ORIGINAL ARTICLES

Renal Dysfunction in Workers Exposed to Inorganic Lead

Y C Lim,*, K S Chia,**_FAMS, MD, FFOM (Lond)_, H Y Ong,***_BSc_, V Ng,****_BSc Hons_, Y L Chew,****_Dip Biotech_

Abstract

Introduction: We studied the relationship between renal dysfunction and occupational lead exposure in a local factory. _Materials and Methods:_ A cross-sectional study with a cohort component was conducted with 55 male workers of a factory producing PVC stabilisers as subjects. Workers from this factory have been followed up with 6-monthly blood lead measurements since 1982. Two indices of overall lead exposure, i.e. the number of times a worker's blood lead exceeded 40 μg/dL (PbB40) and cumulative blood lead index (PbBint), were obtained from this data. Recent blood lead concentration (PbB) was measured. 4-hour creatinine clearance and various other urinary and serum markers of renal dysfunction were used as effect indices. _Results:_ There was no relationship between PbB and any of the renal markers. However, creatinine clearance decreased significantly (P <0.001) with increasing PbB40 and PbBint after adjustment for age and smoking habits. Urinary albumin (Ualb), urinary α-1 microglobulin (Uα1m), urinary β-2 microglobulin (Uβ2m) and urinary retinol-binding protein (URBP) increased significantly with both increasing PbB40 and PbBint. Total urinary activity of N-acetyl-β-D-glucosaminidase (NAG-T) and its heat-stable isoenzyme (NAG-B) increased significantly with increasing PbB40. A significant difference in renal parameters occurred when PbB40 was 1 or more. _Conclusions:_ We have found a positive association between overall lead exposure and renal dysfunction. The renal parameters were significantly higher among those who had at least one episode of blood lead above 40 μg/dL. Our findings also strengthen the case for the use of Ualb, Uα1m, Uβ2m, URBP, NAG-T and NAG-B as early markers of lead nephropathy.

**Ann Acad Med Singapore** 2001; 30:112-7

Key words: α-1 microglobulin, β-2 microglobulin, Creatinine clearnance, N-acetyl-β-D-glucosaminidase

Introduction

Chronic lead exposure causes progressive nephropathy that eventually results in renal failure. Known as chronic lead nephropathy, this disease has been recognised since the 19th century[1] and is occasionally accompanied by hypertension and gout. Mortality studies of lead workers have found excess mortality from chronic renal disease compared to the general population.[2-4] Baker et al[5] found raised blood urea nitrogen (BUN) in 8% to 32% of lead-exposed workers from 3 plants. Lilis et al[6] studied patients with occupational lead poisoning and found significantly lower creatinine clearance amongst those who satisfied various criteria for "chronic lead poisoning". In Queensland, Australia, an endemic of lead poisoning amongst children was associated with a rise in the incidence of chronic renal failure in the same cohort 10 to 40 years later.[7-10] Among patients who suffered from lead poisoning by consuming illegally distilled alcohol, renal failure frequently occurred.[11] Pathological features of kidney specimens from victims of the Queensland epidemic[12] and animal experiments[13,14] are intra-nuclear inclusion bodies and non-specific interstitial nephritis.

With implementation of stricter controls, lower levels of lead exposure are now encountered in occupational settings. It is unclear whether nephropathy occurs at these levels. In animal studies, significant nephropathy was not demonstrated with low dose lead exposure.[15,16] Gennart et al[17] and Roels et al[18] did not find significantly different renal function between exposed workers and control groups, unlike Pinto de Almeida et al[19]. Ehrlich et al[20] found positive exposure-response relations between current PbB, historical PbB and serum creatinine.

Lead nephropathy has none or few clinical signs and glomerular function tests, such as creatinine clearance, only become abnormal after nephropathy is irreversible. Hence, recently there has been much interest in markers that indicate lead nephropathy at an early and reversible stage. Low-molecular-weight proteins such as urinary

   * Undergraduate
     Faculty of Medicine
  ** Associate Professor
 *** Principal Laboratory Officer
**** Laboratory Officer
     Department of Community, Occupational and Family Medicine
     National University of Singapore

Address for Reprints: Dr Chia Kee Seng, Department of Community, Occupational and Family Medicine, National University of Singapore, MD3, Lower Kent Ridge Road, Singapore 119260.

α-1 microglobulin (Uα1m) and urinary retinol-binding protein (URBP), urine albumin, renal antigens such as brush-border antigen, prostaglandins such as thromboxane-A2 and urinary 6-keto-prostaglandin-F, enzymes such as N-acetyl-β-D-glucosaminidase (NAG), growth factors and other analytes have been evaluated, and although some studies have shown correlation between them and lead exposure,[20-24] others have not.[17,18,25]

The aim of this study was to evaluate the relationship between renal dysfunction and occupational lead exposure in workers from a plant in Singapore producing PVC stabilisers.

Subjects and Methods

Subjects

The study population consisted of 80 male and female workers from a factory producing PVC stabilisers using lead ingots as raw materials. Twenty-three subjects were excluded due to a history of diabetes, hypertension or recent ingestion of analgesics, antipyretics or antibiotics. A further 2 subjects, 1 with thalassaemia and the other the only remaining female were excluded, leaving 55 male subjects for analysis. The workers worked 12-hour shifts and were rotated among various departments. Since 1982, workers from this factory have undergone 6-monthly statutory medical examinations. Records of their blood lead levels measured during each examination are kept in the Department of Community, Occupational and Family Medicine of the National University of Singapore. This study utilised these records as well as measurements taken during a routine examination in August 1999. Many of the subjects participated in a similar study 5 years ago.[23]

Field Work

The examination was conducted during the course of a workday. A field investigator enquired about the medical history, smoking history and recent drug ingestion. The workers' weight and height were taken and blood pressure measured. An occupational physician then verified the medical history and enquired about recent changes in working hours or environment as well as health complaints. Blood and urine samples were then taken. Blood samples for blood lead analysis were stored in heparinised lead-free polypropylene tubes. Urine samples for analysis of Uα1m and urinary β-2 microglobulin (Uβ2m) were buffered (pH 7.2). Finally, each worker was issued a bottle for a 4-hour urine collection. We collected all the bottles 4 hours after the last bottle was issued.

Laboratory Analysis

All urine and blood samples were transported within 2 hours to the abovementioned laboratory. Urine samples were transported in an ice box. Blood lead concentration (PbB) was analysed using atomic absorption spectrophotometry (Varian Spectra AA-30) with a graphite furnace. External quality control has been done yearly under the National External Quality Assurance Scheme (England) and Inter-Laboratory Comparison Programme (Canada) with excellent results. Volume of each 4-hour urine collection was measured. An aliquot from each collection was taken for analysis of creatinine. Serum and urine samples were stored at -30^0C until analysis, which was done within 3 months. Urine and serum creatinine concentrations were determined using standard laboratory techniques. Urinary albumin (Ualb), serum α−1 microglobulin (Sα1m), serum β-2 microglobulin (Sβ2m) Uα1m, Uβ2m, and URBP were measured by enzyme-linked immunosorbent assay (ELISA) using commercially available polyclonal antibodies or test kits. Total urinary activity of N-acetyl-β-D-glucosaminidase (NAG-T) was determined using Noto's method.[26] The isoenzymes were separated by heat treatment according to Tan et al[26] and NAG-B activity was measured.

Data Analysis

The number of times each subject's PbB exceeded 40 μg/dL (PbB40) and cumulative blood lead index (PbBint) were obtained from previous PbB values and the present measurement. PbBint was calculated using the following formula:

$$PbBint = 0.5\Sigma(t_{i+1} - t_i)(PbB_i + PbB_{i+1}),$$

where PbB_i and PbB_{i+1} are the ith and (i+1)th PbB levels respectively and $(t_{i+1} - t_i)$ is the time between the 2 readings. Creatinine clearance was corrected for a body surface area of 1.74 m². It was also compared with the predicted creatinine clearance from serum creatinine using the formula proposed by Cockcroft and Gault.[27] NAG-A activity was calculated by subtracting NAG-B from NAG-T. The ratio of NAG-B activity to total NAG activity (NAG-B/NAG-T) was also computed.

Analysis was done using the SPSS statistical software package. Urinary parameters were corrected for urine flow and expressed per gram creatinine. The distributions of PbB40, PbBint, Ualb, Sβ2m, Uβ2m, URBP, NAG-T, NAG-A and NAG-B/NAG-T were highly positively skewed, so logarithmic transformation was used to normalise them. The distributions of Uα1m and NAG-B were less highly skewed; therefore, square-root transformation was used. As Uβ2m is unstable in acidic pH, it was used for data analysis only when urine pH >5.5. Analysis of variance was used to compare means of effect markers. Analysis of covariance was used to adjust the means for smoking habit (ever-smoker, non-smoker) and age.

Results

Study Population

Fifty-five male subjects with a mean age of 36 years were analysed. There was an equal number of smokers and non-smokers. The mean PbB was 24.1 µg/dL, with 4 subjects' PbB exceeding 40 µg/dL. The mean creatinine clearance was 120.9+/-14.9 mL/min/1.73m^2, with 2 subjects' (3.6%) creatinine clearance falling below 90 mL/min/1.73m^2 (Table I).

TABLE I: DEMOGRAPHICS AND EXPOSURE MARKERS IN 55 WORKERS EXPOSED TO LEAD

Age (y)	
Mean	35.73
SD	9.59
Sex	
Male	55
Smoking	
Never-smoked	27 (49.1%)
Ever-smoked	28 (50.9%)
PbB (µg/dL)	
Mean	24.1
SD	9.6
Range	7.7-45.3
PbB40	
Mean (geometric mean)	1.9
Range	0-24
PbBint (µg years/dL)	
Mean (geometric mean)	880.64
Range	136.00-6342.00

PbB: blood lead concentration; PbB40: number of times a subject's blood lead concentration exceeded 40 µg/dL; PbBint: cumulative blood lead index, where PbBint = $0.5\Sigma(t_{i+1} - t_i)(PbB_i + PbB_{i+1})$

Relationships between Exposure and Effect Indices

PbB was less than 20 µg/dL in 18 (32.7%) subjects, between 20 and 30 µg/dL in 23 (41.8%), and greater than 30 µg/dL in 14 (25.5%). There was no significant difference in all effect markers between these groups.

PbB40 was 0 in 24 (43.6%) subjects, 1-4 in 14 (25.5%) and 5 or more in 17 (30.9%). Creatinine clearance decreased significantly with increasing PbB40 before and after adjustment for age and smoking habit. Ualb, Uα1m, URBP, NAG-T and NAG-B increased significantly with increasing PbB40 before and after adjustment. Sα1m and NAG-T increased significantly with increasing PbB40 but this became insignificant after adjustment.

PbBint was less than 300 µg years/dL in 14 (25.5%) subjects, between 300 and 1500 µg years/dL in 23 (41.8%) and greater than 1500 µg years/dL in 18 (32.7%). Creatinine clearance decreased significantly with increasing PbBint

before and after adjustment. Ualb, Uα1m, Uβ2m and URBP increased significantly with increasing PbBint before and after adjustment (Table II).

To further examine the relationship between PbB40 and the renal markers, subjects with PbB40 = 0 (n = 24, 43.6%) and PbB40 ≥1 (n = 31, 56.4%) were compared. Creatinine clearance was significantly lower in the latter group after

TABLE II: RELATIONSHIP BETWEEN PbBint AND RENAL DYSFUNCTION IN LEAD WORKERS

	PbBint (µg years/dL)			
	<300	300-1500	>1500	*P* value
Creatinine clearance (mL/min/1.73m²)				
N	14	23	18	
Adjusted mean*	125.7	124.1	113.2	0.019
Ualb (mg/g creatinine)				
N	14	23	18	
Adjusted mean*	5.08	5.25	11.54	0.008
Sα1m (mg/L)				
N	14	23	18	
Adjusted mean*	47.58	44.49	52.34	0.357
Sβ2m (µg/L)				
N	14	22	17	
Adjusted mean*	4819.48	5023.43	4931.74	0.917
Uα1m (mg/g creatinine)				
N	14	23	18	
Adjusted mean*	4.04	4.19	8.69	0.001
Uβ2m (µg/g creatinine)				
N	6	12	8	
Adjusted mean*	181.13	202.30	437.52	0.007
URBP (µg/g creatinine)				
N	14	23	18	
Adjusted mean*	1.74	1.70	4.10	0.002
NAG-T (U/g creatinine)				
N	14	23	18	
Adjusted mean*	1.79	1.90	2.46	0.243
NAG-B (U/g creatinine)				
N	14	23	18	
Adjusted mean*	0.50	0.49	0.64	0.369
NAG-A (U/g creatinine)				
N	14	23	18	
Adjusted mean*	1.23	1.31	1.66	0.557
NAG-B / NAG-T				
N	14	23	18	
Adjusted mean*	0.26	0.22	0.23	0.718

NAG : N-acetyl-β-D-glucosaminidase
         (T: total, A: heat-labile isoenzyme, B: heat-stable isoenzyme)
PbBint : cumulative blood lead index,
         where PbBint = $0.5\Sigma(t_{i+1} - t_i)(PbB_i + PbB_{i+1})$
Sα1m : serum α-1 microglobulin concentration
Sβ2m : serum β-2 microglobulin concentration
Uα1m : urinary α-1 microglobulin concentration
Ualb   : urinary albumin concentration
Uβ2m : urinary β-2 microglobulin concentration
URBP : urinary retinol-binding protein concentration
* Adjusted for age and smoking habit by ANCOVA

TABLE III: COMPARISON OF RENAL PARAMETERS IN WORKERS WITH PbB40 = 0 AND PbB40 ≥1

	PbB40		
	0	≥1	*P* value
Creatinine clearance (mL/min/1.73m^2)			
N	24	31	
Adjusted mean*	130.4	113.6	0.000
Confidence interval	125.6 – 135.2	109.4 – 117.8	
Ualb (mg/g creatinine)			
N	24	31	
Adjusted mean*	4.98	8.51	0.031
Confidence interval	3.48 – 7.13	6.21 – 11.64	
Sα1m (mg/L)			
N	24	31	
Adjusted mean*	46.85	48.62	0.712
Confidence interval	39.74 – 53.95	42.39 – 54.84	
Sβ2m (μg/L)			
N	24	29	
Adjusted mean*	4920.40	4954.50	0.935
Confidence interval	4365.16 – 5559.04	4446.31 – 5533.50	
Uα1m (mg/g creatinine)			
N	24	31	
Adjusted mean*	2.84	8.03	0.000
Confidence interval	2.03 – 3.79	6.78 – 9.38	
Uβ2m (μg/g creatinine)			
N	11	15	
Adjusted mean*	187.93	308.32	0.050
Confidence interval	129.72 – 272.27	224.91 – 423.64	
URBP (μg/g creatinine)			
N	24	31	
Adjusted mean*	1.59	3.01	0.007
Confidence interval	1.14 – 2.23	2.24 – 4.05	
NAG-T (U/g creatinine)			
N	24	31	
Adjusted mean*	1.66	2.39	0.020
Confidence interval	1.32 – 2.08	1.96 – 2.91	
NAG-B (U/g creatinine)			
N	24	31	
Adjusted mean*	0.40	0.66	0.009
Confidence interval	0.29 – 0.53	0.53 – 0.80	
NAG-A (U/g creatinine)			
N	24	31	
Adjusted mean*	1.12	1.64	0.098
Confidence interval	0.81 – 1.57	1.23 – 2.20	
NAG-B/NAG-T			
N	24	31	
Adjusted mean*	0.22	0.25	0.460
Confidence interval	0.17 – 0.28	0.20 – 0.31	

NAG : N-acetyl-β-D-glucosaminidase (T: total, A: heat-labile isoenzyme, B: heat-stable isoenzyme)
PbB40 : number of times a subject's blood lead concentration exceeded 40 μg/dL
Sα1m : serum α-1 microglobulin concentration
Sβ2m : serum β-2 microglobulin concentration
Uα1m : urinary α-1 microglobulin concentration
Ualb   : urinary albumin concentration
Uβ2m : urinary β-2 microglobulin concentration
URBP : urinary retinol-binding protein concentration
*Adjusted for age and smoking habit by ANCOVA

adjustment. Ualb, Uα1m, Uβ2m, URBP, NAG-T and NAG-B were significantly higher in this group after adjustment. There was no overlap in the 95% confidence intervals of the 2 groups for creatinine clearance, Uα1m and URBP, suggesting that these are more highly correlated with PbB40 (Table III).

Discussion

Exposure

The mean PbB is very low and approximates to that of the control groups in some studies,[17,19] indicating that our population is currently exposed to low levels of lead.

In our study, apart from PbB, PbB40 and PbBint were also used to indicate lead exposure. Due to poor excretion, much of the lead that is absorbed by the body is accumulated over time in the bones and soft tissues.[28] This accumulated lead contributes to systemic blood lead levels even after exposure has ceased,[29] causing adverse effects on various body systems. Total body lead should therefore have better correlation with lead nephropathy than recent blood lead. The ideal indicator of total body lead burden is naturally bone lead, which can be measured by X-ray fluorescence—this, however, requires expensive equipment and skilled expertise. PbBint and PbB40 give indirect measures of total body lead by quantifying overall lead exposure. PbBint has been found to correlate well with tibial bone lead (PbT),[30] and is relatively easily computed from serial PbB measurements. PbB40 is easily obtained from serial measurements, and has the added advantage of being easy to use in clinical practice.

Relationships

There was no dose-response relationship between PbB and any marker of renal dysfunction. This result is similar to that of Gennart et al.[17]

Creatinine clearance decreased significantly (P <0.001) with both increasing PbBint and increasing PbB40. This finding parallels another study[20] where a positive dose-response relationship was found between serum *creatinine* and historical blood lead (calculated by dividing PbBint by duration of exposure). In contrast, Roels et al reported *positive* correlation between PbT and creatinine clearance before and after protein-induced hyperfiltration.

Amongst the putative early markers of lead nephropathy, we found a dose-response relationship between Ualb, Uα1m, Uβ2m and URBP and both PbB40 and PbBint, as well as between NAG-T and NAG-B and PbB40. Again, both similar and contradictory results have been reported in the literature. Chia et al[23] found significantly higher Uα1m amongst exposed workers compared to controls, and also good correlation between PbBint and Uα1m. Bernard and Lauwerys[30] reviewed 5 previous studies and found that

urine NAG appeared to be the only marker elevated in early nephropathy. Ehrlich et al[20] and Endo et al[31] reported the same observation. Chia et al[32] reported that elevation of urine NAG correlates less well with PbBint than recent increase in PbB, suggesting that NAG reflected acute injury rather than chronic lead nephropathy, but Ehrlich et al[20] could not find any association between NAG and recent change in blood lead. Roels et al[18] did not find significant difference in Ualb, Uβ2m and URBP between exposed and controls despite the former group having a three-fold higher PbB and PbT than the latter. To put things in perspective though, the clinical significance of these markers is as yet unknown. Furthermore, our study is limited by the small number of subjects and the lack of a control group. Therefore, we suggest that cohort studies be done to delineate the relationship between these markers and lead nephropathy. If such studies show that changes in these markers precede clinically evident renal dysfunction or abnormal glomerular function tests, their role as early markers of lead nephropathy becomes certain.

Finally, subjects with PbB40 = 0 and PbB40 ≥1 were found to have significantly different creatinine clearance, Ualb, Uα1m, Uβ2m, URBP, NAG-T and NAG-B. This suggests that workers with one or more PbB40 measurements exceeding 40 μg/dL have some degree of renal dysfunction.

Conclusions

We found a positive association between indices of overall lead exposure, PbBint and PbB40, and renal dysfunction. We also found that workers with one or more measurements of PbB exceeding 40 μg/dL have some degree of renal impairment. Our findings also strengthen the case for the use of Ualb, Uα1m, Uβ2m, URBP, NAG-T and NAG-B as early markers of lead nephropathy.

REFERENCES

1. Ritz E, Mann J, Stoeppler M. Lead and the kidney. Adv Nephrol Necker Hosp 1988; 17:241-74.

2. Selevan S G, Landrigan P J, Stern F B, Jones J H. Mortality of lead smelter workers. Am J Epidemiol 1985; 122:673-83.

3. McMichael A J, Johnson H M. Long-term mortality profile of heavily-exposed lead smelter workers. J Occup Med 1982; 24:375-8.

4. Cooper W C, Gaffey W R. Mortality of lead workers. J Occup Med 1975; 17:100-7.

5. Baker E L Jr, Landrigan P J, Barbour A G, Cox D H, Folland D S, Ligo R N, et al. Occupational lead poisoning in the United States: clinical and biochemical findings related to blood lead levels. Br J Ind Med 1979; 36:314-22.

6. Lilis R, Gavrilescu N, Nestorescu B, Dumitriu C, Roventa A. Nephropathy in chronic lead poisoning. Br J Ind Med 1968; 25:196-202.

7. Nye L J J. An investigation of the extraordinary incidence of chronic nephritis in young people in Queensland. Med J Aust 1929; 2:145.

8. Emmerson B T. Chronic lead nephropathy: the diagnostic use of calcium edetate and the association with gout. Aust Ann Med 1963; 12:310.

9. Tepper L B. Renal function subsequent to childhood plumbism. Arch Environ Health 1963; 7:76.

10. Hu H. A 50-year follow-up of childhood plumbism: hypertension, renal function, and hemoglobin levels among survivors. Am J Dis Child 1991; 145:681-7.

11. Morgan J M, Hartley M W, Miller R E. Nephropathy in chronic lead poisoning. Arch Intern Med 1966; 118:17-29.

12. Inglis J A, Henderson D A, Emmerson B T. The pathology and pathogenesis of chronic lead nephropathy occurring in Queensland. J Pathol 1978; 124:65-76.

13. Goyer R A, Rhyne B C. Pathological effects of lead. Int Rev Exp Pathol 1973; 12:1-77.

14. Khalil-Manesh F, Gonick H C, Cohen A H, Alinovi R, Bergamaschi E, Mutti A, et al. Experimental model of lead nephropathy: I. Continuous high-dose lead administration. Kidney Int 1992; 41:1192-203.

15. Goyer R A. Mechanism of lead and cadmium nephrotoxicity. Toxicol Lett 1989; 46:153-62.

16. Khalil-Manesh F, Gonick H C, Cohen A H. Experimental model of lead nephropathy: III. Continuous low-level lead administration. Arch Environ Health 1993; 48:271-8.

17. Gennart J P, Bernard A, Lauwerys R. Assessment of thyroid, testes, kidney and autonomic nervous system function in lead-exposed workers. Int Arch Occup Environ Health 1992; 64:49-57.

18. Roels H, Lauwerys R, Konings J, Buchet J P, Bernard A, Green S, et al. Renal function and hyperfiltration capacity in lead smelter workers with high bone lead. Occup Environ Med 1994; 51:505-12.

19. Pinto de Almeida A R, Carvalho F M, Spinola A G, Rocha H. Renal dysfunction in Brazilian lead workers. Am J Nephrol 1987; 7:455-8.

20. Ehrlich R, Robins T, Jordaan E, Miller S, Mbuli S, Selby P, et al. Lead absorption and renal dysfunction in a South African battery factory. Occup Environ Med 1998; 55:453-60.

21. Staessen J A, Lauwerys R R, Buchet J P, Bulpitt C J, Rondia D, Vanrenterghem Y, et al. Impairment of renal function with increasing blood lead concentration in the general population: the Cadmibel study group. N Engl J Med 1992; 327:151-6.

22. Cardenas A, Roels H, Bernard A M, Barbon R, Buchet J P, Lauwerys R R, et al. Markers of renal changes induced by industrial pollutants: II. Application to workers exposed to lead. Br J Ind Med 1993; 50:28-36.

23. Chia K S, Jeyaratnam J, Lee J, Tan C, Ong H Y, Ong C N, et al. Lead induced nephropathy: relationship between various biological exposure indices and early markers of nephrotoxicity. Am J Ind Med 1995; 27:883-95.

24. Verschoor M, Wibowo A, Herber R, van Hemmen J, Zielhuis R. Influence of occupational low-level lead exposure on renal parameters. Am J Ind Med 1987; 12:341-51.

25. Buchet J P, Lauwerys R, Roels H, Bernard A. Relationship between exposure to heavy metals and prevalence of renal dysfunction. Arch Toxicol Suppl 1980; 4:215-8.

26. Tan C, Cheng L Y, Chia K S, et al. Separation of N-acetyl-β-D-glucosaminidase isoenzymes by heat treatment: its reproducibility. J Med Lab Sci 1991; 5:13-5.

27. Cockcroft D W, Gault M H. Prediction of creatinine clearance from serum creatinine. Nephron 1976; 16:31-41.

28. Philip A T, Gerson B. Lead poisoning—Part I. Incidence, etiology and toxicokinetics. Clin Lab Med 1994; 14:423-44.

29. Erkkila J, Armstrong R, Riihimaki V, Chettle D R, Paakkari A, Scott M, et al. In vivo measurements of lead in bone at four anatomical sites: long term occupational and consequent endogenous exposure. Br J Ind Med 1992; 49:631-44.

30. Bernard A, Lauwerys R. Epidemiological applications of early markers of nephrotoxicity. Toxicol Lett 1989; 46:293-306.

31. Endo G, Horiguchi S, Kiyota I. Urinary N-acetyl-β-D-glucosaminidase activity in lead-exposed workers. J Appl Toxicol 1990; 10:235-8.

32. Chia K S, Mutti A, Tan C, Ong H Y, Jeyaratnam J, Ong C N, et al. Urinary N-acetyl-β-D-glucosaminidase activity in workers exposed to lead. Occup Environ Med 1994; 51:125-9.