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The Nervous System Effects of Occupational Exposure on Workers in a South African Manganese Smelter

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Abstract

Five hundred and nine production workers at a manganese (Mn) smelting works comprising eight production facilities and 67 external controls were studied cross-sectionally for Mn related neuroehavioural effects. Exposure measures from personal sampling included Mn in inhalable dust as cumulative exposure indices (CEI) and average intensity (INT). Biological exposure and biological effect measures included blood (MnB), urine (MnU) manganese and serum prolactin. Endpoints included items from the Swedish nervous system questionnaire (Q16), World Health Organisation neurobehavioural core test battery (WHO NCTB), Swedish performance evaluation system (SPES), Luria–Nebraska (LN), and Danish product development (DPD) test batteries, and a brief clinical examination. Potential confounders and effect modifiers included age, educational level, alcohol and tobacco consumption, neurotoxic exposures in previous work, past medical history, previous head injury and home language. Associations were evaluated by multiple linear and logistic regression modelling. Modelling assumptions were tested. Average exposure intensity across all jobs ranged from near 0 (0.06 μg/m^3) for external controls to 5.08 mg/m^3 for inhalable Mn, and was greater than the ACGIH TLV for 69% of subjects. Results from the large number of tests performed resolved into three groups. Group 1 shows differences between external unexposed referents and all the exposed and/or differences between internal low exposed referents and the rest of the exposed but no further exposure–response relationships. It includes the Santa Ana, Benton and digit-span tests from the WHO NCTB; the hand tapping and endurance tapping tests from the SPES; Luria–Nebraska item 2L; questionnaire items tired, depressed, irritated, having to take notes in order to remember things, and subjects' perception that they had sex less often than normal; a test of clinical abnormality; and increased sway under two conditions (eyes open without foot insulation, eyes open with foot insulation). Group 2 shows the presence of a more substantive exposure–response relationship. It consists of only two tests: and includes the WHO digit-symbol test (although the major impact is at low exposure and therefore counterintuitive, arguably placing this test in group 3) and the LN item 1R which has a step to a poorer score at high exposure. Group 3 contains the overwhelming majority of test results (almost all the questionnaire items, almost all the DPD tests including tremor, sway and diadochokinesia, and serum prolactin) which were either null or counterintuitive (did not make sense). The CEI was the strongest predictor of test abnormalities, except for the clinical test which was more strongly associated with blood manganese. Despite a comprehensive range of endpoints, and levels of exposure ranging from environmental to industrial, this large study of Mn workers found little convincing

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evidence for a continuum of effects, contributing further questions to current debates about the adequacy of the current ACGIH TLV.

Keywords: **Manganese; Neurotoxicity; Occupational; Exposure; Neurobehavioural**

INTRODUCTION

There has been increasing interest in the chronic nervous system effects of long term occupational Mn exposures below the American Conference of Governmental Industrial Hygienists (ACGIH) threshold limit value (TLV) of 0.2 mg/m^3 (ACGIH, 1996), and in environmental exposures at or above the United States Environmental Protection Agency reference concentration (US-EPA RfC) of 0.05 μg/m^3 (IRIS, 1999) sparked by increasing use of the gasoline additive methylcyclopentadienyl manganese tricarbonyl (MMT) (US-EPA, 1994). Iregren (1999) reviewed 13 studies of Mn exposed workers, showing mostly motor effects with tests for finger tapping, diadochokinesometry and Luria–Nebraska (LN) items, along with postural tremor and sway abnormalities. Pegboard, memory, reaction time and cognitive tests were less conclusive. More recently, Lucchini et al. (1999) found Mn effects with World Health Organisation neurobehavioural core test battery (WHO NCTB) and LN tests in smelter workers at levels around 0.1 mg/m^3. Beuter et al. (1999) and Mergler et al. (1999) found subtle neurobehavioural effects and exposure-age interactions with environmental exposures measured as blood manganese (MnB). As part of the same study Bowler et al. (1999) found mood effects, while Hudnell (1999) located these exposure–response relationships at environmental concentrations around the US-EPA RfC. Roels et al. (1999) in a prospective study, showed reversibility of effects below 0.1 mg/m^3. These studies have raised concerns that the ACGIH TLV might be too high to protect against neurobehavioural effects. On the other hand, no effects were found by either Gibbs et al. (1999) at occupational exposures averaging 0.18 mg/m^3 total dust, or by Deschamps et al. (2001) at exposures averaging below 0.15 mg/m^3 over a 20-year period.

Problems with the relatively few existing studies include small numbers of exposed subjects (the largest study had 141 exposed), and non-standard possibly insensitive neurobehavioural tests. Furthermore, there is relatively poor consistency across studies for exposure effects and for the character of exposure–response relationships. There is still insufficient scientific information relating to subjective symptoms and mood effects, especially from prospective study designs at low exposure levels, and to the individual clinical implications of group effects found. This study sought to address some of these problems by studying a large number of subjects within a long term average exposure spectrum ranging from near 0 to 25 times the ACGIH TLV. It was not possible in this study to meaningfully disentangle chronic from acute effects of manganese exposure and accordingly the relationship between the cumulative exposure index (CEI) an integrated intensity–duration exposure measure across jobs and a number of neurobehavioural endpoints was investigated. Selection of endpoints was based upon those reported in the state-of-the-art review of the scientific literature by Iregren (1999) which lists tests with evidence for Mn effects. A companion paper (Myers et al., 2003b, in press) details atmospheric and biological exposure/effect measures and their correlations, and the implications of these for exposure and medical surveillance in the workplace.

This study sought to clarify some of the inconsistencies in the literature and particularly to better understand the nature of the exposure–response relationship. ''Exposure–response relationship'' denotes the existence of structure beyond a simple difference between external referents and all the exposed and/or between the internal referents and the rest of the exposed, while ''continuity of response'' denotes continuing incremental impairment in function as exposure increases. Scientific understanding of the impact of Mn exposure on nervous system function leads to the hypothesis that there should be no adverse impact on function at low exposures, given that Mn is an essential element in human nutrition; that there should be some threshold for exposure above which adverse effects appear; and that these adverse effects should increase in frequency with higher levels of exposure beyond that threshold. This latter phenomenon is a continuity of exposure–response above the threshold.

MATERIAL AND METHODS

A cross-sectional study was conducted on manganese exposed subjects drawn from one of the eight production environments in a Mn smelting works in

South Africa. Different plants or activities at the works were divided into notionally high (three Ferro- and Silico-Mn smelters), medium (one Ferro-Silicon smelter, raw and finished materials handling plants) and low (quality control laboratories, administration and security workers, and a chemical plant making no use of Mn). The low exposure group served as an internal control with no direct Mn exposure. Two hundred subjects were targeted by random selection from mainly low skilled production workers in each of these three exposure groups. Maintenance workers with highly variable exposures were excluded. High exposure was considered to be above 2 mg/m^3, medium between 2 and 0.1 mg/m^3, and low exposure below 0.1 mg/m^3. All of the plants were geographically located in a single works, and even the plant not directly using Mn received exposure from fugitive emissions in the works environment. Consequently, an external unexposed reference group from an electrical fittings assembly plant without any direct or indirect Mn exposure was included in the study.

The study was conducted over a period of 1 year from late 1999 to late 2000. Between five and eight subjects drawn from either morning (starting at 7:00 a.m.) or afternoon (starting at 2:00 p.m.) shifts were seen each day. Four subjects were allocated to each of four testing stations, viz. questionnaire/examination, Swedish performance evaluation system (SPES), World Health Organisation neurobehavioural core test battery (WHO NCTB)/Luria–Nebraska (LN) and Danish product development (DPD) simultaneously and rotated through the other testing stations. A trained neurobehavioural psychologist served as the study coordinator and administered the SPES tests, while two other trained interviewers administered the DPD battery, and the WHO/LN tests, respectively. A single trained interviewer conducted all the questionnaire interviews and brief clinical examinations in the home language of the subjects which included English, Afrikaans, isiXhosa, isiZulu and Sesotho.

There were no refusals to participate in the study although there were some refusals to provide blood samples on cultural grounds. Recruitment of subjects continued over the course of a year until the requisite number of subjects was obtained from each of the three gross exposure categories, viz. low, medium and high.

Sampling strategy, measurement and analysis for atmospheric Mn, as well as for blood (MnB) and urine (MnU) manganese are described elsewhere, as is the calculation of exposure indices (Myers et al., 2003b, in press). Representative exposure intensity estimates were obtained for all Mn exposed production jobs at all plants at the works and at the external reference plant. A cumulative exposure index (mg/m^3 per year) was calculated for each subject by summing the product of the average exposure intensity for each job by the number of years this activity was performed. This was divided by total years of service (LOS) in the smelter works to yield a measure of average exposure intensity across all exposed jobs (INT) (mg/m^3). The cumulative exposure index and average intensity were also categorised using cutpoints using the cutpoints in Table 1.

Table 1
Categorisation of exposure variables

Category names	Exposure ranges (mg/m^3)	Significance	n
Average exposure intensity across all jobs (INT) (mg/m^3)			
0	0	Unexposed external referents	67
1	$0 < x \leq 0.1$	LOAEL = 0.1	105
2	$0.1 < x \leq 0.2$	ACGIH TLV = 0.2	50
3	$0.2 < x \leq 1$	SA OEL for fumes = 1	235
4	$1 < x \leq 2$	Company advisors safe level = 2	59
5	>2		59
Cumulative exposure index across all jobs (CEI) (mg/m^3 per year)			
0	0	External unexposed referents	67
1	$0 < x \leq 1.3$	Bottom quintile of exposed	104
2	$1.3 < x \leq 5.4$	Second last quintile	98
3	$5.4 < x \leq 10.6$	Middle quintile	103
4	$10.6 < x \leq 22.4$	Second to topmost quintile	101
5	>22.4	Topmost quintile of exposed	102

LOAEL: lowest observed adverse effect level; ACGIH TLV: American Conference of Governmental Industrial Hygienists threshold limit value (TWA); SA OEL: South African occupational exposure limit for Mn fume.

First voided urine specimens were brought in by subjects on the day of testing along with 2 weeks' growth of toenail clippings. Venous blood specimens were collected each day during the testing session between 12:00 noon and 2:00 p.m., with precautions taken to avoid contamination. Specimens were kept on dry ice and sent immediately for analysis of MnB/MnU and serum prolactin to the National Centre for Occupational Health and the South African Institute for Medical Research Laboratories, respectively.

Nervous system outcomes were measured by a battery of 46 questionnaire items drawn from the Swedish Q16 instrument (Axelson and Hogstedt, 1988), and the WHO NCTB questionnaire (WHO, 1986) for autonomic nervous system symptoms, subjective symptoms referable to the nervous system, and neuropsychiatric questions aimed at measuring mood. Dummy questions (ankle swelling and earache) were included to measure reporting bias.

Other questionnaire items measured potential confounders and effect modifiers such as age, educational level, home language and alcohol and tobacco consumption. A detailed life history was obtained for neurotoxic exposures in previous work, past relevant medical history including head injury, and nervous system disease.

A neurobehavioural test battery deemed most appropriate from a review of the literature included seven items (1, 2, 3, 4, 21, 22, 23) or 13 subitems (left and right) testing motor function from the Luria–Nebraska battery (Golden et al., 1980). Test results in integers were categorised into three scores ranging from 0 for high performance to 2 for poorest performance. These scores were dichotomized grouping 0 as a good score (0) and scores 1 and 2 together as a poor score (1). Integer scores were also dichotomised with scores 0 and 1 representing good performance (0) and category 2 being a poor performance (1). The Benton visual retention test for memory, digit-span and the digit-symbol tests for cognitive ability, and the Santa Ana pegboard test for motor function were selected from the WHO NCTB. Finger tapping for the dominant and non-dominant hands and finger tapping endurance for motor function, and the simple reaction time tests were selected from the SPES battery (Iregren et al., 1996).

A device produced by Danish product development (DPD, 1994) was used for quantitative neurometric testing comprising the Catsys test for dysdiadochokinesia, the Tremor test for postural tremor, and the Sway test on a force platform for postural sway. Eight Tremor test parameters are derived from analysis of a tremor power spectrum, and include tremor intensity (m/s^2), median frequency (F50); standard deviation of F50 (sF50) indicating the degree of irregularity of tremor, and the harmonic index (HI) comparing the tremor spectrum with that of a single harmonic oscillation which has a $HI = 1.00$.

The Catsys system tests hand pronation/supination, finger tapping and auditory reaction time yielding 20 parameters. Subjects pronate and supinate or tap on a drum to a metronome at slow and fast rhythms. The time distance between each stimulus and response is measured. The maximum frequency at which rhythmical pronation/supination and tapping can be maintained is also measured.

The Sway test was performed under four conditions: eyes open, no insulation under feet; eyes open, insulation; eyes shut no insulation; and eyes shut with insulation (Despres et al., 2000). For each test a graph of sway in two dimensions is recorded and a composite variable created by the length of distance travelled by the stylus on the graph. One parameter is recorded for each condition.

A brief clinical examination was conducted testing the glabellar reflex and observing facial expression, gait and balance of the subject while walking backwards on a line. Gross abnormality of the limbs was also excluded.

Questionnaires and the details of other methods employed are available on request from the author.

An analysis plan for investigating exposure–response relationships involved the following.

(a) Comparing (adjusted for all potential confounders):
  (i) external unexposed referents with all other subjects; and
  (ii) internal low exposed referents exposed at less than the lowest observed adverse effect level ($LOAEL = 0.1\ mg/m^3$) with the rest of the exposed.
(b) Examining the overall exposure–response relationship between outcomes and exposure variables (cumulative, intensity) using smoothed plots with locally weighted robust regression (see, e.g. Cleveland, 1979) for continuous outcomes, and frequency tables for categorical outcomes.
(c) Further examination of the nature of the exposure–response relationship by examination of structure of the point estimates of effect across categories of exposure (CEI) adjusting for confounders.
(d) Modelling the overall exposure–response relationship or trend using multiple linear or logistic regression with continuous and categorical outcome

variables, respectively, and continuous exposure variables (CEI or INT), adjusting for confounders.
(e) Making a judgement based on examination of the panel of all these results for each candidate exposure–response relationship as to the presence, character and significance of the relationship.

Effects were adjusted throughout for age, years of schooling, smoking status, alcohol consumption, past job exposures to neurotoxins, previous head injury and home language. Stata version 6 software was used (STATA, 1999).

The study was approved by the ethics and research committee of the Health Sciences Faculty of the University of Cape Town. Informed consent was signed by all participants. A research reference panel was set up with representatives of workers and their trade unions, management and researchers to oversee all aspects of the study and to assist the research team. The reference panel served as a conduit for stakeholder input to the research process. While the study was mostly funded by the company, independence of the researchers in planning and conducting the research, and in analysing, interpreting and publishing the results was ensured in a research contract.

RESULTS

In the analyses that follow, unless otherwise stated, all conclusions apply to exposure modelled as a cumulative exposure index integrated across all jobs. Repeat analyses were undertaken for exposure considered as exposure intensity (INT) and length of service (LOS) modelled jointly, or as blood manganese (MnB). Where results were different these are indicated below.

Sociodemographic Characteristics

From the smelter works 201 high, 201 medium, and 107 low exposed workers were tested, along with 67 unexposed reference workers. Descriptive information is provided in Table 2.

This also shows the distribution of other factors that might have affected performance on the neurobehavioural testing in the last panel.

Variables from the pretest questionnaire including previous night hours of sleep, past 24 h alcohol, smoking and coffee consumption and visual acuity were included in regression models but did not influence any of the results.

Table 2
Descriptive statistics: comparison of smelter workers and external unexposed referents

Continuous variables	Smelter mean (S.D.)	Unexposed mean (S.D.)
Age (years)	45.1 (8.4)	38.6 (10.3)
School standard passed (years)	4.7 (3.2)	8.0 (2.5)
Length of service (years)	18.2 (7.6)	9.4 (7.0)
Cumulative exposure index (mg/m^3 per year)	16.0 (22.4)	0
Average intensity of exposure (mg/m^3)	0.82 (1.04)	0
Blood manganese (μg/l)	12.5 (5.6)	6.4 (1.7)
Urine manganese (μg/l)	10.5 (20.3)	0.96 (0.81)
Serum prolactin (μg/l)	6.1 (3.0)	6.2 (2.5)
Categorical variables	**Percent**	**Percent**
Previous job involving neurotoxins	14	6
Previous head injury	28	22
Current alcohol drinker	43	58
Past alcohol drinker	17	9
Current smoker	38	60
Past smoker	32	7

Workplace Exposure

Four hundred and forty-two personal inhalable dust samples were measured in various homogeneous exposure zones for jobs in different production locations. These enabled the construction of exposure indices for each subject (Myers et al., 2003b, in press).

Table 2 shows the cumulative exposure index (CEI; mg/m^3 per year), and average lifetime exposure intensity (INT) (mg/m^3) across all Mn exposed jobs at the works.

Mean INT in the highest exposed of the three Mn smelters was 10 times higher than the ACGIH TLV of 0.2 mg/m^3, and one-third of this level in the least exposed (chemical) plant at the works, while the unexposed reference group was exposed at the USA EPA RfC.

WHO NCTB Tests

Table 3 shows that of four independent tests from the WHO NCTB battery, the digit-symbol test scores of exposed workers were different from the external referents, that internal exposure referents were different from the rest of the exposed and that there was a continuing exposure–response relationship across the exposure range. The digit-span test showed differences between the external referents and all of the exposed and less so between internal referents and the rest of the exposed, but no exposure–response relationship across

Table 3
Selected WHO NCTB test result panels

Analysis		Santa Ana		Benton		Digit-span (forward and backwards)		Digit-symbol	
		β	*P*	*β*	*P*	*β*	*P*	*β*	*P*
Unexposed referents	Mean score	49.36		7.09		15.64		33.81	
Dichotomous comparisons	All exposed vs. external referents	−4.51	0.00	−0.70	0.01	−2.31	0.00	−2.76	0.01
	Rest of exposed vs. internal referents	0.14	0.87	−0.13	0.51	−1.25	0.00	−1.59	0.04
Analysis of overall trends	With CEI	−0.007	0.653	−0.005	0.201	−0.017	−0.015	−0.050	0.001
Exposure–response by CEI categories[a]	1	−4.077	0.001	−0.584	0.047	−1.142	0.040	−1.536	0.200
	2	−4.538	0.000	−0.625	0.035	−2.601	0.000	−2.607	0.031
	3	−4.600	0.000	−0.734	0.015	−2.865	0.000	−2.406	0.050
	4	−4.634	0.000	−0.874	0.004	−2.663	0.000	−3.720	0.003
	5	−5.121	0.000	−0.764	0.015	−2.908	0.000	−4.852	0.000

[a] Comparison for each category of cumulative exposure is relative to external referents as baseline.

the exposure categories. The Santa Ana and Benton visual retention tests showed only a difference between external referents and all the exposed. Associations and trends observed were predominantly seen at very low exposure levels (i.e. below the ACGIH TLV). Fig. 1 shows the clearest exposure–response relationship found, i.e. digit-symbol score plotted against the CEI, and demonstrates this point.

SPES Tests

Table 4 shows a panel of results for four SPES tests of which the three tapping tests are not wholly independent. Two of three tap tests (endurance and tapping with the dominant hand) show differences between external referents and all of the exposed. Two (endurance and nondominant hand tapping) tests show differences between the internal referents and the rest of the exposed. There is no overall exposure–response relationship seen for any of the four tests.

DPD: Catsys

An examination of the 18 tests showed that for the right sided rapid hand pronation and supination test there was a significant difference between internal controls versus the rest of the exposed. No differences were detected between external controls and all the

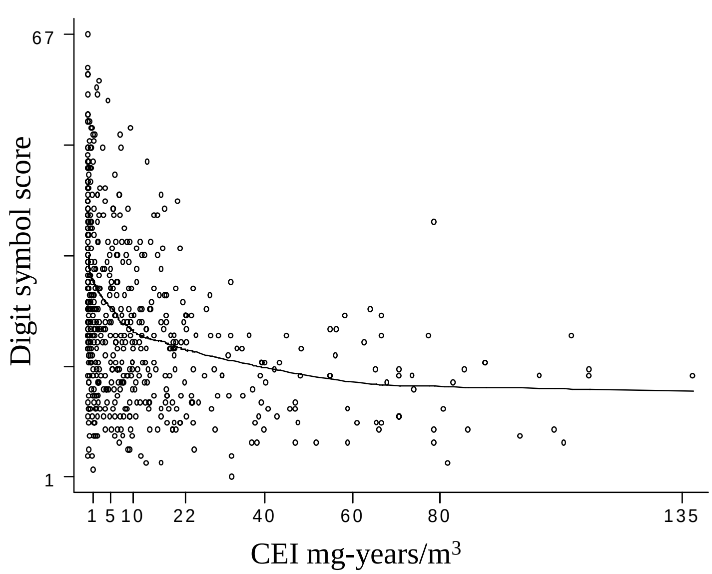

Fig. 1. Digit-symbol score vs. cumulative exposure index (CEI).

Table 4
Selected SPES test result panels

Analysis		Mean reaction time		Endurance		Tapping dominant hand		Tapping non-dominant hand	
		β	P	β	P	β	P	β	P
Unexposed referents	Mean score	266.37		337.06		59.36		54.60	
Dichotomous comparisons	All exposed vs. external referents	11.68	0.06	−13.81	0.02	−3.64	0.00	−2.00	0.06
	Rest of exposed vs. internal referents	2.39	0.63	−9.63	0.03	−1.66	0.06	−1.69	0.05
Analysis of overall trends	With CEI	0.08	0.41	−0.13	0.14	−0.03	0.07	−0.03	0.09
Exposure–response by CEI categories[a]	1	7.78	0.27	−7.99	0.22	−2.59	0.04	−1.00	0.41
	2	6.36	0.37	−13.55	0.04	−4.04	0.00	−2.13	0.08
	3	15.73	0.03	−19.42	0.00	−3.78	0.00	−3.06	0.02
	4	21.56	0.00	−13.75	0.05	−3.57	0.01	−1.05	0.42
	5	12.15	0.12	−20.75	0.01	−5.47	0.00	−4.07	0.00

[a] Comparison for each category of cumulative exposure is relative to external referents as baseline.

exposed, nor between internal controls versus the rest of the exposed for any of the other tests. No exposure–response relationship was observed for any test.

DPD: Tremor

An examination of the eight test parameters showed no differences between external controls and all the rest of the exposed. Left sided median frequency was significantly decreased while left and right sided dispersion around the median frequency were significantly increased for the rest of the exposed versus internal referents. No exposure–response relationship was observed.

DPD: Sway

Mean sway across the four test conditions showed a difference between external controls for the baseline condition (eyes open and feet not insulated) and for the condition with eyes open and feet insulated. No differences between internal referents and the rest of the exposed, nor any exposure–response relationships were found for any of the four conditions, and specifically for the most stressed condition (eyes closed and feet insulated) no effect was evident.

Categorical Outcomes

Questionnaire

An examination of the 42 questionnaire items revealed only five isolated associations. Differences between external referents and all the exposed were evident for irritation (OR = 2.54; 1.3–5.1), depression (OR = 2.25; 1.1–4.5) and having sex less than contemporaries (OR = 3.69; 4.9–277.7). Differences between internal referents and the rest of the exposed were shown for being tired (OR = 1.93; 1.2–3.1), feeling that something bad was about to happen (OR = 1.11; 1.02–1.22) irritated (OR = 1.99; 1.2–3.4), having to take notes to remember things (OR = 2.31; 1.1–5.1) and less sex (OR = 1.80; 1.1–3.1). None of the five associations showed any exposure–response relationship. A notable finding was that compared to the external referents there was a very high adjusted odds ratio of 36.9 (95% CI 4.9–277.7) for having sex less frequently than considered normal (Table 5).

Clinical Examination

Only two subjects had glabellar reflexes that were exaggerated, while four had abnormal gait (three mild, one more marked), none had immobile facies, while 42 had some difficulty walking backwards on a line. A combined clinical abnormality variable had 45 subjects with one or more above abnormalities. This showed no significant difference between external controls and all of the exposed, but did show a significant difference (OR = 3.68; 1.1–13.0) between the internal referents and the rest of the exposed. There was no overall trend with CEI. Examination of the exposure–response relationship by exposure categories showed a suggestive but non-significant trend with increasing exposure. When blood Mn as a continuous variable was used as exposure measure there was a small but significant

Table 5
Selected categorical test results panels

Analysis		Clinical test		Less sex than peers		LN 1R		LN 2L		LN 22R	
		[a]OR	P	OR	P	OR	P	OR	P	OR	P
Proportion abnormal in baseline		0.02		0.02		0.30		0.73		0.24	
Dichotomous comparisons	All exposed vs. external referents	4.17	0.19	36.84	0.00	1.05	0.89	0.94	0.87	0.35	0.01
	Rest of exposed vs. internal referents	3.68	0.04	1.8	0.03	1.68	0.04	2.34	0.00	1.02	0.94
Analysis of overall trend	With CEI	1.01	0.22	1.01	0.02	1.02	0.00	1.79	0.13	1.05	0.90
Exposure–response by CEI categories[b]	1	1.11	0.93	21.77	0.03	0.55	0	0.5	0.19	0.28	0.01
	2	2.1	0.54	33.42	0.07	0.69	0.13	1.02	0.96	0.38	0.03
	3	4.33	0.20	45.3	0.00	0.74	0.43	1.53	0.37	0.19	0.00
	4	6.48	0.10	59.77	0.00	2.27	0.03	0.92	0.85	0.37	0.02
	5	6.67	0.09	43.22	0.00	2.54	0.02	3.26	0.06	0.59	0.23

[a] OR = adjusted odds ratio.
[b] Comparison for each category of cumulative exposure is relative to external referents as baseline.

overall trend OR $= 1.08$ ($P = 0.01$): blood Mn was divided into four categories of equal size and the odds ratios of the last three compared with the first as baseline increased from 2.59 (0.6–11.0) through 3.56 (0.9–14.5) to 4.46 (1.1–17.5) only the last of which attained significance. There was a non-significant trend in the point estimates across exposure categories for both CEI and MnB.

Luria–Nebraska Tests

When categorised (0, ≥ 1), items 1R and 2L showed an effect compared with internal referents. 1R shows an impact only at high levels of exposure (categories 3 and 4) and 2L shows a non-significant difference from baseline for the highest exposure category only. Item 2R shows an overall trend which is only significant because of a change from a counterintuitive protective effect of exposure to a negative effect. 22R, 22L, 23R and 23L all show a protective effect of exposure (which is counterintuitive) when compared with external referents, without any overall trend. 1L, 3R, 3L, 4R, 4L and 21 show no effects.

DISCUSSION AND CONCLUSIONS

This is the largest study of Mn exposed workers to date with such a comprehensive range of nervous system endpoints, where exposures range considerably higher and lower than the ACGIH TLV and specifically include the low end of the exposure range all the way down to near zero exposure. It has consequently been possible to look seriously beyond bivariate comparisons between exposed and unexposed subjects, and also to explore the nature of exposure–response relationships in greater detail across the exposure range, beyond a simple estimate of linear trend.

Results from the large number (more than 100) of comparisons performed resolve into three groups. Group 1 shows differences between external referents and all the exposed and/or differences between internal referents and the rest of the exposed, but no further exposure–response relationship. It includes the Santa Ana, Benton and digit-span tests from the WHO NCTB; the finger tapping and endurance tapping tests from the SPES; Luria–Nebraska item 2L; questionnaire items tired, depressed, having to take notes in order to remember things, feeling that something bad is going to happen, irritated, and subjects' perception that they had sex less often than normal; a test of clinical abnormality; increased sway under conditions eyes open without foot insulation, and eyes open with foot insulation; and right sided fast hand pronation and supination. Group 2 shows the presence of a more substantive exposure–response relationship. It consists of only three tests: and includes the WHO digit-symbol test (although the major impact is at low exposure and therefore counterintuitive which could arguably place this result in group 3, see below), LN item 1R, which has a step to a poorer score at high exposure (categories 3 and 4), and the clinical test where MnB rather than CEI is the exposure measure. Group 3 contains the overwhelming majority of test results (almost all the questionnaire items, almost all the DPD tests including tremor, sway and diadochokinesia, and serum prolactin) which were either null or in a counterintuitive direction. Cumulative or integrated exposure was the strongest predictor of test abnormalities, except for the clinical test for which abnormality was more strongly associated with blood manganese.

Only the digit-symbol test showed a clear exposure–response relationship that exhibited no lower exposure threshold for an effect that appears to be strongest at low exposures (<0.2 mg/m^3) and weaker above the TLV, with evidence of saturation at higher exposures (above an average intensity of 0.5 mg/m^3), levelling off between 1 and 2 mg/m^3. This is difficult to interpret toxicologically.

The DPD tests which were assumed to be likely to be more sensitive to subtle Mn effects contributed disappointingly little. Catsys kinesometry was non-contributory, while effects shown for tremor parameters median frequency and its dispersion were in the opposite direction to those expected, and inconsistent with findings reported by other investigators (Despres et al., 2000; Lucchini et al., 1999). Sway was non-contributory and inconsistent with the findings of Chia et al. (1995). DPD devices are also expensive, very difficult to use logistically in field settings, and have substantial data transfer and data reduction problems. SPES items, too, were more or less non-contributory as well as posing quite severe data extraction difficulties. It appears that these computerised test batteries are designed for clinical purposes generating copious data from which only a small subset needs to be extracted for processing by statistical software in epidemiological analysis. This requires very substantial effort and time to prepare.

Luria–Nebraska test results were only partly consistent with those of Lucchini et al. (1999), and were also counterintuitive. Lucchini found linear exposure–response relationships with items 1, 2, 3, 4 but not 21, while this study found some exposure–response relationships for items 1R and then only at exposure levels greater than 1 mg/m^3. Results for 22 and 23 were nonsensical and there were no effects for 2, 3, 4 and 21. These tests are nevertheless logistically and financially appealing, especially in field settings in developing countries for possible utility as a subclinical screen in high exposure situations.

One explanation for these findings is that the study lacks validity. This is thought to be unlikely as our exposure measures as reported elsewhere (Myers et al., 2003b, in press) are consistent with prior isolated measurements at the smelter works and are comparable with data from other Mn smelters. Additionally, exposure–plant relationships or exposure–production location relationships are as expected. Our biological exposures are comparable with reference values for the unexposed, as well as with other reported levels amongst the exposed. Moreover atmospheric and biological exposures are reasonably well correlated (Myers et al., 2003b, in press). The WHO NCTB results compare very well with previous results found in South African workers similar to those at the smelter works (London et al., 1997; Myers et al., 1999), while the DPD component results compare very well with data published for a Canadian reference population (Despres et al., 2000). SPES results are comparable with other worker populations (Iregren, 1990). Luria–Nebraska scores compare with those reported by Lucchini et al. (1999). Lastly, the proportion of test score variance explained by years of education and age compares very well with previous South African and other international neurobehavioural studies (Myers et al., 1999). Lastly, the findings are consistent with a recent study conducted by the authors (Myers et al., 2003a) on a large number of manganese miners exposed to an average exposure intensity around the ACGIH TLV, in which no Mn effects were detected.

The most likely explanation for few, weak and inconsistent findings with implausible or counterintuitive exposure–response relationships is chance, and it is concluded that this is essentially a negative study, providing only weak and unconvincing evidence for exposure effects in general, or for the notion of a continuum of effect (Mergler et al., 1999) across exposures ranging considerably above and below the current ACGIH TLV. As such it simply adds further questions to current debates about the adequacy of the current ACHIH TLV.

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