BioMetals 1994, **7**, 130–134

Cadmium in blood in Alzheimer's disease and non-demented subjects: results from a population-based study

H. Basun*,¶, B. Lind†, M. Nordberg‡, M. Nordström¶, K. Sparring Björkstén§ & B. Winblad*,¶

*Karolinska Institutet, *Department of Geriatric Medicine, Huddinge University Hospital, †Institute of Environmental Medicine, ‡Department of Environmental Hygiene, §Department of Psychiatry, St Göran's Hospital and ¶Stockholm Gerontology Research Center, Stockholm, Sweden*

Received 11 June 1993; accepted for publication 27 August 1993

Blood cadmium concentrations were studied in Alzheimer's disease (AD) and non-demented subjects. The 29 individuals were randomized from the ongoing population survey on ageing and dementia in Stockholm, the Kungsholmen Project. Smokers had, as expected, higher cadmium levels than non-smokers. Cadmium concentrations in blood were related to diastolic blood pressure in non-smoking, non-demented individuals. In contrast to previous reports no differences in blood cadmium levels were found between AD sufferers and non-demented subjects. Furthermore, there were no correlations between cadmium levels in blood and age or cognitive functions. The importance of quality assurance in sample collection and analysis of cadmium as well as scrutinizing smoking habits is emphasized.

Keywords: Alzheimer's disease, blood, cadmium

Introduction

Contributions to the genetic hypothesis of Alzheimer's disease (AD) have been made during the last years (Goate *et al.* 1991, Mullan *et al.* 1992). They found two different mutations in the amyloid precursor protein gene in families with an early onset type of AD. However, most cases of AD are sporadic and of late onset, where the etiology is unknown. In the ongoing population survey on ageing and dementia in Stockholm, the Kungsholmen Project, four risk factors for AD were found: age, a family history of dementia, alcohol abuse and manual work (Fratiglioni *et al.* 1993). Solvents and aluminum exposure have been found to be associated with impaired cognition (Axelsson *et al.* 1976, Sjögren *et al.* 1990, Rifat *et al.* 1990). Several metals have also been proposed as pathogenic co-factors in AD (for review, see Basun *et al.* 1991). One of these, cadmium, has been found in high concentrations in liver tissue and blood in AD sufferers (Lui *et al.* 1990, Basun *et al.* 1991).

Address for correspondence: H. Basun, Department of Geriatric Medicine, Huddinge University Hospital, S-144 86 Huddinge, Sweden. Fax: (+46) 8 711 1751.

In muscles, as well as in other organs, cadmium accumulates with exposure through food and tobacco smoke during the whole life span. However, at age 40–50 years a maximum is reached in the kidney, which is considered to be the critical organ for long-term low level exposure (Kjellström 1985). Accumulation in liver levels off at age 30. The concentration is 10–30 times higher in the kidney than in the liver at 50 years of age (Nordberg *et al.* 1985). Various biological half-times for cadmium are reported depending on the model used for calculating the half-time (WHO/IPCS 1992). By use of an eight-compartment model, biological half-time was estimated to be 7.5 and 12 years for liver and kidney, respectively (Kjellström & Nordberg 1978). For muscle, the biological half-time is even longer, up to as long as 30 years has been mentioned. The half-times of the tissues are about 15–40% of the life span (Nordberg *et al.* 1985). Even if cadmium concentration is low in the muscles, the total contribution to body burden is extensive due to the large weight of muscles (WHO/IPCS 1992).

Inhalation of tobacco smoke contributes significantly to cadmium concentration in organs. Smokers have about twice the concentration in the kidney compared with non-smokers. The concentration of

cadmium in blood is a useful indicator of ongoing exposure during recent months, but it may also partly be related to body burden. There is a pronounced influence on blood cadmium concentration from the inhalation of tobacco smoke. Among non-smokers, a slight increase with age in the blood cadmium concentration can usually be seen (Friberg *et al.* 1986). Hypertension and cardiovascular disease in relation to cadmium have been debated. A somewhat higher cadmium concentration was observed in liver and kidney in people in the general environment deceased from hypertensive and cardiovascular disease compared with other causes of death (Schroeder 1965, 1967). As in many other studies, smoking habits were not taken into account.

Due to the increased cadmium content in blood of a selected group of AD patients reported by Basun *et al.* (1991), this metal was chosen to be investigated in a population-based study under extreme standardized conditions including quality assurances.

Aims of the investigation

What is the blood cadmium concentration in normal ageing and AD? Is there any relation between cadmium concentrations in blood and cognitive functions, degree of illness or age? Is there any relation between cadmium concentrations in blood and blood pressure, body mass index (BMI) or kidney functions?

Materials and methods

Subjects

Subjects were seen as a part of a population-based study, which has been reported in detail elsewhere (Fratiglioni *et al.* 1991, 1992). Briefly, the project is based on all inhabitants (born 1912 and before) living in an area of Stockholm called Kungsholmen in October 1987. There were 2368 individuals, consisting of 1800 females and 568 males, living at home or in institutional care. The whole population was assessed with the Mini-Mental State Examination (MMSE) (Folstein *et al.* 1975) used as screening test with a cut-off point of 23/24 to discriminate subjects with and without cognitive impairment. All subjects who screened positive (MMSE ≤ 23) and a random sample of subjects who screened negative (MMSE ≥ 24) were extensively investigated with clinical examination, family interview and laboratory tests to reach a final diagnosis of dementia and different types of dementia. The sample was selected from all the screened negatives, matched by gender and age (± 5 years of age) with screened positives. The clinical examination included a medical and social history, physical and neurologic examination, cognitive examination, and assessment of depression using a sub-scale of the Comprehensive Psychopathological Rating Scale (Perris *et al.* 1984). Diet was assessed by an interview with the proband and with a next-of-kin using a structured family interview (Roth *et al.* 1986). Height, body weight and blood pressure were recorded for all individuals.

The dementia diagnosis was based on DSM-III-R criteria (American Psychiatric Association, *Diagnostic and Statistical Manual of Mental Disorders*, revised 3rd edn (DSM-III-R) 1987) with some modifications (Fratiglioni *et al.* 1991).

The investigated patients in the current study were randomized from the above presented group of aged people. Demographic data are given in Table 1. These individuals were further investigated with different psychometric tests, EEG and magnetic resonance imaging of the brain. In this group the diagnosis of AD was based on the criteria from the National Institute of Neurological Disorders Association and Stroke–Alzheimer's Disease and related disorders (NINCDS–ARDRA) (McKhann *et al.* 1984). Of the demented subjects, six were diagnosed as probable AD.

Blood sampling

Whole blood (10 ml) was collected from 29 subjects (21 females and eight males). To avoid contamination from cadmium in tobacco smoke the sample collectors were non-smokers. The skin of the patients was cleaned with mediswab (Pharmax, Bexley, Kent, UK) containing isopropanol before collection of blood for the cadmium analysis via evacuated tubes, Venojet VT 100-H with green stoppers (Terumo, Tokyo, Japan). The tubes contained sodium–heparin and were hence turned 10–15 times for mixing before storage at 4 °C. Then, 1 ml was transferred with a pasteur pipette to an acid-washed tube of polyethylene, and immediately frozen at −20 °C and after 1 week stored at −80 °C.

Table 1. Demographic data in non-demented and demented subjects (those demented subjects diagnosed as AD are also shown separately)

	Non-demented	Demented	AD
No.	19	10	6
Age	82 ± 5[a]	84 ± 6	85 ± 3
Sex (F/M)	12/7	9/1	5/1
MMSE	26 ± 4	21 ± 5	20 ± 5
Height	159 ± 23	160 ± 8	161 ± 9
Body weight	63 ± 12	62 ± 9	61 ± 6
BMI ($kg\,m^{-2}$)	24.1 ± 2.8	23.7 ± 4.2	23.4 ± 2.4
Smoker/ non-smoker	5/12[b]	3/6[b]	1/4[b]

[a]Mean ± standard deviation.
[b]Smoking habits were unknown in two control subjects and in one AD individual.

Analytical methods

Blood samples were also taken for routine analysis of hemoglobin, glucose, leukocytes and sedimentation rate in whole blood, albumin, chlorides, cobalamin, creatinine, folic acid, iron, sodium and thyroid hormones in serum. These analyses were performed according to the routines at the Department of Clinical Chemistry, St Göran's Hospital, Stockholm.

Whole blood was analyzed for cadmium (Elinder *et al.* 1983) after deproteinization (0.3 ml blood +0.5 ml 0.8 M HNO_3) by use of graphite furnace atomic absorption spectrophotometry (GFAAS). The equipment used was a Perkin Elmer 5000 AAS with Zeeman background correction and with a graphite furnace HGA-500, L'vov platform, an automatic sample exchanger AS-40, a two-pen recorder PE-56 and a computer PE-7500 as accessories. The detection limit for cadmium in blood was 0.08, 0.11 and 0.17 ng g^{-1} for the 3 days of analysis. For evaluation of accuracy, a quality control (QC) program was performed as described by Vahter & Friberg (1988). The results of the three QC sets, one for each day of analysis, were all accepted according to the criterion, maximum allowable deviation (MAD): $y = x \pm (0.05x + 0.02)$ ng g^{-1}, used for an international program under WHO. Mean values for the relative standard deviation (RSD) between duplicates were 3, 10 and 3% RSD, respectively.

Data analysis

Comparions between groups were made with Student's *t*-test. Pearson's correlation coefficients were used when analyzing the relationships between cadmium levels and continuous variables. Spearman Rank correlation coefficients were calculated when comparing MMSE scores and cadmium concentrations. Significance levels were set at $P < 0.05$.

Results

Age, sex, number of smokers and non-smokers, MMSE, height, body weight and BMI are presented in Table 1. Non-demented subjects have a higher body weight and BMI compared with demented individuals and AD subjects. One person in the control group was not possible to identify with regard to smoking history and thus not included in the study. Smokers had higher cadmium levels in blood than non-smokers (Table 2).

Non-smokers

Non-demented individuals, demented subjects and AD sufferers had blood cadmium concentrations ($X \pm$ SD) of 0.46 ± 0.17, 0.44 ± 0.06 and 0.49 ± 0.14 ng g^{-1}, respectively. The difference between these groups was not statistically significant (Table 2). There was a positive relationship between blood cadmium concentration in non-demented subjects and diastolic blood pressure, albumin and cobalamin in serum. Furthermore, there was a negative association to leukocytes in blood and height, respectively. No associations with age or MMSE were seen in either of the groups. In Figure 1, diastolic blood pressure is plotted against blood cadmium concentration in the non-demented individuals. Data from the relation between height, blood pressure, leukocytes, albumin, creatinine and cobalamin are given in Table 3.

Table 2. Blood cadmium levels (ng g^{-1}) in smokers and non-smokers in non-demented and demented subjects (those demented subjects diagnosed as AD are also shown separately)

	Non-demented	Demented	AD	*P*
Smokers	1.06 ± 0.97[a]	1.27 ± 1.03	0.54	NS[b]
	($n = 5$)	($n = 3$)	($n = 1$)	
Non-	0.46 ± 0.17	0.44 ± 0.06	0.49 ± 0.14	NS
smokers	($n = 12$)	($n = 6$)	($n = 4$)	
P	< 0.04[c]	< 0.06	NS	

[a]Expressed as mean ± standard deviation.
[b]Non-demented versus demented and AD subjects, respectively. Student's *t*-test.
[c]Smokers versus non-smokers. Student's *t*-test.

Smokers

Non-demented individuals, demented patients and AD sufferers had cadmium blood concentrations of 1.06 ± 0.97, 1.27 ± 1.03 and 0.54 ng g^{-1}, respectively.

Discussion

In the non-smoking, non-demented subjects (mean age 84 and range 76–93 years) the mean cadmium

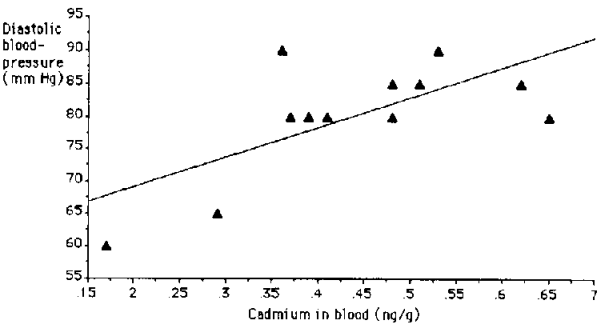

Figure 1. Correlation between blood cadmium levels (ng g^{-1}) and diastolic blood pressure (mm Hg) in non-smoking individuals classified as non-demented

Table 3. Significant correlations with blood cadmium levels in non-smoking individuals classified as non-demented and demented (those demented subjects diagnosed as AD are also shown separately)

	Non-demented	Demented AD
Height	−0.72; $P = 0.01$[a]	
Blood pressure		
systolic	+0.55; $P = 0.06$	
diastolic	+0.68; $P = 0.01$	
Leukocytes	−0.60; $P = 0.04$	
Albumin	+0.57; $P = 0.05$	
Creatinine		−0.95; $P = 0.05$
Cobalamin	+0.60; $P = 0.04$	

[a]Correlation coefficient and probability.

concentration was 0.46 ng g^{-1}, which is to be compared with 0.3 ng g^{-1} reported by Elinder *et al.* (1983) in a somewhat younger population. This may indicate an age-associated increase in cadmium levels, which has been suggested by Friberg *et al.* (1986). However, such a relation could not be observed with increasing age within the current study. This may be due to the rather limited number of individuals taking part in the study (altogether 29) or to the possibility that the cadmium content in blood has reached a maximum at 76 years. This would be in line with the circumstances in kidney and liver.

Cognitive functions as assessed by MMSE were not related to cadmium levels in blood in either of the groups. This may indicate that blood cadmium concentrations lower than the levels of known acute toxicity do not affect cognitive functions. These data, however, must be interpreted with caution, as MMSE is just a screening tool for cognitive dysfunctions.

Concentrations for different metals in plasma and cerebrospinal fluid differed in a selected group of AD sufferers as compared with healthy, aged control subjects (Basun *et al.* 1991). This was most prominent for the high cadmium levels detected in plasma from AD patients. In the current pilot study no difference was seen between the AD and non-demented individuals. Thus, we were not able to confirm the earlier findings of high plasma cadmium in AD sufferers. This may be due to the use of quality assurance in sample collection and analysis of cadmium in the present study. Furthermore, history of smoking habits was carefully scrutinized in this population-based study. This is in contrast to the earlier study, which was performed on a selective group of individuals. However, the current study was done on a restricted number of subjects.

We observed an association between cadmium content in blood and diastolic blood pressure. There was also a trend towards a relationship between systolic blood pressure and the cadmium concentration in blood ($P = 0.06$). This is in accordance with the findings of Schroeder (1965, 1976).

There was a negative correlation between cadmium concentration in blood and creatinin in serum. This may reflect a release from the muscles according to the well known loss of body weight in AD patients (Sandman *et al.* 1987).

More joint research between gerontologists and toxicologists is warranted as too little is known concerning ageing and chemicals with regard to adverse health effects, which has recently been focused upon (WHO/IPCS 1993).

Acknowledgments

We gratefully thank all the members of the Kungsholmen Project Study Group for their collaboration and generous assistance. For skilful laboratory work, Brita Palm and Bo Nilsson are gratefully acknowledged. Funds from Gamla Tjänarinnor, Gadelius, Groschinsky and Karolinska Institutet supported the study.

References

American Psychiatric Association. 1987 *Diagnostic and Statistical Manual of Mental Disorders*, revised 3rd edn (DSM-III-R). Washington DC: American Psychiatric Association.

Axelsson O, Hane M, Hogstedt C. 1976 A case referent study on neuropsychiatric disorders among workers exposed to solvents. *Scand J Work Environ Health* **2**, 14–20.

Basun H, Forsell L-G, Wetterberg L, Winblad B. 1991 Metals and trace elements in plasma and cerebrospinal fluid in normal ageing and Alzheimer's disease. *J Neural Transm (P-D Sect)* **4**, 231–258.

Elinder C-G, Friberg L, Lind B, Jawaid M. 1983 Lead and cadmium levels in blood samples from the general population of Sweden. *Environ Res* **30**, 233–253.

Folstein MF, Folstein SE, Mc Hugh PR. 1975 'Mini-Mental State': a practical method for grading the cognitive state of patients for the clinician. *J Psychiatr Res* **12**, 189.

Fratiglioni L, Grut M, Forsell Y, *et al.* 1991 Prevalence of Alzheimer's disease in an elderly urban population: relationship with age, sex and education. *Neurology* **41**, 1886–1892.

Fratiglioni L, Viitanen M, Bäckman L, Sandman P-O,

Winblad B. 1992 Occurrence of dementia in advanced age: the study design of the Kungsholmen Project. *Neuroepidemilogy* **11** (suppl 1), 29–36.

Fratiglioni L, Ahlbom A, Viitanan M, Winblad B. 1993 Risk factors for late onset Alzheimer's disease: a population-based-case control study. *Ann Neurol* **33**, 258–266.

Friberg L, Kjellström T, Nordberg GF. 1986 Cadmium. In: Friberg L, Nordberg GF, Vouk V, eds. *Handbook on the Toxicology of Metals*, Vol. II. Amsterdam: Elsevier.

Goate A, Chartier-Harlin M-C, Mullen M, *et al*. 1991 Segregation of a missense mutation in the amyloid precursor protein gene with familial Alzheimer's disease. *Nature* **349**, 704–706.

Kjellström T. 1985 Critical organs, critical concentrations, and whole body dose–response relationships. In: Friberg L, Elinder C-G, Kjellström T, Nordberg GF, eds. *Cadmium and Health. A Toxicological and Epidemiological Appraisal*, Vol. II. Boca Raton, FL: CRC Press.

Kjellström T, Nordberg GF. 1978 A kinetic model for cadmium metabolism in the human being. *Environ Res* **16**, 248–269.

Lui E, Fisman M, Wong C, Diaz F. 1990 Metals and the liver in Alzheimer's disease. An investigation of hepatic zinc, copper, cadmium, and metallothionein. *J Am Geriatr Soc* **38**, 633–639.

McKhann G, Drachman D, Folstein M, Katzman R, Price D, Stadlan EM. 1984 Clinical diagnosis of Alzheimer's disease: report of the NINCDS–ADRDA Work Group under the auspices of the Department of Health and Human Services Task Force on Alzheimer's disease. *Neurology* **34**, 939–944.

Mullan M, Crawford F, Axelman K, *et al*. 1992 A pathogenic mutation for probable Alzheimer's disease in the APP gene at the N-terminus of β-amyloid. *Nature Genetics* **1**, 345–347.

Nordberg GF, Kjellström T, Nordberg M. 1985 Kinetics and metabolism. In: Friberg L, Elinder C-G, Kjellström T, Nordberg GF, eds. *Cadmium and Health. A Toxicological and Epidemiological Appraisal*, Vol. I. Boca Raton, FL: CRC Press.

Perris C, Eisemann M, von Knorring L, Perris H. 1984 Presentation of a subscale for the rating of depression and some additional items on the comprehensive psychopathological rating scale. *Acta Psychiatr Scand* **70**, 261.

Rifat SL, Eastwood MR, Crapper McLachlan DR, Corey PN. 1990 Effect of exposure of miners to aluminium powder. *Lancet* **336**, 1162–1165.

Roth M, Tym E, Mountjoy CQ, *et al*. 1986 Camdex. A standardized instrument for the diagnosis of mental disorder in the elderly with special reference to the early detection of dementia. *Br J Psychiatry* **149**, 698–709.

Sandman P-O, Adolfsson R, Nygren C, Hallmans G, Winblad B. 1987 Nutritional status and dietary intake in institutionalized patients with Alzheimer's disease and multiinfarct dementia. *J Am Geriatr Soc* **35**, 31–38.

Schroeder HA. 1965 Cadmium as a factor in hypertension. *J Chron Dis* **18**, 647–656.

Schroeder HA. 1976 Cadmium, chromium, and cardiovascular disease. *Circulation* **35**, 570.

Sjögren B, Gustavsson P, Hogstedt C. 1990 Neuropsychiatric symptoms among welders exposed to neurotoxic metals. *Br Ind Med* **47**, 704–707.

Vahter M, Friberg L. 1988 Quality control in integrated human exposure monitoring of lead and cadmium. *Fresenius Z Anal Chem* **332**, 726–731.

WHO/IPCS. 1992 *Cadmium. Environmental Health Criteria 134*. Geneva: WHO.

WHO/IPCS. 1993 *Principles for Evaluation of Chemical Effects of the Aged Population. Environmental Health Criteria 144*. Geneva: WHO.