packs. Although the mean values for the organs, urine, and β_2-microglobulin were higher for the smokers (Table 1), only the kidney and liver levels of cadmium were significantly elevated (t-test). The amount of cadmium in these tissues of cigarette smokers was, on average, approximately double that of nonsmokers. In this study, the average increment in smokers was 2.7 mg for the kidney and 1.8 μg/g for the liver. When adjusted for smoking history, the increase per pack-year was 0.112 ± 0.05 mg for the kidney and 0.077 ± 0.065 μg/g for the liver. The smokers increased their body burdens by 680 μg per pack-year above the amount of cadmium accumulated through dietary ingestion.

This report presents not only the first direct in vivo measurements of kidney and liver content of cadmium in man, but also the first data for a nonoccupationally exposed population. Data from the group under study demonstrate significant differences for organ cadmium levels between smokers and nonsmokers. The group was not, however, sufficiently large to support analysis of age-related variations. The data suggest that an adult American male nonsmoker (age 52) has a total body burden of approximately 19 mg of cadmium (assuming that kidneys and liver represent 50 percent of the total body burden). A smoker (age 50, with a smoking history of 38.7 pack-years) may have an average total body burden of 35 mg of cadmium. Thus, on the basis of these limited data, it appears that cigarette smoking may double the body burden of cadmium in man.

Once body burdens of cadmium are known, the biological variables of whole-body half-time, dietary absorption, and inhalation absorption from cigarettes can be calculated. A biological half-time of 15.7 years (range 10 to 33 years) for the whole body was derived from total body burden data and daily cadmium excretion data (*15*). The amount of cadmium retained by dietary ingestion is 2.7 μg/day (*16*). If the dietary cadmium intake is 75 to 100 μg/day, then the gastrointestinal absorption would be less than 4 percent (Table 2). Similar calculations based on the additional body burden of 16.2 mg for smokers in this study imply an inhalation dose of 1.9 μg per pack (*17*). Using autopsy samples, Lewis *et al.* (*18*) estimated the absorbed dose of cadmium at 1.4 μg per pack. Conversion of these values to a percent absorption is more complicated than in the case of dietary retention. Any estimate must be based, in part, on animal data and the use of smoking machines. Friberg *et al.* (*1*) estimated that 2 to 4 μg of cadmium is inhaled from smoking one pack of 20 cigarettes. Extrapolation of values from Menden *et al.* (*13*) would lead to 2.0 to 2.4 μg per pack. Lewis *et al.* (*18*) report a range of 0.75 to 3 μg per pack. Our calculations (*19*) agree with those of Friberg *et al.* (*1*). Thus, if we assume that 2 to 4 μg of cadmium is in the inhaled smoke of one pack of cigarettes, the percentage absorption ranges from 47.5 to 95 percent.

In previous attempts to estimate the kidney and liver cadmium levels in living man have been based on indirect measurements of blood and urine cadmium levels. Our study indicates no significant relationship between blood or urine data and kidney or liver burden of cadmium. The in vivo measurement technique appears to be the most reliable means available for the determination of cadmium in the liver and kidney. The application of this technique to the study of patients with chronic diseases may serve to clarify the role of cadmium in these diseases.

KENNETH J. ELLIS, DAVID VARTSKY
ITALO ZANZI, STANTON H. COHN
Medical Department,
Brookhaven National Laboratory,
Upton, New York 11973
SEIICHI YASUMURA
Department of Physiology,
Downstate Medical Center,
Brooklyn, New York 11203

References and Notes

1. L. Friberg, M. Piscator, G. F. Nordberg, T. Kjellstrom, *Cadmium in the Environment* (CRC Press, Cleveland, Ohio, 1974).
2. L. Friberg, *Proc. Int. Cong. Ind. Med.* **9**, 641 (1948).
3. G. Kazantzis, F. I. Flynn, J. S. Spawazi, D. G. Trott, *Q. J. Med.* **32**, 165 (1963).
4. H. M. Perry, Jr., J. A. Pierce, R. N. Hirst, J. E. Purifoy, E. F. Perry, M. Cruz, *Trace Substances in Environmental Health* **VI**, 207 (1973).
5. H. A. Schroeder, *J. Chronic Dis.* **18**, 647 (1965).

6. K. Fulkerson and H. E. Goeller, Eds., *Cadmium: The Dissipated Element* (Oak Ridge National Laboratories, publication No. NSF EP 21, Oak Ridge, Tenn., 1973).
7. H. M. Perry, Jr., G. S. Thind, E. F. Perry, *Med. Clin. North Am.* **60**, 759 (1976).
8. D. Vartsky, K. J. Ellis, N. S. Chen, S. H. Cohn, *Phys. Med. Biol.* **22**, 1085 (1977).
9. J. S. McLellan, B. J. Thomas, J. H. Fremlin, *ibid.* **20**, 88 (1975).
10. E. F. Perry, S. R. Koityohann, H. M. Perry, Jr., *Clin. Chem.* **21**, 626 (1975); F. C. Wright and J. C. Riner, *At. Absorpt. Newsl.* **14**, 103 (1975). The assay utilized the method of additions, with each sample run in triplicate and each sample repeated. Additional samples from several subjects were collected over a period of 1 to 3 months and assayed as outlined above. In subjects with multiple samples, the individual samples were not statistically different from each other, hence an average was used for these subjects. The group means and standard deviations given in Table 1 are not significantly altered if any one sample or the mean value from multiple samples are used. The precision (reproducibility) for cadmium concentrations in the normal range (<3 μg/liter) was $\leq$ 10 percent, with a detection limit below 2 pg.
11. Phadebas β_2-Micro Test Kit (Pharmacia Diagnostics AB, Uppsala, Sweden).
12. Using all 20 subjects as a single group, all permutations of the liver, kidney, blood, urine, and β_2-microglobulin data were tested for possible correlations. Analysis of variance was performed using the SPSS computer package [N. H. Nie, Ed., *Statistical Package for the Social Sciences* (McGraw-Hill, New York, ed. 2, 1975)], in which the level of significance was set at $P < .05$. Since cadmium is considered to be a nonessential trace element, its accumulation in the body may follow a lognormal distribution [J. Aitchison and J. A. C. Brown, *The Lognormal Distribution* (Cambridge Univ. Press, Cambridge, 1976)]. Therefore, normal versus normal, normal versus lognormal, and lognormal versus lognormal correlations were performed. For statistical comparisons between smokers and nonsmokers, the t-test was used on either normal or lognormal distributions whenever appropriate. The level for a significant difference was set at $P < .05$. The results of the t-test are presented in Table 1.
13. E. E. Menden, V. J. Elia, L. W. Michael, H. G. Petering, *Environ. Sci. Technol.* **6**, 830 (1972).
14. Although the unit of "pack-years" has been used, we believe it to be inadequate since it assumes a linear response. For example, a smoking history of one pack per day for 30 years would be identical to a history of five packs per day for 6 years. Even if it were possible to determine the exact number of cigarettes smoked, individual variations such as brand of cigarette, inhalation pattern, and frequency are also important factors.
15. The rate of change of cadmium in the whole body is described by the equation $dC/dt = -\lambda C + R$, where λ = percentage loss per year, C = whole body burden of cadmium (milligrams), and R = yearly exposure (milligrams per year). The loss per year is λC where $\lambda = \ln 2/t_{1/2}$, $t_{1/2}$ = biological half-time for the whole body. Since urinary excretion represents the only significant means of loss from the total body, λC = urinary loss per day $\times$ 365 day/year. In the present study, urinary loss was 2.3 μg/liter $\times$ 1.5 liter/day $\times$ 365 day/year = 1.26 mg/year. Solving for $t_{1/2}$ gives $t_{1/2} = (\ln 2 \times 28.7)/1.26 = 15.7$ years. The variation of $\pm$ 1.2 μg/liter per day of urinary cadmium gives a range of 11 to 33 years for $t_{1/2}$. For nonsmokers, the calculation is $\lambda = (1.66$ μg/liter $\times$ 1.5 liter/day $\times$ 365 day/year)/19,300 μg = 0.047 per year or $t_{1/2} = 14.7$ years. For smokers, $\lambda = (2.72$ μg/liter $\times$ 1.5 liter/day $\times$ 365 day/year)/35,500 μg = 0.042 per year or $t_{1/2} =$ 16.5 years.
16. The rate of change of cadmium is $dC/dt = -\lambda C + R$. Solving the equation gives $R = (\lambda C)(1 - e^{-\lambda t})^{-1}$. In the case of the nonsmokers, R represents the dietary intake, $\lambda = 0.047$ per year, $C =$ 19.3 mg, and $t = 52$ years. Substitutions of these values gives $R = 993$ μg/year = 2.7 μg/day.
17. For smokers, C = additional body burden of 16.2 mg = 35.5 mg $-$ 19.3 mg. R is then the exposure rate due to cigarette smoking. For the present study, $\lambda = 0.042$ per year, $C = 16.2$ mg, and $t = 27$ years (average number of years smoking). Substitution of these values gives $R =$ 1003 μg/year = 2.75 μg/day. The present smokers, however, averaged 1.4 packs per day, thus the absorbed dose is 1.9 μg per pack.
18. G. P. Lewis, W. J. Jusko, L. L. Coughlin, *J. Chronic Dis.* **25**, 717 (1972).
19. Assuming 2 μg per cigarette (*13*) leads to 40 μg per pack. If only 2/3 to 3/4 of any cigarette is smoked and 70 percent of the cadmium is in the smoke, then 19 to 20 μg per pack may be inhaled. The mainstream smoke represents 10 to 20 percent of the total smoke. Thus, the inhaled smoke from one pack of cigarettes may contain 2 to 4 μg of cadmium.
20. We thank A. LoMonte for performing the atomic absorption measurements, J. Rothmann for assisting in the activation procedures, and D. Pion for preparing the manuscript. Supported by U.S. Department of Energy contract EY-76-C-02-0016.

10 October 1978

Ketone Bodies Are Selectively Used by Individual Brain Regions

Abstract. *Close study of 3-hydroxybutyrate uptake by brain suggests that its metabolism is limited by permeability. Furthermore, the permeability characteristics vary from region to region; areas known to have no blood-brain barrier show the highest rate of utilization. The results imply that rather than substitute fuels, ketone bodies should be considered supplements which partially supply specific areas but are incapable of supporting the entire energy requirement of all brain regions.*

Ketone bodies have been shown to be an alternative fuel of brain energy metabolism. Unlike glucose, they do not appear to be metabolized freely by all cerebral areas. Instead, their metabolism appears to be restricted by transport across the blood-brain barrier. This phenomenon may explain several observations and alter some of our current concepts.

In normal, well-nourished sedentary mammals the ketone bodies 3-hydroxybutyrate and acetoacetate are present in low circulating concentrations. Yet there are physiological and pathological circumstances under which their concentrations rise appreciably and they become an important respiratory fuel. In addition to elevation during fasting and diabetes, increased concentrations of ketone bodies occur during pregnancy, during prolonged exercise, in persons eating high-fat diets, in uremia, during the perinatal period, and during infancy (*1, 2*). A novel situation is the recent popularity of diets consisting almost entirely of fats and proteins, where progress is monitored not only by weight loss but by the presence of ketone bodies in the urine (*3*).

Late in the 19th century the detection of ketone bodies in blood and urine of diabetic patients, coupled with the discovery that ketosis occurred when fatty acids were oxidized, led to the belief that ketone bodies were undesirable side products of fatty acid degradation (*4*). The discovery by Lynen and co-workers (*5*) in the 1950's that acetoacetate and 3-hydroxybutyrate were not direct products of fatty acid degradation, but were synthesized by a separate pathway under close biochemical control, led to a reevaluation of the physiological role of these metabolites. In 1961 Krebs (*6*) presented the view that "the finding that ketone bodies are ready substrates of respiration suggests that their presence in the circulating blood serves to supply tissues with a fuel of respiration; their function is analogous to that of glucose and the nonesterified fatty acids." Many laboratories confirmed this statement, and the concept of "physiological ketosis," whereby acetoacetate and 3-hydroxybutyrate are considered to be normal and useful metabolites (*7*), is now generally accepted.

Consumption of ketone bodies diminishes the demand for glucose, reducing the necessity for gluconeogenesis and concomitant degradation of protein. The most notable example of this is the situation in the human brain. Originally it was thought that brain relied only on glucose as a fuel of respiration. However, Owen *et al.* (*2*) pointed out that humans can starve for more than 4 to 6 weeks. They reasoned that if brain used only glucose during this period, all of the protein in the body would be consumed, since body carbohydrate stores are meager and the primary substrate for gluconeogenesis is protein. By determining arteriovenous differences across brains of humans starved for 40 days, Owen *et al.* (*2*) proved that ketone bodies provided a large additional source of energy. In fact, under these circumstances, ketone bodies became the major fuel for brain metabolism, accounting for about 60 percent of the energy requirement. Subsequently, it was shown not to be necessary to postulate that this was an enzymatic adaptation by brain. Williamson *et al.* (*8*) demonstrated conclusively that the necessary enzymes were always present in amounts greater than required (*9, 10*). Furthermore, these enzyme activities were not changed by starvation or diabetes. Physiological experiments showed that ketone bodies could be and were used as soon as they appeared in the circulation, the most important determinant being their plasma concentrations (*11-13*).

Although the data show conclusively that ketone bodies are at least a partial substitute for glucose, it is an open ques-

0036-8075/79/0720-0325$00.50/0