

Air exposure assessment and biological monitoring of manganese and other major welding fume components in welders†

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In a cross-sectional study, 96 welders were compared with 96 control subjects. Also 27 former welders, all diagnosed as having manganism, were examined. Exposure to welding fumes was determined in the 96 welders, while the concentration of elements in whole blood and urine was determined in all subjects. The geometric mean (GM) concentrations of manganese (Mn) and iron in the workroom air were $97 \mu\text{g m}^{-3}$ (range 3–4620 $\mu\text{g m}^{-3}$; $n = 188$) and $894 \mu\text{g m}^{-3}$ (range 106–20 300 $\mu\text{g m}^{-3}$; $n = 188$), respectively. Thus the Mn concentration in the workroom air was on average 10.6% (GM) of that of the Fe concentration. No substantial difference was observed in the air Mn concentrations when welding mild steel as compared to welding stainless steel. The arithmetic mean (AM) concentration of Mn in whole blood (B-Mn) was about 25% higher in the welders compared to the controls (8.6 vs. 6.9 $\mu\text{g l}^{-1}$; $p < 0.001$), while the difference in the urinary Mn concentrations did not attain statistical significance. A Pearson's correlation coefficient of 0.31 ($p < 0.01$) was calculated between B-Mn and Mn in the workroom air that was collected the day before blood sampling. Although the exposure to welding fumes in the patients had ceased on average 5.8 years prior to the study (range 4 years–7 years), their AM B-Mn concentration was still higher than in referents of similar age (8.7 $\mu\text{g l}^{-1}$ vs. 7.0 $\mu\text{g l}^{-1}$). However, their urinary concentrations of cobalt, iron and Mn were all statistically significantly lower.

Introduction

High, long-term occupational exposure to manganese (Mn) can result in manganism, a severe neurological disorder characterized by movement disturbances.¹ Exposure occurs during mining and crushing/milling of Mn ores and in the production of Mn alloys and steel. Manganese is also used in dry-cell batteries, Mn-containing chemicals, in alloys with aluminium and in the fuel additive methylcyclopentadienylmanganese tricarbonyl.² Since Mn constitutes a significant part of the welding electrode composition, welders are by number probably the largest group of exposed workers.

Welders are exposed to aerosols containing a mixture of elements and to gases such as ozone and nitrous oxides.³ The aerosols are formed primarily through homogeneous nucleation of element vapour followed by competing growth mechanisms such as coagulation and condensation.⁴ Individual particles freshly formed near the welding arc are mainly spherical with diameters ranging from 50 to 300 nm.⁴ The aerosol mass distributions are uni-modal with geometric mean aerodynamic equivalent diameters from 0.29 to 0.59 μm .⁵

Particle number measurements show that welding produces a log-normal particle mode with a 120 nm count median.⁶ Thus, these respirable particles will easily penetrate more deeply into the lung. The minimum alveolar deposition efficiency is around 25% for particles with a diameter of about 0.5 μm , with increasing deposition efficiencies at smaller particle diameters.⁷ The mechanisms of particle formation explain the tendency of particles to be formed with different chemical compositions from the core to the surface.⁸ Bulk measurements suggest that Mn(II) and Mn(III) are the most probable oxidation states in welding fumes generated by manual or gas metal arc welding.³ X-ray diffraction studies have identified a variety of Mn-containing compounds in welding fumes; e.g. KMnF_3 , MnO , K_2MnO_4 , $\gamma\text{-Mn}_2\text{O}_3$ and Mn_3O_4 .⁹

Pulmonary absorption of Mn in rats is higher for soluble than for less soluble compounds.^{10,11} This may also be the case in workers producing Mn alloys.¹² Little is known about the solubility of Mn-containing welding fume particles in the lung, but the solubility increases according to the increasing content of fluorine and potassium in the welding fume.⁹

The understanding of the pulmonary uptake mechanisms of welding fume components is insufficient. Transferrin receptors and divalent metal transporter 1 (DMT 1) that are found on the surface of the lung epithelial cells may point to a role of these transporters in mediating the uptake of divalent and trivalent cations.¹³ The observation that rats on an iron (Fe)-enriched diet had lower pulmonary uptake of Mn may support this view.¹⁴ However, evidence also exists that the mechanisms for the pulmonary uptake of Mn are different from those of iron.^{15,16}

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Welders may be exposed to considerable amounts of Mn. Stationary sampling showed mean air concentrations ranging from 0.20 to 1.47 mg m⁻³ of Mn in vehicle production.¹⁷ Stationary sampling was most likely also used in three plants where concentrations ranged from 0.44 to 2.6 mg Mn m⁻³.¹⁸ Personal exposure has been reported ranging from 0.004 to 2.67 mg m⁻³ of Mn in shipyard welders and up to 1.8 mg m⁻³ of Mn during flux cored arc welding.^{19,20} The amounts of welding fume inside a welding helmet may be considerably lower than outside. Therefore assessing the welding fume exposure in the breathing zone *under* the respiratory protection equipment is important (Antonini²¹).

Fewer data are available on Mn-concentrations in blood and urine from welders. Mean whole blood concentrations of Mn (B-Mn) of 300 nmol l⁻¹ in 10 welders, 190 nmol l⁻¹ in the referents and, a nearly fourfold increase in urinary Mn (U-Mn) concentrations were reported.²² Also higher serum concentrations (2.9 µg l⁻¹ vs. 0.7 µg l⁻¹ or 3.4 µg l⁻¹ vs. 1.1 µg l⁻¹) in welders compared to the referents have been reported.^{17,23} A non-significantly higher U-Mn concentration was also reported.²³ Rats exposed to air Mn-concentrations of 1.6 mg m⁻³ or 3.5 mg m⁻³ in welding fumes for two hours daily for 60 days increased their B-Mn from about 12 µg l⁻¹ to 17 and 21 µg l⁻¹, respectively.²⁴ Welders exposed from 0.1 to 1.56 mg m⁻³ of Mn had increased Mn signal intensities in the globus pallidus, the midbrain, the pituitary gland and the putamen of their brains as assessed by magnetic resonance imaging.²⁵

Many cases of manganism among welders have been officially recognised as an occupational disease in Russia. Although fewer new cases are being identified, this is still a serious occupational health problem in a country with more than 1 million people exposed to welding fumes. This work is part of an epidemiological study emphasizing nervous system functions of welders. Exposure characteristics for Mn and other welding aerosol contaminants, and biological levels of selected elements and their association to exposure are described.

Material and methods

Subjects

The welders were recruited from two plants, each employing several thousand workers. One of the plants produces heavy machinery and the other is a shipyard. They are situated in the vicinity of St Petersburg in Russia. The plants have 5 and 2 welding departments, respectively. One welding department was selected at random from each plant. One day-shift of welders from each of the selected departments was eligible for inclusion.

The overall design was a cross-sectional pair-matched study restricted to men. Criteria for inclusion as exposed were at least one year of employment as a welder and being employed at the time when the examinations were carried out. Control subjects were selected among turners/fitters recruited from the same two plants. They were matched 1 : 1 with an exposed subject based on age with a maximum age difference of ±2 years within a pair.

Subjects who had been on a sick leave for more than 14 days at the day of examination were not considered for inclusion.

Among the exclusion criteria were known alcohol (or drug) abuse, larger damage of the dominant hand, current or previous diseases of the central nervous system which is probably unrelated to Mn exposure (*e.g.* brain tumours, transitory ischemic attacks), diabetes mellitus, serious kidney or liver diseases. Occupational exposure to organic solvents for more than three years as painters or spray-painters, previous employment at plants producing solvents or previous occupational exposure to lead or mercury for more than one year led to exclusion as well.

In all, 132 welders were identified, of whom five were not available for inclusion (sick leave, vacation). Eleven welders were excluded based on the records from the occupational health services (OHS) (mainly due to high alcohol intake). Twenty of the remaining 116 eligible welders declined to participate. This resulted in a participation rate of 82.8% among the eligible welders. Among 156 turners/fitters identified as working in the departments that were chosen for recruiting the referents, 14 subjects were not available for the study (sick-leave, vacation). Twenty-two potential referents were excluded owing to the violation of the exclusion criteria (mainly due to high alcohol intake). Thus the reference base consisted of 120 eligible potential referents, of whom 24 refused to participate (participation rate 80.0%).

The Northwest Public Health Research Centre (NWPHRC) in St Petersburg (Russia) has identified 74 patients in their files with the diagnosis of manganism. These cases are officially recognised as occupational diseases in the Russian insurance system. It was *a priori* decided to study half of the patients by selecting every second individual (odd numbers) from an alphabetical list. Two subjects were not available, and four subjects had one or more of the above exclusion criteria. As four patients refused to participate, 27 (participation rate 87.1%) could be included. They were all former welders, and four were females.

All examinations, including a structured interview and sampling of blood and urine, were carried out at the local occupational health clinics of the respective plants. The patients were, however, examined at the NWPHRC. Background characteristics for the welders and referents are shown in Table 1.

The participation in the study was voluntary. All participants were informed about the content of the study, and signed an informed written consent. The study was assessed and approved by the Ethics Committee of St Petersburg Pasteur Institute.

Table 1 Background characteristics of 96 male welders and 96 male controls

	Welders		Controls		<i>P</i>
	AM ^a	Range	AM	Range	
Age/years	36.3	20–65	36.1	18–66	0.90
Education/years	12.7	8–17	12.3 ^b	8–19	0.20
Current smokers (%)	60.4	—	62.5	—	0.77
Alcohol consumption/g year ⁻¹	7700	0–47 300	5300	0–72 800	0.07
Duration of welding/years	13.5	1–40	—	—	—

^a AM: arithmetic mean; ^b Information on 95 subjects.

Strategy of air sampling and collection of biological samples

The exposure to welding aerosols was characterised by personal full shift measurements. Full-shift air samples were collected during the two days directly preceding the collection of blood and urine. Air samples were collected for one day only in four welders (the first sampling day) due to technical failures of the sampling equipment.

The welders were instructed to bring with them to the examinations a first-voided morning urine sample from the morning after the second day of air sampling. The urine was voided directly into a clean plastic cup before transfer to a 25 mL Universal container (Nalge Nunc Int. Corp., Rochester, NY, USA) in order to minimise inadvertent contamination. Heparinized whole blood samples were collected the same morning between 8:30 and 9:30 with 10 mL Venoject tubes (Terumo Corp., Belgium). The biological samples were frozen and stored locally (NWPHERC) at $-20\text{ }^{\circ}\text{C}$ before shipment to the National Institute of Occupational Health (NIOH) in Oslo. The controls and the patients followed the same sampling strategy except that no air samples were collected.

Welding methods

Three basic welding methods were used in this study: (1) In Shielded-Metal Arc Welding the heat generated melts a portion of the electrode tip, its coating and the base metal in the immediate area. Most conventional arc welding is done hand-held by means of a coated consumable electrode. We have termed this method “manual welding”; (2) Gas Metal-Arc Welding shields the weld zone with an external gas or gas mixture. We have termed these methods “semi-automatic”; (3) Fluxed-Core Arc-Welding uses a tubular electrode filled with flux. The emissive fluxes that are used shield the weld arc from surrounding air, or shielding gases are used and non-emissive fluxes are employed. The welding process is easily automated with robotic systems. We have termed this process “automated welding”.

Air sampling

Exposure to welding fumes was assessed by employing 25 mm Millipore plastic cassettes (M000025A0) equipped with 5.0 μm pore-size polyvinyl chloride membrane filters (Millipore, Bedford, MA, USA, PVC502500). These filter cassettes for the measurement of “total” dust were placed in the breathing zone underneath the welding helmet. The pumps employed were SKC’s Sidekick personal units operated at a constant flow of 2.0 L min^{-1} (SKC Ltd, Dorset, UK). The airflow was measured at the beginning and at the end of each sampling period using a rotameter.

Analysis of air filters

The air filters were analysed at NIOH. Air filters were placed in a Teflon autoclave and 2 mL of *aqua regia* and 0.2 mL of hydrofluoric acid were added to dissolve the welding aerosol.

The material collected on the inside surfaces of the Millipore cassette was recovered by washing and decantation with an additional 2 mL of ultrapure water (added 25% HNO_3), which was also transferred to the autoclave. A known quantity of beryllium chloride solution was added as an internal spectroscopic standard before acid digestion. The autoclaves were

heated in a microwave unit (MLS 1200, Teflon Container SV140, 10 bar, Milestone, Sorisole, Italy). The dilution volume for the digest was 15 mL.

Details of the inductively coupled plasma optical emission spectrometric (ICP-OES) measurements of elements in the acid digest and the quality assurance protocol were as described previously.²⁶ In-house prepared reference workroom air filters were employed. These were traceable to international certified reference materials and simulated the occupational exposure limits (OELs) of individual elements. The long-term daily use at NIOH has permitted the achievement of a 2% or better accuracy and reproducibility (day-to-day variation) for the measured elements.

Measurements of elements in blood and urine

The elements were measured at NIOH. For the measurements of cadmium (Cd), mercury (Hg), Mn and lead (Pb) in whole blood, 1.5 mL of 65% ultrapure nitric acid was added to 1 mL of whole blood in a polypropylene digestion tube. The tube was heated to $95\text{ }^{\circ}\text{C}$ for 1 hour. After being cooled to room temperature, 200 μL of an internal standard solution (containing ^{115}In for ^{114}Cd ; ^{60}Ni for ^{55}Mn ; ^{204}Tl for $^{206,207,208}\text{Pb}$ and $^{200,201,202}\text{Hg}$) was added to the sample and diluted to volume (10 mL).

To prevent any risk of laboratory acquired infections and to dissolve urine precipitates, all urine samples were, after thawing, heated for one hour at $95\text{ }^{\circ}\text{C}$ prior to analysis. To 1 mL of urine was added 200 μL of an internal standard solution (containing ^{72}Ge for ^{75}As and ^{78}Se ; ^{115}In for ^{114}Cd , ^{59}Co , ^{52}Cr , ^{60}Ni and ^{55}Mn ; ^{129}I for ^{127}I ; ^{205}Tl for $^{206,207,208}\text{Pb}$ and $^{200,201,202}\text{Hg}$) and diluted to volume (5 mL).

The digested blood and diluted urine samples were analysed by inductively coupled plasma sector field mass spectrometry (ICP-SF-MS) using an Element 2 mass spectrometer (Thermo Electron, Bremen, Germany) calibrated with whole blood and urine matrix matched standard solutions. The instrument was programmed to determine Cd by use of the $^{114}\text{Cd}^+$ ion with automatic mass correction caused by the $^{114}\text{Sn}^+$ ionic interference. Since the Mo concentrations in whole blood is around 1 ng mL^{-1} or lower, any mass interference at ^{114}Cd from the $^{98}\text{Mo}^{16}\text{O}$ was not considered to contribute to the overall signal. Since the Mo concentrations in urine may be 100-fold higher than in whole blood, it is of paramount importance to correct for the $^{98}\text{Mo}^{16}\text{O}$ mass interference when measuring ^{114}Cd in urine.

Seronorm™ Trace Elements human whole blood and urine quality control materials were used for quality assurance of the element measurements. The obtained analytical results are depicted as an electronic attachment. Creatinine in urine was measured by the Jaffe reaction using a SFA-200 flow injection analyzer (Burkard Scientific Ltd., Uxbridge, UK).

Statistics

Most of the measured variables had a skewed distribution by visual inspection. They were log-transformed if the skewness exceeded 2.0. For those variables the geometric mean (GM) concentrations are given, while the arithmetic means (AM) are otherwise presented. Analysis of variance was used for group comparisons of continuous variables, and the least square

differences calculated in order to separate between groups when more than two groups were compared. Dunnett's test was used when comparing the controls with several subgroups.

Least square regression analysis, yielding the Pearson's correlation coefficient, was used to assess univariate associations. The level of statistical significance was set two-tailed at $p < 0.05$. The statistical calculations were performed on a PC using SPSS[®], version 11.5.

Results

The Mn-content of the steel on which it was welded was known for 175 of the 188 sampled welding days. The AM content was 1.3% (up to 2.6%). For most of the remaining sampling days, more than one steel type was welded by the welder. The main elemental constituents of the welding aerosol in 188 collected workroom air samples were Fe, followed by the flux elements Ca and Si. For all collected air samples, the GM concentration of Mn was $97 \mu\text{g m}^{-3}$ (range 3–4620 $\mu\text{g m}^{-3}$) (Table 2). The applied welding methods were known for 177 days of air sampling (Table 2). For the remaining 11 welding days more than one method was used by the welder. The elemental air concentrations were generally lower for automated welding; the GM concentration of Mn was $19 \mu\text{g m}^{-3}$. No substantial differences in the welding aerosol compositions were observed between semi-automatic welding and manual welding, except for the flux components Ca, Na and K. Those concentrations were about 3 to 4-fold higher in the manual welding process.

The chromium (Cr) content of the steel was known for 160 welding days (Table 3). Stainless steel ($n = 54$) had an AM Cr content of 18.4% (range 12.5–23.2) in contrast to the AM of 0.05% (range 0–0.5) for the mild steel ($n = 106$). The GM air concentration of Mn was non-significantly lower when welding stainless steel compared to mild steel, the difference being of statistical significance for automated welding.

High Pearson's correlations coefficients (not tabulated) were calculated between the concentrations of Fe and Mn in the welding aerosol for semi-automatic ($r = 0.80$; $p < 0.001$; $n = 58$), manual welding ($r = 0.79$; $p < 0.001$; $n = 90$) and automated welding ($r = 0.82$; $n = 29$).

The corresponding correlation coefficients between air-Mn and air-Cr and air-Ni, respectively, were 0.32 ($p < 0.05$; $n = 58$) and 0.18 (not significant; $n = 58$) for semi-automatic welding and 0.41 ($p < 0.001$; $n = 90$) and 0.39 ($p < 0.001$; $n = 90$) for manual welding. Fig. 1 shows the association between Mn and Fe in all air samples.

The Mn concentration in the workroom air was on average 10.6% (GM) (95% CI; 9.5 to 11.9; $n = 177$) of that of the Fe concentration when considering all samples where the welding methods were known (not tabulated). The corresponding figures for automated welding were 6.5% (95% CI 5.0 to 8.4; $n = 29$), for semi-automatic welding 11.3% (95% CI 9.2 to 13.8; $n = 58$) and for manual welding 11.9% (95% CI 10.3 to 13.9; $n = 90$). When stratifying the material according to welding stainless steel (GM 12.6%; 95% CI 9.8–16.2; $n = 54$) or mild steel (GM 10.6%; 95% CI 9.4–12.0; $n = 106$), the percentage did not differ significantly between the welding of the two steel types ($p = 0.23$).

The AM concentration of B-Mn was nearly 25% higher in all welders when compared to the controls (Table 4). Also higher B-Pb concentrations were measured. The GM concentration of U-Mn was of borderline significance higher in the welders, while U-Fe was almost identical in the two groups. The expected observation of higher U-Ni and U-Cr in the welders was in contrast to the observation of the highly statistically significantly lower U-Co.

The associations between the welding aerosol components from one day before and two days before (and the mean thereof) the collection of biological samples and the concentration of elements in blood and urine are shown in Table 5. The best fit between air-Mn and B-Mn was found when air samples collected one day prior to the biological sampling

Table 2 The geometric mean (GM) air concentrations and range (in $\mu\text{g m}^{-3}$) of welding aerosol components in 188 full-shift air samples collected among 96 male welders during two successive days, and according to welding method. The 90th percentile is shown for all samples. The calculated p -values refer to comparisons made between welding methods

	All		Automated $N = 29$ GM (range)	Semi-automatic $N = 58$ GM (range)	Manual $N = 90$ GM (range)
	$N = 188$	90th perc.			
Mn ^{ab}	97 (3–4620)	470	19 (3–265)	131 (7–1510)	121 (4–4620)
Fe ^{ab}	894 (106–20300)	3394	297 (110–2180)	1165 (106–6290)	1013 (208–20300)
Cr ^{ab}	13 (1–976)	165	6 (2–18)	12 (2–387)	16 (1–976)
Ni ^{ab}	15 (1–270)	59	6 (1–29)	15 (1–270)	18 (2–240)
Pb ^{ab}	2.3 (<0.9–79)	6.9	1 (<0.9–7)	3 (<0.9–10)	3 (<0.9–79)
Al ^{ab}	57 (12–2280)	144	79 (13–2280)	53 (15–341)	48 (12–290)
Cu ^{ab}	11 (2–168)	35	3 (2–31)	16 (2–127)	13 (2–168)
Zn ^{ab}	19 (1–2490)	203	5 (1–203)	23 (2–335)	25 (1–2490)
Ti ^{abc}	15 (1–787)	69	4 (1–29)	14 (1–112)	23 (1–787)
Mg ^{abc}	17 (3–1580)	54	36 (7–1160)	11 (3–175)	15 (3–106)
Ca ^{abc}	215 (26–8630)	864	203 (46–5450)	100 (26–343)	319 (65–8630)
Si ^{ab}	190 (25–3460)	527	112 (25–3460)	194 (40–939)	205 (33–3300)
Na ^{bc}	89 (2–8380)	632	31 (13–143)	41 (2–354)	177 (7–8380)
K ^{abc}	75 (5–9880)	527	22 (10–66)	39 (8–406)	149 (5–9880)

^a $p < 0.05$ automated vs. semi-automatic. ^b $p < 0.05$ automated vs. manual. ^c $p < 0.05$ semi-automatic vs. manual.

Table 3 The geometric mean (and range) air concentrations (GM) of Fe, Mn, Cr and Ni (in $\mu\text{g m}^{-3}$) according to the chromium (Cr) content of the steel

	>1% Cr		<1% Cr		P
	GM	Range	GM	Range	
All samples	N = 54		N = 106		
Cr content (in %) ^a	18.4	12.5–23.2	0.05	0–0.52	
Air-Fe	579	106–12900	1050	117–20300	<0.001
Air-Mn	73	3–3930	112	4–4620	0.08
Air-Cr	37	3–976	8	1–315	<0.001
Air-Ni	21	2–270	11	1–78	0.004
Semi-automatic	N = 16		N = 30		
Cr content (in %) ^a	18.3		0.09		
Air-Fe	746	106–3900	1840	615–6290	<0.001
Air-Mn	153	7–741	205	27–1510	0.42
Air-Cr	73	7–387	7	2–67	<0.001
Air-Ni	28	2–270	10	1–78	0.03
Manual	N = 24		N = 61		
Cr content (in %) ^a	16.6		0		
Air-Fe	785	208–1290	1060	224–20300	0.15
Air-Mn	126	18–3930	115	4–4620	0.75
Air-Cr	57	5–976	10	1–315	<0.001
Air-Ni	34	3–240	14	2–59	0.001
Automated	N = 14		N = 15		
Cr content (in %) ^a	21.7		0.20		
Air-Fe	256	110–577	341	117–2180	0.37
Air-Mn	12	3–53	30	6–265	0.04
Air-Cr	9	3–18	4	2–11	<0.001
Air-Ni	7	2–25	5	1–29	0.33

^a Arithmetic mean.

were considered. The air-Mn concentrations measured two days before the blood sampling were not quite significantly associated with B-Mn. The association between air-Mn collected one day prior to biological sampling and U-Mn was of borderline significance. The welders were stratified into three equally large groups according to their air-Mn concentration the day before the collection of the biological samples. The highest exposed welders had nearly $3 \mu\text{g l}^{-1}$ higher AM concentration of B-Mn than the controls (Fig. 2). B-Mn was statistically significantly higher in the highest exposed subjects compared to the controls (Dunnett's test).

U-Cr was also associated with current exposure (Fig. 3). The lowest GM concentration of U-Cr was found in the controls, followed by the former welders. Increasing concentrations of U-Cr in relation to increasing current Cr exposure was observed in the welders.

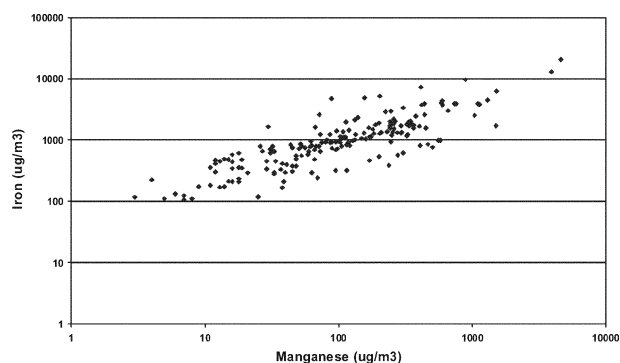


Fig. 1 The relationship between Mn and Fe in 188 full-shift air samples collected during welding.

The exposure to welding fumes had ceased on average 5.8 years (range 4–7) earlier in the 27 former welders. The youngest ex-welder was 41 years old. Therefore all controls and current welders ≥ 41 years of age were used for comparison. The ex-welders had significantly higher AM concentration of B-Mn than the controls (Table 6). The mean concentrations of U-Mn, U-Fe and U-Co were significantly lower in comparison to the controls, but U-Cr was higher.

The mean concentration of the essential trace elements U-I and U-Se were similar in the two groups, as was also the case for the electrolytes U-Mg and U-Ca.

Discussion

This investigation describes some characteristics of exposure and associations between Mn in workroom air during welding and Mn concentrations in biological samples. The issue of element concentrations in biological samples of former welders who have received the diagnosis of manganism is also addressed. This has to our knowledge not been previously reported. The controls used in the present investigation work in the same plants as the welders. As they are also factory workers of the same age and with similar education, it can be assumed that they represent a well-suited group for comparison.

The work tasks were only welding related. Thus, the collected air samples represent their total air exposure, with the welding operation as the only significant source of aerosol generation.

The aerosol generated during welding consists of respirable particles. The sampling characteristics of the 25 mm sampling

Table 4 The geometric mean concentrations (GM), ranges and the 90th percentiles of elements in whole blood and urine of 96 male welders and 96 male controls

	Welders			Controls			<i>P</i>
	GM	Range	90th perc.	GM	Range	90th perc.	
Whole blood/ $\mu\text{g l}^{-1}$							
B-Mn ^a	8.6	3.7–24	13.4	6.9	2.5–14	9.8	<0.001
B-Cd ^a	1.1	0.1–5.4	2.4	0.9	0.1–4.4	2.0	0.30
B-Hg	1.7	0.1–25	4.5	1.5	0.2–8.1	3.8	0.30
B-Pb	47	18–162	95	37	11–208	75	0.001
Urine/ $\mu\text{g g}^{-1}$ cre ^b)							
U-Mn	0.17	0.03–5.5	0.82	0.12	0.02–10.2	0.66	0.07
U-Fe	48	19–436	74	51	17–1760	132	0.62
U-Cr	2.6	0.14–51	11	0.35	0.04–13	1.3	<0.001
U-Ni	4.0	0.68–28	16	2.3	0.46–111	5.7	<0.001
U-Hg	0.17	0.02–1.8	0.54	0.13	0.02–1.9	0.37	0.07
U-As	17	1.1–294	112	18	2.7–448	101	0.76
U-Cd	0.22	0.02–2.6	0.77	0.24	0.02–2.7	1.0	0.60
U-Zn	224	3.4–2390	712	193	4.7–1250	698	0.46
U-Co	0.25	0.03–11	0.62	0.39	0.09–12	1.0	<0.001
U-Ca ^a	81	2.1–423	157	90	12–346	175	0.32
U-Mg	70	6.9–389	148	67	12–234	126	0.58
U-Se	16	3.4–53	24	16	3.4–53	21	0.77
U-I	83	17–687	230	87	17–687	193	0.63

^a AM: arithmetic mean. ^b Cre: creatinine.

cassette that we used, and that it could be mounted in the breathing zone of the welders underneath the welding helmet, means that the cassette is well-suited for welding fume exposure measurements.⁷ Since all particles of the welding aerosol are part of the respirable aerosol fraction, the sampling efficiency of this “total” sampler is comparable with a respirable cyclone, thus simplifying the air exposure assessment considerably.

Two samples for each welder were collected, one on each of two successive days. The range in the air-Mn concentrations was large, from 3 to 4620 $\mu\text{g m}^{-3}$, and the intra-individual variation between days could be considerable. The measured concentrations are comparable with previously reported air-Mn concentrations from welding.^{17,19,20} The exposure to Mn was much lower when automated welding was compared to manual or semi-automatic welding, while the exposure levels were similar for the latter two methods. The mean Mn content of the welded steel was 1.3%, thus suggesting that the exposure

reflects welding on steel with “normal” Mn-content. No large difference in the mean air concentrations of Mn was measured when welding stainless steel as compared to mild steel, suggesting that the risk for high exposure to Mn may be largely independent of working with these two types of steel. As expected, the air-Cr concentrations were higher when welding stainless steel. A high correlation between Fe and Mn was found in the welding aerosol. If this association is true and universal, one may use measured Fe concentrations in welding fumes as a proxy estimate of exposure to Mn in epidemiological studies if measurements of Mn are not available.

The welders had about 25% ($1.7 \mu\text{g l}^{-1}$) higher mean concentration of B-Mn than the controls. Although the group mean was higher, the B-Mn of the individual welder was generally within the range of the controls. An increase of

Table 5 The correlation coefficients (Pearson’s *r*) between selected biomarkers of exposure and the air concentrations of the respective elements (all log-transformed) assessed two days before sample collection (air sample 1), one day before sample collection (air sample 2) and the average between the two days (air sample mean)

	Air sample 1	Air sample 2	Air samples mean
Whole blood/ $\mu\text{g l}^{-1}$			
B-Mn	0.19 ^a	0.31 ^c	0.22 ^b
B-Pb (log)	−0.01	0.20 ^a	0.18 ^a
Urine/ $\mu\text{g g}^{-1}$ cre			
U-Mn (log)	0.16	0.19 ^a	0.16
U-Cr (log)	0.21 ^a	0.32 ^c	0.33 ^c
U-Ni (log)	0.47 ^d	0.36 ^d	0.39 ^d

^a $p < 0.10$. ^b $p < 0.05$. ^c $p < 0.01$. ^d $p < 0.001$.

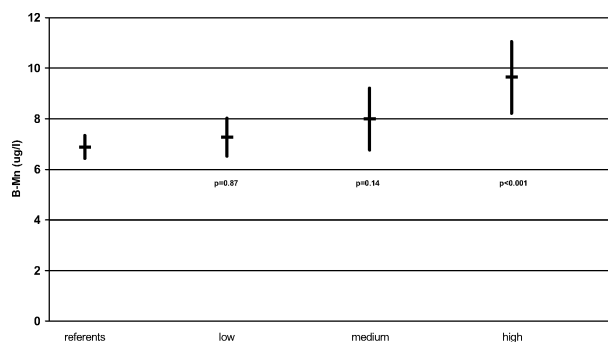


Fig. 2 The arithmetic mean (and 95% CI) concentration of B-Mn in controls ($n = 96$) and in welders stratified into low ($n = 30$), medium ($n = 31$) and highly ($n = 30$) exposed according to their level of air exposure to Mn on the day before blood sampling. The GM (range) air concentrations were $27 \mu\text{g m}^{-3}$ (5–66), $122 \mu\text{g m}^{-3}$ (67–239) and $495 \mu\text{g m}^{-3}$ (247–4620), respectively. The *p*-values are referring to Dunnett’s test comparing the exposed subgroups to the controls.

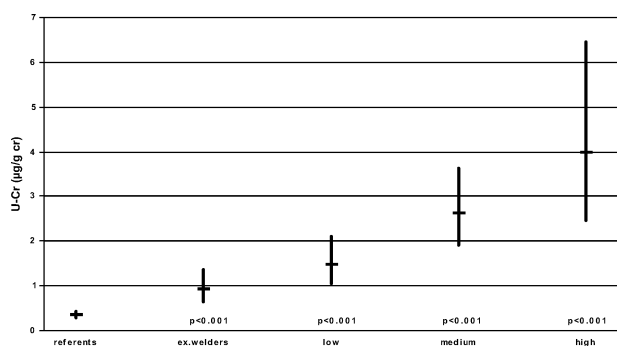


Fig. 3 The geometric mean (and 95% CI) concentration of U-Cr in controls ($n = 96$), in former welders ($n = 26$) and in welders stratified into low ($n = 31$), medium ($n = 31$) and highly ($n = 29$) exposed according to their level of air exposure to Cr on the day before urine sampling. The GM (and range) air concentrations were $4.2 \mu\text{g m}^{-3}$ ($0.9\text{--}6.5$), $8.8 \mu\text{g m}^{-3}$ ($6.7\text{--}13$) and $100 \mu\text{g m}^{-3}$ ($16\text{--}976$), respectively. The p -values refer to Dunnett's test comparing the currently and previously exposed subgroups to the controls.

Mn in serum of about $2 \mu\text{g l}^{-1}$ has been reported recently in welders.^{17,23} Whether the increase found in the present study may be related to the serum or to the cellular compartment of the blood cannot be determined. Little is known about the kinetics of Mn in the blood of occupationally exposed humans. A study of one miner could indicate at least a two compartment kinetic model in whole blood, but the data are sparse.²⁷ A correlation was found on an individual basis between B-Mn and Mn in air sampled the day before the collection of the blood samples. We have not found any reports on the association between air-Mn and B-Mn in welders. However, we have previously reported a weak in-

dividual association between semi-soluble Mn compounds in the respirable aerosol fraction sampled approximately at the time of blood sampling and B-Mn in the Mn-alloy producing industry.²⁸ Two other recent studies reported associations between Mn in "total" dust and B-Mn,^{29,30} thus increasing the evidence that B-Mn is associated with the current air exposure to Mn. Other studies have, however, not found this association on an individual basis.³¹

B-Mn was also higher in the former welders when compared to the controls of similar age. They have an average exposure to welding fumes of 23.1 years, and have received the diagnosis of manganism. Thus, one could assume that they have been substantially exposed to Mn-containing welding fumes, and that they have had higher B-Mn as they still were exposed. This result may suggest long-term accumulation of Mn that can be reflected in their B-Mn concentration even several years after the cessation of exposure. This has to our knowledge previously not been observed in welders, and could suggest the existence of a compartment with a very long elimination half-life of Mn. Information on the anatomical location of potential Mn-deposits in former welders is presently scarce, but some evidence exist indicating Mn-deposits in the lungs.³² This could indicate accumulation of Mn in addition to accumulation of Fe. Pulmonary Fe accumulation has been known to be associated with siderosis in welders for some time.³³

The welders exposed to the lowest amounts of Mn in air had B-Mn concentrations hardly above the level of the controls, while the highest exposed welders had about 40% higher B-Mn ($3 \mu\text{g l}^{-1}$). This higher B-Mn was associated with an average air concentration of Mn of $495 \mu\text{g m}^{-3}$ (range $247 \mu\text{g m}^{-3}$ – $4620 \mu\text{g m}^{-3}$). Slightly higher B-Pb was also found in the welders which is compatible with the presence of some lead in the welding aerosol.

Table 6 The geometric mean concentrations (GM) of elements in the urine of 27 patients with manganism, and 42 welders and 42 controls ≥ 41 years of age

	Patients		Welders		Controls		$p\text{-value}_{\text{ANOVA}}$
	Range	GM	Range	GM	GM	Range	
Age ^a /years	50.7	41–58	46.8	41–65	48.4	41–66	—
Years of welding	23.1	15–30	21.4	1–40	—	—	—
Whole blood/ $\mu\text{g l}^{-1}$							
B-Mn ^{acd}	8.7	5.2–19	8.5	4.7–22	7.0	3.8–14	0.05
B-Hg	1.8	0.25–6.5	1.9	0.2–8.8	1.8	0.17–8.1	0.96
B-Pb ^{bd}	31	13–83	48	18–141	36	16–208	0.002
B-Cd ^{ab}	0.59	0.08–2.3	0.90	0.05–3.4	0.70	0.12–2.0	0.12
Urine/ $\mu\text{g g}^{-1} \text{cre}^e$							
U-Fe ^{bcd}	34	12–85	49	19–141	69	26–1770	<0.001
U-Mn ^{bc}	0.07	0.03–0.17	0.12	0.03–1.1	0.19	0.02–10	0.001
U-Cr ^{bcd}	0.93	0.19–8.1	2.7	0.15–23	0.52	0.06–13	<0.001
U-Ni ^b	2.2	0.82–5.8	3.7	1.1–28	2.8	0.46–111	0.06
U-Cd	0.32	0.05–2.5	0.28	0.02–2.6	0.35	0.03–2.2	0.58
U-Hg	0.11	0.01–5.2	0.16	0.04–1.8	0.17	0.06–1.9	0.12
U-As	29	4.0–602	24	1.2–294	26	2.7–448	0.80
U-Zn	124	6.3–762	185	3.4–2390	203	8.5–1230	0.35
U-Co ^{cd}	0.28	0.10–4.7	0.23	0.03–1.1	0.42	0.14–5.9	0.002
U-Mg	56	4.1–147	58	6.9–150	60	25–148	0.91
U-Ca ^a	96	17–203	83	2.1–423	106	12–346	0.36
U-I	82	33–488	89	17–687	97	21–567	0.69
U-Se	14	9.0–27	16	3.4–53	15	9.8–24	0.59

^a AM: arithmetic mean. ^b $p < 0.05$ between patients and welders. ^c $p < 0.05$ between patients and controls. ^d $p < 0.05$ between welders and controls. ^e Cre: creatinine.

A non-significantly higher concentration of U-Mn in the current welders than in the controls was found. This is compatible with the recent observation in Chinese welders who also had non-significantly higher U-Mn than the controls.²³ The concentrations of U-Ni and U-Cr were significantly higher in the welders, as previously shown.^{34–37} The increase in B-Mn and the dose–response association between air-Mn and B-Mn in the welders are strong indicators of Mn actually being taken up by the body of the welders. In this context it is interesting that the former welders had substantially, and statistically significantly, lower (about 50%) concentrations of Fe, Mn and Co in their urines, although their B-Mn was higher. The divalent cations of these elements (and Ni(II)) are all known to be transported by DMT 1.³⁸

The urinary elemental concentrations were not generally lower in the patients, and for instance the concentrations of the essential trace elements such as Se and I or the electrolytes Mg or Ca did not differ between the patients and the controls (or the current welders). These elements, however, are not known to be transported by DMT 1. The *a priori* expected higher U-Cr concentration was also observed. The higher U-Cr excretion may be related to long-term deposits of Cr in the former welders. It should also be emphasized that the concentrations of U-Mn in the current welders were related to the duration of welding, however in the negative direction (not shown). This suggests that being exposed to Mn-containing welding fumes at high levels for many years contributes to lower U-Mn (actually also U-Co and U-Fe) during and/or after the cessation of exposure. These results could point to a dysregulation in the urinary excretion of divalent cations that are known to be transported through DMT 1, but we have not found any reports addressing this issue.

Little is known about the urinary excretion mechanisms of trace elements in humans. DMT 1 may be involved in the renal handling of Fe. Large amounts of mRNA encoding DMT 1 are present in the kidney of rats and mice.^{39–42} The glomerularly filtered Fe is reabsorbed mainly in the loop of Henley and the collecting duct in rats, and Mn, Cu and Zn may reduce Fe reabsorption.³⁹ Dietary Fe alters the renal DMT 1 expression and the urinary Fe excretion in rats. Low dietary Fe resulted in increased renal DMT 1 expression accompanied by decreased urinary Fe excretion, while high dietary Fe had the converse effect.⁴¹ There are, however, indications that DMT 1 acts as a divalent cation reuptake system at the brush border of the kidney proximal tubule cells in mice, thus suggesting differences between mice and rats in the anatomical location of the DMT 1-associated re-uptake.⁴⁰ Further complicating the issue does a study of Belgrade rats that carry a mutation in the gene encoding for DMT 1, suggesting that a lack of DMT 1 may be compensated by other, as yet unknown, mechanisms.⁴² It is currently not possible mechanistically to explain our finding of a reduced excretion of U-Mn in the patients.

In conclusion, welding on steel containing “normal” Mn-levels results in a dose-related increase in B-Mn. No substantial difference in exposure to Mn was found when welding stainless steel compared to mild steel. On average about ten times more Fe than Mn was measured in the work room air. The results further suggest that long-term high exposure to welding fumes may result in alterations of the urinary excretion of certain cations known to be transported through the DMT 1 transport system.

tion of certain cations known to be transported through the DMT 1 transport system.

References

- 1 D. E. McMillan, *Neurotoxicology*, 1999, **20**, 499.
- 2 A. H. Reides, in *Ullman's Encyclopedia of Industrial Chemistry*, ed. B. Elvers, S. Hawkins and G. Schulz, VCH Verlagsgesellschaft GmbH, Weinheim, 5th edn, 1990, vol. A16, pp. 123–143.
- 3 J. M. Antonini, M. D. Taylor, A. T. Zimmer and J. R. Roberts, *J. Toxicol. Environ. Health*, 2003, **67**, 233.
- 4 A. T. Zimmer, *J. Environ. Monit.*, 2002, **4**, 628.
- 5 P. Hewett, *Am. Ind. Hyg. Assoc. J.*, 1995, **56**, 128.
- 6 D. Stephenson, G. Seshadri and J. M. Veranth, *Am. Ind. Hyg. Assoc. J.*, 2003, **64**, 516.
- 7 J. H. Vincent, in *Aerosol Science for Industrial Hygienists*, Elsevier Science Ltd, Oxford, UK, 1st edn, 1995, pp. 238–303.
- 8 A. T. Zimmer and P. J. Biswas, *J. Aerosol Sci.*, 2001, **32**, 993.
- 9 V. Voitkevich, in *Welding Fumes—Formation Properties and Biological Effects*, Cambridge, Abington, England, 1995, pp. 18–71.
- 10 D. B. Drown, S. G. Oberg and R. P. Sharma, *J. Toxicol. Environ. Health*, 1986, **17**, 201.
- 11 H. Roels, G. Meiers, M. Delos, I. Ortega, R. Lauwerys, J. P. Buchet and D. Lison, *Arch. Toxicol.*, 1997, **71**, 223.
- 12 D. G. Ellingsen, S. M. Hetland and Y. Thomassen, *J. Environ. Monit.*, 2003, **5**, 84.
- 13 J. L. Turi, F. Yang, M. D. Garrick, C. A. Piantadosi and A. J. Ghio, *Free Radical Biol. Med.*, 2004, **36**, 850.
- 14 K. Thompson, R. Molina, T. Donaghey, J. D. Brain and M. Wessling-Resnick, *Toxicol. Appl. Pharmacol.*, 2006, **210**, 17.
- 15 E. Heilig, R. Molina, T. Donaghey, J. D. Brain and M. Wessling-Resnick, *Am. J. Physiol.: Lung Cell. Mol. Physiol.*, 2005, **288**, 887.
- 16 E. A. Heilig, K. J. Thompson, R. M. Molina, A. R. Ivanov, J. D. Brain and M. Wessling-Resnick, *Am. J. Physiol.: Lung Cell. Mol. Physiol.*, 2006, **290**(6), 1247.
- 17 L. Lu, L.-L. Zhang, G. J. Li, W. Guo, W. Liang and W. Zheng, *Neurotoxicology*, 2005, **26**, 257.
- 18 S. V. Chandra, G. S. Shukla and R. S. Srivastava, *Clin. Toxicol.*, 1981, **18**, 407.
- 19 H. Sińczuk-Walczak, M. Jakubowski and W. Matczak, *Int. J. Occup. Med. Environ. Health*, 2001, **14**, 329.
- 20 W. Matczak and M. Przybylska-Stanisławska, *Med. Pr.*, 2004, **55**(6), 481 (in Polish).
- 21 J. M. Antonini, *Crit. Rev. Toxicol.*, 2003, **33**, 61.
- 22 J. Järvisalo, M. Olkinuora, M. Kiilunen, H. Kivistö, P. Ristola, A. Tossavainen and A. Aitio, *Int. Arch. Occup. Environ. Health*, 1992, **63**, 495.
- 23 G. J. Li, L.-L. Zhang, L. Lu, P. Wu and W. Zheng, *J. Occup. Environ. Med.*, 2004, **46**, 241.
- 24 I. J. Yu, J. D. Park, E. S. Song, K. S. Song, K. T. Han, J. H. Han, Y. H. Chung, B. S. Choi, K. H. Chung and M. H. Cho, *Neurotoxicology*, 2003, **24**, 777.
- 25 Y. Kim, K. S. Kim, J. S. Yang, I. J. Park, E. Kim, Y. Jin, K.-R. Kwon, K. H. Chang, J.-W. Kim, S.-H. Park, H.-S. Lim, H.-K. Cheong, Y. C. Shin, J. Park and Y. Moon, *Neurotoxicology*, 1999, **20**, 901.
- 26 Y. Thomassen, E. Nieboer, D. Ellingsen, S. Hetland, T. Norseth, J. O. Odland, N. Romanova, S. Chernova and V. P. Tchachtine, *J. Environ. Monit.*, 1999, **1**, 15.
- 27 G. C. Cotzias, K. Horiuchi, S. Fuenzalida and I. Mena, *Neurology*, 1968, **376**.
- 28 D. G. Ellingsen, S. M. Hetland and Y. Thomassen, *J. Environ. Monit.*, 2003, **5**, 84.
- 29 R. Lucchini, P. Apostoli, C. Perrone, D. Placidi, E. Albin, P. Migliorati, D. Mergler, M.-P. Sassine, S. Palmi and L. Alessio, *Neurotoxicology*, 1999, **20**, 287.
- 30 P. Apostoli, R. Lucchini and L. Alessio, *Am. J. Ind. Med.*, 2000, **37**, 283.
- 31 H. Roels, R. Lauwerys, P. Genet, M. J. Sarhan, M. de Fays, I. Hanotiau and J.-P. Buchet, *Am. J. Ind. Med.*, 1987, **11**, 297.
- 32 T. Kraus, H.-J. Raithel and K.-H. Schaller, *Zentralbl. Hyg. Umweltmed.*, 1989, **188**, 108.
- 33 M. J. Doherty, M. Healy, S. G. Richardson and N. C. Fisher, *Occup. Environ. Med.*, 2004, **61**, 82.

-
- 34 A. Mutti, A. Cavatorta, C. Pedroni, A. Borghi, C. Giaroli and I. Franchini, *Int. Arch. Occup. Environ. Health*, 1979, **43**, 123.
- 35 B. Åkesson and S. Skerfving, *Int. Arch. Occup. Environ. Health*, 1985, **56**, 111.
- 36 I. C. Stridsklev, B. Hemmingsen, J. T. Karlsen, K. H. Schaller, H. J. Raithel and S. Langård, *Int. Arch. Occup. Environ. Health*, 1993, **65**, 209.
- 37 J. L. Edmé, P. Shirali, M. Mereau, A. Sobaszek, C. Boulenguez, F. Diebold and J. M. Haguenoer, *Int. Arch. Occup. Environ. Health*, 1997, **70**, 237.
- 38 M. D. Garrick, K. G. Dolan, C. Horbinski, A. J. Ghio, D. Higgins, M. Porubcin, E. G. Moore, L. N. Hainsworth, J. N. Umbreit, M. E. Conrad, L. Feng, A. Lis, J. A. Roth, S. Singleton and L. M. Garrick, *Biometals*, 2003, **16**, 41.
- 39 M. Wareing, C. J. Ferguson, R. Green, D. Riccardi and C. P. Smith, *J. Physiol.*, 2000, **524**, 581.
- 40 F. Canonne-Hergaux and P. Gros, *Kidney Int.*, 2002, **62**, 147.
- 41 M. Wareing, C. J. Ferguson, M. Delannoy, A. G. Cox, R. F. T. McMahon, R. Green, D. Riccardi and C. P. Smith, *Am. J. Physiol. Renal Physiol.*, 2003, **285**, 1050.
- 42 C. J. Ferguson, M. Wareing, M. Delannoy, R. Fenton, S. J. McLarnon, N. Ashton, A. G. Cox, R. F. T. McMahon, L. M. Garrick, R. Green, C. P. Smith and D. Riccardi, *Kidney Int.*, 2003, **64**, 1755.