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Toluene and Ethanol Effects on Baboon Match-to-Sample Performance: Possible Synergistic Action[1]

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GELLER, I., R. J. HARTMANN, V. MENDEZ AND E. M. GAUSE. *Toluene and ethanol effects on baboon match-to-sample performance: Possible synergistic action.* PHARMACOL BIOCHEM BEHAV 22(4) 583–588, 1985.—Four juvenile male baboons were trained to respond for banana pellet rewards on a match-to-sample discrimination task. Exposure of the animals to a range of concentrations of either toluene or ethanol vapor resulted in a slowing of response times and a reduction in the percent trials attempted for some concentrations of either vapor. When behaviorally ineffective (subthreshold) concentrations of each vapor were combined, effects upon response times and trials attempted were similar to the effects produced by the higher concentrations of the individual vapors. However, while high concentrations of ethanol vapor produced errors in half of the subjects, combinations of ethanol and toluene did not increase this effect. This information suggests an ethanol potentiation of toluene effects, rather than the reverse.

Delayed match-to-sample discrimination Toluene Ethanol Baboons Response times

"PRACTICALLY any volatile organic solvent possessing the capability of lipid solubility, can produce intoxication" [1]. Self administration of organic solvents (inhalant abuse) as well as exposure to them in an industrial setting has been well documented [1,7]. Individuals who are inhalant abusers have been reported to co-abuse other drugs such as hypnotics, sedatives, stimulants, hallucinogens, marijuana and alcohol [9]. Not only has heavy abuse of alcohol been reported for inhalant abusers [15], but it has also been reported that alcohol is used extensively by individuals exposed to organic solvents in the workplace [12]. Toluene, a major constituent of many organic solvent mixtures is known to be abused alone or as a solvent mixture [11]. Recent animal studies have shown a synergistic action of toluene and diazepam [5]. It has also been shown that exposure of rats to toluene produces an increased intake of ethanol [6]. These observations strongly suggest the need for further studies to investigate possible underlying mechanisms for the interaction of substances that are co-abused. Investigators have reported that exposure of humans to toluene at concentrations above 200 ppm impaired reaction times and perceptual ability [10,14]. Memory impairment has also been reported for individuals exposed to organic solvent mixtures in the workplace [7]. The baboon delayed match-to-sample (MTS) discrimination task has been shown to be of value for the study of organic solvents insofar as it allows one to measure changes in reaction or response times as well as short term memory or accuracy of performance [3]. The purpose of the present investigation was to determine if exposure to toluene and ethanol vapors would produce qualitatively similar effects on the baboons' performance of the MTS task and the possible effects of exposure to combinations of ineffective concentrations of toluene and ethanol.

METHOD

The subjects were four male juvenile baboons (*Papio cynocephalus*) ranging in age from 14 to 21 months when obtained from the breeding colony of Southwest Foundation for Biomedical Research. They were housed in behavioral test chambers with an intelligence panel on one wall. The panel contained a row of three translucent discs (levers) on which visual stimuli could be projected. Under the appropriate experimental conditions, pressing either side disc activated a feeder which delivered a banana pellet reward. Experimental sessions of 2-hr duration were conducted on Monday through Friday of each week. The procedure for training the baboons on the MTS task was as follows. When a session timer was activated a variable-interval (VI) programming tape was set in motion. The tape programmed the occurrence of a stimulus on the center lever on the average of once every 3 minutes. The VI tape was inoperative during each trial, which began with the illumination of one of the stimuli on the center lever or probe stimulus. This stimulus was terminated at the end of a 30-sec period or by a response on the lever. Termination of the stimulus activated a timer for 2 min, called the delay interval. At the end of the delay interval, stimuli appeared on both levers adjacent to the cen-

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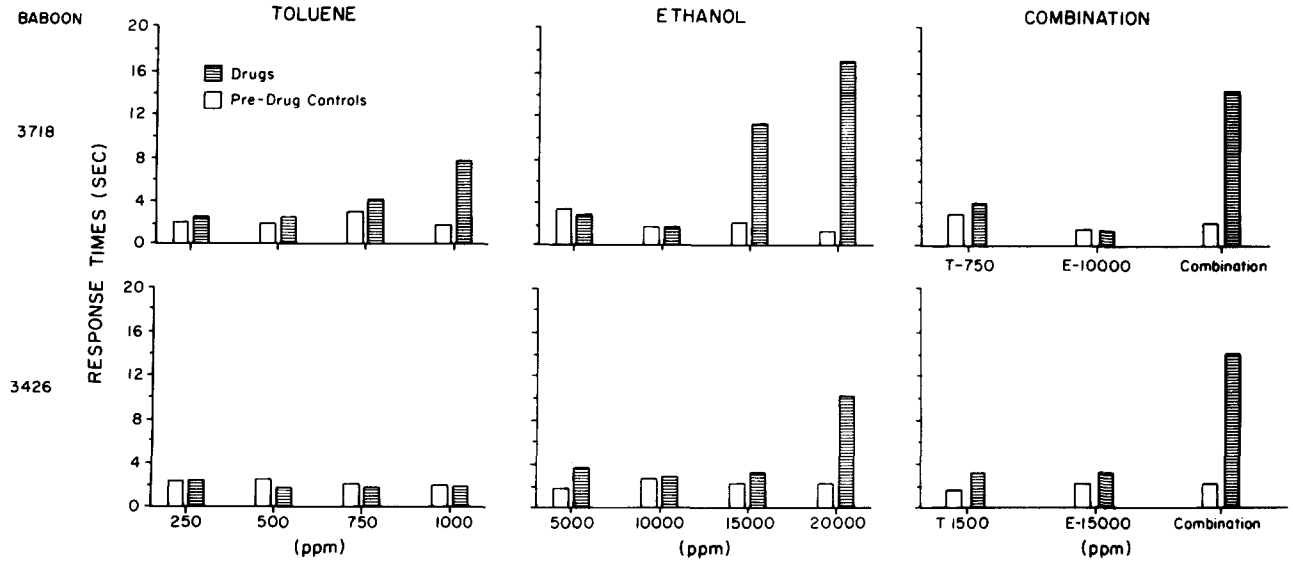

FIG. 1. Effects of toluene and ethanol alone and in combination on mean response times of baboons on a match-to-sample discrimination task. Open bars show pre-exposure control data; shaded bars show exposure data. These two baboons received all ethanol exposures first.

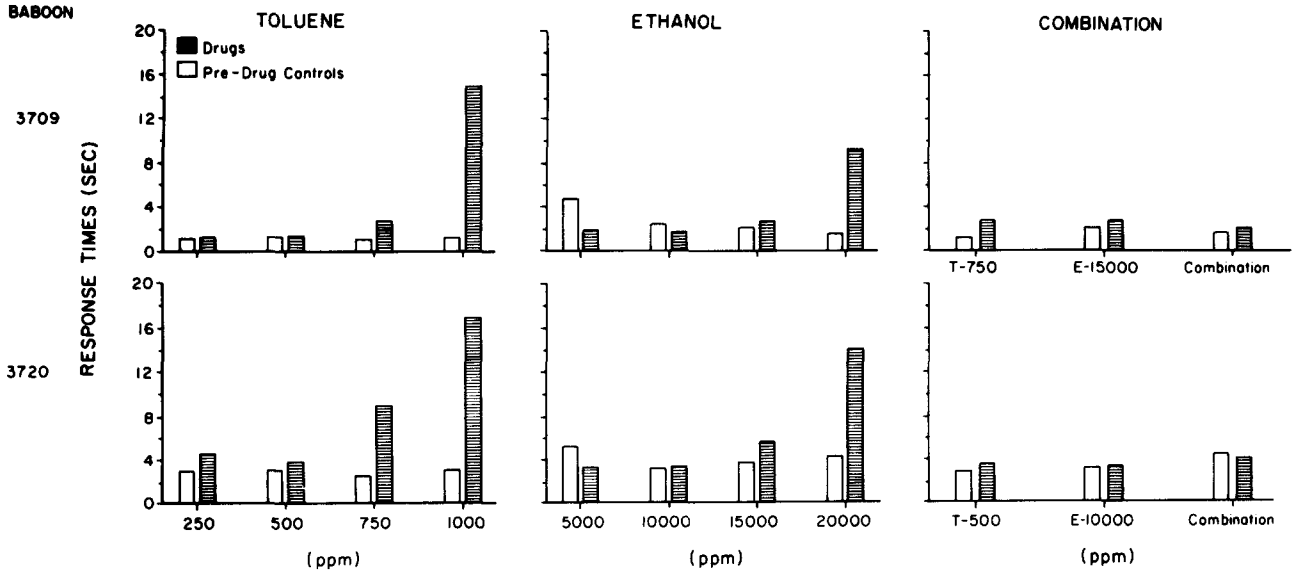

FIG. 2. Effects of toluene and ethanol alone and in combination on mean response times of baboons on a match-to-sample discrimination task. Open bars show pre-exposure control data; shaded bars show exposure data. These two baboons received all toluene exposures first.

ter lever. The correct matching stimulus was varied between these two levers in a mixed order. A response on the correct lever, when the stimulus matched the center lever stimulus, terminated the stimuli, activated the feeder and produced a banana pellet reward. Responses on the incorrect lever simply terminated the stimuli and again set the VI tape in motion.

A record was kept of the number of probe stimuli presented during each 15-min segment of a 2-hr session, the number of incorrect matching responses on the right and left levers and the number of extra responses that occurred on any of the three levers in the absence of discriminative stimuli or during the delay interval. Also measured were the percent of trials worked by the baboon and the time it took the baboon to respond, once a stimulus had been activated (response time). A 20-sec time limit was imposed so that if an animal did not respond in 20 seconds, the matching stimuli were terminated.

The exposures were conducted in large stainless steel and glass chambers that contained the behavioral test cages. These chambers have been described previously [4]. The baboons were exposed to toluene and ethanol vapors under

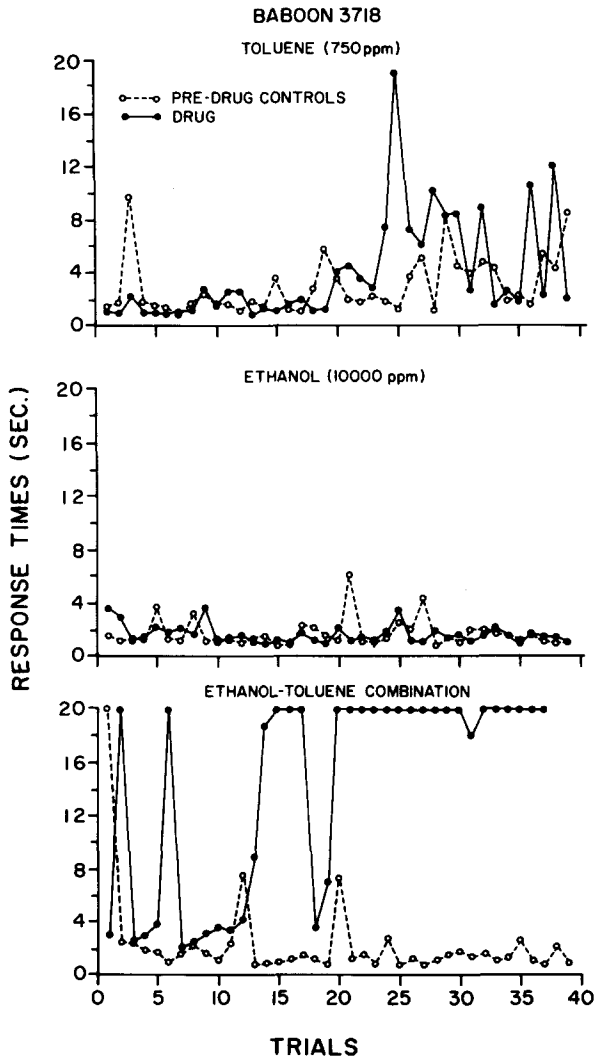

FIG. 3. Response time data for each trial during pre-exposure and exposure sessions to 750 ppm toluene, 10,000 ppm ethanol and a combination of these concentrations of toluene and ethanol. Broken lines show pre-exposure control data; solid lines show exposure data.

dynamic flow conditions whereby atmospheres of gases were generated by the vapor saturation technique [2], a method which can be used for substances liquid at room temperature. Air was bubbled through a gas washing bottle containing the liquid to be vaporized, in a constant-temperature bath. The vapor-saturated air was ducted into the air intake parts of the exposure chamber. Manipulation of either the bath temperature or the flow-rate of the gas allowed the production of a wide range of vapor concentrations in the exposure chambers. The chamber atmosphere was then monitored by periodic sampling for gas chromatographic analysis.

After the baboons were trained to 90–100% efficiency on the discrimination task, acute exposures to ethanol or toluene were conducted. Exposures of 3-hr duration were begun 1 hr prior to a behavioral test session on Wednesdays or Thursdays, no more frequently than once a week. Two of the baboons were first exposed to toluene in order of increasing concentrations of 250, 500, 750 and 1,000 ppm. The other two baboons were exposed to ethanol in order of increasing concentrations of 5,000, 10,000, 15,000 and 20,000 ppm. The animals were then switched so that the toluene animals received the series of ethanol exposures and the ethanol animals received the series of toluene exposure. After this phase of the research was completed, the baboons were exposed to several combinations of ethanol and toluene at concentrations that were ineffective during exposure to the individual vapors.

RESULTS

Figures 1 and 2 show the effects of toluene and ethanol alone and in combination on mean response times per session. The open bars represent the pre-drug control data and the shaded bars the exposure data. The data shown in Fig. 1 are for the baboons that received the series of ethanol exposures first and the data in Fig. 2 are for the baboons that received the toluene first. Toluene produced little change in response times for baboon 3718 at dose levels up to 1,000 ppm where mean response times slowed from a control value of 1.7 sec to 7.59 sec under the drug (Fig. 1). Mean response times were increased from 2.1 sec on control to 11.17 sec under 15,000 ppm ethanol and from 1.29 sec on control to 16.88 sec under 20,000 ppm ethanol. Exposure to a combination of relatively ineffective concentrations of 10,000 ppm ethanol and 750 ppm toluene produced a slowing of response times from 2.26 sec on control to 14.30 sec under the drug combination.

Similar data for baboon 3426 indicate a slowing of response times during exposure to 20,000 ppm ethanol but no effect during exposure to toluene at concentrations ranging from 250–1,000 ppm. An additional exposure to 1,500 ppm toluene (not shown) was also without effect. Exposure to the combination of the ineffective concentrations of 1,500 ppm toluene and 15,000 ppm ethanol also produced a slowing of response times.

Figure 2 shows a much greater slowing of response times under 1,000 ppm toluene for baboons 3709 and 3720 than was seen for the two animals in Fig. 1. Response times slowed from a control value of 1.44 to 15.15 sec for baboon 3709 and from 2.99 to 16.77 sec for baboon 3720 (Fig. 2). Response time under 750 ppm for baboon 3720 slowed from 2.33 sec on control to 8.75 sec. Ethanol increased response times for these two baboons only at the highest exposure concentrations of 20,000 ppm. Combinations of ineffective concentrations of ethanol and toluene did not increase response times for these baboons.

Figure 3 shows response time data for baboon 3718 for each trial during pre-drug (broken lines) and drug sessions (solid lines). Exposure to 750 ppm toluene produced a slowing of response times during the last 15 trials of the session while exposure to 10,000 ppm ethanol had no effect on response times. However, exposure of the baboon to 750 ppm toluene in combination with 10,000 ppm ethanol produced a slowing of response times which remained at the 20-sec maximum during the second half of the experimental session.

Figures 4 and 5 show the total percent trials attempted by each baboon during pre-drug control sessions (broken lines) and during each drug session (solid lines). It can be seen from Fig. 4 that exposure of baboon 3718 to 1,000 ppm toluene reduced the percent trials attempted by 16%. Since all

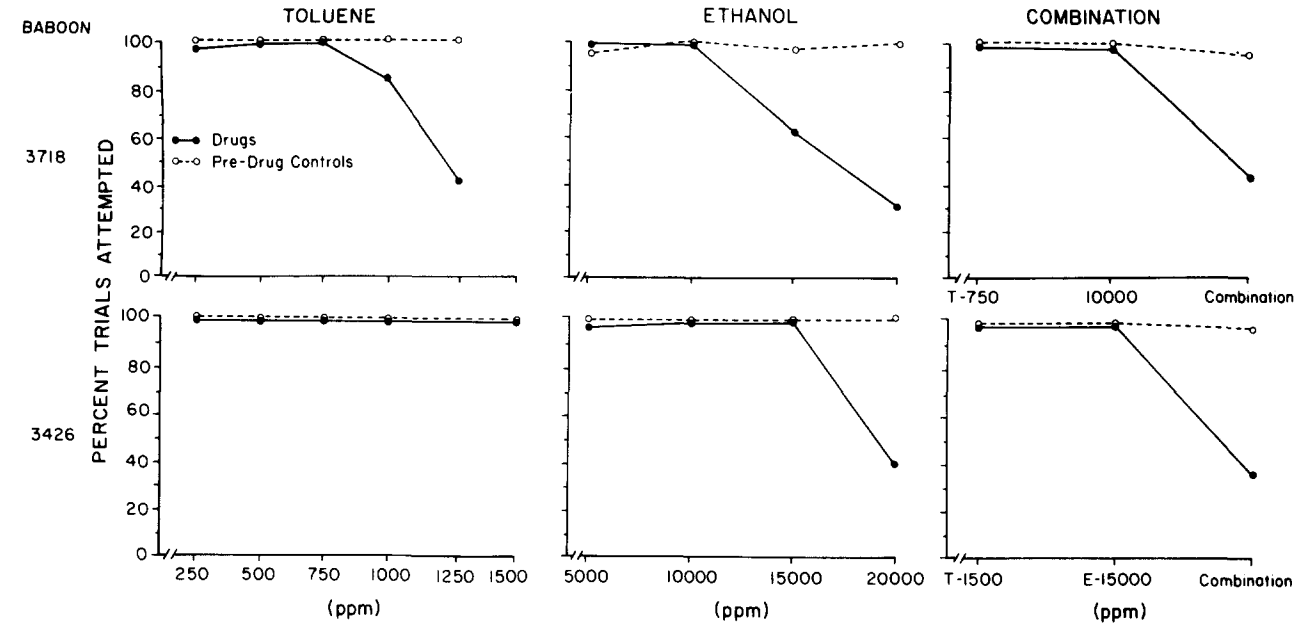

FIG. 4. Effects of toluene and ethanol alone or in combination on the percent trials attempted on a baboon match-to-sample discrimination task. Broken lines show pre-exposure control data; solid lines show exposure session data. These two animals received all ethanol exposures first.

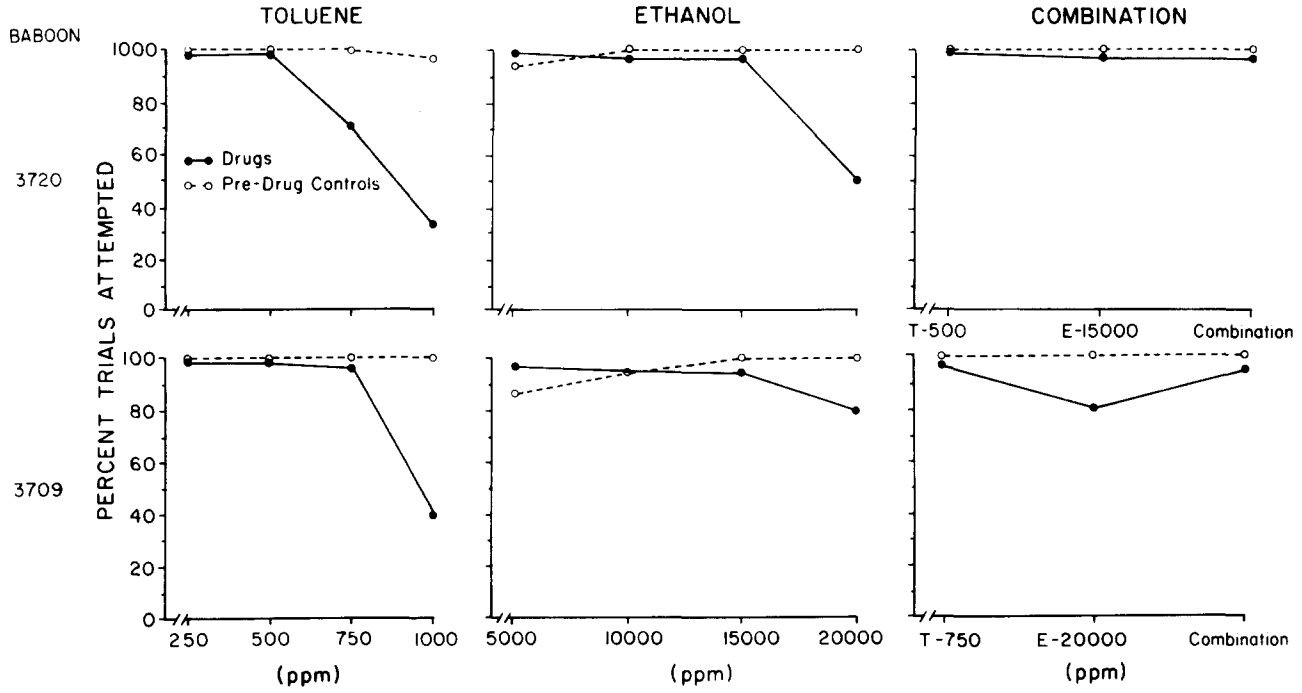

FIG. 5. Effects of toluene and ethanol alone or in combination on the percent trials attempted on a baboon match-to-sample discrimination task. Broken lines show pre-exposure control data; solid lines show exposure data. These two animals received all toluene exposures first.

toluene effects were minimal, an additional exposure to 1,250 ppm was conducted. During this session the percent trials were reduced by 59%. The percent trials attempted by baboon 3426 (Fig. 4) were not reduced under toluene even at an additional exposure to a higher concentration of 1,500 ppm. Ethanol at 20,000 ppm decreased the percent trials attempted by 70% for baboon 3718 and by 61% for baboon 3426. Exposure to the combination of the ineffective concen-

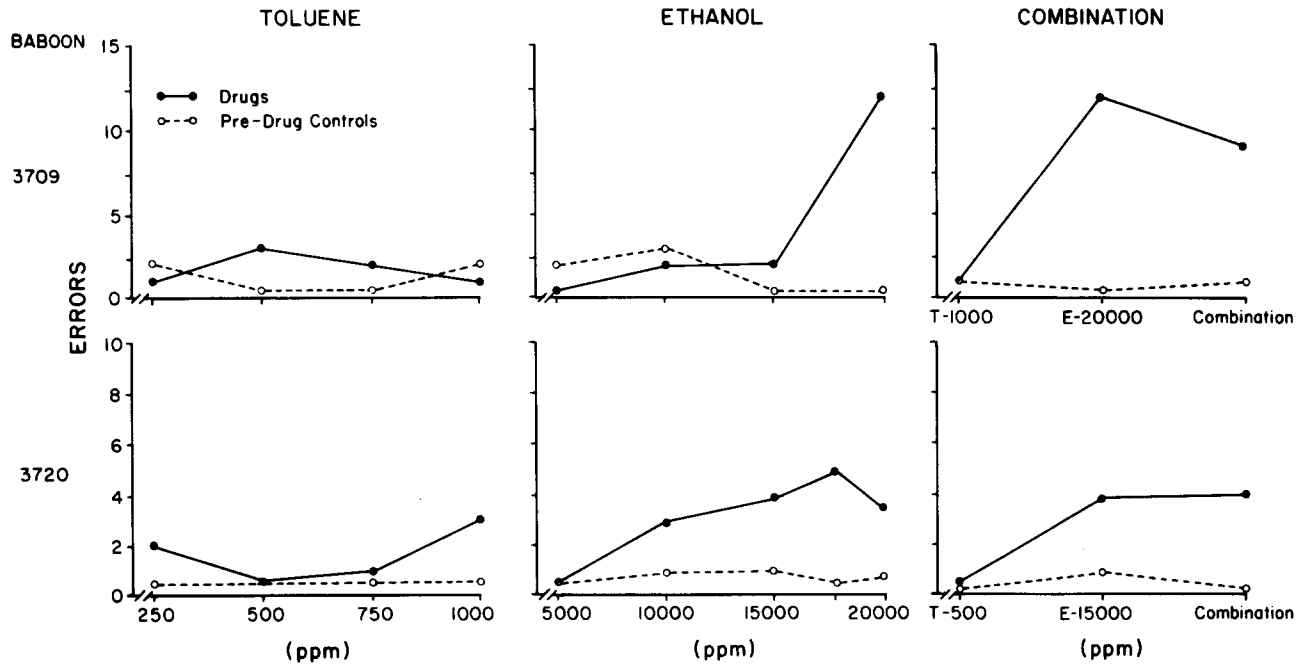

FIG. 6. Effects of toluene and ethanol alone and in combination on a baboon match-to-sample discrimination task. Broken lines show pre-exposure control data; solid lines show exposure session data.

trations of 750 ppm toluene and 10,000 ppm ethanol for baboon 3718 reduced the percent trials attempted from 97 during the pre-drug control session to 40 during the drug session. Similarly, the combination of the ineffective concentrations of 1,500 ppm toluene and 15,000 ppm ethanol for baboon 3426 reduced the percent trials attempted from 97 during the pre-drug control session to 36 during the drug session.

Data for the other two baboons, shown in Fig. 5, reveal a reduction in percent trials attempted at the highest exposure concentrations of toluene or ethanol. However, combinations of the individually ineffective concentrations of toluene and ethanol didn't affect the percent trials measure for either animal.

Only two baboons made errors on the MTS task during exposure to ethanol. These data are shown in Fig. 6. Baboon 3709 made 12 errors during exposure to 20,000 ppm. Baboon 3720 made 4 errors under 15,000 ppm ethanol and 3 errors under 20,000 ppm ethanol; an additional exposure conducted at 17,500 ppm ethanol resulted in five errors by this baboon. Since no baboons made errors during exposures to combinations of ineffective concentrations of toluene and ethanol, it was decided to expose these two animals to an ineffective toluene concentration in combination with an effective concentration of ethanol. When exposed to 1,000 ppm toluene in combination with 20,000 ppm ethanol, baboon 3709 made nine errors, three less than were produced by ethanol alone. Similarly, baboon 3720 made four errors during exposure to a combination of 500 ppm toluene and 15,000 ethanol, as well as only four errors during exposure to this concentration of ethanol alone.

DISCUSSION

Exposure of juvenile baboons to some concentrations of either toluene or ethanol vapors during a 3-hr period slowed response times and reduced the percent of trials attempted by the animals working on a MTS discrimination task. These observations agree in part with previous clinical reports that exposure to 200 ppm toluene increased reaction times [10] or produced an impairment of coordination and reaction time [14]. Although qualitatively similar dose-effects were observed for four baboons, the extent of effects differed between animals. Baboons 3709 and 3720 were more sensitive to the effects of toluene; response times for these baboons slowed to approximately 16 sec during exposure to 1,000 ppm (Fig. 2). However, the maximum effect for baboon 3718 was a slowing of response times to 8 sec under 1,000 ppm toluene, a dose that was without effect for baboon 3426 (Fig. 1). Since the latter two animals received the ethanol exposures first, perhaps the early ethanol treatments may have decreased the subsequent pharmacological actions of toluene.

Errors were made by two of the baboons and only during exposure to ethanol at the highest concentrations (Figs. 5 and 6). The presence or absence of this effect may also be attributable to the order in which the animals received exposures.

An apparent synergistic action between ineffective doses of toluene and ethanol occurred on the response time and percent trial measures but not on errors. Since only ethanol produced errors, the toluene-ethanol interaction is most likely a potentiation of toluene effects by ethanol rather than a toluene induced increase of ethanol effects. This speculation derives support from human and animal studies. Ingestion of a moderate dose of ethanol prior to exposure to approximately 147–274 ppm xylene caused a marked alteration in the kinetics of xylene clearance [12]. The blood xylene level rose 1.5- to 2-fold and urinary methylhippuric acid, a xylene metabolite, decreased by 50%, thereby suggesting

that ethanol decreased the rate of metabolic clearance of xylene. The authors speculated that the disturbance in xylene kinetics was probably due to an inhibition of microsomal metabolism by ethanol. In a study of xylene-ethanol interactions in rats, behavioral evaluation also indicated a potentiation of xylene effects by ethanol [13].

The fact that the presently-described synergistic action was not observed in all animals is probably explainable insofar as the amount of ethanol in the body with other chemicals may be a critical determinant of whether it has a stimulating or inhibiting effect on drug-metabolizing enzymes [8]. Unfortunately, blood ethanol levels were not obtained for these baboons during the course of these studies because of possible trauma to the animals and possible impairment of behavioral baselines. Future studies will be designed to provide blood levels of toluene and ethanol under the appropriate experimental conditions in order to ascertain those levels that are critical for inducing synergism.

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