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Neurobehavioral Effects of Acute and Chronic Mixed-Solvent Exposure in the Screen Printing Industry

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This 2-year prospective study examined the neurobehavioral effects of acute and chronic exposure to mixed solvents in workers in a screen printing business. A total of 30 subjects participated in the study in two field testings over a 12 month period. Each subject completed a detailed medical and occupational questionnaire, had a neurological examination, and underwent a battery of neuropsychological tests. Industrial hygiene investigation identified the following chemical exposures as present: toluene, methyl ethyl ketone, mineral spirits, β-ether, methylene chloride, and acetic acid. Different departments and jobs had varying degrees of exposure to these chemicals, the highest exposures being in the ink mix area and the screen washroom area. However, all exposure levels were below recommended threshold limit values. Persons categorized as having higher acute exposure demonstrated significantly impaired test performance on tasks involving manual dexterity, visual memory, and mood. Those with higher chronic exposure demonstrated significantly poorer performance on visual memory tasks and mood. Results suggest that the mixed solvents used in the screen printing industy have an effect on central nervous system functioning in the absence of obvious clinical disease.

Key words: neuropsychological tests, solvents, occupational exposure, printing industry

INTRODUCTION

Neuropsychological deficits in workers with acute and chronic solvent exposures have been well documented for a variety of mixed and single solvents. Such deficits can occur even when air concentrations of solvents are well within occupational standards [Johnson, 1987; Hanninen et al., 1976]. Recently increasing atten-

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tion has been paid to the phenomenon of "subclinical solvent encephalopathy" as defined by measurable neuropsychological changes accompanying solvent exposure in the absence of obvious clinical disease [Baker et al., 1985; White, 1986; White and Feldman, 1987; White et al., 1990].

Several investigators have assessed functioning using neuropsychological tests in solvent-exposed workers. In these investigations, neuropsychological tests were administered to nonexposed worker controls and to workers exposed to a variety of single solvents, including trichloroethylene [Grandjean et al., 1955], toluene [Lindstrom, 1981; Hanninen et al., 1987], and styrene [Lindstron, 1980, 1981], and to solvent mixtures such as those experienced by house painters [Hane et al., 1977; Lindstrom and Wickstrom, 1983], car painters [Hanninen et al., 1976; Lindstrom, 1981], and workers in other industries [Maizlish et al., 1985; Cherry et al., 1985; Ekberg et al., 1986; Fidler et al., 1987; Baker et al., 1988; Mikkelson et al., 1988; White et al., 1994]. These studies differed somewhat in methodology and specific findings, but several reported that workers with chronic mixed-solvent exposures showed poor performance on tests of higher order reasoning, visuospatial functioning, mood, and short-term memory. Certain cognitive capacities, such as language, were consistently found to be unaffected by solvent exposure.

Methodologically, there have been some limitations to the prior studies. Exposed populations have consistently been composed of workers from widely divergent workplaces and have thus been markedly variable in the type and intensity of exposure to agents under investigation. In addition, control groups often consisted of workers from another trade and may have differed in basic population parameters from the exposed workers, which would contribute to error variance in comparing the two groups.

The present investigation focused on screen printers, an occupational group that has not been systematically studied in regard to occupational central nervous system (CNS) disturbances, but that is exposed to mixed solvents with known neurotoxic properties [McCann, 1979], such as methyl ethyl ketone, hexane, heptane, toluene, methanol, and ethanol [Kay, 1976]. In order to improve upon prior methodology, higher exposed workers were compared to a low or unexposed group of workers within the same plant, and they were followed longitudinally. A specialized test battery was designed [White and Proctor, 1992], which included tasks to address the hypothesis that solvents preferentially affect functioning of the frontal region of the brain [White et al., 1992]. It was predicted that workers with higher mixed-solvent exposures would perform less well on tests involving executive function, attention, visuospatial processing, short-term memory, and psychomotor speed, and have more mood complaints than workers with low to no exposure.

METHODS

This was a 2-year prospective study. In year 1, 24 of the plant's 27 employees were tested (89%). In year 2, 26 of the plant's 31 employees participated (84%), and all of the year 1 participants who were still working in the plant agreed to be retested. A total of 30 subjects (21 males and 9 females) completed the testing over the 12-month period. The mean age of the subjects at entry into the study was 34.0 years (SD = 10.5) and ranged from 21 to 62 years. The mean educational level was 13.3 years (SD = 2.2), ranging from 8 to 18 years.

At the outset of the study, all subjects underwent screening of occupational, demographic, medical, and neurological status and history by completing a questionnaire and undergoing a neurological examination performed by a neurologist. From the questionnaire, which had been pretested in a prior 3-year study of lead-exposed workers [Baker et al., 1984], information regarding occupational history, work habits, hobbies, family medical history, smoking, alcohol and drug use, hospitalizations, and medical and neurological symptoms was gathered. The neurological examination assessed handedness, mental status, cranial nerve function, motor and sensory function, and reflexes [Feldman and Travers, 1984]. Repeated general screening was completed prior to the second testing session on each subject. The interim screening questionnaire was a shortened version of the initial questionnaire and addressed work habits; smoking, alcohol, and drug use; medical and cognitive symptoms; recent illness and surgery; and changes in work duties and chemical exposures.

Solvent Operations and Exposure Assessment

Production methods have changed dramatically over recent years with the introduction of ultraviolet (UV) ink systems, which have replaced the conventional solvent-based ink systems. UV technology improves upon the conventional ink systems since it has useful physical and chemical properties that assist in chemical resistance, gloss, abrasion, and lower production costs in the speed of curing, space, and energy. In the UV ink system, the screen printer mixes the color following a recipe and the ink base is produced by reacting epoxy or urethane resins with acrylic acid. Then the ink is thinned with a dilutent (acrylate ester or monomer); and additives, pigments, and photoinitiators or catalysts are added. Lastly, the film is cured by light. With conventional ink systems, the resin is mixed with additives and solvents for consistency and heat is applied to dry the paper. The process uses more quantities of solvents and is more fire-prone since paper is passed under heat.

In this plant, the owner estimated that the UV ink system accounted for two thirds of the company's output and that a 70% reduction in solvent use had occurred between 1981 and 1987 (personal communication). However, at the time of study there was only one production line (line #2) dedicated to the UV system and an older production line still dedicated to the conventional solvent-based system (line #4). The remaining two production lines (#1, 3) could do either system; and over the 4 days spent in the plant in year 2, two to three shifts between ink systems per day were observed.

Air monitoring studies were completed over several days by the project industrial hygienist (D.E.) at the time of the second field testing. The sampling method used was the standard NIOSH method for organic solvents, PC & M 1500. Samples were collected on a charcoal tube using a sampling train and were analyzed by Clayton Environmental Consultants, Inc., a certified laboratory in Novi, Michigan. Twenty air samples were collected, 12 on individual workers and 8 from different work areas. The sampling strategy included (1) sampling at least one worker in each job category and one from each of the different job tasks by personal air monitors, and (2) area sampling of the separate work areas in the plant. Sampling was done while both UV and conventional ink systems were in operation. Based on a review of the plant's past production practices by the study industrial hygienist, it was concluded that exposure levels in year 1 were similar to those measured in year 2.

Based on review of the material safety data sheets, the following chemicals were identified as being used in the plant in various jobs: toluene, methyl ethyl ketone (MEK), mineral spirits, β-ether, methylene chloride (MC), diacetone alcohol, acetic acid, and lead. β-cellosolve was also identified but could not be analyzed in samples, along with diacetone alcohol, due to conflicting analytical methodology. A measure of total hydrocarbon content (THC) was also done on the submitted samples to include a measure of the solvents not identified in the above-mentioned list. All area and individual-sampled exposures were within recommended 1994 American Conference of Governmental Hygienists (ACGIH) threshold limit values (TLVs) [ACGIH, 1994] (Table I). The highest solvent exposures were in the ink mix area sample (toluene, MEK, mineral spirits, THC) and in the screen washroom area sample (methylene chloride, toluene, MEK, mineral spirits, THC). Methylene chloride levels were measured to estimate TLV ceiling (TLV-C) in the screen wash room area. Three Draeger tube instantaneous spot samples of methylene chloride taken over a 15 min period measured 25–50 ppm at the start of cleaning, 50–75 ppm while cleaning, and 75–100 ppm 10 min after the start of cleaning. The usual length of time spent washing screens was under 20 min, about six times a day.

Determination of the worst case estimate and best estimate of the fraction of the TLVs present in the samples was made using an additive model for a mixture of components with similar toxicological effects:

$$C_1/TLV_1 + C_2/TLV_2 + C_3/TLV_3 + \ldots = 1.$$

The *worst case estimate* of exposure was calculated with THC generically equivalent to n-hexane, which is a relatively toxic solvent and therefore is a conservative estimate of risk. The *best estimate* of the fraction of the TLV was made by substituting a more likely solvent exposure for the unknown THC. The decision concerning the more likely solvent exposure was made by the industrial hygientist's walk-through observations. A fractional TLV value equal or greater than 1 was considered overexposure to the solvent mixture. For example, in the line #3 personal sample for the individual working on both solvent and UV processes (sample #1), 1.3 ppm MEK, 1.3 ppm toluene, and 11 mg/m^3 total HC were measured. Eighteen percent of the total HC measured could not be explained by concentrations of other compounds measured. To determine the worst case, total HC was assumed to be n-hexane: $(11/180\ \text{mg/m}^3)_{\text{n-hexane}} = 0.06$ TLV. To determine the best estimate, the unaccounted portion of the total HC was assumed to be Aromatic 100, a light naphtha consisting of predominately C9 HC, where Exxon's recommended TLV is 50 ppm or 245 mg/m^3: $(1.3/200\ \text{ppm})_{\text{MEK}} + (1.3/50\ \text{ppm})_{\text{toluene}} + (2/245\ \text{mg/m}^3)_{\text{naphtha}} = 0.04\ \text{TLV}_{\text{mix}}$.

In the area sample taken in the ink mix area (sample #9), 24 ppm MEK, 28 ppm toluene, 1.1 ppm mineral spirits, and 190 mg/m^3 total HC were measured. Thirteen percent of the total HC measured could not be explained by concentrations of other compounds measured. To determine the worst case, total HC was assumed to be n-hexane: $(190/180\ \text{mg/m}^3) = 1.06$ TLV. To determine the best estimate, the unaccounted portion of the total HC was assumed to be Aromatic 100, a light naphtha consisting of predominately C9 HC, where Exxon's recommended TLV is 50 ppm or 245 mg/m^3: $(24/200\ \text{ppm})_{\text{MEK}} + (28/50\ \text{ppm})_{\text{toluene}} + (1.1/100\ \text{ppm})_{\text{mineral spirits}} + (11.57/245\ \text{mg/m}^3)_{\text{naphtha}} = 0.74\ \text{TLV}_{\text{mix}}$.

TABLE I. Fraction of Exposure Threshold Limit Values (TLV) Present at Screen Printing Workplace

Samples analyzed for solvents	Fraction of TLV[a] Worst case[b]	Fraction of TLV[a] Best estimate[c]	Unspecified exposure fraction based on THC measures
Individual samples			
Line #3: toluene, MEK, mineral spirits, b-ether, THC	0.06	0.04	18%
Line #2: toluene, MEK, mineral spirits, THC		0.02	0
Line #1: toluene, MEK, mineral spirits, THC		0.02	0
Line #4: mineral spirits, THC, b-ether	0.06	0.05	85
Sample lost			
Line #3: toluene, MEK, mineral spirits, THC		0.04	0
Line #2: toluene, MEK, THC		0.06	—
Line #4: toluene, MEK, mineral spirits, THC	0.07	0.04	11
Ink tech.: toluene, MEK, mineral spirits, THC	0.22	0.17	35
Screen clean area: MC, toluene, MEK, mineral spirits, THC	0.07	0.06	99
Set up: MEK, THC	0.01	0	
Shipping: THC	0.13	0.13	100
Area samples			
Ink mix area: toluene, MEK, mineral spirits, THC	**1.06**	**0.74**	**13**
Art room area: THC	0.13	0.06	100
Washroom B—not analyzed			
Setup area: MEK		0.01	0
Line #1 area: diacetone alcohol, THC		0.01	
Dark room: acetic acid		0.05	
Washroom A: MC, toluene, MEK, mineral spirits, THC	**0.48**	**0.43**	**99**
Line #1 area (analyzed for lead)		0.40	0

MC = methylene chloride; MEK = methyl ethyl ketone; THC = total hydrocarbon.
[a]Calculated based on 1994 ACGIH TLVs.
[b]Calculated using unspecified THC generically equivalent to n-hexane; value >1.0 is considered overexposed.
[c]Calculated using unspecified THC as a more likely solvent exposure based on industrial hygienist's walk-through observations. Value >1.0 is considered overexposed.

Neuropsychological Assessment

Workers were scheduled for neurobehavioral testing randomly throughout their work day, based on their work schedules. The person who scheduled the testings was blind to the workers' exposure levels. A standard battery of neuropsychological tests (Table II) was administered during each of the two field testing periods, 1 year apart. All tests were given in a predetermined order by technicians trained and experienced in administration of these tests. The testing took approximately 45 min.

TABLE II. Neuropsychological Battery

Tests: Both years	Function tested
Vocabulary subtest, WAIS-R[a]	Verbal ability (hold test)
Similarities subtest, WAIS-R[a]	Verbal reasoning
Block design, WAIS-R[a]	Visual construction, reasoning, organization
Digit span, WAIS-R[a]	Attention
Digit symbol, WAIS-R[a]	Motor speed, visual short-term memory
Continuous performance test[b]	Attention, reaction time
Visual reproductions, WMS[c]	Visual memory (immediate, delayed recall)
Paired associate learning, WMS-R[d]	Verbal short-term memory
Santa Ana formboard test[e]	Manual dexterity and speed
Profile of mood states[f]	Mood
Year 1 only	
Controlled oral word association[g]	Cognitive flexibility, planning
Trail making test[h]	Attention, tracking, sequencing
Year 2 only	
Wisconsin card sorting test[i]	Cognitive flexibility

[a]Wechsler D (1981): "Wechsler Adult Intelligence Scale-Rev." New York: Harcourt, Brace, Jovanovich.
[b]Rosvold H, Mirsky A, Sarason I, et al. (1956): A continuous test of brain damage. J Consult Clin Psychol 20:343–350.
[c]Wechsler D, Stone C (1945): "Wechsler Memory Scale." New York: The Psychological Corp.
[d]Wechsler D (1987): "Wechsler Memory Scale-Revised." New York: The Psychological Corp.
[e]Fleishman EA (1954): Dimensional analysis of psychomotor abilities. J Exp Psychol 48:437–454.
[f]McNair DM, Lorr M, Droppleman LF (1971): "Profile of Mood States." San Diego, CA: Educational and Industrial Testing Service.
[g]Benton AL, Hamsher S deS (1976, manual revised 1978): "Multilingual Aphasia Examination." Iowa City, IA: University of Iowa.
[h]Halstead WC (1947): "Brain and Intelligence." Chicago, IL: University of Chicago Press.
[i]Grant D, Berg EA (1948): "The Wisconsin Card Sorting Test." Department of Psychology, University of Wisconsin.

Data Analyses

For purposes of analysis, the industrial hygienist categorized each worker as having either high or low *acute* solvent exposure in the plant. Based on the results of the air sampling and a walk-through analysis of the plant, the screen washroom workers and ink technicians had the highest exposures, the press operators had a moderate degree of exposure, and several workers (i.e., foremen, part-time screen setup workers) had intermittent exposures. These persons were categorized in the high acute exposure group. The artists, shipping workers, and clerical workers were considered to have low acute exposures. Classifications of acute mixed solvent exposures were based on individual and area samples, site of work station, use of personal protection, and specific job tasks handled by each worker at time of air sampling. The industrial hygienist also classified each worker as having either high or low *chronic* solvent exposure in the plant. The industrial hygienist did not participate in the neuropsychological testing and thus was blind to the test results when rendering these classifications. Chronic mixed solvent exposure classifications were based on years on the job, history of job categories in which each worker had been employed, and exposure measures at the time of the study. Descriptive statistics and analysis of variance (ANOVA) models adjusted for the effects of age, education, and gender were performed to examine the relationships between solvent exposure and neurobehavioral test performances.

TABLE III. Characteristics of High and Low Exposure Groups of Screen Printing Workers

	Low exposure	High exposure
Acute exposure	(n = 18)	(n = 12)
Mean years of age (SD)	34.9 (12.0)	32.6 (7.9)
Mean years of education (SD)	14.0 (1.8)	12.3 (2.5)[a]
% Female	50	0[a]
Chronic exposure	(n = 14)	(n = 16)
Mean years of age (SD)	34.1 (11.9)	33.8 (9.5)
Mean years of education (SD)	13.4 (1.4)	13.2 (2.8)
% Female	50	12.5[a]

[a]$p < 0.05$.

RESULTS

Of the 30 subjects, 12 were classified as having high acute exposure and 16 were classified as having high chronic exposure. In general, acute and chronic exposure indices were highly correlated ($r = .764$, $p < 0.0001$). However, there were four workers in management positions at the time of the study with low acute exposure, who had spent time in the printing shop in prior years and thus had a high level of chronic exposure. Descriptive analysis between high and low acute and chronic exposure groups showed a significant difference in gender, with all the nine female workers being in the low acute exposure group and only two of the women being in the high chronic exposure group. Mean years of education, but not age, were significantly different between the high and low acute exposure groups (Table III). Examination of the correlations between age and individual test performances showed that age was not significantly correlated with any test scores. Years of education were significantly positively correlated with better performance on similarities (scaled score), vocabulary, visual reproductions, controlled word association, and Santa Ana with the dominant hand ($p < 0.05$).

Higher acute exposure was found to be associated with a poorer performance on tasks relying on visual short-term memory and manual motor dexterity (digit-symbol test, WAIS-R; Santa Ana, nondominant hand) and on mood changes (depression and confusion subscales of the Profile of Mood States (POMS)) when adjusted for the effects of age and education. When further adjustment for gender was made, only a poorer performance on the Santa Ana test remained significant. In addition, a significantly poorer performance on visual reproductions, another test of visual short-term memory, was observed (Table IV).

Chronic exposure effects were evaluated by: (1) examining the relationship between level of chronic exposure and test performance; (2) examining the relationship between number of years working with neurotoxicants and test scores. Only the anger and confusion subscales of the POMS were significantly associated with chronic exposure after adjusting for age and education. With further adjustment for gender, the mood changes were no longer significant, but visual reproduction was significant. For 26 of the subjects with clear work history data regarding past neurotoxicant exposure, number of years working with neurotoxicants was significantly correlated only with the test scores for the vigor subscale of the POMS. In addition, there were no significant differences in mean neurobehavioral test scores when com-

TABLE IV. Results of Analyses Between High and Low Exposure Groups: 30 Screen Printing Workers

	Mean Scores		Significance adjusted for age + education	Significance adjusted for gender also
	Low	High		
Acute exposure	(n = 18)	(n = 12)		
Digit symbol, raw scores	58.1	47.7	F = 4.1; p = .05	F = 3.9; p = .06
Santa Ana, nondom. hand	21.3	17.7	F = 6.5; p = .02	F = 4.8; p = .04
Visual reproductions	10.7	8.8	F = 1.6; p = .21	F = 6.3; p = .02
POMS depression t-score	38.2	44.4	F = 5.8; p = .02	F = 2.3; p = .14
POMS confusion t-score	37.4	41.5	F = 4.1; p = .05	F = 1.8; p = .19
Chronic exposure	(n = 14)	(n = 16)		
Visual reproductions	10.7	9.3	F = 2.5; p = .13	F = 6.4; p = .02
POMS anger t-score	42.1	49.3	F = 5.6; p = .03	F = 3.9; p = .06
POMS confusion t-score	36.6	41.2	F = 4.7; p = .04	F = 2.6; p = .12

paring those persons with greater than 10 years work with neurotoxicants (n = 8) to those with less than 10 years (n = 18).

On neurological examination, none of the subjects demonstrated signs of neurological disease. However, higher acute exposure was found to be associated with a significantly lower degree of knee reflex response, when adjusted for the effects of age and education ($F(1,22) = 6.1$, $p = 0.02$). When additional adjustment for gender was made, both knee and ankle reflex response were significantly lower in those with higher acute exposure ($p < 0.05$).

Attempts to look at the data longitudinally were made. However, the high or low acute or chronic exposure level classifications did not change for any of the 20 people tested at both time 1 and time 2 over that time. For all tests, scores increased across time (i.e., performance improved) (Table V).

DISCUSSION

The present study in the screen printing industry suggested an effect of exposure to mixed solvents on neurobehavioral test performance on tasks involving visual short-term memory and manual speed and dexterity. Significantly more feelings of depression, confusion, and anger were also observed in those categorized as having higher exposures, but the relationship was not significant when adjusted for the additional effect of gender.

Due to the small sample size in this study, there was only significant power ($1 - \beta = 0.80$, alpha = 0.05) to enable detection of greater than 20–25% differences in mean test scores between exposure groups. However, the functional domains affected by exposure in this study are those predicted by the brain-behavior hypothesis concerning mixed solvents and neurobehavioral effects. Further study on a larger sample size is needed to support these findings.

It is possible that other workplace factors (e.g., work shift, overtime, type of work) or methodological issues (e.g., scheduling of testing) not accounted for in the study design may have affected the study results. Some experimental and epidemiological studies have demonstrated that cumulative fatigue, resulting from changing work shift schedules [Rosa et al., 1989; Rubin et al., 1991] or increased numbers of

TABLE V. Mean Test Scores (SD) for Screen Printing Workers Tested in Both Year 1 and Year 2 (n = 20)

Test	Year 1	Year 2
Similarities, scaled score	8.9 (1.5)	10.2 (1.5)[a,d]
Digit span, scaled score	11.0 (2.1)	11.1 (2.1)
Vocabulary	21.3 (4.8)	21.8 (4.9)
Digit symbol, scaled score	9.5 (2.1)	10.5 (2.5)[c]
Block design, scaled score	10.4 (2.2)	11.1 (2.7)
Visual reproductions	10.0 (2.3)	11.7 (1.6)[d]
Associate learning	12.2 (2.1)	12.2 (2.1)
Santa Ana, dominant hand	21.9 (3.1)	23.6 (3.0)[d]
Santa Ana, nondominant hand	20.6 (3.4)	21.2 (2.8)
Santa Ana, both hands	30.8 (4.7)	33.4 (6.2)
POMS tension, t-score	42.0 (8.9)	38.8 (6.7)[b,d]
POMS depression, t-score	40.7 (8.5)	38.7 (5.5)
POMS anger, t-score	46.4 (9.5)	45.1 (6.3)
POMS fatigue, t-score	47.8 (8.4)	44.6 (6.5)[d]
POMS confusion, t-score	38.9 (6.7)	37.0 (5.8)[d]

[a]A higher score in year 2 indicates a better performance.
[b]A lower score in year 2 indicates less mood symptoms.
[c]$p \leq 0.05$.
[d]$p \leq 0.01$.

overtime hours [Proctor, 1992]; and diurnal variation [Monk and Embrey, 1981; de Vries-Griever and Meijman, 1987] can affect cognitive function as measured by neurobehavioral tests, as well as mood. Lack of individual control on the job has been shown to increase occupational stress [Karasek and Thoerell, 1990], which may affect motivation and cognitive function. All subjects in this study worked the same shift (8 AM to 3 PM), and there was no indication that persons were working overtime hours at the plant. However, it is possible that some had second jobs. There were differences in time of day and day of the week that subjects were tested, which could affect the results if most persons with low exposure were tested at different times from most persons with high exposure. This situation was not likely, though, as in order for the plant to continue operation throughout the day during time of testing, subjects were removed from their worksites on a staggered schedule, e.g., not all people from one area were removed at the same time.

It was interesting to note probable practice effects in two of the tests used in this battery (similarities subtest, WAIS-R; visual reproductions subtest, WMS). These tasks have been used many times in epidemiologic studies of neurobehavioral dysfunction following neurotoxicant exposure, and data should be carefully considered when using the tasks in other studies in which practice effects may be a significant factor. It is possible that such practice effects could obscure subtle or mild dysfunction secondary to exposure.

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