Cadmium, NAG activity, and β_2 -microglobulin in the urine of cadmium pigment workers

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ABSTRACT Cadmium (Cd), N-acetyl- β -D-glucosaminidase (NAG) activity, β_2 -microglobulin (BMG), and creatinine (cr) in urine were measured during April and September 1986 in workers exposed to cadmium pigment dust (maximum exposure $3.0 \mu\text{g}/\text{m}^3/8 \text{ h}$ for respirable dust). In April and September urinary Cd ranged from 0.2 to 9.5 and from 0.5 to 7.0 $\mu\text{g}/\text{g cr}$ with a geometric mean of 0.7 and 1.2 $\mu\text{g}/\text{g cr}$, respectively. The correlation coefficient between Cd and NAG was 0.261 ($n = 61$) in April and 0.389 ($n = 50$) in September. The correlation coefficient between Cd and BMG was 0.241 ($n = 63$) in April and 0.115 ($n = 50$) in September. It appears that urinary Cd concentrations have a closer relation with urinary NAG than urinary BMG, even when urinary Cd concentrations are less than 10 $\mu\text{g}/\text{g cr}$. It is concluded that NAG is a more sensitive indicator of Cd absorption than BMG even at urinary Cd concentrations of less than 10 $\mu\text{g}/\text{g cr}$.

The renal cortex is a critical organ after long term exposure of cadmium (Cd) and the excretion of low molecular weight protein in the urine after tubular dysfunction is considered to be the earliest sign of Cd poisoning.^{1,2} The histopathological change in the kidney associated with Cd is an increase in the size and number of lysosomes in the tubular cells.³ N-acetyl- β -D-glucosaminidase (NAG) is a lysosomal enzyme abundant in the renal proximal tubules with a molecular weight of 120 000; it is normally not filtered from the glomerulus.

NAG activity in the urine increases with renal damage, during treatment with some drugs, in tubulointerstitial disease and glomerular disorders, and after the development of diabetic microangiopathy and renal hypertension. It is also a good indicator for the follow up of activity of patients with renal transplants. β_2 -Microglobulin (BMG), a major component of the low molecular weight proteins in the urine of Cd workers (Mol wt = 11 800), is freely filtered from the glomerular basement membrane and completely resorbed by the proximal tubules. It increases with renal tubular dysfunction, in acute pyelonephritis, after surgical or accidental trauma or burn injuries, and the administration of certain drugs.

In the present study we have measured concentrations of urinary Cd, NAG, and BMG in workers employed at a pigment manufacturing factory to assess the usefulness of urinary NAG activity and

BMG concentrations as biological indicators of Cd absorption.

Subjects and methods

All the subjects were workers in a factory manufacturing Cd pigments, CdS and CdSe. They were divided into five groups (A to E) in April and four groups (B to E) in September according to the jobs they performed. Table 1 gives the number in each group, their ages, and job histories in April. Group A consisted of young newcomers on a rotation scheme and group B workers not exposed to Cd; both groups were non-exposed. In April in Japan newcomers enter a company and work in a rotation scheme. In this study in September the workers were assigned to sections, thereby eliminating group A. Group C consisted of synthetic resin manufacturing workers exposed to atmospheric Cd to a negligible degree. Groups D and E were Cd pigment manufacturing workers. Group D was placed in a quality control section, where the Cd content in the atmosphere was far less than for group E.

In May 1986 the atmospheric concentration of Cd, measured at seven points on group E worksite, ranged from 3 to 350 $\mu\text{g}/\text{m}^3$. It was highest at the canning process (group E). In December 1986 daily individual exposure to Cd was estimated for one worker of group D and two workers of group E. The mean respirable concentrations of Cd dust of groups D and E workshops were 0.18 $\mu\text{g}/\text{m}^3$ and 3.0 $\mu\text{g}/\text{m}^3$, respectively. The highest exposure to Cd was observed in group E.

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Table 1 Some characteristics of workers exposed (groups C, D, E) and non-exposed (groups A and B) to cadmium (April 1986) (Arithmetic mean $\pm$ 1 SD in parentheses)

Group	Exposed				Non-exposed		
	Total	C	D	E	Total	A	B
No of workers	29	10	11	8	35	18	17
Mean age (y)	34.6 (9.26)	31.1 (8.99)	34.7 (8.86)	38.8 (9.48)	25.5 (6.18)	21.8 (2.48)	29.5 (6.53)
Duration of factory employment (y)	12.8 (8.35)	9.6 (8.10)	13.5 (6.93)	15.9 (9.96)			9.2 (6.52)

followed by groups D and C in decreasing order of exposure.

Cd in the urine was measured by a flameless atomic absorption spectrophotometer (Hitachi, type 180-30) using the method of Koizumi *et al.*⁴ BMG was measured by enzyme immunoassay using a kit obtained from Fujirebio Inc and NAG with the Shionogi NAG test kit.⁵ Cr was measured by the Folin-Wu method. Urine specimens were stored at 4-6°C and measured within a few days.

Results

BIOLOGICAL STUDIES IN APRIL

Table 2 gives the geometric mean values, geometric standard deviations of Cd, NAG activities, and BMG of the five groups. The mean concentration of Cd in the urine increased with exposure to Cd. There was a significant difference between the means of the non-exposed groups and all exposed groups except between groups B and C.

There was a significant correlation between age and urinary Cd concentration for the non-Cd-exposed groups (groups A and B), indicating that urinary Cd increases with age ($r = 0.504$; $p < 0.005$). This was also true of the exposed groups (groups C, D, and E) ($r = 0.433$; $p < 0.05$). The urinary Cd concentration of the exposed groups was higher than of the non-exposed groups. The regression equation for urinary Cd ($Y \mu\text{g/g cr}$) against age (X years) was found to be $Y = 0.0195X + 0.004$ for the non-exposed groups and $Y = 0.0447X - 0.223$ for the exposed groups.

Fig 1 Relation between urinary cadmium ($\mu\text{g/g cr}$) and NAG activity levels. (Geometric mean and SD are also shown (in April 1986)).

The slope of the regression line for the latter was twice that of the former.

A scattergram of the urinary Cd concentrations and NAG activity is shown in fig 1 with a log scale. There was a low but significant correlation between urinary Cd and NAG ($r = 0.261$; $n = 61$) with urinary Cd

Table 2 Geometric mean value of biological indicators of cadmium pigment workers divided into five groups according to level of exposure. (Geometric mean value $\pm$ 1 geometric SD in parentheses (in April 1986))

Group	No of subjects	Cd-U $\mu\text{g/g cr}$	NAG-U U/g cr	BMG-U $\mu\text{g/g cr}$
Total	64	0.7 (0.3~1.6)	2.1 (1.2~3.7)	82 (40~167)
Non-exposed	35	0.5 (0.3~0.8)	1.8 (1.1~3.0)	72 (44~118)
A	18	0.4 (0.3~0.6)	1.9 (1.1~3.2)	69 (45~106)
B	17	0.6 (0.3~1.1)	1.7 (1.0~2.8)	75 (42~132)
Exposed	29	1.1 (0.5~2.5)	2.6 (1.5~4.5)	97 (40~236)
C	10	0.9 (0.5~1.4)	2.8 (1.7~4.8)	131 (59~291)
D	11	1.0 (0.6~1.9)	2.1 (1.3~3.4)	92 (47~182)
E	8	1.7 (0.5~5.8)	3.0 (1.6~5.8)	72 (22~231)

Table 3 Geometric mean value of biological indicators of cadmium pigment workers divided into four groups according to level of exposure. (Geometric mean value $\pm$ 1 geometric SD in parentheses (in September 1986))

Group	No of subjects	Cd-U $\mu\text{g/g cr}$	NAG-U U/g cr	BMG-U $\mu\text{g/g cr}$
Total	50	1.2 (0.7~2.3)	2.9 (1.7~5.0)	125 (62~251)
Non-exposed = B	18	1.0 (0.8~1.2)	2.3 (2.1~2.6)	130 (107~158)
Exposed	32	1.4 (0.8~2.7)	3.3 (1.9~6.0)	122 (54~276)
C	13	1.1 (0.9~1.3)	3.3 (3.0~3.7)	106 (79~142)
D	9	1.8 (1.1~2.9)	2.7 (1.3~5.6)	188 (116~305)
E	10	2.3 (1.3~4.1)	4.1 (3.2~5.3)	98 (29~332)

concentrations of 10 $\mu\text{g/g cr}$ or less. There was also a low but significant correlation between urinary Cd and BMG ($r = 0.241$; $n = 63$).

BIOLOGICAL STUDIES IN SEPTEMBER

Table 3 shows geometric mean values, geometric standard deviations of Cd, NAG activities, and BMG of the four groups. The mean concentration of Cd in urine increased with exposure to Cd and the means of groups B (non-exposed group) and E ($p < 0.01$) differed significantly.

Again, a significant correlation was found between age and urinary Cd concentration for the groups exposed to Cd (groups C, D and E) ($r = 0.490$; $p < 0.005$). The regression equation of urinary Cd ($Y \mu\text{g/g cr}$) against age (X years) was $Y = 0.0777X - 0.781$ for the exposed groups. In this survey there was no correlation between age and urinary Cd concentration of the non-exposed groups.

Fig 2 Relation between urinary cadmium ($\mu\text{g/g cr}$) and NAG activity levels. (Geometric mean and SD are also shown (in September 1986)).

The correlation between log transformed urinary Cd concentrations and NAG activity is shown in fig 2. A significant correlation was evident between urinary Cd and NAG ($r = 0.389$; $n = 50$) but none was found between urinary Cd and BMG ($r = 0.115$; $n = 50$).

Discussion

We found a significant correlation between urinary Cd and NAG activity with urinary Cd concentrations about 1 $\mu\text{g Cd/g cr}$ on two occasions. Sugihira *et al* observed that urinary NAG activity of people living in a Cd polluted area correlated significantly with urinary Cd concentration⁶ but the mean Cd concentration in urine in their study was in excess of 5 $\mu\text{g/g cr}$. Tsuchiya *et al* also observed a significant correlation between BMG and Cd in the urine from inhabitants in a Cd polluted area⁷ with a mean urinary Cd greater than 6 $\mu\text{g/l}$.

Harada *et al* by contrast, observed that the increase in BMG in the urine does not occur below urinary Cd concentrations of 12 $\mu\text{g/g cr}$ and found no correlation between them⁸ but their data were obtained from 11 subjects only. Bernard *et al* found BMG to be significantly correlated with urinary Cd ($r = 0.74$, $n = 42$)⁹ with urinary Cd concentrations above a mean of about 10 $\mu\text{g Cd/g cr}$ but not at lower concentration. This conclusion is supported by Buchet *et al*.¹⁰ In our study an increase in BMG in the urine was not found when Cd concentrations were 10 $\mu\text{g Cd/g cr}$ or less. There was a low but significant correlation between Cd and BMG in April but not in September.

In the present study urinary Cd concentrations were less than 10 $\mu\text{g Cd/g cr}$ with a geometric mean of 0.7 $\mu\text{g/g cr}$ in April and 1.2 $\mu\text{g/g cr}$ in September. We conclude that: (1) significant correlations exist between urinary Cd under 10 $\mu\text{g/g cr}$ and NAG activity on the basis of two observations; (2) a significant correlation between urinary Cd under 10 $\mu\text{g/g cr}$ and BMG level was not reproduced on the second occasion, and (3) the correlation between NAG activity and Cd is higher than that between urinary Cd and BMG level. Measurement of urinary NAG activity is not only sensitive but relatively simple, inexpensive, and reliable. A urinary NAG assay is thus

recommended for the biological monitoring of exposure to Cd.

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