



Feasibility of Biological Effective Monitoring of Chrome Electroplaters to Chromium through Analysis of Serum Malondialdehyde

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Abstract

Background: Great concern about occupational exposure to chromium (Cr [VI]) has been reported due to escalated risk of lung cancer in exposed workers. Consequences of occupational exposure to Cr (VI) have been reported as oxidative stress and lung tissue damage.

Objective: To investigate the feasibility of biological effect monitoring of chrome electroplaters through analysis of serum malondialdehyde (MDA).

Methods: 90 workers directly involved in chrome electroplating—categorized into three equal groups based on their job as near bath workers, degreaser, and washers—and 30 workers without exposure to Cr (VI), served as the control group, were studied. Personal samples were collected and analyzed according to NIOSH method 7600. Serum MDA level was measured by HPLC using a UV detector.

Results: Median Cr (VI) exposure level was 0.38 mg/m³ in near bath workers, 0.20 mg/m³ in degreasers, and 0.05 mg/m³ in washers. The median serum MDA level of three exposed groups (2.76 µmol/L) was significantly ($p < 0.001$) higher than that in the control group (2.00 µmol/L). There was a positive correlation between electroplaters' level of exposure to Cr (VI) and their serum MDA level (Spearman's ρ 0.806, $p < 0.001$).

Conclusion: Serum MDA level is a good biomarker for the level of occupational exposure to Cr (VI) in electroplaters.

Keywords: Chromium; Electroplating; Chromium hexavalent ion; Occupational exposure; Biological assay; Malondialdehyde

Introduction

Occupational exposure to chromium (Cr) compounds occurs in a wide range of industrial processes including chrome plating, mining and chromite ore processing, production and use of alloys, chromate chemicals, wood pre-

servatives, pigments, cement dusts, and welding of stainless steel.^{1,2} The element Cr has a wide range of valences. The adverse health effects of Cr exposure depend on its valence as well as the level and route of exposure. Cr compounds have diverse toxicities including carcinogenicity and skin sensitization.²⁻⁷ According to epide-

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miological studies, occupational exposure to Cr (VI) is associated with 18–80-fold increase in the risk of lung cancer.⁸

The most commonly used solution in chrome plating bath is chromic acid (soluble hexavalent Cr compound). During the work, a considerable amount of mists are released into the air. According to Chen, *et al*,⁹ the occupational exposure level of hard chrome electroplaters to chromic acid Cr (VI) mist ranged from 4.40 to 96.0 mg/m³. In a study by Kuo, *et al*,¹⁰ the mean occupational exposure to Cr (VI) of near bath workers was 6.68 (SD 1.98) mg/m³.

Biological monitoring is the most valuable tool for acquiring knowledge of integrated exposure of working groups. It could provide reliable data for the evaluation of the risk of toxic compounds such as Cr (VI).^{11,12} American Conference of Governmental Industrial Hygienist (ACGIH) recommends measurement of total Cr in urine as the biological exposure index for Cr (VI).¹³ This index is based on reduction of Cr (VI) to Cr (III) after its transportation from the cell membrane. However, appreciable amounts of Cr (III) are received through diet ranging from 8.4 to 23.7 µg/1000 cal;¹⁴ it is also excreted as Cr (III) in urine. Some authors stated biological monitoring of Cr (VI) exposure could be biased due to nutritional habits.^{15–17}

Cr (VI), a potent oxidant, can cross the cell membrane and cause membrane damage by lipid oxidation.¹⁸ Malondialdehyde (MDA) is produced as a result of membrane lipid peroxidation and can be measured in serum or urine as a biological indicator of exposure to oxidant compounds.¹⁹

Chrome electroplating workers are highly exposed to Cr (VI). Exposure occurred because of airborne chrome electroplating vapors in breathing zone. Work steps and tasks include:

1. Degreasing to remove heavy soiling (Degreaser workers): In electroplating there are two stages for degreas-

ing (hot degreasing and cold degreasing). The solvents used in degreasing are caustic. In hot degreasing, which is done first, a bath is filled with the degreasing solvent. The solvent is then heated for use. Workers submerge the metallic parts in the bath for a while and then pull them out. Cold degreasing will start later using another caustic degreasing solvent. The approach is similar to that of hot degreasing except for heating the solvent.

2. Placement into the chrome plating vat (Near bath workers): Near bath workers submerge the metallic parts, hanged on a copper hook, in a chrome electroplating bath. They are responsible to check the temperature, electrical current and solvent concentration of the bath. This task, hold them near bath till electroplating is finished.
3. Washing (washing workers): Using 10% sulfuric acid solution and cold water, the metallic parts are then rinsed by washers. At the last stage, the metallic parts are washed with cold water and packaged.

We therefore conducted this study to evaluate personal monitoring along with investigating oxidative stress of chrome electroplaters through measurement of their serum MDA level as a relevant biological effective dose measure.

Materials and Methods

In a cross-sectional study, 90 out of 127 workers who directly involved in chrome electroplating as well as 30 non-exposed workers working in the same metal manufacturing facilities (quality control personnel) were selected by stratified random sampling using a random number table and personnel's ordered list. The exposed group workers were selected from three different jobs: near bath workers (n=30), degreaser (n=30), and washers (n=30),

that had different exposure levels.

The work was done in a common place by the three exposed groups. The work shift was eight hours. Demographic data (*eg*, age, work hours, work experience, smoking, health status, and use of personal protective equipment) were obtained by face-to-face interview. Diet similarity was confirmed through studying their two free meals per day at the same metal manufacturing plant (which were mainly composed of a dish of meat and vegetables with rice and dairy products).

Because, Tehran, the city where the study was conducted, was a metropolis and commuting was not easy, workers generally live in the close vicinity of their workplace and extraprofessional exposure was fairly identical for the exposed and non-exposed workers. Only non-smoking healthy workers with at least one year of work history were included in this study.

The study research protocol was confirmed by a research ethics committee considering the Declaration of Helsinki code of ethics. All study participants were assured that the results of the research will be treated anonymously.

The personal monitoring of all participants were performed in the morning shift (8:00 to 17:00) according to the NIOSH method No.7600 20. Sampling was done using polyvinyl chloride filter 5.0- μm pore size, 37-mm diameter (Sigma Co.), mounted in a polystyrene cassette filter holder (MSA Co.) connected to a personal pump (SKC Co.) with a flow of 2 L/min for a period of 240 min. Filters were removed, and extracted with 0.5 N sulfuric acid. Subsequently diphenylcarbazide reagent solution was added. Soluble Cr (IV) was measured by a spectrophotometer (Cecil 2041) with a detection limit of 0.01 mg/m³.

For biological monitoring, whole blood was collected from participants and poured into ETDA-lined 15 mL tubes. Sera were obtained within 15 min by centrifugation

TAKE-HOME MESSAGE

- Chromium (Cr) compounds have diverse toxicities including carcinogenicity and skin sensitization. Therefore, occupational exposure to Cr (VI) is important.
- During the work, a considerable amount of mists are released into the air.
- Malondialdehyde (MDA) is produced as a result of membrane lipid peroxidation and can be measured in serum or urine as a biological indicator of exposure to oxidant compounds.
- In chrome electroplaters, serum MDA level is closely correlated with the level of exposure to Cr (VI).

at 5000 RPM for 10 min, kept at -20 °C and analyzed for MDA. A 50- μL aliquot of serum was added to 250 μL 0.1 M perchloric acid, 700 μL deionized water was added and the solution was then centrifuged at 5000 RPM for 10 min. A 20- μL aliquot of the prepared solution was then injected for high performance liquid chromatography (HPLC) analysis (mobile phase: 65% methanol, and 30 mM 35% KH_2PO_4). The flow rate was 1.0 mL/min through RP18 column; the detector was set at 254 nm and MDA was analyzed with detection limit of 1.2×10^{-8} mol/L.²¹

Statistical Analysis

SPSS® for Windows® ver 16 and MedCalc® ver 13.3.0.0 were used for data analyses. Normality of Cr (VI) exposure data was examined by Shapiro Wilk's test. Because of non-normality of Cr (VI) exposure data, Kruskal-Wallis test was used to compare medians among studied groups. Spearman's ρ was used to explore the correlation between studied variables. The accuracy of measuring serum MDA level to discriminate exposed from non-exposed workers was evaluated by receiver operating characteristic (ROC) curve analysis. To predict Cr (VI) exposure level by measuring serum

Table 1: Level of Cr (VI) exposure and serum MDA concentrations in three exposed (n=90) and unexposed (n=30) groups. Figures are either median (IQR).

Group	Cr (VI) exposure (mg/m ³)	MDA serum level (µmol/L)
Exposed (n=90)	0.21 (0.29)	2.76 (0.96)
Near bath workers (n=30)	0.38 (0.013)	3.54 (0.41)
Degreasers (n=30)	0.20 (0.08)	2.75 (0.43)
Washers (n=30)	0.05 (0.06)	2.37 (0.38)
Unexposed (n=30)	Nil*	2.01 (0.83)

*Lower than Cr (VI) detection limit of 0.01 mg/m³

MDA level, a linear regression model (after curve estimation test) was used. A p value <0.05 was considered statistically significant.

Results

The mean age of exposed, and unexposed workers was 28.0 (SD 6.0), and 29 (SD 5.9) years, respectively. The median work experience of the exposed, and unexposed participants was and 4.5 (IQR 4.2), and 6.0 (IQR 4.0) years, respectively. Exposed and unexposed participants did not have any significant differences in terms of age and work experience. The level of exposure and serum MDA concentration in three exposed workers and unexposed participants are presented in Table 1. The level of exposure and serum MDA concentration did not follow a normal distribution.

Exposure level of 87% of chrome elec-

troplaters exceeded the ACGIH threshold limit value-time weighted average (TLV-TWA) of 0.05 mg/m³.¹³ Median serum MDA level in exposed workers was significantly (p<0.001) higher than that in non-exposed workers. Median serum MDA level was significantly (p<0.001) different among the three exposed groups.

Correlation coefficients between MDA level and other variables such as Cr (VI) exposure level, age, work experience, are shown in Table 2 for exposed and unexposed groups.

The correlation between serum MDA level and Cr (VI) exposure level was significant (p<0.001). The correlation between serum MDA level and age or work experience was not significant, neither in the exposed nor in the control group (Table 2).

The area under the ROC curve (AUC) was 0.91 (95% CI 0.85 to 0.96) (Fig 1). Youden's index was used to figure out the

Table 2: Correlation coefficients between serum MDA level and level of exposure to Cr (VI), age, and work experience in the exposed and unexposed groups

Variable	Exposed Group (n=90)		Unexposed Group (n=30)	
	Spearman's ρ	p value	Spearman's ρ	p value
Cr (VI)	0.806	<0.001	—	—
Age	0.090	0.398	0.038	0.844
Work experience	0.155	0.144	-0.113	0.553

most appropriate cut-off value for serum MDA level. Using a cut-off value of 2.31 $\mu\text{mol/L}$ for serum MDA level was corresponding to a sensitivity of 81.1% (95% CI 71.5% to 88.6%) and specificity of 93.3% (95% CI 77.9% to 99.2%) and an overall accuracy of 91.0% (95% CI 84.3% to 95.4%). The corresponding number needed to misdiagnose (NNM),²² a measure of diagnostic test effectiveness, was estimated at 11.1 (95% CI 6.4 to 21.7). This means if one used serum MDA level as a test for determining the level of exposure of workers to Cr (VI), one out of 11.1 (10 of 111) workers tested will be misdiagnosed (either false positive or false negative results).²²

A linear regression model was used to model serum MDA level against Cr (VI) exposure level (Fig 2). The regression equation developed was:

$$\text{MDA level} = 2.095 + 3.534 \times \text{Cr (VI) exposure}$$

The serum MDA level corresponding to the acceptable TLV-TWA of 0.05 mg/m^3 is therefore 2.27 mmol/L .

Discussion

In this study, Cr (VI) exposure level as well as serum MDA level of chrome electroplaters working under three conditions with various exposures including near bath workers, degreasers, and washers, was investigated. The maximum exposure level was measured in near bath workers. All near bath workers and degreasers had exposure higher than the TLV-TWA set by ACGIH.¹³ Exposure level of all near bath workers and degreasers was higher than exposures reported by Chen, *et al*, (0.025 mg/m^3).⁹ However, the level of exposure was lower than that reported in a study from Taiwan.¹⁰

The association between the risk of lung cancer and cumulative Cr (VI) exposure level has been reported in many studies.^{23,24} Considering the level of exposure

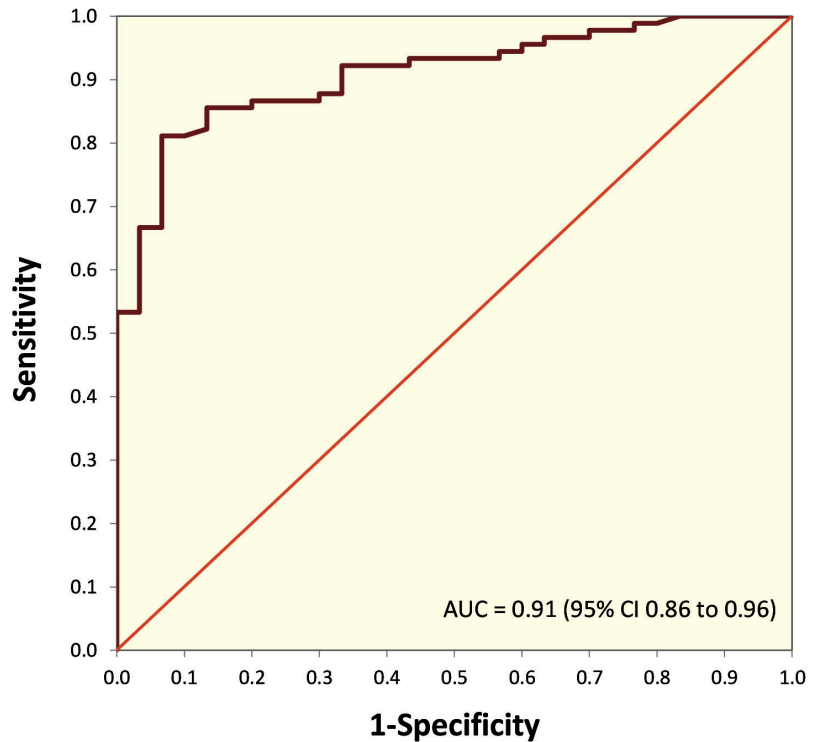


Figure 1: Receiver operating characteristic (ROC) curve for different cut-off values of serum malondialdehyde (MDA) concentrations as a diagnostic test for determining level of exposure to Cr (VI)

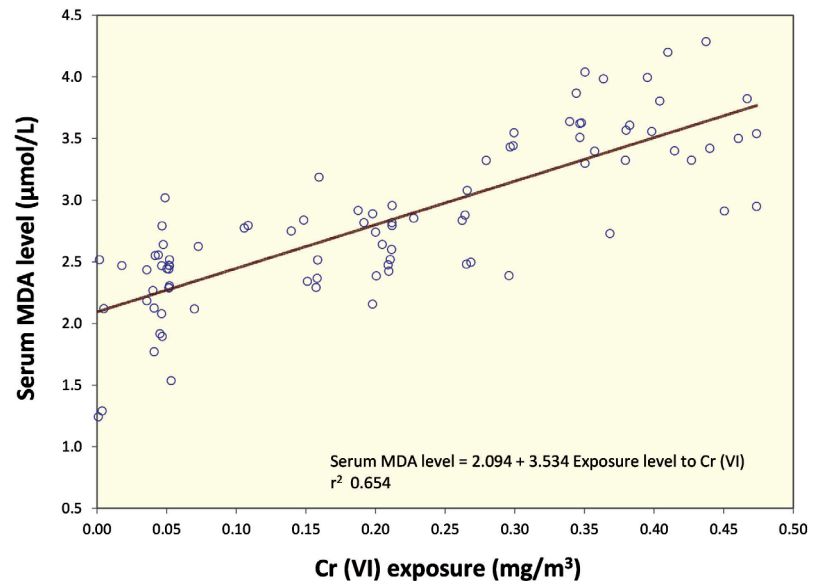


Figure 2: Linear regression model for serum malondialdehyde (MDA) level as a function of the level of occupational exposure to Cr (VI)

of chrome electroplaters to Cr (VI) in our study, based on risk models introduced by Park, *et al*,²⁵ and comparing the concept of acceptable risk criteria introduced by OSHA²⁶ and the German Committee on Hazardous Substances AGS,²⁷ the studied workers carried an unacceptable risk.

The median serum MDA levels of the three exposed groups and the unexposed group were significantly different. It was in line with previous reports by Huang, *et al*, in chrome-plating factory workers,²⁸ and Khan, *et al*,²⁹ in a population exposed to Cr. However, in another study, the mean serum MDA level was not significantly different between tannery workers exposed to high and low levels of Cr.³⁰ This result could be attributed to the low exposure level recorded in tannery workers compared with chrome electroplating workers.

ACGIH recommends total urinary chromium as a biological exposure index for Cr (VI).¹³ However, for nutritional intake of Cr compounds other than Cr (VI), this measure could be biased.¹⁵⁻¹⁷ Few investigators have also recently suggested measurement of Cr in red blood cells as a potential biomarker for long-term exposure to Cr.³¹ According to the results of this study and few other recent studies,³²⁻³⁵ serum MDA would be a potential biomarker effective dose for chrome electroplaters with excessive exposure to Cr (VI).

Use of serum MDA as a biomarker effective dose for Cr (VI) exposure has the advantage that it merely refers to Cr (VI) level and excludes Cr (III). However, there are few conditions associated with increased serum MDA including lung cancer and smoking.^{36,37} In the current study, we controlled potential confounders. Considering the regression model used, serum MDA can be used as a nonspecific biological exposure index for Cr (VI) exposure in chrome electroplaters within the range of exposure of 9.0×10^{-4} to 47.0×10^{-2} mg/m³.

The chrome electroplaters in our study

were exposed to a considerable amount of Cr (VI). More studies on workers with other levels of exposure are recommended.

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Conflicts of Interest: None declared.

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