Plasma Metallothionein Antibody and Cadmium-Induced Renal Dysfunction in an Occupational Population in China

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It has been reported that anti-metallothionein (a metallothio- nein antibody) is present in the circulation of healthy subjects and in patients suffering from atopic dermatitis. The aim of this study was to investigate whether cadmium-induced renal dysfunction is related to the presence of the plasma metallothionein antibody (MT-Ab) in workers exposed to cadmium (Cd) occupationally. Plasma metallothionein antibody was determined by enzyme linked immuno sorbent assay (ELISA) techniques, and both exposure assessment and risk assessment were conducted in cadmium-exposed workers in China. We demonstrate that there is a significantly increased prevalence of renal dysfunction with respect to the level of urinary cadmium in a dose-dependant manner. We found no significant correlations between the levels of MT-Ab and the external or internal exposure doses of cadmium (p > 0.05), but the levels of MT-Ab did correlate positively with two biomarkers of renal dysfunction—urinary β2-microglobulin (UB2M; r = 0.218, p < 0.05) and N-acetyl-β-D-glucosaminidase (UNAG; r = 0.302, p < 0.001)—in the cadmium-exposed workers. Workers who have high levels of MT-Ab display cadmium-induced tubular nephrotoxicity more frequently than those possessing low levels of MT-Ab; odds ratio (OR) 4.2; 95% confidence intervals 1.2–14.5 (p < 0.05). This study suggests that subjects that have higher MT-Ab levels more readily develop cadmium-induced renal dysfunction. Thus, the levels of plasma MT-Ab can be used as a biomarker of susceptibility to renal dysfunction in occupational cadmium exposure.

Key Words: cadmium; metallothionein; renal dysfunction; autoantibody; susceptibility.

INTRODUCTION

Metallothioneins (MTs) are a family of stress proteins containing a high content of cysteine and divalent metals. Metallothionein genes are expressed ubiquitously in many tissues on basal levels. Their expression is induced by many factors, especially by metal ions such as cadmium, zinc, and copper. A number of other agents—including free radicals, irradiation, acute phase cytokines (e.g., interleukin-1, interleukin-6, and tumor necrosis factor-α), inflammatory agents [e.g., lipopolysaccharide (LPS)], and alkylating agents (Haq et al., 2003; Kagi, 1991)—can induce the synthesis of MTs. The physiological function of MTs remains under study. Metallothioneins have been suggested to play a role in the homeostasis of essential trace elements such as zinc and copper, in the detoxification of certain toxic metals, and as scavengers of free radicals. Studies on the metabolism of some toxic metals, especially cadmium, have identified the role of MTs in the absorption, transportation, and excretion of metals (Jin et al., 1998; Nordberg et al., 1971; Nordberg, 1998; Nordberg and Nordberg, 2000). Four major forms, MT 1–4, have been identified to date, as have a number of isoforms of human MT-1. Although MT-1 and MT-2 are expressed in many tissues, they are found mostly in liver and kidney tissue; MT-3 is found in the brain, and MT-4 is expressed in keratinocytes.

Cadmium, an environmental pollutant, causes a number of adverse health effects (Nordberg et al., 1971; WHO, 1992), particularly toward the kidneys (WHO, 1992). When taken up in the body, cadmium circulates initially in plasma primarily bound to albumin; cadmium then enters the liver where it is bound to metallothionein and is later released into the bloodstream (Nordberg et al., 1971; WHO, 1992). Metallothionein-bound cadmium is readily filtered through the renal glomerulus and reabsorbed from the glomerular filtrate by proximal tubule cells. Cadmium bound to metallothionein is degraded in tubular lysosomes to release free cadmium, which then induces the synthesis of renal cell metallothionein. Renal damage is believed to occur once free cadmium or an excessive concentration of non-MT-bound cadmium increases within the cell. The ability of an individual to synthesize or utilize MT after exposure to cadmium affects his or her susceptibility to cadmium toxicity. Lu et al. (2001, 2004, 2005) found that individuals displaying a low induced expression of MT in peripheral lymphocytes were more susceptible to cadmium-induced toxicity.

The authors certify that all research involving human subjects was done in full compliance with all government policies and the Helsinki Declaration.

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nephrotoxicity upon both occupational and environmental exposure. Animal studies (Klaassen et al., 1999) indicated that MT knockout animals that cannot synthesize MT are more susceptible to cadmium nephrotoxicity.

Many reports have indicated that antibodies against heat shock proteins, which are another class of stress proteins, are present in the sera of healthy subjects and workers subjected to toxic chemical stress (Kindas-Mugge et al., 1993; Pockley et al., 1998; Wu et al., 1996, 1998). The antibody to MT is present in human serum, as are antibodies to heat shock proteins, particularly in patients suffering from atopic dermatitis who have metal allergies (Jin et al., 2003). Jin GB et al. (2002a) investigated the effect that MT-Ab induction has on mercury-induced bone injury. BALB/c mice were injected with MT to induce MT-Ab and then they were treated with HgCl2. Metallothionein immunization in conjunction with HgCl2 treatment dramatically decreased the bone mineral density (BMD), the humoral bone formation indices, the alkaline phosphatase (ALP) activity, and the osteocalcin levels. These results indicate that MT-Ab induction increases the susceptibility to mercury-induced bone injury in BALB/c mice and enhances the occurrence of mercury-related immune disorders. The relationship between MT-Ab, cadmium exposure, and cadmium-induced renal dysfunction, however, has yet to be studied. The aim of this study was to investigate the effect that MT antibody has on the renal dysfunction induced by cadmium in a workplace in China; we measured the levels of plasma MT antibody using enzyme-linked immunoabsorbent assay (ELISA) methods and evaluated the relationship between the levels of MT antibody and the indicators of renal dysfunction.

MATERIALS AND METHODS

Studied population. Eighty-five workers in a cadmium smelter located in east Hunan Province, central China, were selected to be the exposed population. Twenty-nine health care officers in the factory hospital were selected to be the control group. All of the subjects studied were men between 25 and 55 years of age. Each participant was interviewed by a trained and supervised interviewer and also completed a detailed questionnaire. Personal data—such as living customs, social and economic conditions, and lifestyles, including smoking habits, drinking habits, and history of medicines taken and metal exposure—were collected. Blood and urine samples were collected for biological measurements.

This study was carried out with the permission of both the local authorities and the Ethics Committee of Fudan University, and with the consent of each participant. The responsible Chinese principal investigators stress that participation was on a completely voluntary basis.

Bioanalysis and estimation of cadmium uptake. Spot urine samples were collected in acid-washed containers and stored at −70°C until required for analysis. Several of the urine containers were checked by atomic absorption spectrometry (AAS) to confirm the absence of cadmium contamination. The cadmium concentrations in urine and blood were analyzed through graphite furnace atomic absorption spectrometry (GF-AAS). Analytical quality assurance was taken into account by using both calibration standards and one run-through of reference materials (Seronorm Trace Elements Urine; Nycomed, Oslo, Norway) during every assay. The recoveries were 87.8–113.1%, and the coefficients of variation (CV) were 4.7–13.7%. Urinary albumin (UALB), N-acetyl-β-D-glucosaminidase (UNAG), and urinary β2-microglobulin (UB2M) were used as indices of renal dysfunction. The levels of UB2M and UALB were measured by means of an ELISA; the levels of UNAG were measured as described by Tucker et al. (1975). The levels of creatinine (Cr) were measured using the Jaffe reaction method. Urinary MT levels were determined through an ELISA (Cousins, 1991). All urinary parameters were adjusted for the levels of creatinine in urine. Quality control followed procedures similar to those described in a previous study (Jin et al., 2002b).

The cadmium uptake was estimated using the procedure described previously (Jin et al., 2004). Cadmium exposure occurred through both occupational and environmental exposure. The total uptake of cadmium (TTCd) is equal to the sum of the occupational and environmental uptakes. Cadmium uptake from occupational exposure was calculated on the basis of the average cadmium concentration in the air at the workplace; the environmental uptake was calculated based on the levels of rice and cigarette consumption.

Measurement of MT-Ab. Ninety-six-well microtiter plates (Corning Costar, Corning, NY, USA) were coated overnight with purified human fetus liver metallothionein (100 μl/well)—which was purified in our laboratory according to a previously described method (Nordberg et al., 1975b)—at 1 μg/ml in 0.05 mol/l carbonate buffer (pH 9.6) at 4°C. After plates were washed with phosphate-buffered saline (pH 7.4) containing 0.05% (v/v) Tween-20 (PBST), phosphate-buffered saline (200 μl/well) containing 10% (v/v) fetuin bovine serum was added to both the human metallothionein-coated and non-coated plates, they were incubated for 1 h at 37°C to block any nonspecific binding. The plates were then washed with PBST, and the tested plasma (1:100; 100 μl/well) was added to both the metallothionein-coated and non-coated plates, which were then incubated for 1 h at 37°C. After five washes with PBST, peroxidase-conjugated goat anti-human IgG Fab2 (Vector Laboratories, Burlingame, CA, USA) was added (at 1:10,000 dilution), and the plates were incubated for 30 min at 37°C. The plates were then washed five times with PBST and developed by the addition of orthophenylenediamine (Sigma, St. Louis, MO, USA) in the presence of 0.003% (v/v) H2O2. The enzyme reaction was stopped with 2 mmol/l H2SO4 (50 μl/well), followed by reading absorbance at 492 nm (EIA reader Model ELX-800, Bio-Tek Instruments, Inc., Winooski, VT, USA). The optical density (OD) of the non-MT-coated plate was subtracted from that of the coated plate. All samples were assayed in duplicate. A prescreened high-titer sample was used as a positive control; it was assigned an optical density of 1.0 at 492 nm. Urinary MT levels were determined through an ELISA (Cousins, 1991). All urinary parameters were adjusted for the levels of rice and cigarette consumption.

Statistical methods. Analyses were undertaken using SPSS 11.5 statistical analysis software. Distributions of the biological measurements were normalized through logarithmic transformation. The data are expressed in terms of geometric means and 95% confidence intervals for geometric means. For comparisons between more than two groups, one-way analysis of variance (ANOVA) was used. A cut-off p value for statistical significance was set at p < 0.05.

RESULTS

Cadmium Exposure and Renal Dysfunction in the Study Population

Table 1 presents the main characteristics and results of exposure and effect measurements in both the control and exposed groups. Some of the basic characteristics—such as age and smoking and drinking habits—were comparable between the two groups. In this study, we used UCd, BCd, and TTCd as
indicators of cadmium exposure and retention. The levels of UCd and BCd and the total cadmium uptake in the exposed group were statistically higher than those in the control group (\(p < 0.01\)). The American Conference of Governmental Industrial Hygienists and China Health Criteria recommended a biological exposure index for cadmium exposure (UCd concentration) of 5 \(\mu g/g\) creatinine. In this study, most of the UCd values were below 5 \(\mu g/g\) Cr in the control group; only one was above this value. In contrast, the UCd levels were above 5 \(\mu g/g\) Cr in 23 of the 85 cadmium-exposed workers. Pearson correlation analysis indicated that the correlation coefficients between UCd and both BCd and TTCd were 0.56 and 0.39 (\(p < 0.01\)), respectively; the coefficient between BCd and TTCd was 0.40 (\(p < 0.01\)).

In this study, we used UNAG, UB2M, UMT, and UALB as the biomarkers of cadmium-induced renal damage. Table 1 lists the levels of these urinary biomarkers and provides a comparison of their levels within the control and exposed groups. We observe that the excretions of UB2M, UMT, and UALB in the exposed group were statistically higher than those in the control group (\(p < 0.05\)).

**Relationship between Urinary Cadmium and Renal Dysfunction**

Figure 1 displays the levels of the urinary indicators of renal dysfunction at different values of UCd. We found that in the group having UCd of up to 2–5 \(\mu g/g\) Cr, the levels of UMT were significantly higher than those found within the group having UCd < 2 \(\mu g/g\) Cr. The levels of UB2M and UNAG increased substantially at UCd ≥5 \(\mu g/g\) Cr; UALB increased significantly at UCd >10 \(\mu g/g\) Cr. There were significant correlations between UCd and each of UNAG, UB2M, UMT, and UALB: the Pearson correlation coefficients were 0.36, 0.33, 0.45, and 0.24 (\(p < 0.01\)), respectively.

In this study, we define the cut-off points (abnormal values) for the criterion variables (B2M, NAG, UMT, and ALB) as the 90% upper limit values in the control group. The cut-off points for UNAG, UB2M, UMT, and UALB were 9.84 U/g Cr, 187.57 \(\mu g/g\) Cr, 388.78 ng/g Cr, and 13.45 mg/g Cr, respectively. We calculated the prevalence of hyper-NAGuria, hyper-B2Muria, hyper-MTuria, and hyper-ALBuria in all subjects at different levels of urinary cadmium (Fig. 2). When the UCd concentration was >5 \(\mu g/g\) Cr, we found that the prevalence of hyper-NAGuria, hyper-B2Muria, and hyper-MTuria increased significantly when compared with their prevalence at UCd <2 \(\mu g/g\) Cr. The prevalence of hyper-ALBuria increased dramatically, however, only when UCd was ≥10 \(\mu g/g\) Cr. This finding demonstrates clearly that there was a significantly increased, dose-dependent prevalence of hyper-NAGuria, hyper-B2Muria, hyper-MTuria, and hyper-ALBuria. These increases were statistically significant in the chi-squared test for the trend of relationship between the urinary cadmium levels and the prevalence of each renal dysfunction (\(p < 0.01\)).

**Cadmium Exposure and Plasma MT-Ab**

We observed no significant differences between the levels of plasma MT-Ab in the control and exposed groups (138.4 \(\pm\) 145.7 (Table 2). To further elucidate the relationship between the levels of MT-Ab and the cadmium exposure dose, we compared the values of the MT-Ab titers within various ranges of TTCd (0–50, 50–100, 100–150, >150 mg), BCd (0–5, 5–10, 10–20, >20 \(\mu g/l\)), and UCd (0–2, 2–5, 5–10, >10 \(\mu g/g\) Cr). Table 2 indicates that there were no statistically significant increases in the levels of MT-Ab with respect to the levels of UCd, BCd, or TTCd. According to the optimal density of MT-Ab, we divided the subjects into high (HMTAb, +) and low (LMTAb, −) levels of MT-Ab groups. The prevalence of HMTAb did not undergo any statistical increase as a result of
We performed Pearson correlation analyses between the levels of MT-Ab and the cadmium dose indicators. We did not observe any significant correlation between the levels of MT-Ab and those of BCd, UCd, TTCd, and UMT (Table 3). After correction for other factors that might affect the MT-Ab titer—such as age, number of working years, and smoking and drinking habits—we performed partial correlation analyses between the levels of MT-Ab and those of BCd, UCd, TTCd, and UMT. We did not observe any significant correlation between the levels of MT-Ab and the cadmium exposure dose as expressed through the levels of BCd, UCd, TTCd, and UMT (Table 3).

**Plasma MT-Ab Titer and Cadmium-Induced Renal Dysfunction**

We used Pearson correlation analyses to evaluate the potential for a relationship between the levels of MT-Ab and the occurrence of cadmium-induced renal dysfunction. There was a significant positive correlation between the levels of MT-Ab and those of UB2M \( (r = 0.186; p < 0.05) \) and UNAG \( (r = 0.228; p < 0.05) \) in all of the subjects, but we detected no such phenomenon between the levels of MT-Ab and UALB \( (r = 0.152; p > 0.1) \).

In addition, we analyzed these associations within both the exposed and the control groups. Figure 3a and 3b display plots of the correlations between the levels of MT-Ab and those of UNAG and UB2M, respectively, in the exposed workers. There was a significant positive correlation between the levels of MT-Ab and UNAG \( (r = 0.302; p < 0.001) \). This finding indicates that the levels of MT-Ab correlated very well with those of UNAG. We observed a similar significant correlation (Fig. 3b).
between the levels of MT-Ab and UB2M in the exposed workers ($r = 0.218, p < 0.05$). It was clear from Figure 3 that the levels of UCd, MT-Ab, UNAG, and UB2M all increased markedly. In the control group, the levels of UNAG and UB2M did not correlate significantly with the levels of MT-Ab ($r = 0.140$ and $0.047$, respectively; $p > 0.05$). We detected no obvious association between the levels of MT-Ab and UALB in either the exposed group or the control group ($r = 0.177$ and $0.063$, respectively; $p > 0.05$). We performed Pearson correlation analyses between the levels of MT-Ab and UMT in both the exposed and control groups. The coefficients between MT-Ab and UMT were $0.005$ ($p > 0.5$) in the exposed group and $0.286$ ($p > 0.1$) in the control group.

Next, we compared the prevalence of renal dysfunction between the high- and low-level MT-Ab groups. Table 4 indicates that the prevalence of hyper-B2Muria and hyper-NAGuria in the HMTAb group was statistically higher than that in the low MT-Ab group ($\chi^2 = 4.087$ and $6.904$, respectively; $p < 0.05$). In this study, we used UB2M and UNAG as biomarkers of renal tubular dysfunction. When the values of UB2M or UNAG were higher than the cut-off points, we counted the subjects as cases of renal tubular dysfunction. These cases were matched with normal subjects lacking renal tubular dysfunction based on their UCd levels; at the same time, we matched some other factors, such as smoking habits and age. We found that subjects displaying renal tubular dysfunction had significantly elevated values of MT-Ab titer than those displaying normal renal function (167.86 vs. 117.68, $p < 0.05$). Subsequently, a case-control study was performed to explore the relationship between different values of MT-Ab titer and renal tubular dysfunction (Table 5). This study indicates that subjects with higher levels of MT-Ab are more susceptible to cadmium-induced renal tubular dysfunction than are those in the low-level group ($\chi^2 = 5.438; p = 0.020; OR = 4.200; 95\% CI: 1.213–14.541$).

### DISCUSSION

In the present study, we demonstrate clearly that the smelter workers experienced a relatively high level of ongoing exposure to cadmium. The estimated total cadmium uptake, UCd and BCd, of the exposed workers was statistically higher than that of the control population. Within the occupationally
exposed group, 27.06% of the subjects had a level of urinary cadmium above 5 \( \mu g/g \) Cr, which is the recommended biological exposure limit in China. This exposure was high enough to cause a slight renal dysfunction (Table 1). When urinary cadmium exceeded 5 \( \mu g/g \) Cr, the levels and the prevalence of UB2M, UNAG, and UMT all increased dramatically; the level of UALB increased when the urinary cadmium dose reached \( \geq 10 \mu g/g \) Cr. These results on cadmium exposure and renal dysfunction are in accordance with the results of previous studies (Jin et al., 2002b, 2004).

It is known that toxic metals can interfere with the immune system (Cooper et al., 1999), yet it is noteworthy that metals such as mercury, lead, and cadmium can display immunostimulatory effects toward developing autoimmune disease (Ohsawa, 1997). It has been reported that elevated levels of circulating autoantibodies are found in animals and workers exposed to toxic metals, including cadmium (Bernard et al., 1984, 1987; Frenkel et al., 1994; Ohsawa et al., 1988). Workers heavily exposed to cadmium and nickel displayed high titers of anti-oxidized DNA base autoantibodies (Frenkel et al., 1994). Bernard et al. (1987) found that anti-laminin autoantibodies were more prevalent in cadmium-exposed workers who had urinary cadmium levels \( \geq 20 \mu g/g \) Cr. Jin et al. (2003) first reported that high levels of MT-Ab exist in healthy subjects and in patients suffering from atopic dermatitis, either with or without a metal allergy. Those patients having a metal allergy had an extremely high prevalence of MT-Ab. The precise mechanism for the presence of MT autoantibodies in the sera of healthy people and atopic dermatitis patients having a metal allergy remains unclear, but it may be attributed to the release of intracellular MT into the circulation or to structural alteration under excessive metal exposure (Jin et al., 2003). In fact, some autoantibodies—the so-called natural autoantibodies (NAAs)—can be present in the sera of healthy individuals in the absence of deliberate immunization with any antigen (Lacroix-Desmazes et al., 1998). In both human and mouse, NAAs are known to bind to a broad range of evolutionarily conserved cell surfaces, intracellular and circulating antigens, such as glutamic acid decarboxylase, interferons, factor VIII, and glomerular basement membrane, among others (Lacroix-Desmazes et al., 1998; Poletaev and Osipenko, 2003). Natural autoantibodies may be encoded by germline genes and produced in the early stages of life, independent of stimulation by foreign antigen (Lacroix-Desmazes et al., 1998). According to our research, none of the subjects in this study had ever used metallothionein products, and none had a history of metal allergies or hereditary diseases. Thus, we cannot attribute the small variances in the levels of MT-Ab in these subjects to their exposure to xenogenic antigens. In addition, in this study we did not observe any significant association between cadmium exposure and the levels of MT-Ab, even though it is known that cadmium exposure can increase the release of metallothionein into blood plasma in Cadmium-exposed workers (Nordberg et al., 1982). It seems that the increased levels of plasma metallothionein did not stimulate an autoimmune reaction in this case. We believe that an explanation for this phenomenon could exist. The level of cadmium exposure in the present study was not as high as that reported in Bernard’s study on anti-laminin autoantibodies; thus, it might be not high enough to evoke an immune response to metallothionein. Bernard et al. (1987) reported that anti-laminin autoantibodies were only present in those workers whose levels of urinary cadmium exceeded 20 \( \mu g/g \) Cr. In the present study, the average level of urinary cadmium was much lower than that reported by Bernard et al. (1987): only 12 of the workers had urinary cadmium values above 10 \( \mu g/g \) Cr, and only 5 of them had a level above 20 \( \mu g/g \) Cr. Whatever the cause, the precise relationship between the degree of cadmium exposure and the levels of MT-Ab remains to be elucidated.

It is interesting that in this study we observed that the MT-Ab titer displayed a positive correlation with the levels of UNAG
and UB2M in all of the subjects and in the group of cadmium-exposed workers. We observed statistically significant increases in the levels of UNAG and UB2M upon increases in the levels of urinary cadmium and plasma MT-Ab. When we compared the MT-Ab titers between groups divided by their tubular function and matched with their degrees of cadmium exposure, ages, and smoking habits, we found that workers who had tubular dysfunction possessed higher levels of MT-Ab. That is to say, workers displaying a high titer of MT-Ab were more susceptible to cadmium-induced tubular dysfunction. This report is the first to demonstrate such a relationship between the levels of MT-Ab and the degree of cadmium-induced nephrotoxicity.

Studies on the metabolism and toxicity of cadmium have clearly indicated that MTs play an important role in the detoxification of cadmium (Nordberg et al., 1975a; Klaassen et al., 1999). Renal damage is believed to occur when the localized cadmium, or an excessive concentration of cadmium, is unbound to metallothionein (Jin et al., 1998). Pretreatment with CdCl₂, which can effectively induce the synthesis of metallothionein in renal cells, was found to prevent acute nephrotoxicity from occurring after an injected challenge dose of Cd-metallothionein (Jin et al., 1987). Metallothionein gene knockout mice (MT-null mice) have been created that lack the expression of the MT protein (Masters et al., 1994); their susceptibility to renal damage induced by cadmium is enhanced markedly when compared with that of wild-type (WT) mice (Liu et al., 1998, 2000). These observations confirm that MT plays a critical role in protecting animals and humans against cadmium-induced nephrotoxicity. Considering the importance of MT in the detoxification of cadmium, the ability of the critical organ to synthesize MTs upon cadmium exposure determines the outcome of the cadmium exposure, that is, the susceptibility. As an antibody against metallothionein, it is reasonable to speculate that MT-Ab may interact with metallothionein, but the details of the mechanism underlying the effect that MT-Ab has on renal dysfunction in subjects exposed to cadmium is still unclear. Several toxic metals (such as cadmium, mercury, and lead) may interfere with the immune system and lead to the development of immune type glomerulonephritis in rats (Ohsawa, 1997). If an individual is producing antibodies to their own MT, then that suggests that cadmium exposure and MT production could lead to an autoimmune disorder. There could be a second mechanism operating involving circulating antibodies interacting with the glomeruli and statistically increasing the filtration of proteins in general. If such is the case, MT-Ab should show a positive association not only with low molecular biomarkers but also with high molecular biomarkers, i.e., UALB in this study.

Although, we did not detect a statistically significant increase in UALB (Table 4), such a phenomenon cannot be ruled out, because the study power could be limited by the sample size. Further studies with larger sample size should examine such a possible autoimmune mechanism.

In addition, we did not observe any association between the levels of MT-Ab and UMT, which may also be used as an indicator of tubular dysfunction (Shaikh and Tohyama, 1984). The MT-Ab is present mainly in the circulation, and it can bind with plasma metallothionein. Because of its high molecular weight, the MT-Ab–Ag complex is difficult to filter through the renal glomeruli, which increases the time that MT remains in the circulation. Only the free, unbound metallothionein can be filtered readily and be excreted in the urine. It seems that there is no relationship between the levels of MT-Ab in the

| TABLE 4 | Distribution of Renal Dysfunction among Workers at Different Levels of MT-Ab |
|----------|---------------------------------|---------------------------------|---------------------------------|---------------------------------|
|          | hyper-B2Muria (%)              | hyper-NAGuria (%)              | hyper-ALBuria (%)              | hyper-MTuria (%)                |
| HMTAb    | 12                              | 29                              | 29.27                          | 15                              |
| LMTAb    | 10                              | 63                              | 13.70                          | 11                              |
| χ²       | 4.087                           | 6.904                           | 0.236                          | 2.332                           |
| p        | 0.043                           | 0.009                           | 0.627                          | 0.127                           |
| OR       | 2.607                           | 3.252                           | 1.297                          | 2.067                           |
| 95% CI   | (1.011–6.724)                  | (1.318–8.020)                  | (0.453–3.714)                  | (0.805–5.304)                  |

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<th>TABLE 5</th>
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<td>Tubular dysfunction</td>
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circulation and MT in the urine. Upon immunization with MT, BALB/c mice can induce a high titer of MT-Ab (Jin et al., 2002b). Metallothionein immunization can impede the mercuric chloride-induced increase of MT expression in the liver and lead to increases in the level of mercury in both the serum and liver while decreasing the level found in the kidneys. Metallothionein immunization in conjunction with mercuric chloride treatment dramatically increases the susceptibility to bone toxicity (Jin et al., 2002a). We hypothesize that the MT-Ab may interfere with the detoxification by MT in persons exposed to cadmium, especially in the renal tubular cells. Most of the evidence available at present indicates that cadmium’s toxicity to the kidneys is related to the balance between the levels of toxic “free” non-MT-bound Cd and CdMT in renal cells (Jin et al., 1998; Nordberg and Nordberg, 2000). The effect of the MT-Ab may arise from changes in the conformation of MT that occur after binding. It is known that metallothionein can bind to seven metals because of the presence of its thiol residuals. After binding, the MT-Ab may influence both the conformations and the metal-binding capacity of MT. On the other hand, the MT-Ab may inhibit in several ways the ability to induce MT upon cadmium exposure. We believe that the level of MT-Ab could reflect, to some extent, the functional state of the individual in response to some environmental stress, such as exposure to the metal cadmium, although the exact relationship remains unclear.

In summary, our study of a Chinese occupational population demonstrates that cadmium exposure can induce renal dysfunction in a dose-dependent manner. Elevated levels of MT-Ab in cadmium-exposed workers were associated with increased levels of biomarkers of tubular damage, but there was no clear relationship between MT-Ab levels and the degree of cadmium exposure. The subjects displaying a relatively increased titer of MT-Ab thus were more susceptible to cadmium-induced renal tubular dysfunction. Further studies on MT-Ab are warranted to increase our understanding of the effects of MT-Ab in relation to metallothionein and cadmium toxicity.

SUPPLEMENTARY DATA

Supplementary data are available online at http://toxsci.oxfordjournals.org/.

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