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REPORT DOCUMENTATION PAGE

Form Approved
OMB No 0704-0188
Exp Date Jun 30, 1986

AD-A212 660

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1b. RESTRICTIVE MARKINGS

3. DISTRIBUTION/AVAILABILITY OF REPORT
Approved for public release distribution unlimited.

5. MONITORING ORGANIZATION REPORT NUMBER(S)

6a. NAME OF PERFORMING ORGANIZATION
Dept. of Medical Neurosciences
WRAIR

6b. OFFICE SYMBOL (If applicable)

7a. NAME OF MONITORING ORGANIZATION
Walter Reed Army Institute of Research

6c. ADDRESS (City, State, and ZIP Code)
Walter Reed Army Institute of Research
Washington, DC 20307-5100

7b. ADDRESS (City, State, and ZIP Code)
Washington, DC 20307-5100

8a. NAME OF FUNDING/SPONSORING ORGANIZATION
USAMRDC

8b. OFFICE SYMBOL (If applicable)

9. PROCUREMENT INSTRUMENT IDENTIFICATION NUMBER

8c. ADDRESS (City, State, and ZIP Code)
Fort Detrick
Frederick, MD 21701-5012

10. SOURCE OF FUNDING NUMBERS

PROGRAM ELEMENT NO.	PROJECT NO.	TASK NO.	WORK UI ACCESSIO NO
62787	A875	BB	

11. TITLE (Include Security Classification)
Effects of atropine and azaprophen on matching and detection in rhesus monkeys.

12. PERSONAL AUTHOR(S)
R.F. Genovese, T.F. Elsmore

13a. TYPE OF REPORT

13b. TIME COVERED FROM _____ TO _____

14. DATE OF REPORT (Year, Month, Day)

15. PAGE COUNT

16. SUPPLEMENTARY NOTATION

17. COSATI CODES

FIELD	GROUP	SUB-GROUP

18. SUBJECT TERMS (Continue on reverse if necessary and identify by block number)
Atropine; azaprophen; chlinergic; learning; memory; operant conditioning; primate; [illegible]

19. ABSTRACT (Continue on reverse if necessary and identify by block number)
Azaprophen (6-methyl-6-azabicyclo[3.2.1]octan-3 ol 2,2-diphenylpropionate) (6) is a confc mationally restricted analog of atropine. The antimuscarinic characteristics of azaprophe have been investigated in a number of in vitro preparations. For example, azaprophen has found to be substantially more potent than atropine for inhibiting carbachol-induced amyl release (6, 7, 13) and for inhibiting acetylcholine-induced contractions in guiena pig ile (6,7). In contrast, azaprophen has been reported to be slightly less potent than atropine for attenuating carbachol induced inhibition of prolactin (5) and has been found to be bot more potent (7) and less potent (13) than atropine for inhibiting [3H]N-methylscopolamine binding, depending on the cell line studied. In a single behavioral assay, azaprophen wa slightly less potent than atropine for producing response suppression under a simple sche ule of reinforcement in rats and, unlike other benzilates, failed to produce response rate increases (13). It has been suggested that, because of differences in the pharmacological profile (e.g., potency relationships in vitro), azaprophen may interact with muscarinic receptors in a novel manner (13). (keywords)

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Pharmacology Biochemistry & Behavior, Vol. 32, pp. 495–498. Pergamon Press plc. 1989. Printed in the U.S.A. 0091-3057/89 $3.00 + .00

Effects of Atropine and Azaprophen on Matching and Detection in Rhesus Monkeys[1]

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Received 16 June 1988

GENOVESE, R. F. AND T. F. ELSMORE. *Effects of atropine and azaprophen on matching and detection in rhesus monkeys.* PHARMACOL BIOCHEM BEHAV 32(2) 495–498, 1989.—The effects of the anticholinergic atropine and azaprophen, a novel, conformationally restricted analog of atropine, were examined in rhesus monkeys using delayed match-to-sample and detection tasks. Both compounds (0.01–0.32 mg/kg) produced dose-dependent decreases in the rate of responding under both tasks. Drug effects on the match-to-sample task correlated with drug effects on the detection task. Both compounds produced decreases in the percentage of correct responses on the match-to-sample task when choice trials occurred 4 or 16 sec, but not 0.01 sec, following sample presentation. Doses of atropine and azaprophen decreasing accuracy on the match-to-sample task also decreased the number of responses on the task. In general, atropine was slightly more potent than azaprophen on both tasks. These results further characterize azaprophen's anticholinergic effects.

Atropine Azaprophen Cholinergic Learning Memory Operant conditioning Primate

AZAPROPHEN (6-methyl-6-azabicyclo[3.2.1]octan-3-α-ol 2,2-diphenylpropionate) (6) is a conformationally restricted analog of atropine. The antimuscarinic characteristics of azaprophen have been investigated in a number of in vitro preparations. For example, azaprophen has been found to be substantially more potent than atropine for inhibiting carbachol-induced α-amylase release (6, 7, 13) and for inhibiting acetylcholine-induced contractions in guinea pig ileum (6,7). In contrast, azaprophen has been reported to be slightly less potent than atropine for attenuating carbachol-induced inhibition of prolactin (5) and has been found to be both more potent (7) and less potent (13) than atropine for inhibiting [^{3}H]N-methylscopolamine binding, depending on the cell line studied. In a single behavioral assay, azaprophen was slightly less potent than atropine for producing response suppression under a simple schedule of reinforcement in rats and, unlike other benzilates, failed to produce response rate increases (13). It has been suggested that, because of differences in the pharmacological profile (e.g., potency relationships in vitro), azaprophen may interact with muscarinic receptors in a novel manner (13).

We further investigated the behavioral effects of azaprophen and atropine in rhesus monkeys using concurrent delayed match-to-sample and detection tasks. The involvement of the cholinergic system in learning and memory has been the focus of a great deal of investigation [e.g., (3, 8, 10)]. Results from a number of studies (1, 2, 9, 11) suggest that anticholinergics, including atropine (12), produce a selective disruption of learning and memory processes in nonhuman primates. Therefore, we were interested in determining whether atropine and azaprophen could be differentiated on the basis of their effects on a "memory sensitive" task like the match-to-sample. We were also interested in determining whether atropine and azaprophen would have similar effects on a simple operant task in rhesus monkeys (i.e., detection) as demonstrated previously with rats (13).

METHOD

Subjects

Four adult male rhesus monkeys (*Macaca mulatta*) weighing between 8.5–11.0 kg were used. Monkeys were individually housed in aluminum primate cages housed in a temperature-controlled environmental room under a 12-hr light-dark cycle. A water bottle was attached to each chamber and was filled regularly. All food, with the exception of daily fruit and vitamin supplements, was presented during the behavioral tasks. To insure the stability of body weights, monkeys were weighed regularly throughout the experiment. All four monkeys had previously been trained

[1]In conducting the research described in this report, the investigators adhere to the "Guide for the Care and Use of Laboratory Animals," as promulgated by the Committee on Care and Use of Laboratory Animals of the Institute of Laboratory Animal Resources, National Research Council. The views of the authors do not purport to reflect the position of the Department of the Army or the Department of Defense (para 4-3, AR 360-5).

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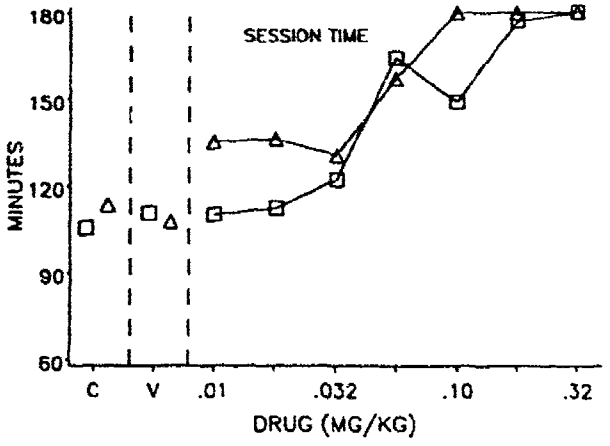

FIG. 1. Average effects of atropine (squares) and azaprophen (triangles) on session duration of the match-to-sample task in four rhesus monkeys. Ordinate: Session length in minutes. Abscissa: Drug dose in mg/kg. Points above V represents data from vehicle injections. Points above C represent the mean of four noninjection control sessions.

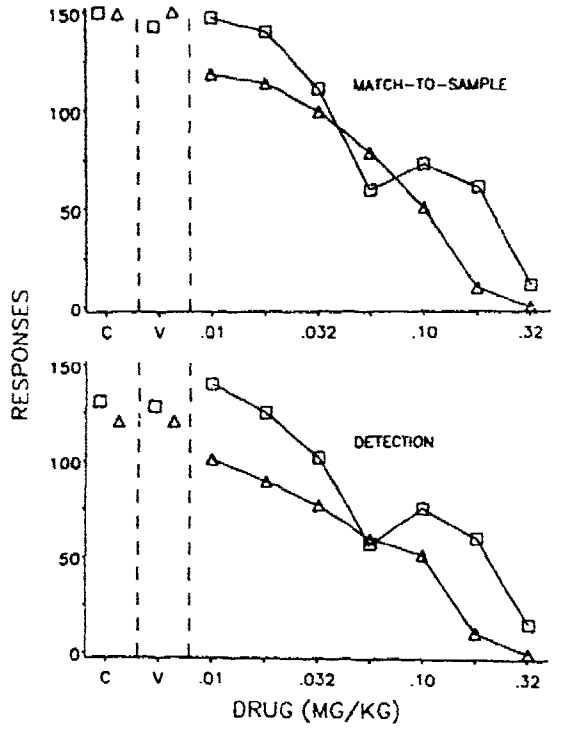

FIG. 2. Average effects of atropine (squares) and azaprophen (triangles) on the number of choice responses on the match-to-sample (top) and detection (bottom) tasks in four rhesus monkeys. Data are from the first 60 min of the sessions. Ordinate: Number of responses. Abscissae: Drug dose in mg/kg. Points above V represent data from vehicle injections. Points above C represent the mean of four noninjection control sessions.

on the concurrent match-to-sample and detection tasks and had pharmacological experience with scopolamine, aprophen and physostigmine.

Apparatus

The rear wall of each primate chamber was equipped with an intelligence panel connected to solid-state controlling equipment and a PDP-11/73 computer located in an adjacent room. Each panel contained five press keys (Coulbourn model E36-15), three keys were mounted horizontally 60 cm above the cage floor (upper keys) and two keys (lower keys) were mounted horizontally 45 cm above the cage floor. Each upper press key could be transilluminated with three stimulus colors and each lower press key could be transilluminated with one stimulus color. A food dispenser, equipped with a stimulus light and capable of dispensing 750 mg banana flavored food pellets (BioServe), was mounted in the center of the panel, 30 cm from the cage floor. Experimental events were controlled and monitored using the SKED-11 operating system (State Systems, Kalamazoo, MI).

Behavioral Procedure

Monkeys responded on concurrent delayed match-to-sample and detection tasks. Trials on the matching task were initiated when monkeys pressed the center upper key (initiating response) within 30 sec after it was illuminated white. Following the initiating response, the center upper key was illuminated either red or green (sample stimulus) for up to 30 sec. Monkeys were required to make eight presses (FR8, on the center upper key while the sample stimulus was present. When the FR8 response requirement was met the center upper key went dark and, following a delay of 0.01, 4, or 16 sec, the upper left and upper right keys were illuminated either red or green (choice stimuli) for a maximum of 30 sec. A choice response occurred when monkeys made a single press on either the left or right upper keys while they were illuminated. If the choice response was on the key illuminated with the same color stimulus (red or green) as the previous sample stimulus, a correct response was considered to occur and a food pellet was presented and the food hopper was illuminated for 1 sec.

When a correct choice was made, the next trial started after 5 sec. When an incorrect choice was made, or the response requirements for initiating a trial, selecting the sample stimulus, or making a choice, was not met, the next trial started after 30 sec. The delay interval, sample stimulus color, and position of correct choice stimulus, was randomly determined for each trial. Sessions lasted for 180 min or until 150 food pellets were earned on the match-to-sample task. Sessions were conducted daily and started at 1300 hr.

A simple detection task was presented concurrently with the match-to-sample task. With an average frequency of one in twenty sec, one of the two lower press keys was illuminated white for up to 2 sec. A single press on the illuminated key produced a food pellet with a probability of 0.25. When food pellets were presented on the detection task the food hopper was illuminated for 1 sec. The position of the illuminated key (either right or left) was randomly determined.

Data Analysis

When a response or an experimental event occurred, the elapsed time during the session was recorded. From these data the following measures for the first 60 min of each session were calculated: 1) total choice responses on the

TABLE 1

POTENCIES OF ATROPINE AND AZAPROPHEN FOR PRODUCING RESPONSE SUPPRESSION ON THE MATCH-TO-SAMPLE AND DETECTION TASKS

	Atropine		Azaprophen	
Monkey	Match-to-Sample	Detection	Match-to-Sample	Detection
7	0.496	0.509	0.706	0.752
8	0.127	0.268	0.230	0.278
9	0.286	0.186	0.579	0.553
10	0.293	0.306	0.297	0.313
Mean	0.301	0.317	0.453	0.474

Data are ED_{50} values in μm/kg based on the weight of the base form of the drugs.

match-to-sample task; 2) total responses on the detection task; 3) percent correct choice responses under each delay condition of the match-to-sample task. The first 60 min of the session was chosen for analysis in order to facilitate comparisons of drug effects by minimizing the contribution of potential differences in the duration of action of atropine and azaprophen as well as individual subject differences in the time required to complete the task. The total session time was also calculated. In order to quantify the comparison of the two compounds, ED_{50} values for response suppression under both tasks were interpolated from dose-effect functions, fitted by least-squares estimation procedures, obtained from data from individual monkeys. Thus, ED_{50} values represent the dose of drug producing response rates of 50% of control rates.

Pharmacological Procedure

Doses of atropine SO_4 (mol.wt.=676.8) and azaprophen HCl (mol.wt.=391.9) (United States Army Medical Research Institute of Chemical Defense) were dissolved in distilled water and distilled water was used for vehicle injections. Injections were IM, about the leg muscles, in a volume of 0.05 ml/kg body wt., 45 minutes before the start of the sessions. Drugs were administered on Tuesdays and Fridays, and data from Thursdays were treated as noninjection control. Drug doses were administered in a mixed order and azaprophen was examined before atropine.

RESULTS

Figure 1 presents the average effects of azaprophen and atropine on the length of time required to complete the match-to-sample task. Both compounds produced dose-dependent increases in session length and a dose of 0.32 mg/kg of either drug increased session length to the maximum allowable duration. Although certain doses of azaprophen (i.e., 0.01 and 0.018 mg/kg) produced small increases in the average session length, whereas equivalent doses of atropine did not, in general, both compounds had similar potencies on this measure.

Atropine and azaprophen produced dose-dependent decreases in the number of responses occurring on the match-to-sample and detection tasks (see Fig. 2). A dose of 0.32 mg/kg of either drug produced a complete or nearly complete suppression of responses under both procedures in all four monkeys. Table 1 presents the ED_{50} values for response suppression under the match-to-sample and detection tasks, in individual monkeys. In general, atropine was slightly more potent than azaprophen for producing response suppression on both of these measures. Additionally, drug effects on the match-to-sample task were correlated with drug effects under the detection task. That is, each drug suppressed responding on the match-to-sample task to approximately the same extent as on the detection task.

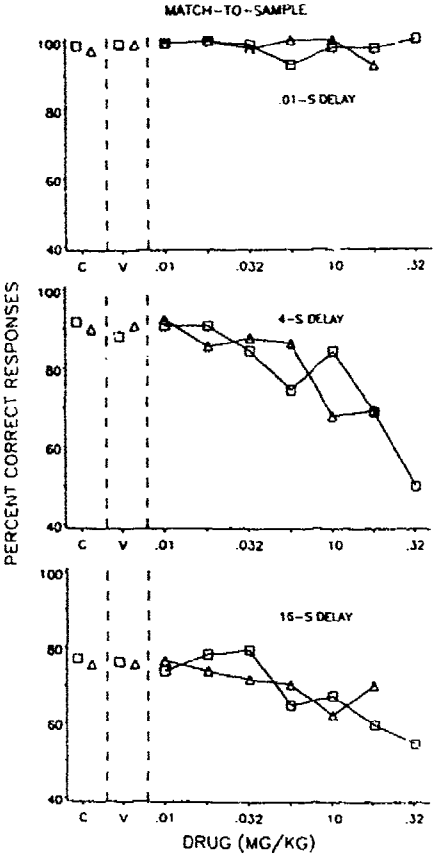

FIG. 3. Average effects of atropine (squares) and azaprophen (triangles) on the percentage of correct choice responses on the 0.01-sec (top), 4-sec (middle), and 16-sec (bottom), delay conditions of the match-to-sample task, in four rhesus monkeys. Data are from the first 60 min of the sessions. Ordinates: Percentage of correct choice responses. Abscissae: Drug dose in mg/kg. Points above V represent data from vehicle injections. Points above C represent the mean of four noninjection control sessions. Sessions containing less than five responses have been eliminated.

Figure 3 presents the effects of atropine and azaprophen on the average percentage of correct choices on the match-to-sample task for each of the three delay intervals. Under baseline conditions, accuracy on the match-to-sample task depended on the delay interval. That is, average percent correct responding was near 100% on 0.01-sec trials, approximately 90% on 4-sec delay trials, and, approximately 77% on 16-sec delay trials. Neither atropine nor azaprophen had any substantial effect on percent correct responding on 0.01-sec

delay trials at any dose. Certain doses (i.e., 0.10, 0.18, 0.32 mg/kg) of both drugs produced decreases in percent correct responding on 4-sec and 16-sec delay trials. Doses producing decreases in percent correct responses also produced substantial decreases in the total number of responses.

DISCUSSION

The major effect observed with both azaprophen and atropine in the present study was to suppress responding. Both compounds produced dose-dependent decreases in the rate of responding on the match-to-sample and detection tasks. Additionally, both drugs produced dose-dependent increases in session length. The similarity between the effects of atropine and azaprophen further characterizes azaprophen's anticholinergic properties.

Atropine was slightly more potent than azaprophen for response suppression on both tasks. The relative potency of atropine and azaprophen observed in the present study is consistent with previous results obtained with in vitro tests of carbachol-induced inhibition of prolactin (5) and [^{3}H]N-methylscopolamine binding (13) assays, but is in contrast with results from carbachol-induced α-amylase release (6, 7, 13) and acetylcholine-induced contractions of guinea pig ileum (6, 7, 9) preparations. These results are also consistent with and extend previous results with schedule-controlled behavior in rats (13).

Both atropine and azaprophen decreased the accuracy of response on the match-to-sample task when the choice delay was 4 or 16 sec, but not when the delay was 0.01 sec. These results are consistent with results reported by Penetar and McDonough (12) for atropine in rhesus monkeys, using a similar procedure. It is notable that, in the present study, all doses of atropine or azaprophen that produced decreases in correct responses on the match-to-sample task also produced a substantial degree of response suppression. It is possible that nonspecific drug effects contributed to the observed decreases in accuracy and thus, in the present study, the match-to-sample task does not appear to be sensitive to drug effects on memory processes. It is clear, however, that atropine and azaprophen have similar effects on the match-to-sample task.

ACKNOWLEDGEMENTS

The authors thank Jeffrey Witkin for helpful comments on the manuscript and Donald Conrad and Lisa King for technical assistance with the conduct of the experiments.

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