Toxicology Letters, 43 (1988) 31–49
Elsevier

TXL 02043

Effects of short duration exposures to acetone and methyl ethyl ketone

Robert B. Dick[1], William D. Brown[1], James V. Setzer[1], Bobby J. Taylor[1] and Rakesh Shukla[2]

[1]*U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control, National Institute for Occupational Safety and Health, Division of Biomedical and Behavioral Science, Cincinnati, OH (U.S.A.), and* [2]*Biostatistics-Epidemiology Laboratory, Department of Environmental Health, University of Cincinnati Medical Center, Cincinnati, OH, (U.S.A.)*

(Received 20 January, 1988)
(Revision received 26 May, 1988)
(Accepted 23 June, 1988)

Key words: Acetone; Methyl ethyl ketone; Human exposures; Neurobehavioral testing; Biochemical measurement

SUMMARY

Workers are commonly exposed to mixtures or combinations of chemical agents, and these mixtures often consist of solvents. One group of solvents that has been extensively studied for its neurotoxic properties has been the ketones. However, previous research has focused on neuropathies produced by extended exposures and not on the simple pharmacokinetics or the reversible central nervous system (CNS) effects from short-duration exposures. In this research, 137 volunteers were recruited and tested for neurobehavioral performance changes and biochemical indicators during and after a short-duration (4-h) exposure to either acetone at 250 ppm, methyl ethyl ketone (MEK) at 200 ppm, acetone at 125 ppm with MEK at 100 ppm, or a chemical-placebo. Ethanol (95%, 0.84 ml/kg) was used as a positive control. Testing took place in an environmental chamber with four test stations. The computer-controlled test regimen took 10 h, and several measures were collected: (1) biochemical measurements of venous blood and alveolar breath; (2) psychomotor tests of choice reaction time, visual vigilance, dual task (auditory

---

The mention of product names in this publication in no way constitutes endorsement by the National Institute for Occupational Safety and Health.

Address for correspondence: Robert B. Dick, U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control, National Institute for Occupational Safety and Health, Division of Biomedical and Behavioral Science, 4676 Columbia Parkway, Cincinnati, OH, U.S.A.

Abbreviations: MEK, methyl ethyl ketone; RELs, recommended exposure limits; VDT, video display terminal; POMS, profile of mood states; MANOVA, multivariate analysis of variance; LSM, least mean squares; BAC, blood alcohol concentration; HHE, health hazard evaluation; CNS, central nervous system; THC, tetrahydrocannabinol; GC/MS, gas chromatography/mass spectrometry.

tone discrimination and tracking), and memory scanning; (3) one sensorimotor (postural sway) test; and (4) one psychological (Profile of Mood States [POMS]) test. Blood and breath concentrations during and after exposure did not demonstrate any interaction between the two solvents, nor were statistically significant sex differences present during uptake or elimination. The 250-ppm acetone exposure produced small but statistically significant differences from controls in two measures of the auditory tone discrimination task, and on the anger–hostility scale (males only) of the POMS test. The other chemical exposure conditions, MEK at 200 ppm and combination MEK with acetone, produced no consistent statistically significant results, which suggests there was no potentiation of the acetone effects with the co-exposure to MEK or vice versa under these test conditions. Ethanol at 0.07–0.08% blood alcohol concentration (BAC) caused significant decrements on both the auditory tone and tracking tests in the dual task.

INTRODUCTION

In the daily work setting, individuals are often exposed to combinations of industrial chemicals. Exposures can occur during manufacturing processes (adhesives, plastics, chemicals), degreasing operations, paintings, coatings, and drycleaning [1]. Often, the exposures contain concentrations of several volatile solvents, which could pose potential health risks for workers. The purpose of this research was to study a combination exposure from the same chemical group, test for possible neurobehavioral effects, and profile the biochemical measurements of venous blood and alveolar breath. The chemical group chosen was the ketones, of which several billion pounds are produced annually. Two members of this class, acetone and methyl ethyl ketone (MEK), were selected for study. The selection was based upon their frequency of occurrence in the NIOSH Health Hazard Evaluation (HHE) reports and MEK's property of potentiating certain neurotoxic effects of other ketones (methyl *n*-butyl ketone) [2,3].

The primary routes of exposure from acetone and MEK are inhalation and skin contact, with the former being the most common due to the volatility of most ketones at room temperature. Principal toxic effects from exposure are (progressively) eye, nose, and throat irritation; headache; nausea; vertigo; incoordination; central nervous system (CNS) depression; narcosis; and, with extremely high exposures, cardiorespiratory failure [4]. Neurobehavioral studies involving only single, short-duration exposures to acetone and MEK have been reported in the literature. The studies are not numerous, although some marginal effects have been reported at levels below the OSHA-PEL [5]. Table I summarizes these few studies.

In the present study, single exposures to acetone (250 ppm) and MEK (200 ppm) followed the NIOSH-recommended exposure limits (RELs) [6]. Combination exposures (acetone 125 ppm/MEK 200 ppm) followed the OSHA additivity formula [5]. All exposures occurred in a simulated 8-h workday, and neurobehavioral and biochemical measurements were taken before, during, and after exposures.

TABLE I

HUMAN STUDIES WITH NEUROBEHAVIORAL EXPOSURES TO KETONES

Report	Exposure			Tests[a]	Results
	Chemical	Concentration	Duration		
Matsushita et al. [14]	Acetone	500 ppm	6 h/day for 6 days	Simple RT	10% increase
		250 ppm	6 h/day for 6 days	Simple RT	5% increase
Nakaaki [18]	Acetone	170–690 ppm	4 h	Times estimated	Increase >450 ppm
	MEK	90–270 ppm	4 h	Times estimated	No change
Dick et al. [15]	MEK	200 ppm	4 h	Choice RT	No significant difference
				Visual-vigilance	No significant difference
				Pattern recognition	No significant difference

[a]RT, reaction time.

MATERIALS AND METHODS

One hundred thirty-seven participants were tested over a 1-year period. They ranged in age from 18 to 32 years, and were required to pass a physical exam. Pre-existing medical conditions (viz., pregnancy, diabetes, hypertension) or evidence of recent or chronic drug/alcohol use (verified by urinalysis and breath analysis) were grounds for exclusion from testing or subsequent data analysis. Six subjects were eliminated only from the psychomotor and psychological data analysis because of suspected marijuana usage (9-carboxy-tetrahydrocannabinol (THC) confirmed by gas chromatography/mass spectrometry (GC/MS) > 50 ng/ml), and one subject was eliminated from all data analysis for excessive ethanol (BAC > 0.03). Usable data also are reduced through equipment failures, which caused some imbalance in the number of subjects in the various conditions (see Table II).

Experimental design and procedures

The experiment was a mixed model (split-plot factorial) design, with subjects treated as a random factor. There were six separate treatment conditions, but the chemical and ethanol conditions were analyzed separately because of route of administration (inhalation vs. ingestion) differences. The treatment conditions were (1) acetone, 250 ppm; (2) MEK, 200 ppm; (3) acetone, 125 ppm/MEK, 100 ppm;

TABLE II

NUMBER OF SUBJECTS IN EACH TREATMENT CONDITION BY SEX AND CHEMICAL AND SUBJECTS' RATINGS OF THEIR EXPOSURE[a]

Condition	Male		Female		Total	
	n	%[b]	*n*	%[b]	*n*	%[b]
Acetone (250 ppm)	11(11)	100	11(9)	82	22(20)	91
Acetone (125 ppm)/ MEK (200 ppm)	8(8)	100	11(10)	91	19(18)	95
MEK (200 ppm)	12(10)	83	13(7)	54	25(17)	68
Chemical-placebo	11(11)	100	10(6)	60	21(17)	81
Ethanol	9(9)	100	11(10)	91	20(19)	95
Ethanol-placebo	11(4)	36	11(4)	36	22(8)	36
Total	62(53)	85	66(45)	68	129(99)	77

[a]Number in parentheses refers to the number of subjects who reported they were exposed to a chemical or who reported they had a drink containing ethanol.

[b]Percent of subjects reporting they were exposed or had ingested a drink containing ethanol.

(4) chemical-placebo; (5) 95% ethanol, 0.84 ml/kg; and (6) ethanol-placebo. Subjects were assigned to one of the treatment conditions following a forced randomization scheme. The two requirements were to test equal numbers of males and females in each group and that subjects be at least 21 years of age for the ethanol conditions. Two placebo groups were used, one as a control for the chemical conditions and the other as an ethanol control. Ethanol was used as a treatment condition to verify test sensitivity and for magnitude of effect comparisons. The chemical-placebo consisted of short (2 min), 25-ppm exposures of an acetone/MEK mixture presented twice during the 4-h exposure period. The ethanol-placebo was the ethanol drink mixture without ethanol. Table II shows the number of subjects in each treatment condition and their self-report ratings of what their exposure/ingestion was.

Experimental sessions were conducted double blind (subjects and experimenter). Testing procedures were in accordance with the guidelines of the NIOSH Human Subjects Review Board and the ethical principles of the American Psychological Association. Volunteers were paid $ 135 for their participation and were required to be drug free (except for medication approved by the medical officer) and to abstain from alcohol for 24 h prior to the 2 days of testing. Testing and exposures took place at one of four test stations inside an Environmental Chamber (Forma-Scientific) with interior dimensions of 2.5 m wide by 5.3 m long by 2.2 m high. Test stations were configured with a Hewlett-Packard 1311B video display terminal (VDT), a laboratory-made reaction time panel, one cylindrical microswitch (Switchcraft E-19), two box-mounted toggle switches (Switchcraft 41306), earphones (Realistic Pro-IIA), and a pressure-type joystick (Measurement Systems 735DC).

The testing regimen began with a 2-h practice session on the afternoon before the exposure day and ended with a 2.5-h test session the day following exposures. Exposure day testing took 8 h and was divided into four 2-h test periods. The 4-h exposure, which occurred in the middle two periods, was continuous, except for body burden sampling. The test periods were labeled as Pre:1–2 h, Exp:3–4 h, Exp:5–6 h, Post:7–8 h, and Post:23–24 h. The Post:23–24 h period was used to assess potential learning and exposure carry-over effects. Exposure testing commenced at 8 a.m., with lunch breaks at 0.5 h prior to the Exp:3–4 h period and at the end of the Exp:5–6 h period. Five breath samples and four blood samples were collected from each subject over the 3-day test session. During the 4-h exposure session, blood samples were staggered with one-half of the subjects giving a sample at 2 h and the others at 4 h. Fig. 1 displays the test regimen, showing the times of the blood and breath samples.

Subjects were tested for 10 h with the neurobehavioral tests described below. In each 2-h test period, approximately 32 measurements were used for purposes of analysis. These measurements were derived from four psychomotor tests, one sensorimotor test, and one psychological test. These tests are labeled in Fig. 1, along with their order of appearance and time of administration. Included in the test regimen was an eyeblink reflex test that will be reported in a separate paper. While the actual testing of subjects took 1 year, control over seasonal effects was accomplished through randomization. In any 3-month period, the six treatment conditions occurred at least once.

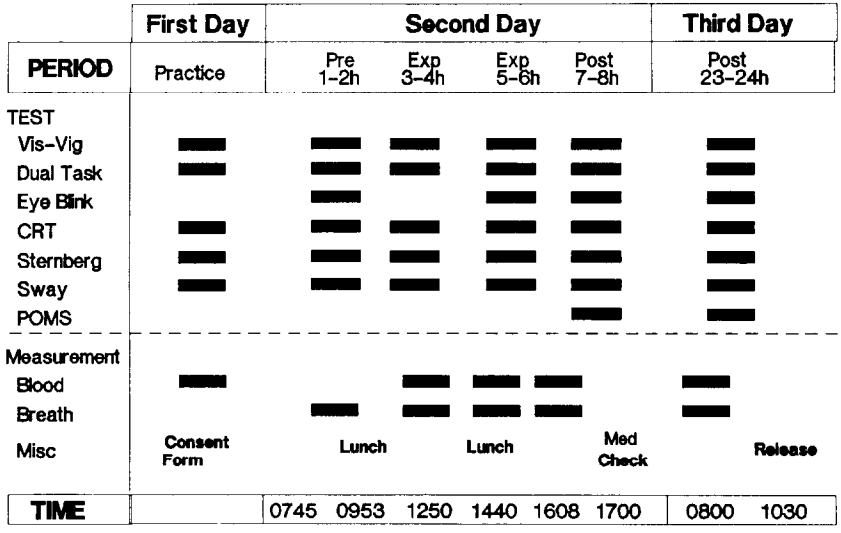

Fig. 1. Test regimen (neurobehavioral performance tests were presented in order from top to bottom).

Performance tests

All tests, with the exception of the mood test, were administered to subjects in the environmental chamber. An IBM Series I was used for the computer-controlled tests except the postural sway test, which used a Northstar Horizon computer. Software was both user-developed and proprietary.

Subjects were administered tests at the same time, and the order of test presentation was the same in each period and succeeding periods. However, trial order within a test was different for each subject, and the trial order changed with each successive administration of the same test. All subjects were required to reach criterion levels or they received a set number of practice trials on the psychomotor tests during the 2-h practice session the day before exposure and testing. Speed and accuracy were equally emphasized in the instructions for all the psychomotor tasks. Only brief descriptions of the tests follow*.

Visual–vigilance. This test was a non-memory type computerized version of the Mackworth Clock Test [7]. Centered on the VDT screen were 30 spokes radiating from a central hub, and computerized graphics created the appearance of a moving clock hand jumping from one spoke to the next. Movement speed was 60 jumps/min, with the critical signal (i.e., the one to be detected) being a jump of two spokes (24°). The test ran 36 min and there were 28 double jumps (critical events). Time (ms) to correct responses and number of correct and incorrect responses were recorded for analysis purposes.

Dual task. This task ran for 30 min and encompassed single presentations of an auditory tone discrimination task and a compensatory tracking task, followed by the simultaneous presentation (dual) of both tasks. Each test segment took 10 min, and there was a 1-min rest between the single and dual tasks. The auditory tone test required the detection of a 760-Hz tone from a series of 750-Hz tones.

In the compensatory tracking task, subjects used a pressure-type joystick to reposition the moving arrow underneath the stationary arrow. Three sine wave forcing functions creating tracking task difficulty levels (hard, medium, easy). The tracking error measurement computed was a 1-min average modulus mean error at each level.

In the dual task presentation, the auditory tone discrimination test was paired with the tracking task. Tracking error, time (ms) to correct responses and correct and incorrect responses were recorded.

---

* This paper represents the condensation of two separate papers, one prepared by the first author and one prepared by the second author. Each of these papers has been submitted to separate journals for publication and present the research findings in much greater detail than is covered in this paper. The article relevant to the blood and breath analysis has been published in *Journal of Occupational Medicine* (1987) 29(11), 877–883. The article relevant to the neurobehavioral effects is scheduled to be published in the *British Journal of Industrial Medicine*.

Choice reaction time test. Arrayed on a 16 × 25-cm response panel in a semicircular pattern were eight momentary push-button switches with red translucent covers. Centered at the bottom of the panel was a single green button. Subjects kept the green button depressed at all times, removing it only when a red button was lit. There were 80 trials, and two reaction times (release time and movement time) were calculated on each trial.

Memory scanning. This test was a computerized version of the Sternberg Short Term memory scanning test [8]. Subjects were presented 90 trials (lists) of numbers and each trial consisted of either 2, 4, or 6 digits (monosyllable digits 1–9). Following successive presentation of each trial, another single digit (probe digit) appeared. During this time subjects responded (yes or no) whether or not the probe digit matched a number in the previously presented list. A varied set procedure was used (each trial presented a different list of digits to be remembered). For data analysis purposes, six mean reaction times for each set size (list length) and probe type (yes or no) were used.

Postural sway (steadiness) test. A measure of postural steadiness was used to determine whether chemical exposures would affect any CNS processes controlling body posture. A computerized biomechanics platform system manufactured by Advanced Mechanical Technology (AMTI) was used for testing. Two 30-s samples, one with eyes closed and one with eyes open, were recorded during every 2-h test period. Data were collected at the rate of ten samples/s.

Profile of mood states (POMS). This test measures six mood or affective states from a factor-analyzed inventory. The test is derived from research by McNair et al. [9]. The six affective states measured are tension–anxiety, depression–dejection, anger–hostility, vigor–activity, fatigue–inertia, and confusion–bewilderment. Subjects were instructured to 'include their feelings during the past week including today' (1-week POMS) in filling out the test, and it was scored using the 'college norm' profile sheet.

Atmosphere generation and body burden measurement

Atmospheres were generated by drawing acetone and MEK, 99.43% and 99.46% pure, respectively, from a reservoir through a dual high pressure metering pump (Eldex-Model AA) into a 2 liter, three-vertical-neck mixing flask. Chamber airflows provided two fresh air changes per hour, with a constant temperature of 22 ± 1°C and humidity of 50 ± 5%. Chamber atmospheres were sampled from ports calibrated to agree with the subjects' breathing zones at each of the four stations. The atmospheres were monitored by two methods: (1) continuous monitoring via Miran 1A infrared analyzers (850 nm) using an Apple IIe computer to control sampling and record readings, and (2) monitoring at 30-min intervals by gas

TABLE III

CHAMBER CONCENTRATIONS FOR CHEMICAL EXPOSURE CONDITIONS

Condition measure	Average sample measurements[a]				
	Hour 1 (ppm)	Hour 2 (ppm)	Hour 3 (ppm)	Hour 4 (ppm)	4 h mean (ppm)
Acetone (250 ppm)					
Exposures[b] = 9					
Mean	204.6	250	245.2	249.4	237.4
SD	17.6	4.3	3.9	4.3	4.4
Methyl ethyl ketone (200 ppm)					
Exposures = 8					
Mean	155.1	197.0	194.5	198.5	186.3
SD	26.3	4.7	9.4	6.7	4.8
Combination (acetone and MEK)					
Exposures = 8					
Acetone (125 ppm)					
Mean	100.5	120.7	118.4	121.2	115.2
SD	17.2	11.7	7.9	3.2	5.6
Methyl ethyl ketone (100 ppm)					
Mean	72.6	94.6	91.6	93.3	88.1
SD	13.2	9.7	9.9	13.0	9.3

[a]SD = standard deviation.

[b] Exposures refer to the number of times this treatment condition was run.

chromatography (Perkin-Elmer 3920). Table III summarizes the chamber concentrations by sampling times using data from the infrared analyzers.

Breath samples were collected from subjects immediately upon exit from the chamber in 5-liter Mylar sampling bags using a 30-s breath-holding method [10]. The samples were analyzed within 15 min. Blood samples were collected from the antecubital vein upon exit from the chamber in 5-ml heparinized glass tubes and analyzed within 24 h. Analysis was performed by gas-liquid chromatography using the head space technique.

Ethanol administration

Ethanol was administered in a cocktail that contained 0.84 ml/kg 95% ethanol, combined with 100 ml quinine water, 75 ml of orange juice, two drops of tobasco sauce, and crushed ice. Additional quinine and orange juice replaced ethanol in the placebo cocktail.

RESULTS

Blood and breath analysis

The results of the blood and breath analysis are presented in Figs. 2 and 3 and

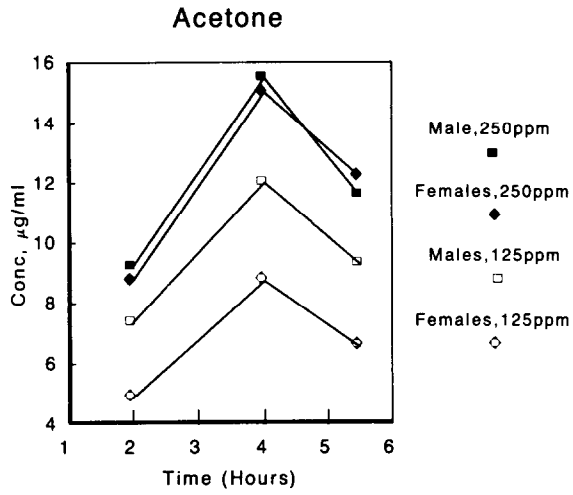

Fig. 2. Acetone concentrations in venous blood during and after 4-h inhalation exposures. Time begins with onset of exposure. Data points are means.

in Table IV. Sample sizes in Table IV represent usable blood and breath samples from only the chemical and ethanol conditions. Endogenous concentrations of acetone were detected in most of the pre-exposure blood and breath samples from subjects. Pre-exposure blood concentrations averaged 1.9 µg/ml (SD = 1.0), and breath concentrations averaged 0.3 ppm (SD = 0.7) for the chemical control group. The other treatment groups showed minor but not significant variations from the

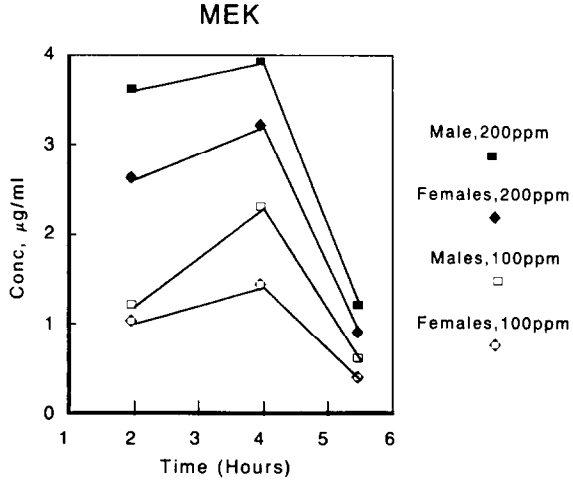

Fig. 3. Methyl ethyl ketone (MEK) concentrations in venous blood during and after 4-h inhalation exposures. Time begins with onset of exposure. Data points are means.

TABLE IV

BLOOD AND BREATH CONCENTRATIONS

Condition	Blood concentration (μg/ml or w/v (%))[a]				
	Pre	2 h	4 h	1.5 h Post	20 h Post
Acetone (250 ppm) n[b] = 22					
Mean	2.0	9.0	15.3	11.9	1.5
SD	2.0	2.0	2.9	2.6	1.0
MEK (200 ppm) n = 26					
Mean	ND[c]	3.1	3.5	1.0	ND[c]
SD		1.1	1.5	0.6	
Acetone (125 ppm) n = 22					
Mean	1.9	6.2	10.4	8.0	1.6
SD	1.0	1.8	2.4	2.1	1.3
MEK (100 ppm) n = 22					
Mean	ND[c]	1.1	1.9	0.5	ND[c]
SD		0.3	0.9	0.3	
Ethanol[d] (0.84 ml/kg) n = 21					
Mean	<0.002	0.062	No sample	0.0072	<0.001
SD		0.1		0.007	
Condition	Breath concentration (ppm)				
	Pre	2 h	4 h	1.5 h Post	20 h Post
Acetone (250 ppm) n = 22					
Mean	0.4	21.5	25.8	12.8	0.6
SD	0.6	4.4	4.0	2.9	0.8
MEK (200 ppm) n = 26					
Mean	ND[c]	11.4	11.9	0.7	ND[c]
SD		3.0	3.0	1.1	
Acetone (125 ppm) n = 22					
Mean	0.3	12.9	15.3	8.0	0.6
SD	0.7	3.3	2.7	2.4	0.8
MEK (100 ppm) n = 22					
Mean	ND[c]	5.1	5.3	0.7	ND[c]
SD		2.9	1.9	1.3	
Ethanol[d] (0.84 ml/kg) n = 21					
Mean	<0.1	179.1	85.8	19.0	<.01
SD		28.3	38.1	16.4	

[a] Values are means and standard deviations (SD) of the tested chemicals and correspond to the treatment condition. Sample times are from the beginning of exposure or ingestion of ethanol, except for the pre-exposure samples. Pre-exposure blood samples were taken on the day previous to exposure and pre-exposure breath samples were taken the morning of exposure. All subjects were tested for ethanol, MEK, and acetone. Pre- and post-acetone values for the non-acetone-exposed groups corresponded to the control values reported for the chemical control group. Likewise, MEK and ethanol values for the other groups were non-detectable (ND).

[b] n = number of subjects.

[c] 100% of values below detectable limits.

[d] Ethanol values are given in w/v (%), which corresponds to percent blood alcohol concentration. Ethanol normally peaks 1 h after ingestion; however, the values in the table are from samples 2 h after ingestion. Assuming a standard 15 mg %/h elimination, peak values averaged higher.

control group. Pre-exposure breath levels of ethanol were detected in trace amounts (see Table IV) in a few subjects. MEK was not detected in any pre-exposure blood or breath samples.

Steady-state breath levels of 8–12% for acetone and 5–6% for MEK of the chamber concentration were obtained at the 2-h sample perod. Steady-state levels for blood were not reached for either chemical within the 4-h exposure period, although the slope of the uptake curve for MEK was less pronounced than the acetone uptake curve. Significant correlation coefficients were obtained between blood and breath measurements for both chemicals. For MEK, it was 0.78 ($P<0.001$) and for acetone it was 0.61($P<0.001$).

Although graphically (Figs. 2 and 3) the results show a consistent trend toward lower blood concentrations in females than males for both solvents, statistically significant differences were not consistently obtained. Significance was only evident in the acetone/MEK combination exposure with the 2-h sample (Kruskal-Wallis = 4,05, $P = 0.04$) and 90-min post-exposure sample (Kruskal-Wallis = 4.47, $P = 0.03$).

For the analysis of all blood sample results involving chemical exposures, pre-exposure acetone values were subtracted from exposure sample values because there were significant sex differences in pre-exposure blood concentrations. Females had lower concentrations than males (Kruskal-Wallis = 4.45, $P = 0.035$). Blood and breath ethanol measurements corresponded to our predicted values and are summarized in Table IV.

Behavioral performance measurements

SAS version 5.15 [11] programs were used to analyse the data. The performance measurements were analyzed using multivariate analyses of variance (MANOVAs) of difference scores, with the chemical groups analyzed separately from the ethanol groups. The MANOVA test statistic reported is the Wilks' Lamda, and statistically significant results were determined using the Hummel-Sligo procedure [12]. With this procedure, significance is necessary on both the multivariate and univariate analysis at a predetermined alpha (0.05) for rejection of the null hypothesis. For measurements that demonstrated significance using this procedure, least-squares means (LSM) comparisons were used to identify treatment group differences on an a priori basis. The only comparisons considered were the placebo groups versus their respective treatment groups for the same test period. The above-described approach was taken to minimize the experiment-wise (type I) error rate, while still retaining enough statistical power to detect treatment effects. However, the possibility of a spurious significant finding cannot be completely eliminated.

Initial analysis of all the behavioral tests indicated that performance test results from males and females were qualitatively and statistically not different. As a result, except for the POMS test which has shown sex differences on some of the factor scores [9], the sexes were combined for analysis. Preliminary analyses also revealed

that tracking task difficulty in the dual task did not interact with the treatment condition, so the difficulty factor was dropped from further analyses. Only two tests, the choice reaction time test and the memory scanning test, showed learning effects over time.

For the chemical treatment groups, only the acetone-exposed group showed statistically significant differences from controls that indicated a possible treatment effect. These differences were evident only in the dual task and on the POMS test. In the dual task, only the MANOVA for the auditory tone discrimination task in single presentation showed significance ($F(9,197) = 2.52$, $P<0.009$). Subsequent univariate analyses showed significant effects on response time ($P<0.05$) and false alarm percent ($P<0.005$). Chemical treatment group LSM comparisons with controls showed significant differences only with the acetone group for response time in the Exp:3-4 h period ($P<0.002$) and in the Post:7-8 h period ($P<0.006$). Response time increases from baseline averaged 12% and 11% over controls for the only two significant test periods. All chemical treatment groups differed significantly from the control group on false alarm percent in all three test periods ($P<0.007$-0.05), but the acetone group was the most dramatically divergent. Averaging scores across all three test periods, the acetone group's false alarm increased by 4%, whereas the control group decreased 16%. The other two chemical groups, acetone/MEK and MEK, both decreased like the control group, but by a lesser magnitude (4–5%). These differences between the groups more clearly indicate a chemical effect for the acetone group only. Fig. 4 graphically shows these results with the corresponding venous blood measurements for each chemical group.

The POMs test showed no significant chemical main effects, but there was a significant chemical × sex interaction ($F(18,207) = 1.76$, $P<0.03$). The only significant univariate was on the anger–hostility scale ($P<0.02$). Condition × sex LSM comparisons revealed the effect was significant only for males exposed to acetone ($P<0.001$).

In the ethanol group's analyses, the dual task measurements in both single and dual presentation showed several statistically significant differences. In addition, the false alarm rate in the visual-vigilance test was increased over controls, but at a significance level (0.058) slightly above the chosen alpha cutoff. In the auditory tone discrimination task, ethanol effects were present in single presentation (MANOVA, condition $F(3,38) = 6.39$, $P<0.001$). Ethanol also produced significant MANOVAs in dual presentation (auditory tone and tracking for condition ($F(4,37) = 4.79$, $P<0.003$) and condition × period ($F(8,152) = 2.02$, $P<0.048$). Ethanol effects were present on all response variables in the auditory tone single presentation, but only for response times and percent correct hits in the auditory tone dual presentation. Single auditory tone presentation results averaged across the three periods for the ethanol group showed a 10% increase in response time, a 12% decrease in correct hits, and a 10% decrease in false alarm percent over controls for comparable periods. In dual presentations, significant effects from univariate

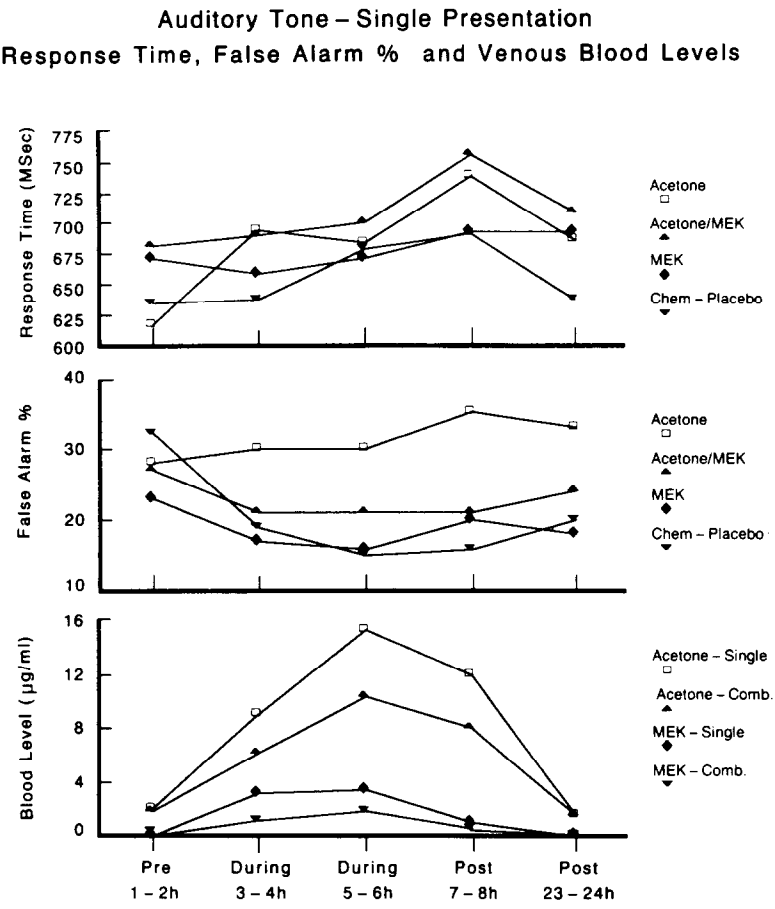

Fig. 4. Response time, false alarm percent rate on the auditory tone discrimination task (single presentation) and venous blood levels for each chemical condition.

analysis ($P<0.05$ to $P<0.001$) occurred with response times, percent correct hits, and tracking error. The significant (LSMs) effects were in the two exposure periods for response times and tracking errors and in all three periods for percent correct hits. Increases averaged about 14% for response time in the two exposure periods compared to only a 2% increase for controls. Tracking error scores increased 23% for the ethanol group in the two periods, while controls only increased 2–3%. Correct hits averaged a 13% drop for exposed vs. a 2% increase for controls across all three periods. Figs. 5 and 6 depict the effects of ethanol on the dual task.

Other tests, such as choice reaction time, postural sway, and memory scanning, did not reveal any statistically significant differences in the chemical conditions. Ethanol did show some marginal effects on the choice reaction time test and in some

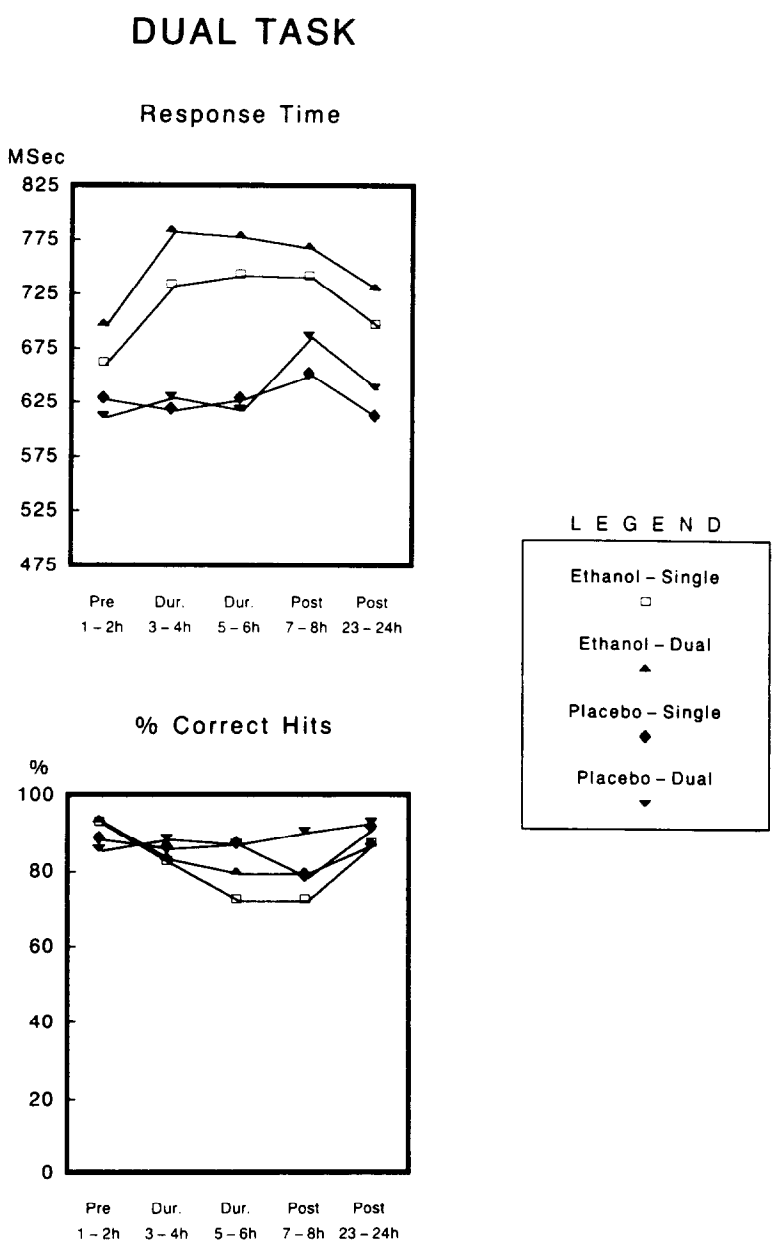

Fig. 5. Ethanol response measurements for the dual task (response time and percent correct hits). Data points are group means.

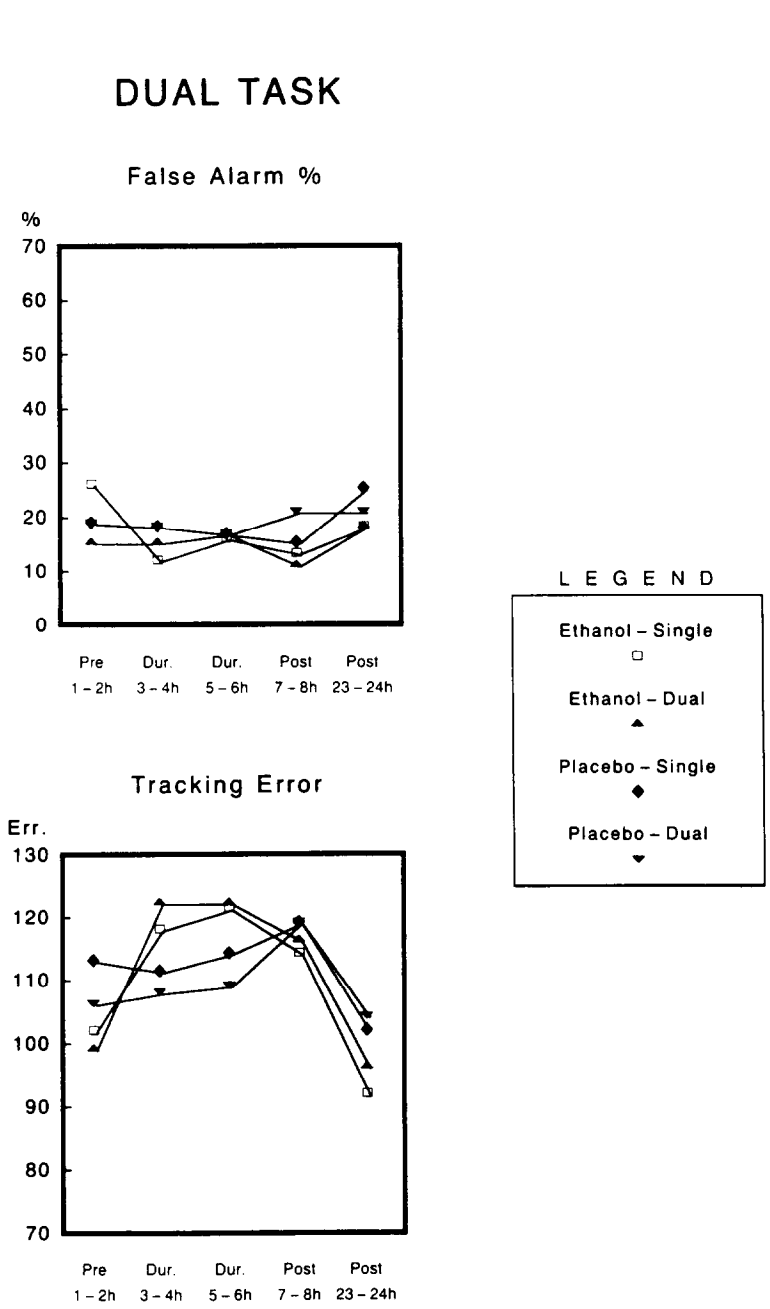

Fig. 6. Ethanol response measurements for the dual task (false alarm percent and tracking error). Data points are group means.

of the postural sway tests (0.09 MANOVA). The CRT results were improved movement time in the Post:7–8 h period only. Postural sway measurements of area, length, and velocity of sway showed increases in the ethanol group over the corresponding ethanol control group. The POMS test was unaffected by ethanol.

DISCUSSION

The biochemical indicators for blood and breath, which are summarized in Table IV, indicate that the desired body burden levels were obtained. In the 4-h exposure used in this study, the average blood concentrations of acetone and MEK continued to rise, although less so for MEK than acetone. The slopes also were very consistent when single exposures were contrasted with combination exposures. This probably indicates the lack of any significant metabolic interaction with these two chemicals at the tested exposure concentration and duration. The acetone blood level results are in agreement with two previous studies [13,14], although some extrapolation is necessary. MEK blood levels were in agreement with our previous work [15]. Alveolar breath samples had generally reached steady-state levels by the 2-h sample, stabilizing at 8–12% of the exposure concentration for acetone and 5–6% for MEK. These results are consistent with research involving acetone [16] and with a previous study in our laboratory on MEK [15].

The limited number of blood samples obtained from each subject prevents the accurate calculation of an overall elimination half-life. However, if first-order kinetics is assumed, the estimated half-lives for MEK and acetone are 49 min and 3.9 h, respectively. With some speculation, individuals exposed to these solvents for up to 8 h in a normal workday might have residual body burden concentrations of acetone the next day, but MEK should have been completely eliminated. Our present study showed no significant increases of acetone over endogenous levels the morning after exposure. A study by Matsushita et al. [14], using a 6-h exposure duration for 6 consecutive days, did show a residual build-up with a 500-ppm exposure concentration. In that study, the 250-ppm concentration did not show any build-up over the 6-day exposure. Results from our study showed no detectable levels of MEK in the blood or breath the morning following exposure, and more than half of the subjects had non-detectable levels of MEK in the breath samples taken 90 min post-exposure.

While not statistically confirmed by the results in this study, there is the strong suggestion of sex differences in the uptake of acetone and MEK. Examination of Figs. 2 and 3 tends to show lower uptake curves for females than males. The differences were only statistically significant in the combination exposure to acetone (125 ppm) and only when corrected for sex differences in baseline acetone blood concentrations. Small sample sizes precluded the use of more powerful statistical tests, thereby limiting the interpretation of these results.

Neurobehavioral performance test results indicated some mild, but detectable, effects caused by one of the chemical treatment conditions in the auditory tone

discrimination task, and in one scale on the POMS test. The results were only significant with the single acetone exposure to 250 ppm. In the auditory tone discrimination test (single presentation), the acetone-exposed group had increases in response time to correct hits and in the false alarm percent rate. When compared to the other chemical groups, the acetone group scores on the false alarm rate reached a plateau and did not improve. Similar delays in test score improvement with an acetone exposure were shown by the Matsushita et al. [14] 6-day exposure experiment. With the POMS test, the statistically significant difference occurred only for males on the anger–hostility scale. With the large number of statistical tests calculated in this study, and with no consistent trend indicated on the POMS test, this is one finding that may be due to chance.

Ethanol caused statistically significant differences from controls on all auditory tone discrimination measures in the single presentation mode, and on all measures except false alarm percent in the dual presentation mode. The other test in the dual task, tracking, was only affected by ethanol in dual presentation. This latter result is consistent with other dual task research involving ethanol [17]. Although not significant at the predetermined alpha of 0.05, the visual–vigilance test ($P = 0.058$) and some postural sway measures ($P = 0.09$) suggest ethanol effects.

The strength of the acetone effects should be interpreted cautiously. Statistically significant results occurred in only one of the neurobehavioral tests (the dual task), and on only one scale of the POMS test. The measurable differences were only on two of the several neurobehavioral measures analyzed. For the two performance measures affected (dual task–response time and false alarm percent), the decrements compared to the control group were roughly 11–12%. Notably, significant performance decrements from exposures to acetone occurred with some consistency throughout the two exposure periods and even into the Post:7–8 h period. The results also paralleled the blood concentrations for acetone (see Fig. 4), which rose throughout the 4-h exposure and were still close to the 2-h sample 90 min after cessation of exposure. The other chemical groups did not have blood concentrations this high, although the combination exposure behaved similarly because of the presence of acetone.

In summary, some small but statistically significant effects were detected during and after a 4-h exposure to 250 ppm acetone. The performance decrements from the acetone treatment were not as dramatic as an ethanol ingestion, which caused more severe decrements on the psychomotor tests. Performance changes from the acetone exposures demonstrated some persistence and paralleled the venous blood levels of acetone, indicating that tolerance did not develop. The results from this study agree with an earlier report by Matsushita et al. [14] that also found some mild decrements in psychomotor performance with 250-ppm exposures to acetone. Acetone is considered to be one of the less toxic solvents and currently has a relatively high OSHA-PEL of 1000 ppm [6]. The findings of this study suggest that further research may be necessary to determine whether exposures to acetone in concentra-

tions approaching 1000 ppm for at least 4 h may produce more distinct CNS performance decrements.

Exposures to 200 ppm MEK showed no interpretable statistically significant results. Furthermore, there were no significant interaction effects with the combination exposure of acetone (125 ppm) and MEK (100 ppm) for either the neurobehavioral or biochemical measures at the concentrations tested in this study.

ACKNOWLEDGEMENTS

The following individuals contributed significantly to the outcome of the study: Jay Klemme, M.D.; Richard L. Hawks, Ph.D.; Amit Bhattacharya, Ph.D.; Randy Smith, M.S.; Pam Schumacher; Margaret Regan.

REFERENCES

1 Curtis, M.F. and Keller, L.W. (1987) Exposure issues in the evaluation of solvent effects: report of the workshop session on exposure issues. In: J.M. Cranmer and L. Goldberg (Eds.), Proceedings of the Workshop on Neurobehavioral Effects of Solvents. Neurotoxicology 7(4), 5–24.
2 Saida, K., Mendell, J.R. and Weiss, H.S. (1976) Peripheral nerve changes induced by methyl *n*-butyl ketone and potentiation by methyl ethyl ketone. J. Neuropathol. Exp. Neurol. 35, 207–225.
3 Spencer, P.S., Schaumberg, H.H., Sabri, M.I. and Veronesi, B. (1980) The enlarging view of hexacarbon neurotoxicity. CRC Crit. Rev. Toxicol. 7, 279–356.
4 Krasavage, W.J., O'Donoghue, J.L. and DiVincenzo, G.D. (1982) Ketones. In: G.D. Clayton and F.E. Clayton (Eds.), Patty's Industrial Hygiene and Toxicology, John Wiley & Sons, New York, pp. 4709–4800.
5 Code of Federal Regulations 29 CFR 1910.1000, Table Z-1, Z-2 (June 1981) U.S. Printing Office, Washington, DC.
6 U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control. (Sept. 26, 1986) NIOSH. Recommendations for Occupational Safety and Health Standards. Morbidity and Mortality Weekly Report-Suppl. (35(1s).
7 Mackworth, N.H. (1961) Research on the measurement of human performance. In: H.W. Sinaika (Ed.), Selected Papers on Human Factors in the Design and Use of Control Systems, Dover, New York, pp. 174–331.
8 Sternberg, S. (1975) Memory scanning: new findings and current controversies. Q.J. Exp. Psychol. 27, 1–32.
9 McNair, D.M., Lorr, M. and Droppleman, L.F. (1981) EITS Manual for the Profile of Mood States, Educational and Industrial Testing Service, San Diego, CA.
10 Jones, R.H., Elliott, M.F., Cadigan, J.B. and Gaensler, E.A. (1958). The relationship between alveolar and blood carbon monoxide concentrations during breath-holding: simple estimate of CoHB saturation. J. Lab. Clin. Med. 51, 553–564.
11 SAS Institute (1985) SAS User's Guide: Statistics, Version 5, SAS Institute, Inc., Cary, NC.
12 Barker, H.R. and Barker, B.M. (1984) Multivariate analysis of variance (MANOVA). University of Alabama Press, Alabama University, Tuscaloosa, AL.
13 Wigaeus, E., Holm, S. and Astrand, I. (1981) Exposure to acetone: uptake and elimination in man. Scand. J. Work Environ. Health 7, 84–94.
14 Matsushita, T., Goshima, E., Miyakaki, H., Maeda, K., Takeuchi, Y. and Inoue, T. (1979) Ex-

perimental studies for determining the MAC value of acetone. 2. Biological reactions in the 'six-day exposure' to acetone. Sangyo Iqaku 11(10), 507–515.

15 Dick, R.B., Setzer, J.V., Wait, R., Hayden, M.B., Taylor, B.J., Tolos, B. and Putz-Anderson, V. (1984) Effects of acute exposure to toluene and methyl ethyl ketone on psychomotor performance. Int. Arch. Occup. Environ. Health 54, 91–109.

16 Tada, O., Nakaaki, K. and Fukabori, S. (1972) An experimental study on acetone and methyl ethyl ketone concentrations in urine and expired air after exposure to those vapors. J. Sci. Labour 48(6), 305–336.

17 Moskowitz, H. (1973) Psychological tests and drugs. Pharmakopsychiatrie 6, 114–126.

18 Nakaaki, K. (1974) An experimental study on the effect of exposure to organic slower vapor in human subjects. J. Labour Sci. 50(21), 89–96.