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ORIGINAL ARTICLE

Measurement of Urinary Cadmium in Glazers Using Solid Phase Extraction Followed by Inductively Coupled Plasma Atomic Emission Spectroscopy

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ABSTRACT

Glazers are exposed to a variety of heavy metals in the ceramic industry, causing adverse effect on the body systems. Cadmium is one of the major raw materials for production of colored glazes. To evaluate occupational exposure to cadmium, spot urine samples were collected from 49 tile and pottery glazers in Yazd City in 2010 at the beginning and end of the work shift (98 samples). Totally, 55 office workers were also evaluated as control group. Samples were prepared using solid phase extraction followed by Inductively Coupled Plasma Atomic Emission Spectroscopy. All the participants filled out a self administered questionnaire comprises questions about work shift, kind of job, use of mask, ventilation, work history, overtime work, age, weight, and height. The lung function tests were performed in a standing position according to the American Thoracic Society recommendation on both control and cadmium exposed individuals. Analysis of covariance (ANCOVA) was used to evaluate the data. The mean values of cadmium levels before and after shift in study group was 3.88 and 10.85 µg/gcreatinine, respectively. The mean values of cadmium levels at the end of the work shift in the glazers urine samples was almost 3.53 times higher than the control group and 2.17 times higher than the ACGIH biological exposure indices (5 µg/g creatinine). In addition the lung functions of glazers was significantly lower than the office workers ($p<0.001$). Exposure to cadmium in ceramic industry can lead to the reduction of respiratory capacity. Hygienic behaviors such as using protective mask and working in efficient ventilated workplaces can decrease the rate of occupational exposure to cadmium.

Keywords: *Urine cadmium, Ceramic industry, Glaze, ICP-AES, Spirometry*

INTRODUCTION

Industries in which high levels of cadmium have been found include the refining of metal ores, the manufacture and use of corrosion-resistant coatings, PVC products, photographic, color pigment, nuclear power plants, nickel-cadmium batteries, and phosphate fertilizers [1, 2]. In ceramic industry, coloring powders particularly cadmium as a commodity is used extensively to produce good coloring, high opacity, and tinting strength. Excellent heat stability and the brilliant colors of cadmium pigments make this metal to be

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Table 1. Population characteristics of the study groups

Variables	Glazer (n = 49)		Control (n = 55)		
	Mean	S.D	Mean	S.D	*p* value
Age (yr)	30.67	5.88	32.6	7.1	0.12
Height (cm)	174.5	8.6	176.42	9.06	0.26
Weight (kg)	77.44	11.7	78.25	10.6	0.7
BMI	25.59	4.39	25.33	4.29	0.76
Overtime (h)	10.05	14.9	0.83	2.74	<0.001
Work history (month)	51.53	33.44	76.32	67.92	0.02
FVC (liter)	4.24	0.67	4.74	0.66	<0.001
FEV1(liter)	3.5	0.7	4.25	0.7	<0.001
FEV1/FVC (percent)	80.99	8.16	88.85	5.6	<0.001
FEF $_{25-75}$ (liter)	3.47	0.8	4.49	0.69	<0.001

indispensable in the ceramic factories [3]. Glaze contain cadmium and other toxic materials applied to the clay surface in a various ways such as brushing, sponging, and dripping methods by tillers and potters. In the tile industry a certain amount of coloring agents is added to the clay and mixed in a ball mill. Cadmium sulpho-selenides is used as component of many glazes for production of yellow, orange, red, and maroon colors. Exposure to cadmium occurs mainly in the ceramic process during the mixing, storing, handling and other activities where large amounts of respirable dust are generated [4, 5].

Cadmium is a toxic metal with extremely long biological half-time of 15-20 years in humans. Kidney is exceptionally sensitive to the effects of cadmium toxicity [6-9]. The mortality studies show an increased risk of mortality from prostate cancer in workers exposed to cadmium [10-12]. Considerable studies have illustrated the relationship between exposure to cadmium and the risk of lung cancer [13-16]. The International Agency for Research on Cancer (IARC) has classified cadmium as carcinogenic in humans (Group 1) [2].

The purpose of this study was to determine the cadmium level in urine of glazers of a ceramic industry and to investigate whether a relationship exists between work history, job, work shift, breathing masks, overtime work, ventilation, age, weight, height, and the urinary concentration of cadmium and also to evaluate the relation exist between occupational exposure in glazers and the lung function. It is worth mentioning that, many approaches have been performed to prepare biological and environmental samples before instrumental analysis [17-18]. Through this study, solid phase extraction developed in our previous work was used to concentrate and purify the samples [19-24].

MATERIALS AND METHODS

This case-control study was performed on the glazers in comparison to control group who were working at the same factories. Urine levels of cadmium were studied in fifteen ceramic factories and several pottery workplaces in Yazd City in 2010. Thirty three tile glazer, 16 pottery glazers, and a control group consisting of 55 office workers were enrolled. In glazers, the age range was from 22 to 50 years with average of 30.67 years, and for the control group the range was from 23 to 50 years with an average of 32.6 years. The control group had never been occupationally exposed to cadmium. The work shift length was 8 hours with two shifts per day. Information was recorded on questionnaires including overtime work, work history, work shift, kind of job, and use of mask, ventilation system, age, weight, and height. There were not significant differences between glazers who were exposed to cadmium and the control concerning age, weight, and height.

Spot urine samples were collected at the start and the end of the work shift of glazers (98 samples). The control individuals were sampled just through the shift (55 samples).

Samples were collected before starting work and also at the end of the work shift when workers removed their working clothes. They were advised to wash their hands before sampling in order to reduce the potential contamination. Twenty ml urine samples were collected in acid-washed polyethylene containers and were stored at +4°C until the analyses time.

A solid phase extraction system was used for separation and preconcentration of cadmium cartridges were filled up with 500 mg amberlite XAD-7 resin. In order to remove organic and inorganic contaminants, amberlite XAD-7 adsorption resin was washed with ethanol, water, and 1 M HCl. 2.5 ml urine sample was adjusted at pH 9 and the cadmium present in the samples was chelated with ammonium pyrrolidine dithiocarbamate (APDC). Then, samples were diluted to 25 ml with distilled water. The solution was then passed through the sorbent at a flow rate of 5 ml/min under

Table 2. Comparison of mean values of urinary cadmium between before-and after-shift samples in the glazers and control group

Group	Mean±SD (µg/gcreatinine)	t	*p* value
Before shift glazer Control	3.88±2.02 3.07±7.24	0.93	0.35
After-shift glazer Control	10.85±12.88 3.07±7.24	2.18	0.03
Before shift glazer After-shift glazer	3.88±2.02 10.85±12.88	-3.3	0.002

gravity. The column was then washed out with water and 1 M NaOH. Then, the retained analyte was eluted by 15 ml of 2 M HNO_3. Determination was performed by inductively coupled plasma-atomic emission spectroscopy (ICP-AES) (SPECTRO, ARCOS, and Germany). Urine samples were sent to the Noor Pathobiology Laboratory in Tehran City for creatinine concentrations determination. Then, final concentration of cadmium was measured in terms of µg/g creatinine**.**

The lung function tests were performed on each control and cadmium exposed subject with vitalograph spirometer (model 2120). The spirometer was calibrated daily with one liter calibration syringe and operated within a temperature range of 20-25°C. Spirometric tests were performed in a standing position according to the American Thoracic Society (ATS) recommendation. Forced vital capacity (FVC), forced expiratory volume in one second (FEV1), forced expiratory flow FEF_{25-75} were measured. The FEV1/FVC ratios were calculated in terms of percentages.

Statistical analysis was performed using the SPSS 11.5 software. Comparisons between control and glazers were made with independent samples *t*-tests .paired t-tests was used to assess the changes between before- and after-shift measurements for the glazers. Analysis of covariance (ANCOVA) was used to study the effect of each factor on after-shift measurement after controlling for the before shift.

RESULTS

Demographic characteristics and the results of the spirometric test for glazers and controls are shown in Table 1. There were no significant differences for age, height or weight between groups. Mean values of FVC, FEV1, FEV1/FVC, and FEF_{25-75} were lower in the exposed group compare to the control group (Table 1). The lung function tests revealed significant differences in all spirometric parameters between the glazers and the control group ($p<0.001$) (Table 1).

Glazes used in the fifteen tile factories and several pottery workplaces had a highly variable cadmium content. Urinary cadmium levels at the start of the work shift ranged from 1.64 to 12.02 µg/g creatinine with a mean value of 3.88 µg/g creatinine and at the end of the work shift ranged from 2.2 to 52.46 µg/g creatinine with a mean value of 10.85 µg/g creatinine and in the control group was 3.07 µg/g creatinine. The significant difference of cadmium values was seen before and after shift work (p=0.002, Table 2). There were significant differences in the mean concentration of cadmium at the end of the work shift and the control group. However, no differences in cadmium levels were found between samples at the start of the work shift and the control group (Table 2).

The values of the urinary cadmium concentrations regarding to each variable have been summarized in Table 3. The results showed that, concentrations of urinary Cd were increased during the shift. On the other hand the highest values of cadmium were observed at the end of the work shift. Glazers who were working in the morning shift showed higher urinary cadmium compare to the evening shift (14.41µg/g creatinine versus 7.44µg/g creatinine). ANCOVA showed that, after adjusting for before shift cadmium, the effect of shift on after-shift cadmium was not significant (Table 4).

This study showed that glazers without protective masks and local exhaust ventilation during mixing or handling of glazes had higher levels of cadmium compare to the glazers who occupationally protected themselves (11.87 µg/g creatinine versus10.31 µg/g creatinine and 11.35 µg/g creatinine versus 9.73 µg/g creatinine respectively). ANCOVA showed that, after adjusting for before shift cadmium, the effect of mask and ventilation on after-shift cadmium was not significant (Table 4).

Results indicated a slightly higher exposure level in tile glazers compared to the pottery glazers (10.93 µg/g creatinine versus 10.61 µg/g creatinine). ANCOVA showed that, the effect of job on after-shift cadmium was not significant (Table 4). Increase in urinary cadmium was seen based on the work history (15.34 versus 9.69). Work history was ranged from less than 24 months to more than 61 months. ANCOVA showed that, the effect of work history on after-shift cadmium was not significant (Table 4). No significant relation was observed regarding to the overtime work. Although there was no significant trend in urinary levels according to the age and BMI, the cadmium levels were higher in the older men and higher BMI (Table 4).

Table 3. Urinary cadmium concentrations of glazers, before-after-work shifts

Parameters		Number	Before shift Mean±S.D (µg/g creatinine)	After-shift Mean±S.D (µg/g creatinine)
Shift	Morning	23	4.01±1.7	14.41±16.35
	Evening	26	3.77±2.29	7.44±7.16
Mask	Yes	32	3.66±2.49	10.31±13.02
	No	17	4.02±1.69	11.87±12.95
Job	Pottery glazer	16	4±2.7	10.61±12.28
	Tile Glazer	33	3.83±1.63	10.93±13.24
Ventilation	Non standard	35	4.08±2.22	11.35±13.67
	Standard	14	3.39±1.34	9.73±11.27
Work history (month)	≤24	15	3.13±1.22	9.69±11.72
	60-25	18	4.58±1.84	8.3±10.55
	≥61	16	3.8 ±2.58	15.34±16.08
Overtime work (hour)	No	29	3.89±2.3	11±15.34
	1-20	11	4. 2±1.66	8.56±9.72
	21-60	9	3.46±1.5	11.79±13.55
BMI	<20	6	3.2 ±2.53	9.05±11.75
	25.99-20	20	3.67±1.4	9.4±12.32
	26-30.99	18	4±1.7	11.8±13.64
	31-40.99	5	5.13±4.03	15.16±16.7
Age (yr)	≤25	10	3.17±1.74	4.57±3.1
	26-32	23	3.88±1.89	12.2±13.49
	≥32	16	4.34±2.33	12.35±14.77

DISCUSSION

Cadmium compounds are extensively used as colored agents in glaze production. The amount of coloring agent added to the glaze depends on the characteristics of the glazes. The mean value of urinary cadmium levels was 2.17 times greater than the biological exposure indices (5 µg/g creatinine) recommended by American Conference of Governmental Industrial Hygienists [25] and 3.53 times greater than the control group (3.07 µg/g creatinine).

In this study, the mean values of cadmium have demonstrated the high concentration of cadmium in the glazing units. However, Vahter et al. showed that, no evidence of increased blood cadmium was found because of a low level of exposure in children who occasionally used batteries containing Cd for the production of glazed tiles [26]. Highly variable cadmium content in the glazers was observed because of the difference in the factories structure, ventilation systems, machines maintenance, and cleaning operations.

In all glazing processes, the glazers were prepared in the morning shift; therefore, cadmium exposure in the morning shift was higher than the evening shift.

The results obtained in this study were in agreement with findings reported by Bache et al. who have reported that, exposure to cadmium decreased with the use of ventilation system and protective masks [27]. In

Table 4. Results of ANCOVA for after-shift urine cadmium by independent factors controlled for before shift cadmium

Source	F	*p* value
Shift	3.08	0.09
Mask	0.61	0.44
Job	0.08	0.78
Ventilation	0.02	0. 88
Work history (month)	0.12	0.89
Overtime work (hour)	0.52	0.6
BMI	0.67	0.57
Age (yr)	1.91	0.16

the present study glazers who worked in well ventilated places, and wore protective mask have lower concentration of cadmium compare to those without occupational protection. Achieved results confirmed the role of personal hygiene in prevention of exposure. There were no significant differences in any of these levels between the two groups probably due to the small size of glazers.

The higher urinary cadmium level was found for tile glazers probably due to the use of more cadmium in the glazes as well as the difference in workplace environments.

Urinary cadmium was higher in work history. The influence of duration of exposure on the blood cadmium in teachers and school children were 2.5-3 times higher in adult as compared to the children [28]. As mentioned beforehand, no significant relation was observed regarding to the overtime work. Similar to the previous studies, there were no significant relation between the higher BMI and elevated urine cadmium [29, 30]. However, lack of knowledge for understanding the effects of the cadmium exposure followed by non-hygienic behavior such as eating in the workplace without washing hands, increase in the age of worker may cause the higher urinary Cd concentration.

Simultaneously, Olsson et al. have reported increasing urinary cadmium regarding the age factor [31]. However, Nordberg et al. observed no relations between cadmium and this factor [32].

Through this study, difference was seen in all spirometric measurements between the two groups, in which, difference in FEV1 and FEV1/FVC was higher in those who exposed to cadmium compared to those who were not exposed. Obtained results show activation in the glazing unit associated with a decline in lung function mainly in the obstructive syndrome type, however, cadmium is one of the heavy metals that can increase this syndrome. In addition to exposure to coloring agents such as cadmium, exposures to raw materials applied for the ceramic production particularly silica may occur which is related to the reduced lung function, so, glazers had been exposed simultaneously to heavy metals and silica dusts. Many studies in ceramic industry have been carried out on effect of silica in tile or pottery workers and have shown a significant difference in lung function measurements between those who have exposed to the silica dust and those who have not exposed. Halvani et al. and Bahrami et al. both have reported exposure to silica related to reduction in pulmonary function and chronic obstructive pulmonary disease (COPD) [33, 34]. Respiratory and pulmonary function disorders were observed in ceramic factory workers in Iran [35]. In these studies, interaction between heavy metals and silica in prevalence impaired lung functions has not been discussed. However, exposure to cadmium alone has also been shown in several studies associated with COPD [36, 37].

Improper formulation and firing practices of glazes can release cadmium to the acidic food substances during household use. Several studies have found cadmium to be leached from glazed dinnerware, for example, Sheets has reported the release of cadmium in some of the porcelain dinnerware into food substances [38, 39].

For reduction of cadmium exposure, respiratory protection, protective clothing, engineering controls of ventilation, work practice controls, hygiene facilities, replacement of cadmium by another substance, pre-employment and periodic medical test of workers, and employee education on the hazards of cadmium have to be organized. The simultaneous effects of heavy metal-containing dusts and silica in prevalence of impaired lung functions have to be discussed by research studies in the future.

CONCLUSION

Results showed the higher urinary cadmium concentration in the glazers. Hygienic behaviors such as using protective mask and working in efficient ventilated workplaces can decrease the rate of occupational exposure to the toxic heavy metals such as cadmium.

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