THE MONITORING OF CADMIUM, ZINC AND COPPER IN THE KIDNEYS AND LIVER OF HUMANS DECEASED IN THE REGION OF CRACOW (POLAND)*

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Abstract. Cadmium, zinc and copper levels were determined in the renal cortex and liver of 60 inhabitants of Cracow, Poland. Cadmium levels in the renal cortex were contained in broad limits of 5–176 μg/g, mean 50.6 μg/g (wet weight). Maximum levels were found in the age group of about 50–60 years. The levels were slightly higher in men (53 μg/g) than in women (45 μg/g), with no effect of location within the region. The levels in smokers (62 μg/g) were much higher than in non-smokers (32 μg/g). The above relations were less pronounced for cadmium levels in the liver. Whole body retention of cadmium followed the pattern of cadmium in renal cortex. The level of zinc in renal cortex reflected those of cadmium. A significant proportion of the population (54% in smokers, 9% in non-smokers) showed cadmium levels in renal cortex exceeding the reference level of 50 μg/g recently accepted for general population. In the view of the authors the exposure to cadmium of the population of Cracow is excessive and calls for attention.

1. Introduction

Because of its exceptional environmental persistence and ability to cumulate in the human organism throughout the whole life span, cadmium (Cd) is considered one of the most dangerous chemical pollutants, and is contained in the global (UNEP, 1992) and national priority lists (Report, 1988).

In chronic exposure, kidneys are the critical organ. Above a concentration of 200 μg/g (wet weight) of the reneal cortex, tubular kidney damage may occur. This value is considered proper for healthy medium-age individuals, as is the case with industrial workers. For the general population, comprising elderly women, the critical renal concentration of Cd may be much lower, about 50 μg/g (Buchet et al., 1990).

For people not exposed professionally and non-smokers, food is the main source of Cd. The cumulation of the metal occurs for almost the whole life–span of humans, mainly in the kidneys and liver where it is bound with a low molecular weight protein, metallothionein (MT). Apart from Cd, this protein also contains zinc (Zn) and copper (Cu).

For the assessment of environmental exposure to Cd, biological monitoring (kidneys, liver) seems most appropriate. In the years 1986–93 such monitoring

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studies were performed in the Dept. of Toxicological Chemistry, Medical University of Lodz for the Central region of Poland (Łódź) (Bem et al., 1993a), Upper Silesia (Katowice) (Bem et al., 1993b), Białystok (Bem et al., 1993c) and Lublin (Orłowski et al., 1995). The results obtained indicate that the biological levels of Cd in Poland are high and deserve attention.

The work presents data for the region of Cracow, the old historical capital of Poland. The town of 744000 inhabitants encompasses a new industrial quarter, Nowa Huta (steel works), built in the 1950s, now constituting a major source of environmental pollution in the region. About 80% of the area of Cracow country is classified as ecologically endangered. In the west, this region borders with the Upper Silesian ecologically endangered area. Both areas are abundant in mining and metallurgic industries. In a complex ranking based on contamination, degradation, population exposure to chemicals and the state of health, this region occupies the eighteenth position among the 27 ecologically endangered areas of Poland (Dutkiewicz, 1994). For comparison, areas subject to our earlier studies, Upper Silesia (Bem et al., 1993b) and Łódź (Bem et al., 1993a), occupy the top positions, whereas Białystok (Bem et al., 1993c) is considered as a reference area.

In Cracow, the mean yearly Cd concentration in air, measured in 1971 and 1986, was 14–17.7 ng/m^3, a value three-fold higher than in a control area 60 km away (Bem and Turzyńska, 1992). More recent data obtained in 1988–90 by the Sanitary Epidemiological Station were collected at four stationary points within the township area and the mean yearly values were 1–7 ng/m^3, with upper mean monthly values not exceeding 18 ng/m^3 (PIOŚ-PIS, 1994. Nonetheless, all these concentrations are below the WHO recommended value of 10–20 ng/m^3 (WHO, 1987). The concentration of Cd in drinking water (1985–90) was in the range of <2–3 μg/l (Bem and Turzyńska, 1992). The content of Cd in vegetables in this area was 3–480 μg/kg, and in potatoes (an important component of Polish diet) 50–900, mean 65 μg/kg (Zawadzka et al., 1990).

For the population of Cracow there are also some data on biological levels of Cd. The mean concentration in blood of eight young women was 0.72 μg/l, and in the urine 0.51 μg/g creatinine (Osman et al., 1992). Similar values were found in another monitoring project: in the urine of 76 smokers, 154 nonsmokers and 99 children, the geometric means were 1.49, 0.52 and 0.29 μg/l, respectively. In the blood, geometric means of 1.15, 0.65, and 0.28 μg/l, respectively were found (Jakubowski, 1995).

2. Materials and Methods

The tissues (liver, kidneys) were obtained from the Departments of Anatomo-Pathology and of Forensic Medicine, Jagiellonian University in Cracow. Tissues were obtained from subjects not exposed occupationally to heavy metals, in which the macroscopic examination did not reveal kidney and liver damage. From the

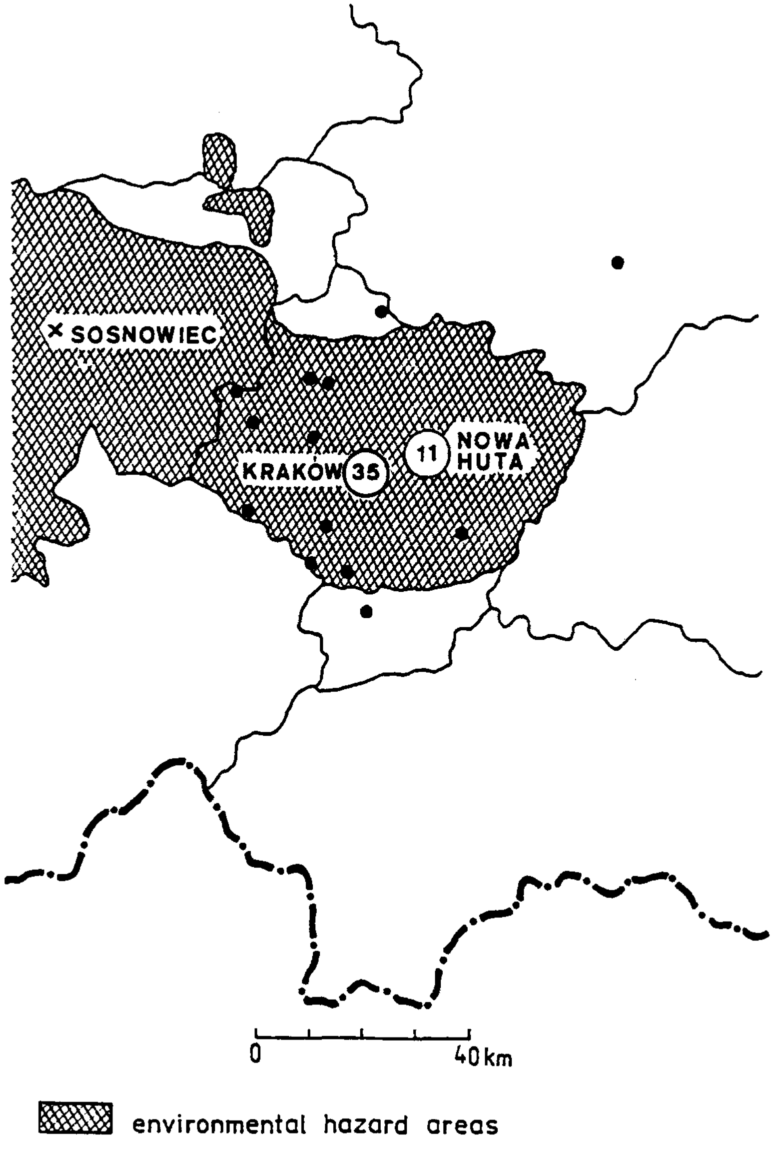

Fig. 1. Geographical distribution of sample sources with indication of province and state boundries.

kidneys, the lower pole of left kidney was taken; from the liver, the upper part of the left patch. In the kidneys, cortex was separated from medulla. Immediately after section, the tissue samples were located in polyethylene containers (washed for 24 h in 10% nitric acid) and freezed. The samples were transported in dry ice. The time lapse from death to section was contained within 8–48 h (mean 27±10). During this time the corpse was preserved in a cold room.

In a questionnaire data were supplied on sex, age, time-lapse from death to section, smoking habit, liver and kidneys weight, profession, place of residence, and clinical and pathology diagnosis. The data were supplied by the pathologist examining the case, and were based on hospital cards and on interviews with the family. The data on occupational exposure and smoking habit are burdened with some uncertainty. Retrospective data on employment were not always complete, a fact that may be of importance in Cracow, closely adjacent to the Silesian heavy industry area. The smoking habit may not have been declared on admission to the hospital for psychological reasons. On the whole such errors in qualifying the subject could, in our judgment, not exceed 5% of cases.

Generally, the study was based on 60 subjects, in which 42 were males, 18 females; 37 smokers and 23 nonsmokers; 48 town dwellers and 11 village dwellers. The majority of people studied were residents of Cracow (75%) and individual cases from other localities (Figure 1). Mean age was 48±18 years. The dominant causes of death were accidents and injuries (31 cases), heart and circulation illness (20), poisonings (5), and inflammatory disease of the abdominal cavity (3).

The levels of metals (Cd, Zn, Cu) were determined by flame atomic absorption spectrometry (Pye-Unicam SP-192) with deuterium background correction; previously, tissue samples were mineralized by a mixture of concentrated acids (nitric, sulphuric and perchloric) (Bem et al., 1986). The limit of detection was: 0.02 μg Cd/ml, 0.01 μg Zn/ml and 0.04 μg Cu/ml. For each sample three parallel independent determinations were made and the mean value was accepted.

The whole body retention was computed under the assumption that the kidneys and liver together bind 50% of the total body burden of Cd, and that the quotient of concentrations of Cd in renal cortex and mean renal concentration is 1.25 (Svartengren, 1986). Our own data on this ratio conformed well with this figure, and the value amounted to 1.25±0.08 (Orłowski, in press). The statistical calculations were made by the SYSTAT 5.30 programme (Wilkinson, 1990). The Bartlett test for homogeneity of group variances was used. The differences between related parameters were evaluated using the Tukey HSD multiple comparison test.

During the period of monitoring our laboratory was subject to inter-laboratory quality control for Cd determinations in the tissues (Veterinary Institute, Puławy). The error of Cd determinations was ±8%, with extreme differences not exceeding 14%. The intra-laboratory quality control was based on the standard 1577b-bovine liver, supplied by the National Institute of Standards and Technology (NIST), USA. In each series of determinations four parallel control samples were analysed. In this intra-laboratory quality control the mean error of determinations was: Cd ± 3.3%, Zn ± 10.9%, Cu ± 3.7%.

Table I

Statistical characteristics of metal levels [μg/g] in the renal cortex and liver as well as whole body retention of cadmium [mg] for various subpopulations

	Renal cortex				Liver			Cd body burden
	Age	Cd	Zn	Cu	Cd	Zn	Cu	
Total (N=60)								
Minimum	19.0	5.2	25.2	0.7	0.3	29.1	1.2	3.4
Maximum	94.0	175.5	92.0	6.8	15.1	85.5	9.4	120.2
Mean	48.2	50.6	49.0	2.5	3.5	52.8	4.8	37.3
Standard dev.	17.6	34.7	15.3	1.3	2.9	14.7	2.0	26.3
Skewness (G1)	0.5	1.2	0.9	1.1	2.4	0.2	0.5	1.5
Kurtosis (G2)	–0.4	1.5	0.5	1.1	6.6	–0.9	–0.4	2.2
Median	46.0	40.6	46.6	2.2	3.0	52.2	4.6	29.6
Female (N=18)								
Mean	57.7	45.0	47.9	2.6	3.4	52.5	4.3	27.3
Standard dev.	18.9	28.0	12.1	1.5	2.3	15.3	1.6	13.5
Median	57.5	40.1	45.3	2.2	3.2	55.0	4.2	25.7
Male (N=42)								
Mean	44.0	53.0	49.4	2.4	3.5	52.9	5.0	41.5
Standard dev.	15.5	37.2	16.6	1.2	3.2	14.6	2.2	29.2
Median	41.0	40.6	48.6	2.3	2.7	52.2	4.7	37.0
Non-smokers (N=23)								
Mean	53.7	31.6	42.7	2.4	3.1	57.5	5.1	23.2
Standard dev.	22.0	18.8	10.0	1.5	2.2	17.0	2.0	12.3
Median	57.0	27.0	43.1	1.9	3.0	64.4	5.1	19.4
Smokers (N=37)								
Mean	44.6	62.4	52.9	2.5	3.7	49.9	4.6	46.0
Standard dev.	13.3	37.2	16.8	1.2	3.3	12.5	2.1	28.9
Median	42.0	56.4	55.0	2.3	2.9	50.0	4.5	40.4
Towns (N=48)								
Mean	50.8	50.3	49.0	2.5	3.2	50.9	4.6	35.6
Standard dev.	17.5	29.7	14.2	1.3	2.5	14.4	1.9	22.1
Median	51.0	43.4	49.2	2.2	3.0	49.6	4.5	33.1
Villages (N=11)								
Mean	37.0	55.1	49.9	2.7	4.9	61.4	5.5	47.0
Standard dev.	14.7	53.2	20.2	1.3	4.4	14.0	2.6	40.0
Median	36.0	28.1	44.8	2.4	3.5	61.6	5.8	24.2

Table I
Continued.

	Renal cortex			Liver			Cd body burden
	Cd	Zn	Cu	Cd	Zn	Cu	
Age <40 (N=21)							
Mean	45.6	48.2	2.4	3.1	57.7	5.5	32.8
Standard dev.	36.7	12.0	1.3	3.0	15.9	2.4	24.6
Median	39.5	47.3	2.3	2.3	56.5	5.9	24.2
Age 40–60 (N=25)							
Dean	62.4	54.2	2.7	3.6	50.7	4.4	47.4
Standard dev.	34.4	16.9	1.4	3.4	13.1	1.7	30.1
Median	56.8	53.7	2.6	2.8	51.8	4.4	44.2
Age >=60 (N=14)							
Mean	40.6	42.2	2.3	3.8	48.9	4.4	28.1
Standard dev.	28.4	14.2	1.1	2.2	15.1	1.9	15.9
Median	30.7	39.3	1.9	3.4	45.7	4.2	24.2

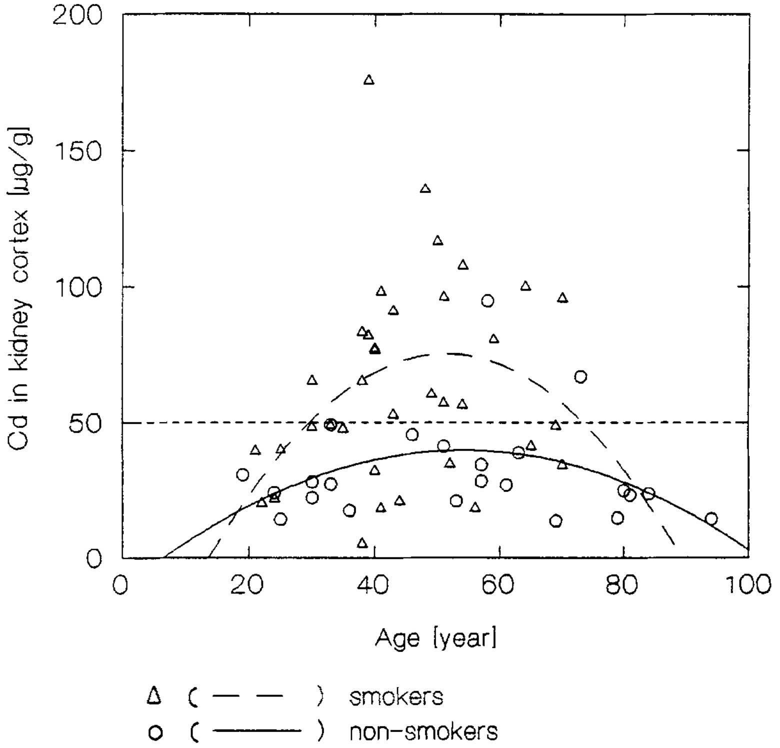

Fig. 2. Age-dependent distribution of Cd in renal cortex.

3. Results and Discussion

A broad range of Cd levels in the renal cortex was found (5.2–175.5), mean 50.6 μg/g (Table I). In conforming with data of other authors, an increase of these concentrations was found with age, up to about the age of 50 years, with a decline thereafter (Figure 2). In smokers, the Cd levels in the renal cortex were twice those in non-smoking persons (**p**<0.001), whereas the sex and residence within the region did not influence the levels.

The levels of Cd in the liver (mean 3.5 μg/g) were slightly higher than reported in other countries. This parameter is correlated with the Cd levels in the renal cortex (r=0.57, p<0.001); however, the levels in smokers, although higher, are not significantly different from those in non-smokers. Also, no differences were found related to sex and residence. From the data in Table I it is apparent that the hepatic levels of Cd do increase with age.

The whole body retention of Cd, computed from the contents of kidneys and liver, also shows a broad range of values (3.4–120.2), mean 37.3 mg Cd and is higher in smokers (p<0.001). Also the age-dependent structure (not presented) is similar to that of renal cortex levels. For both the liver-Cd and whole body retention there are no independent reference values.

The levels of Zn and Cu in the renal cortex and in the liver (Table I) do not deviate from data of other authors (Chung et al., 1986; Heilmaier et al., 1987; Honda and Nogawa, 1987; Takâcs and Tatâr, 1991). The level of Zn in the renal cortex is correlated with the level of Cd in the renal cortex (r=0.84), Cu in renal cortex (r=0.38), Cd in liver (r=0.36) and with whole body Cd retention (r=0.70). Zinc is, apart from Cd, a basic constituent of human renal metallothionein, and both these metals occur in equimolar proportions. Similarly as for Cd, the highest levels of Zn in the renal cortex are found in subjects of middle age.

From the data presented an evident influence of tobacco smoking on the biological levels of Cd is apparent. This is especially striking if the absolute levels of Cd in the renal cortex are compared (difference of about 30 μg/g) (Figure 3, Table I). Compared with data from other countries this phenomenon is more pronounced; we ascribe it to the high levels of Cd in Polish cigarettes: 1.3–3.2 μg Cd/cigarette (Bronisz et al., 1983; Zawadzka et al., 1989). Apart from Cd in the renal cortex and whole body retention of Cd, tobacco smoking elevates Cd levels somewhat in the liver and Zn content of the renal cortex; this is, however, not significant statistically.

For the general toxicological assessment of the situation of special importance are data related to non-smoking individuals who were not exposed to Cd occupationally in the past. The construction of such a group is, however, burdened with some uncertainty, especially in highly industrialized regions.

From the data presented it may be seen that for Cracow the Cd renal cortex level in non-smokers amounts to 27.0 μg/g (median). Analogical values for regions studied earlier were: 27.3, 33.0, 16.5, 20.6 μg/g for Łódź, Katowice, Białystok and Lublin respectively (Bem et al., 1993a–c; Orłowski et al., 1995). Only the value

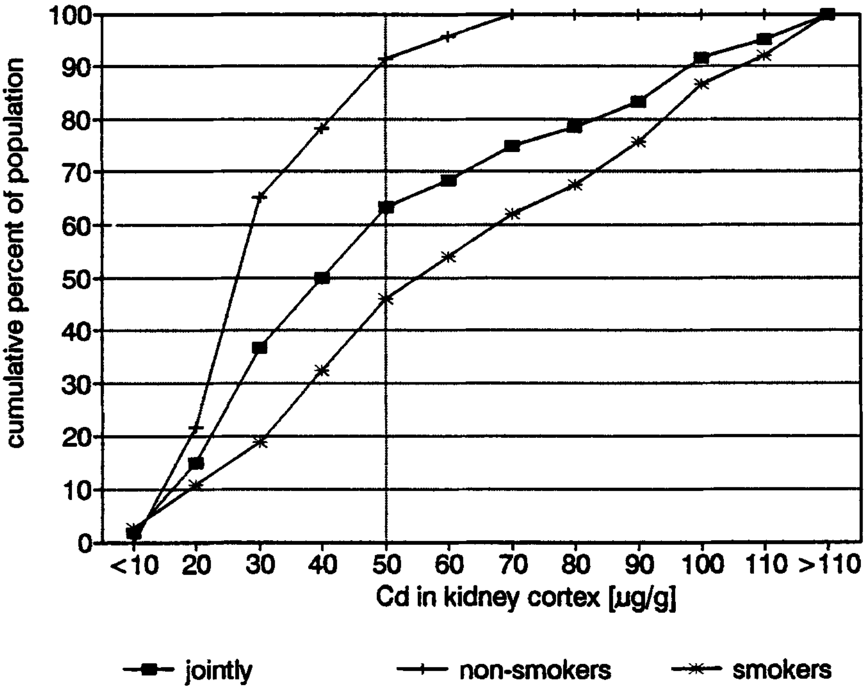

Fig. 3. Cumulative percent of population vs. cadmium levels in renal cortex.

for Białystok (agriculture area) is significantly lower from the data for Cracow (as well as from Łódź and Katowice). This may point to higher contamination by Cd of the southern and central regions of Poland.

The levels of Cd in the renal cortex of the inhabitants of Cracow are among the highest values in the world (Vahter, 1982). Accepting as a reference value the level of 50 μg/g (Buchet et al., 1990), the exceedence is 9, 37 and 54%, for non-smokers, whole population and smokers, respectively. These figures are deeply disquieting. This situation has been assessed in Poland by the Commission on Toxicology, Sanitary-Epidemiological Advisory Board (Report, 1995) and a decision was taken to continue biological monitoring of Cd in the organs of decreased persons and, in parallel, in the blood and urine of living subjects. Emphasis is given to obtaining consistence in interpreting both types of data.

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References

Bem, E. M., Tegegnework, H. and Piotrowski, J. K.: 1986, 'The Choice of the Optimal Mineralization Method of Biological Samples for Zinc and Copper determination', *Bromat Chem Toksykol* **XIX**, 37–41, (in Polish).

Bem, E. M. and Turzyńska, E.: 1992, 'Human Exposure to cadmium. Part I. Cadmium Level in Various Components of the Environment', *Bromat. Chem. Toksykol.* **XXV**(4), 361–371.

Bem, E. M., Kaszper, B. W., Orłowski, Cz., Piotrowski, J. K., Wójcik, G. and Zołnowska, E.: 1993a, 'Cadmium, Zinc, Copper and Metallothionein Levels in the Kidney and Liver of Humans From Central Poland', *Environ. Monit. Assess.* **25**, 1–13.

Bem, E. M., Orłowski, Cz., Piotrowski, J. K., Januszewski, J. and Pajak, J.: 1993b, 'Cadmium, Zinc, Copper and metallothionein, Levels in the Kidney and Liver of Humans from Upper Silesia (Poland)', *Int. Arch. Occup. Environ. Hlth* **65**, 57–63.

Bem, E. M., Piotrowski, J. K. and Turzyńska, E.: 1993c, 'Cadmium, Zinc, and Copper Levels in the Kidneys and Liver of the Inhabitants of North-Eastern Poland', *Pol. J. Occup. Med. Environ. Hlth* **6**, 133–141.

Bronisz, H., Szost, T., Lipska, M. and Zawada, M.: 1983, 'Cadmium Content in Cigarettes', *Bromat. Chem. Toksykol,* **XVI**, 121–127, (in Polish).

Buchet, J. P., Lauwerys, R., Roels, H., Bernard, A., Bruaux, P., Claeys, F., Ducoffre, G., de Plaen, P., Staessen, J., Amery, A., Lijnen, P., Thijs, L., Rondia, D., Sartor, F., Saint Remy, A. and Nick, L.: 1990, 'Renal Effects of cadmium Body Burden of the General Population', *Lancet* **336**, 699–702.

Chung, J., Nartey, N. O. and Cherian, M. G.: 1986, 'Metallothionein Levels in Liver and Kidney of Canadians – A Potential Indicator of Environmental Exposure to Cadmium', *Arch. Environ. Hlth* **41**(5), 319–323.

Dutkiewicz, T.: 1994, 'Methodology and Evaluation of Large Areas of Ecological Hazard in Poland', *Bromat. Chem. Toksykol.* **XXVII**, 287–306, (in Polish).

Heilmaier, H. E., Drasch, G. A., Ketschmer, E. and Summer, K. H.: 1987, 'Metallothionein, Cadmium, Copper and Zinc Levels of Human and Rat Tissues', *Toxicol. Lett.* **38**, 205.

Honda, R. and Nogawa, K.: 1987, 'Cadmium, Zinc and Copper Relationships in Kidney and Liver of Humans Exposed to Environmental Cadmium', *Arch. Toxicol.* **59**, 437–442.

Jakubowski, M.: 1995, 'The Biological Levels of Cadmium in the Inhabitants of Poland', Med. Pracy Supl. 5, 1995, 83–96, (in Polish).

Orłowski, C., Piotrowski, J. K., Bem, E. M. and Szumiło, J.: 1995, 'The Monitoring of Cadmium, Zinc and Copper in the Kidneys and Liver of Humans Deceased in the Region of Lublin', *Acta Pol. Toxicol.* (in press).

Orłowski, C., Piotrowski, J. K. and Swiercz, R.: 'Binding of Cadmium in the Renal Medulla of Humans Not Exposed Occupationally to Cadmium', *Acta Pol. Toxicol.* (in press).

Osman, K., Bjorkman, L., Lind, B. and Nordberg, M.: 1992, 'Biological Monitoring of Metals in Children and Pregnant Women in Poland', *Int. J. Environ. Health Res.* **2**, 212.

PIOS-PIS: 1994, *Air Pollution in Poland, 1993.* The Sate Environmental Protection Inspection and State Sanitary Inspection. The Library of Environmental Monitoring, Warsaw. (in Polish).

'Report of the Commission on Environmental Toxicology, Committee of Human Ecology, Polish Academy of Sciences, on priority environmental toxic chemicals in Poland', *Bromat. Chem. Toksykol.* **XXI**, 309–313, 1988 (in Polish).

Report of the Toxicological Commission, Sanitary – Epidemiological Advisory Board, *Cadmium Hazards in Poland,* Łódź, 1995, Med. Pracy, Supl. 5, 7–23, (in Polish).

Svartengren, M., Elinder, C. G., Friberg, L. and Lind, B.: 1986, 'Distribution and Concentration of Cadmium in Human Kidney', *Environ. Res.* **39**, 1–7.

Takãcs, S. and Tatár, A.: 1991, 'Trace Elements in the Environment and in Human Organs: Analysis According to Domicile and Sex', *Z. Gesamte Hyg.* **37**(2), 53–55.

UNEP: 1992, *Chemical Pollution: A Global Overview*. United Nations Environment Programme, Geneva.

Vahter, M. (ed.): 1982, *Assessment of Human Exposure to Lead and Cadmium Through Biological Monitoring*, UNEP/WHO Stockholm.

WHO: 1987, World Health Organization – Regional Office for Europe: *Air Quality Guidelines for Europe*, WHO Regional Publication, European series No 23, Copenhagen.

Wilkinson, Leland: 1990, *SYSTAT: The System for Statistics*, Evanston, IL: SYSTAT, Inc.

Zawadzka, T., Wojciechowska-Mazurek., Starska, K., Brulińska-Ostrowska, E., Cwiek, K. and Starska, K.: 1989, 'Cadmium and Lead Levels in Domestic and Imported Cigarettes', *Rocz. PZH* **40**(2), 145–151, (in Polish).

Zawadzka, T., Mazur, H., Wojciechowska-Mazurek, M., Starska, K., Brulińska-Ostrowska, E., Cwiek, K., Umińska, R. and Bichniewicz, A.: 1990, 'The Content of Metals in Vegetables from various Regions of Poland in the years 1986–1988. Part I. Lead, Cadmium, Mercury', *Rocz. PZH* **XLI**,, 111–131, (in Polish).