

Field Study

Occupational Exposure to Manganese-containing Welding Fumes and Pulmonary Function Indices among Natural Gas Transmission Pipeline Welders

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Abstract: Occupational Exposure to Manganese-containing Welding Fumes and Pulmonary Function Indices among Natural Gas Transmission Pipeline Welders: Hamid HASSANI, *et al.* Department of Occupational Health Engineering, School of Public Health, Tehran University of Medical Sciences, Iran—**Objectives:** The objectives of this study were to evaluate manganese (Mn)-containing welding fumes' exposure, assess urinary Mn as a biomarker for Mn exposure and investigate the correlation of Mn in air, total fumes and urinary Mn with pulmonary function indices in 118 welders and 37 unexposed controls from two regions in Iran, Assaluyeh and Borujen. **Methods:** Air samples were collected on mixed cellulose ester membrane filters in personal air samplers and then analyzed using inductively coupled plasma atomic emission spectroscopy (ICP-AES) (NIOSH Method 7300). For all participants, urine samples were collected during the entire work shift, and Mn in urine was determined by graphite furnace atomic absorption spectroscopy according to NIOSH Method 8310. Spirometric measurements were also done for participants. **Results:** The maximum exposures to airborne Mn and total fumes were 0.304 ± 0.256 mg/m³ and 21.52 ± 9.40 mg/m³, respectively. The urine Mn levels in the various groups ranged between 0.77 to 7.58 µg/l. The correlation between airborne Mn and urinary Mn was significant for total whole participants. Some values of spirometric indices were statistically lower in welders rather than controls. **Conclusions:** Our results indicate

that many welders have been exposed to higher concentrations of Mn-containing welding fumes. Urinary Mn can be used as a biomarker for Mn exposure. There were weak inverse correlations between Mn-containing welding fumes and pulmonary function indices, and the inverse correlation between urinary Mn with forced vital capacities (FVC) and forced expiratory volume in 1 s (FEV1) was significant.

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Key words: Biomarker, Manganese, Natural gas transmission pipeline welders, Pulmonary disorders, Welding fumes

Natural gas is one of the main sources of energy in the world and is a cleaner fuel than fuel oil¹. Development and planning of natural gas transmission pipelines have considerable impact on the economies of many countries such as Iran, because Iran holds the world's second largest reserves after Russia². One of the most important processes in natural gas transmission is the welding of gas transmission pipelines.

Welding is a common process used to join metals by heating them to welding temperature^{3,4}. Welding processes produce hazardous agents including fumes, gases, vapors, heat, noise and ultraviolet and infrared radiation. The fumes generated during welding are considered to be the most harmful compared with other byproducts of welding. Welding fumes can induce adverse health effects, such as neurological and respiratory problems^{5,6}. Welding fumes are composed of complex metal oxides that form during the welding process. Metals that are commonly found in welding fumes may include iron, manganese (Mn), chromium,

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nickel, silicon and copper⁴). Most of the materials in welding fumes come from the consumable electrode during the welding process. Other factors such as shielding gases, fluxes or surface coatings on the electrode and base metal also can influence the composition of the welding fumes⁷). A significant number of welding workers are exposed to welding fumes throughout the world⁸). According to an estimate of the United States Department of Labor (Bureau of Labor Statistics), about 462,000 full-time welders are employed in the United States, and this number is likely to increase annually⁹). Welding fumes can cause numerous health problems. Bronchitis, metal fume fever, transient lung function changes, siderosis, cancer and neurological disorders have all been reported in welders⁴). Studies performed show that Mn in welding fumes causes neurological disorders^{3, 10}). Mn is a component used in most steels to increase hardness and strength, prevent steel from cracking during manufacture and improve metallurgical properties. It is present in many welding rods and wires as an oxidizing agent^{3, 7}). Some of the most common uses of Mn include iron and steel production, manufacture of dry cell batteries, manufacture of glass, textile bleaching, welding rods, matches and fireworks and tanning of leather. Organic compounds of Mn are present in fuel additives and many other products¹¹). However, Mn is an essential nutrient that can be neurotoxic when inhaled¹²). Manganese intoxication is sometimes referred to as “manganism”¹³). Manganism induces central nervous system abnormalities and neuropsychiatric disturbances, so it is very similar to Parkinson’s disease¹⁴). Inhalation of Mn in the workplace may cause a specific clinical central nervous system syndrome known as occupational manganism⁶). It should be noted that Mn can also affect other systems such as the respiratory, cardiac, liver and reproductive systems^{15, 16}). Welding rods contain Mn, so welders are exposed to mixed metal fumes that contain a small percentage of Mn⁷). Inhalation of welding fumes can also cause respiratory problems such as bronchitis, pneumoconiosis and lung cancer in welders. Spirometry is a helpful tool

to assess worker’s respiratory function^{4, 17}).

This study had three objectives: first, to evaluate welders’ exposure to Mn-containing welding fumes in natural gas transmission pipeline welders; second, to assessing of urinary Mn as a biomarker for Mn exposure; and third, to investigate the correlation of airborne Mn and total fumes with pulmonary function indices in the study population.

Material and Methods

Study population

A total of 118 welders were randomly selected from natural gas transmission pipeline welders in two regions of Iran, Assaluyeh (located in Bushehr Province) and Borujen (a city located in Chaharmahal and Bakhtiari Province), as the exposed group, and 37 office workers were selected as unexposed controls. The task groups in this study included: foreman, fitter, co-fitter, full pass, filling, filling cap, back weld, grinder and office work (these workers are referred to as the control group in this paper). All participants in this study were male. Characteristics of the study population are shown in Table 1.

Work description

The following steps are done in development of natural gas transmission pipelines: 1) selection of a proper line using provided maps, 2) setting pipes in lines, 3) fitting pipes together by fitters and co-fitters, 4) performance of the first layer of welding by full pass workers, 5) welding of 2 to 5 layers by filling welders, 6) welding of the next layers to fill the welding bond by filling cap welders and 7) welding of the inside of pipes by back weld workers to ensure the integrity of the weld. Also, grinders ensure that the locations of welds are uniform after any layer welding. Moreover, all of the tasks are supervised by an experienced individual acting as a foreman. Each worker performs one task.

Welding type

The most common type of welding processes used in gas transmission pipelines welding in Iran is

Table 1. Characteristics of the study population

Characteristics	Welders (n=118) (mean ± SD)	Controls (n=37) (mean ± SD)	<i>p</i> value*
Age (yr)	29.61 ± 6.78	32.13 ± 7.82	0.060
Work history (yr)	3.78 ± 1.51	2.80 ± 1.92	0.170
Height (cm)	173.16 ± 8.28	172.97 ± 17.07	0.925
Weight (kg)	74.77 ± 10.03	76.81 ± 11.75	0.303
No. of smokers (%)	54 (45.8%)	8 (21.6%)	-

*Significant differences between the welders and control group were assessed by the Student’s *t*-test. *p*<0.05 indicates Statistical significance.

Manual Metal Arc Welding (MMAW), also known as Shielded Metal Arc Welding (SMAW). In this method an electrode rod is used to create an electric arc that produces a high temperature, which melts the base metal and the electrode to form a strong bond between the parent metal¹⁸.

Air sampling and analysis

Welding fumes released by the welding process were collected on mixed cellulose ester membrane filters (0.8 mm pore size, 25 mm diameter; SKC Corp) in personal air samplers. All pumps were calibrated before and after use. SKC pumps (224-PCMTX8 model, SKC, UK) operated at a constant flow rate of 2.0 l/min were used for the sampling. Analysis was performed using inductively coupled plasma atomic emission spectroscopy (ICP-AES) according to NIOSH analytical method 7300¹⁹.

Urine Mn

Biological monitoring in this study was performed by determining Mn in urine. For all participants, urine samples were collected in plastic containers during the entire work shift (8 h). Mn in urine was determined by graphite furnace atomic absorption spectroscopy (GFAAS) according to NIOSH analytical method 8310²⁰. The relationship between Mn concentration in the air and urine as well as urine Mn with pulmonary function indices was investigated.

Spirometry test

We used a Spirolab II (MIR Medical Research International S.r.l, Rome, Italy) for spirometry tests. Values of height and weight were measured, and it is necessary to mention that all participants were trained before the spirometry test. Tests were performed by an occupational medicine specialist. Spirometric measurements including FVC, FEV1 and FEF 25–75 were obtained before and after the work shift.

Statistical analyses

We used the Student's *t*-test to determine differences among means for characteristics of some welders and the control group. A Pearson correlation analysis was used to determine the correlation among urine Mn levels and airborne Mn concentrations. Also, a partial correlation analysis was carried to investigate the correlation of airborne Mn, total fumes and urinary Mn with pulmonary function indices. Comparisons between means of exposure to Mn among various tasks were made using a One-way ANOVA. Also, One-way ANOVA was used to analyze spirometry data. Statistical significance was established when $p < 0.05$. The SPSS (v 17) software was used for all statistical analyses.

Results

The characteristics of welders and control individuals were compared (Table 1). There were no significant differences between welders and control individuals in terms of age, work history, height and weight ($p > 0.05$).

The results of air monitoring shows that back weld workers had the maximum exposure to total fumes among welders, and their arithmetic mean exposure was 21.51 ± 9.40 mg/m³ (Table 2). This value is 4.3 times more than the threshold limit value-time weighted average (TLV-TWA) recommended by the American Conference of Governmental Industrial Hygienists (ACGIH)⁸. The values for exposure to total fumes of the full pass, filling and filling cap workers were also more than the TLV-TWA ACGIH (Table 2). Analysis of the welding fumes showed that back weld workers had the maximum exposure to Mn among welders, and their arithmetic mean exposure was 0.304 ± 0.256 mg/m³ (Table 2). Exposure to Mn in other groups was less than the TLV-TWA of the ACGIH. The values calculated for percent of Mn in total fumes are shown in Table 2 and are all less than

Table 2. Concentrations of total fumes and manganese and percentage of Mn in total fumes in various task categories

Tasks	n	Total fumes	Manganese	% of Mn in total fumes
		(mean \pm SD) (mg/m ³)	(mean \pm SD) (mg/m ³)	
Foreman	12	1.25 \pm 0.46	0.011 \pm 0.012	0.88
Fitter	6	1.40 \pm 0.29	0.022 \pm 0.029	1.50
Co-fitter	8	2.53 \pm 0.73	0.037 \pm 0.035	1.46
Full pass	21	9.44 \pm 3.17	0.150 \pm 0.208	1.57
Filling	20	11.23 \pm 5.09	0.149 \pm 0.152	1.32
Filling cap	35	9.80 \pm 4.37	0.128 \pm 0.124	1.30
Grinder	10	3.81 \pm 3.60	0.053 \pm 0.045	1.36
Back weld	6	21.52 \pm 9.40	0.304 \pm 0.256	1.41

Bold numbers: The values are higher than the TLV-TWA (ACGIH). The TLV-TWA (ACGIH) values for total fumes and Mn are 5 mg/m³ and 0.2 mg/m³, respectively.

2% (Table 2).

Urine samples were collected on the same day as air sampling. The Mn concentrations in urine ranged from 0.77 to 7.58 $\mu\text{g/l}$ in the various groups (Table 3). Back-weld workers had the maximum excretion values, with an arithmetic mean of $7.58 \pm 3.95 \mu\text{g/l}$, and comparisons of Mn concentrations in urine between back-weld, full-pass, filling and filling cap workers with the control group were significant ($p < 0.05$) (Table 3). There were significant correlations between Mn in air and Mn in urine for the full-pass, filling and filling cap groups as well as for total whole participants ($p < 0.05$) (Table 4).

Height and weight were normally distributed in welders and controls, and 45.8% of welders and 21.6% of controls were smokers (Table 1). Table 5 shows predicted post-shift values for the pulmonary function indices of welders and controls based on task types. Significant differences were indicated for percent predicted values of post-shift welders and controls in some welding tasks (Table 5). The mean forced vital capacities (FVC) of the filling and grinder groups were significantly lower in comparison with the controls ($p < 0.05$). Also, the forced expiratory volume in 1 s (FEV1) values were significantly lower in the full-pass, filling, filling cap and grinder groups

compared with the controls ($p < 0.05$). Forced expiratory flow at 25–75% (FEF 25–75%) values were not statistically different in welders and controls ($p > 0.05$). One-way ANOVA to compare mean lung incidences among task groups showed no differences among task groups ($p > 0.05$) except for the FEV1 index between the co-fitter and filling groups ($p < 0.05$).

Partial correlation of airborne Mn concentrations, total fumes and urinary Mn with various pulmonary function indices was investigated. It should be noted that partial correlation was adjusted for potential covariates including age, work history and smoking. There were weak inverse relations, and the correlation coefficients ranged from 0.051 to 0.286. The p values showed that there were no statistically significant relationships between airborne Mn and total fumes and pulmonary function indices ($p > 0.05$), but the inverse correlation between urinary Mn and FVC and FEV1 was significant ($p < 0.05$) (Table 6).

Discussion

Occupational exposure to Mn-containing welding fumes has been an issue among occupational health specialists. Thus, the number of studies on workers exposed to Mn has been increasing during recent years. In this study, the findings showed that Mn is a

Table 3. Mn concentrations in urine ($\mu\text{g/l}$) and comparison of values between welders and controls

Tasks	n	Mean ($\mu\text{g/l}$)	SD	Minimum	Maximum	Comparison with control
Foreman	12	1.78	1.12	1.00	4.00	NS
Fitter	6	1.90	1.55	0.80	3.00	NS
Co-fitter	8	2.10	0.75	1.37	3.00	NS
Full pass	21	4.24	2.73	2.00	12.00	S ($p=0.002$)
Filling	20	4.90	3.92	1.00	13.00	S ($p<0.001$)
Filling cap	35	5.01	3.51	1.00	16.00	S ($p<0.001$)
Grinder	10	2.87	1.63	1.00	5.50	NS
Back weld	6	7.58	3.95	3.00	13.00	S ($p<0.001$)
Control	37	0.77	1.05	0.00	4.10	-

S, significant ($p < 0.05$); NS, not significant.

Table 4. Correlation between Mn in air and Mn in urine based on task types

Tasks	n	Mn in air (mg/m^3)	Mn in urine ($\mu\text{g/l}$)	r	p value	Correlation*
Foreman	12	0.011 ± 0.012	1.78	-0.429	0.396	No
Fitter	6	0.022 ± 0.029	1.90	0.794	0.186	No
Co-fitter	8	0.037 ± 0.035	2.10	0.823	0.177	No
Full pass	21	0.150 ± 0.208	4.24	0.831	<0.001	Yes
Filling	20	0.149 ± 0.152	4.90	0.624	0.013	Yes
Filling cap	35	0.128 ± 0.124	5.01	0.645	<0.001	Yes
Grinder	10	0.053 ± 0.045	2.87	-0.435	0.282	No
Back weld	6	0.304 ± 0.256	7.58	-0.348	0.566	No
Total	118	0.125 ± 0.150	4.38	0.598	<0.001	Yes

*Correlations are based on Pearson product-moment calculations. $p < 0.05$ indicates Statistical significance. Mn, manganese.

Table 5. Post-shift pulmonary function indices values in various task groups

Pulmonary function indices	Task groups	n	Work history (yr) (mean ± SD)	Predicted values (mean ± SD)	Percent of predicted (%) (mean ± SD)	Comparison with control
FVC (l)	Foreman	12	5.03 ± 3.04	4.96 ± 0.39	93.80 ± 8.68	NS
	Fitter	6	6.75 ± 4.27	4.83 ± 0.32	97.81 ± 9.82	NS
	Co-fitter	8	4.32 ± 3.28	4.82 ± 0.44	99.27 ± 6.60	NS
	Full pass	21	9.33 ± 7.17	4.68 ± 0.49	89.29 ± 9.32	NS
	Filling	20	3.98 ± 2.42	4.75 ± 0.41	88.08 ± 9.85	S (p=0.004)
	Filling cap	35	6.75 ± 2.56	4.89 ± 0.35	94.07 ± 12.28	NS
	Grinder	10	2.68 ± 3.01	4.83 ± 0.32	89.41 ± 7.83	S (p=0.049)
	Back weld	6	4.40 ± 4.98	4.87 ± 0.26	92.65 ± 5.19	NS
	Control	37	3.75 ± 2.81	4.97 ± 0.52	100.6 ± 10.16	-
FEV1 (l)	Foreman	12	5.03 ± 3.04	4.12 ± 0.30	89.66 ± 3.76	NS
	Fitter	6	6.75 ± 4.27	4.01 ± 0.31	88.90 ± 11.45	NS
	Co-fitter	8	4.32 ± 3.28	4.04 ± 0.37	99.88 ± 4.27	NS
	Full pass	21	9.33 ± 7.17	3.89 ± 0.46	88.44 ± 9.58	S (p=0.016)
	Filling	20	3.98 ± 2.42	4.02 ± 0.31	84.09 ± 9.00	S (p<0.001)
	Filling cap	35	6.75 ± 2.56	4.11 ± 0.28	91.04 ± 11.61	S (p<0.001)
	Grinder	10	2.68 ± 3.01	4.05 ± 0.26	85.12 ± 7.48	S (p<0.001)
	Back weld	6	4.40 ± 4.98	4.10 ± 0.19	93.34 ± 4.27	NS
	Control	37	3.75 ± 2.81	3.95 ± 0.45	101.66 ± 11.60	-
FEF 25–75 (l/s)	Foreman	12	5.03 ± 3.04	4.58 ± 0.23	73.06 ± 13.27	NS
	Fitter	6	6.75 ± 4.27	4.47 ± 0.36	66.96 ± 18.48	NS
	Co-fitter	8	4.32 ± 3.28	4.59 ± 0.32	91.56 ± 19.14	NS
	Full pass	21	9.33 ± 7.17	4.42 ± 0.53	67.86 ± 32.64	NS
	Filling	20	3.98 ± 2.42	4.70 ± 0.15	73.82 ± 29.90	NS
	Filling cap	35	6.75 ± 2.56	4.68 ± 0.22	59.60 ± 36.86	NS
	Grinder	10	2.68 ± 3.01	4.60 ± 0.27	57.52 ± 41.69	NS
	Back weld	6	4.40 ± 4.98	4.70 ± 0.11	64.55 ± 36.52	NS
	Control	37	3.75 ± 2.81	4.72 ± 0.47	84.68 ± 33.54	-

S, significant ($p < 0.05$); NS, not significant. For each pulmonary function index, significant differences between task types and controls were assessed by the ANOVA test. FVC, forced vital capacities. FEV1, forced expiratory volume in 1 s. FEF, forced expiratory flow.

Table 6. Partial correlation* of airborne Mn, total fumes and urinary Mn with various pulmonary function indices

Indices (l)	Airborne Mn=0.13 ± 0.15 (mg/m ³)		Total fumes=8.18 ± 6.01 (mg/m ³)		Urinary Mn=4.38 ± 3.29 (μg/l)	
	r	p value	r	p value	r	p value
FVC=4.60 ± 0.71	-0.051	>0.05	-0.152	>0.05	-0.283	<0.05
FEV1=3.74 ± 0.62	-0.107	>0.05	-0.102	>0.05	-0.286	<0.05
FEF 25–75=3.34 ± 1.72	-0.057	>0.05	-0.130	>0.05	-0.139	>0.05

*Adjusted for age, work history and smoking. $p < 0.05$ indicates Statistical significance. FVC, forced vital capacities. FEV1, forced expiratory volume in 1 s. FEF, forced expiratory flow.

small percent of welding fumes, ranging from 0.88 to 1.57% of total fumes. Antonini *et al.*⁷⁾ suggested that “the amount of Mn in welding rods can range from 1 to 20% of the metals present. Thus, most welders are exposed to mixed metal fumes that contain a small percentage of manganese (<5% per total metal present).” In our study, airborne Mn concentrations varied from 0.011 to 0.304 mg/m³ for various task groups. Previous studies presented different concentrations, such as the air Mn level of 0.14 mg/m³ recorded in a study by Chang *et al.*²¹⁾; Ellingsen *et al.*²²⁾, in a study

on welders, reported 0.003 to 4.26 mg/m³ exposure to Mn. The mean time-weighted average of Mn in air ranged from 0.11 to 0.46 mg/m³ according to Bowler *et al.*²³⁾. Similarly, our obtained concentrations are comparable with most of studies. However, the TLV-TWA recommended by the ACGIH for exposure to Mn is equal to 0.2 mg/m³⁷⁾, while Chang *et al.*²¹⁾ noted that “many studies have shown that an exposure threshold for subclinical neurological effects would be 0.25–1.7 mg/m³”. In the present study, back-weld welders had the maximum exposure to Mn-containing

welding fumes due to welding inside pipes. Other groups were in outdoor environments. Therefore, outdoor welders were exposed to less Mn-containing welding fumes than back-weld welders. The mean air Mn value was lower than the absolute SD value in some task groups (foreman, fitter, full pass and filling) (Table 4). These results may be due to many potential reasons including working in an outdoor environment, wind and its direction as well as its intensity, distance between workers and source of pollution, workers' movements and so on. Also, one limitation of this study was that the individual air Mn measurement for just one day may not represent actual individual exposure or body burden. Many previous studies have used blood and urine Mn as biological measurements to reflect recent exposure. We used urine Mn sampling because measurement of Mn in urine is a noninvasive method and is convenient for participants. Of course, it should be noted that Mn is mostly excreted into feces, and only a small percentage is excreted through urine¹⁸. However, our data shows a significant correlation between airborne Mn levels and Mn concentrations in urine for some task groups including the full-pass, filling and filling cap groups as well as for total participants ($p < 0.05$), but in other task groups (foreman, fitter, co-fitter, grinder and back-weld groups), the relation was insignificant ($p > 0.05$). The number of samples in each task group can influence the significance of results. The values of blood Mn and urinary Mn may reflect several days of exposure, so urinary Mn may reflect more long-term Mn exposure rather than airborne Mn measured on the same day in our study. This reason may be a factor that led to insignificant results for some task groups (foreman, fitter, co-fitter, grinder and back-weld groups). Moreover, other factors such as worker diet, work habits and location of the air sampling devices in relation to the worker's breathing zone may affect the results. There was no correlation between airborne Mn and urinary Mn ($r = 0.084$) in a study by Nastiti *et al.*¹⁸. Also, there are many studies that obtained significant correlation between airborne Mn levels and Mn concentrations in blood and urine¹⁵. A statistically significant rank correlation was found by Roels *et al.*²⁴ between average current Mn concentrations in air (log values) and the geometric mean of urinary Mn ($r_s = 0.83$ with both total and respirable dust, ($p < 0.05$)). It is also worth noting that, Wongwit *et al.*²⁵ suggested that blood concentrations of Mn can be used as a biological indicator. Laohaudomchok *et al.*²⁶ examined the correlation between Mn exposure and Mn concentrations in toenails, blood and urine. They concluded that urinary Mn and blood Mn were not correlated with Mn exposure over a typical work shift but that toenail Mn was very well correlated

with cumulative Mn exposure at 7 to 9, 10 to 12 and 7 to 12 months. As a result, blood and urine concentrations of Mn have not so far been proven to be reliable biomarkers of exposure^{16, 26}.

Negative or inverse correlation means a relationship between two variables in which an increase in the value of one variable results in a decrease in the value of the other variable. So, our results show that an increase in airborne Mn, total fumes and urinary Mn results in a decrease in pulmonary function indices. However, correlation coefficients showed a weak inverse correlation between airborne Mn and total fumes and pulmonary function indices, and the relationships were not significant statistically ($p > 0.05$); however, the inverse correlation between urinary Mn with FVC and FEV1 was significant statistically ($p < 0.05$). This significant finding could be due to the fact that urinary Mn may be relevant to several days of exposure, but air Mn is not like this. Although work history is a significant factor¹⁷, it had no influence on pulmonary functions in our study, since most of the workers were young and their work histories were relatively short. Some previous studies reported unaffected spirometric parameters after exposure to welding fumes. Wolf *et al.*²⁷ reported that FVC and FEV1 were unchanged compared with controls, although the welders reported more pulmonary symptoms than the controls. In contrast, Pourtaghi *et al.*²⁸ suggested that FVC and FEV1 were significantly lower in welders compared with controls. In the present study, pulmonary function indices were weakly correlated with airborne Mn and total fumes, but the association between urinary Mn with FVC and FEV1 was significant. Therefore, we recommend continuing air monitoring as well as biomonitoring, applying control methods such as portable local exhaust ventilation systems and using respiratory protective equipment to protect workers from developing health problems.

Conclusions

In summary, our results indicate that many welders have been exposed to higher concentrations of Mn-containing welding fumes. Urinary Mn can be used as a biomarker for Mn exposure. There were weak, but not significant statistically, inverse correlations between Mn-containing welding fumes and pulmonary function indices, and the inverse correlation between urinary Mn with FVC and FEV1 was significant.

Conflicts of interest: The authors declare that there are no conflicts of interest.

Ethical issues: Ethical issues have been handled appropriately by the authors, and the study was

approved by ethics committee at Tehran University of Medical Sciences.

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