Pharmacology Biochemistry & Behavior, Vol. 30, pp. 119–122. © Pergamon Press plc, 1988. Printed in the U.S.A. 0091-3057/88 $3.00 + .00

Nicotine Impairs Acquisition of Radial Maze Performance in Rats

WILLIAM R. MUNDY* AND EDGAR T. IWAMOTO†[1]

Department of Pharmacology† College of Medicine, the Graduate Center for Toxicology and the Tobacco and Health Research Institute, University of Kentucky, Lexington, KY 40536*

Received 9 July 1987

MUNDY, W. R. AND E. T. IWAMOTO. *Nicotine impairs acquisition of radial maze performance in rats.* PHARMACOL BIOCHEM BEHAV **30**(1) 119–122, 1988.—The effects of nicotine (NIC) and scopolamine (SCOP) on radial maze acquisition were examined using an 8-arm radial maze. In Experiment 1, food-deprived Sprague-Dawley rats were trained to eat food pellets located at the ends of each arm of the radial maze without repeating arm choices. Both NIC (0.45 mg/kg, SC) and SCOP (0.25 mg/kg, IP) impaired acquisition when they were administered before, but not after the daily training sessions. Experiment 2 examined the effect of nicotine on working and reference memory in rats trained to a criterion of 3 correct choices out of the first 4 choices with only 4 of the 8 arms baited. NIC (0.1–0.45 mg/kg) had no effect on working memory (reentry into baited arms) or reference memory (entry into unbaited arms) errors. It is concluded that NIC impairs processes involved in the acquisition but not maintenance of radial maze performance. Neither NIC nor SCOP affects post-training consolidation processes.

Nicotine Scopolamine Radial maze Working memory Reference memory

OLTON and Samuelson [29] developed an apparatus, the radial arm maze, which has been used to study spatial memory in rats. The apparatus consists of a central platform from which the arms of the maze extend like the spokes of a wheel. In this task animals are trained to "remember" the spatial location of food hidden at the end of the maze arms. Subsequent studies by Olton and co-workers demonstrated that accurate choice behavior was dependent on extra-maze cues [27,30]. In addition, a number of lesion studies have suggested that cholinergic systems are critical in maintaining the integrity of spatial memory in rats. Disruption of cholinergic input into the hippocampus [16, 18, 25, 31, 32] or damage to cholinergic cells of the nucleus basalis [1, 6, 16, 22, 35] have been reported to impair radial maze performance.

Pharmacologic studies have also implicated cholinergic systems in the mediation and/or maintenance of spatial memory. The muscarinic antagonist scopolamine has been reported to produce deficits in radial maze performance in well trained rats [3, 4, 7, 13, 17, 23, 37] and disrupt acquisition of the task by naive animals [34,36]. Methylscopolamine, a muscarinic antagonist which does not cross the blood-brain barrier, was used as a control in two of these studies [7,17]. Methylscopolamine did not impair radial maze performance, suggesting that the actions of scopolamine occur at central cholinergic site. These studies suggest that muscarinic cholinergic pathways play an important role in spatial memory.

In contrast, little has been reported regarding nicotinic cholinergic influences on spatial memory. Nicotine has been shown to affect learning and memory in a number of procedures including active avoidance [9–12, 26], passive avoidance [15,23], maze learning [2, 5, 11], and habituation [33] tasks. Recently, we have reported that nicotine impairs the acquisition of an autoshaped lever-touch response in rats [20].

Despite the literature demonstrating nicotine-induced changes in learning and memory processes, the effects of nicotine on spatial memory have not been extensively studied. Therefore, the present experiments were designed to evaluate the effects of nicotine on spatial memory in the rat using the radial arm maze. The initial experiment examined the effects of nicotine on radial maze acquisition. Groups of rats received nicotine either before (pre-session) or immediately after (post-session) daily sessions on the 8-arm radial maze with all arms baited. Nicotine was administered at a dose that had previously been shown to impair acquisition of an autoshaped lever-touch response [20]. The effects of scopolamine on radial maze acquisition were also examined. While it has been previously shown that pre-session scopolamine administration impairs radial maze acquisition [34,36], the effect of post-session scopolamine has not been studied. In the second experiment, the effect of nicotine on working and reference memory was examined in rats trained to asymptotic performance on the 8-arm radial maze with only 4 arms baited. Using this baiting method, the unbaited arms serve as a reference memory component to the task since information that food will not be found at the end of these arms is useful over all trials. The baited arms serve as the working memory component. Within a trial, repeated

[1]Requests for reprints should be addressed to Dr. Edgar T. Iwamoto, Department of Pharmacology, Albert B. Chandler Medical Center, University of Kentucky, Lexington, KY 40536.

entries into the baited arms are indicative of a working memory deficit, while entries into unbaited arms indicated a reference memory deficit.

METHOD

Animals

Adult male Sprague-Dawley rats were obtained from Harlan Industries (Indianapolis, IN) with initial free feeding weights of 275–300 g. The animals were quarantined for two weeks in groups of four, and then were individually housed and gradually food-deprived and maintained at 80–85% of their initial body weight with free access to water. Rats were kept in a temperature contolled (21°C) environment on a 12:12 hr light/dark schedule, with lights on at 0700 hr. Experiments were performed between 1200 and 1700 hr.

Apparatus

Behavioral testing utilized an 8-arm radial maze elevated 50 cm above the floor. The maze was constructed of wood coated with polyurethane. Each arm (65×10 cm wide) extended from an octagonally shaped central platform (22 cm wide). Circular food wells (0.5 cm deep, 1 cm diameter) were located 1 cm in from the ends of each arm. The maze was centered in an enclosed room illuminated by fluorescent bulbs. Visually distinct cues (e.g., door, poster, wall sockets, camera, ceiling beam) surrounded the maze and remained in the same position during the course of the experiments. Animal movements were observed via a video camera mounted above the center of the maze.

Procedure

Experiment 1. Twenty-four rats were initially adapted to the maze by placing them individually in the maze for 10 min per day and allowing them to explore and eat food pellets (45 mg, Bioserve, Frenchtown, NJ) scattered along all arms of the maze. After three days of adaptation rats were randomly assigned to nicotine treated (n=8), scopolamine treated (n=8), and saline control (n=8) groups and acquisition training commenced on the next day. Rats received one daily session on the maze with all 8 arms baited. At the start of each training session, each arm was baited by placing one 45 mg food pellet in the food well. The rat was then placed on the center platform facing a particular arm (the same for all sessions and for all rats) and allowed to choose arms until all 8 pellets were consumed or 10 min elapsed. The maze was thoroughly washed between sessions. All arm entries and their order of entry were recorded for each rat. An arm entry was scored when an animal traveled at least halfway down an arm. Correct choice was defined as entering an unchosen arm for that session. Training continued until all animals in the saline group attained the criterion of 7 correct choices out of the first 8 choices for at least three consecutive sessions. Drug treated animals were injected with nicotine base (0.45 mg/kg, SC; Eastman Kodak Co., Rochester, NY) or scopolamine (0.25 mg/kg, IP; Sigma Chemical Co., St. Louis, MO) 15 min prior to daily sessions. Control animals received SC injections of normal saline. An additional 24 animals were subjected to an identical training procedure except that nicotine and scopolamine were administered immediately after completing each daily session.

Experiment 2. Fourteen experimentally naive rats were adapted and trained on the 8-arm radial maze in a manner similar to that described in Experiment 1, except that a predetermined subset of only 4 arms was baited with the 45 mg food pellets. The baited arms remained the same throughout the experiment but varied from rat to rat. Training was continued until rats attained a criterion of 3 correct choices out of the first 4 choices on three consecutive sessions. Correct choice was defined as an entry into a baited arm. The first entry into an unbaited arm was recorded as a reference memory error. Reentry into any arm previously visited during a session was recorded as a working memory error. The time to complete the first 4 choices was recorded, with a maximum time of 10 min. Four rats not developing consistent performance during training were removed from the experiment. Drug testing for the remaining 10 rats was conducted 7 days a week. Rats received SC injections of 0.1, 0.25, and 0.45 mg/kg nicotine base 15 min prior to testing. Drug tests were conducted every fourth day; during the intervening days performance on the radial maze was evaluated in the usual manner but no drug was administered. Each rat was tested once at each dose level with the order of treatment counterbalanced among subjects using a latin square design.

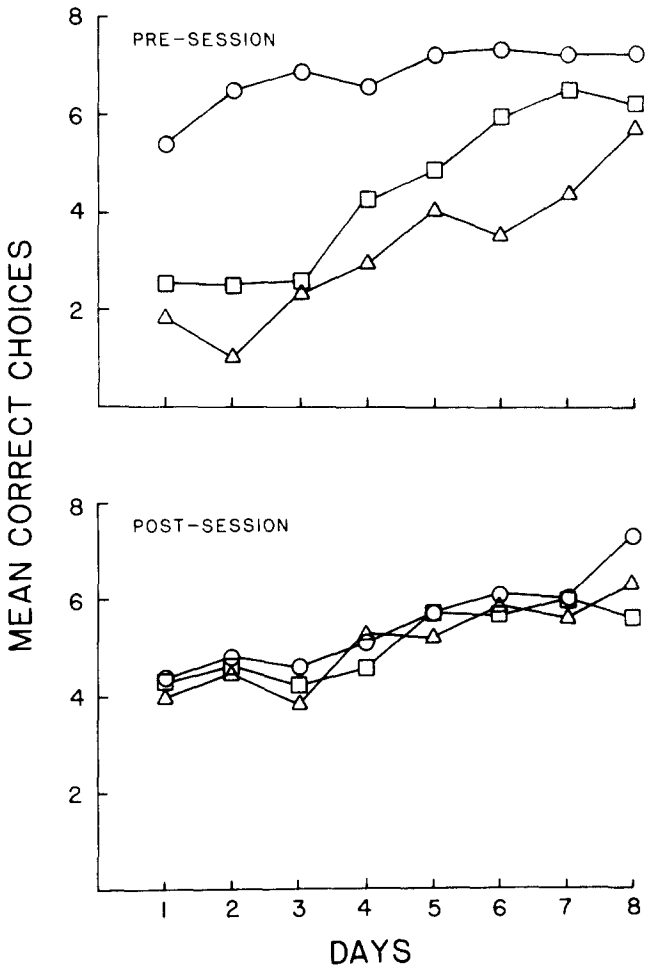

FIG. 1. Mean number of correct choices per session during acquisition of radial maze performance. Rats were injected with 0.45 mg/kg of nicotine (squares), 0.25 mg/kg of scopolamine (triangles), or saline (circles) either 15 min before (top graph) or immediately after (bottom graph) the daily training sessions. N=8 per group.

TABLE 1

MEAN NUMBER OF ERRORS PER SESSION IN RATS TREATED WITH NICOTINE*

		Nicotine (mg/kg)		
Error Type	Saline	0.1	0.25	0.45
Working memory	0.10 (0.10)	0.20 (0.13)	0.10 (0.10)	0.20 (0.13)
Reference memory	0.70 (0.21)	0.80 (0.20)	1.00 (0.30)	0.80 (0.29)

*Drug or saline administered SC 15 min prior to testing. N = 10 per group.

Values are means with one SEM in parentheses.

Statistics

For Experiment 1 the number of correct choices in the first 8 choices were analyzed using a two-way repeated measures analysis of variance, with treatment as one factor and days as the other. Post hoc comparisons were made using Tukey's studentized range test at a significance level of $p<0.05$. The type of error (working or reference) and the time to complete the first 4 choices were analyzed in Experiment 2 using a one-way analysis of variance. In the case of a significant effect of treatment, post hoc comparisons were made using Tukey's studentized range test. Separation of working memory errors as to reentry into baited arms and reentry into unbaited arms had no effects on the outcome of the statistical analysis.

RESULTS

Experiment 1

The effect of pre-session nicotine and scopolamine administration on acquisition of the radial maze task is shown in Fig. 1 (top). Analysis of the number of correct choices in the first 8 choices revealed a significant effect of treatment, $F(2,21)=8.05$, $p<0.005$, and days, $F(7,147)=12.55$, $p<0.0001$, but no significant interaction between treatment and days. All groups showed an improvement in performance over the eight-day training period. Animals in the saline group attained the criterion of at least 7 correct choices out of the first 8 choices by day 5. Post hoc analysis on the significant effect of treatment showed that both nicotine (0.45 mg/kg) and scopolamine (0.25 mg/kg) significantly decreased the number of correct choices compared to saline controls ($p<0.05$).

The effect of post-session nicotine and scopolamine administration is shown in Fig. 1 (bottom). Analysis of the number of correct choices in the first 8 choices revealed no significant effect of treatment or treatment × day interaction. All groups showed an improvement in performance over the eight day training period, which is reflected in the significant effect of days, $F(7,147)=4.08$, $p<0.0004$. Post-session injections of nicotine or scopolamine had no effect on the number of correct choices on any day compared to saline controls.

Experiment 2

The effect of nicotine working and reference memory in trained animals is shown in Table 1. Under the saline condition animals made an average of 0.10 working memory errors and 0.70 reference memory errors. Nicotine at doses of 0.10–0.45 mg/kg did not significantly alter the number of working or reference memory errors compared to the control condition. Nicotine also had no significant effect on the time to complete the first 4 choices (data not shown).

DISCUSSION

Both nicotine and scopolamine injected 15 min prior to training sessions resulted in a profound impairment of acquisition of radial maze performance. These results are consistent with previous studies showing that scopolamine can impair acquisition of radial maze performance when administered before training [34,36]. Nicotine, when administered before training in sufficiently high doses, has been shown to impair acquisition in a number of operant procedures [5, 9, 20, 26]. In the present study nicotine impaired acquisition when administered prior to daily training sessions in naive rats (Fig. 1), but had no effect on working or reference memory errors when administered prior to testing in trained rats (Table 1). Previous studies in our laboratory have also shown that nicotine at doses of 0.1–0.8 mg/kg failed to affect choice performance in trained rats using an 8-arm radial maze with all 8 arms baited [19]. This pattern of results suggests that nicotine may be affecting mechanisms involved in the aquisition but not the maintenance of spatial memory. However, the present experiments based on pre-training drug administration cannot exclude possible drug-induced changes in motor, perceptual, and motivational processes which may have contributed to the acquisition deficits observed after both nicotine and scopolamine treatment.

The results from Experiment 1 indicate that neither drug had an effect on post-training memory consolidation processes related to spatial learning. Acquisition of the radial maze task was not altered by immediate post-session injections of either nicotine or scopolamine compared to saline-treated controls. Other researchers using spatial tasks have also reported negative findings when cholingergic muscarinic antagonists were administered after the training session [13,14]. Recently, we reported that both nicotine and scopolamine caused a dose-related impairment in the acquisition of an autoshaped lever-touch response in rats [20,21]. In these studies the magnitude of impairment was greater after post-session drug administration compared with the same doses administered pre-session, and it was concluded that nicotine and scopolamine were affecting post-training consolidation processes. These results suggest that the effects of post-session nicotine and scopolamine on learning and memory are different for spatial and nonspatial tasks. This is not surprising since Olton and co-workers developed the radial maze task to take advantage of the rats preferential use of spatial stimuli during discrimination learning [29,30]. Rats are extremely well adapted to learn spatial tasks, and acquisition of the radial maze takes place rapidly. The acquisition of an operant may involve more complex behavioral substrates. In addition, it has been shown that substantial learning can take place during the habituation trials [30]. Thus, it might be hypothesized that post-training drug injections in rats would be less likely to affect the acquisition of spatial tasks which are rapidly learned and part of a species-specific response pattern. Further experimentation comparing the effects of post-training drug administration in spatial and nonspatial tasks will be necessary to test this hypothesis.

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