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Effects of Sodium Barbitone on Learning and Memory-Storage of an Appetitive and an Aversive Task

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TOMAZ, C. A. B., D. F. VENTURA AND J. R. LEITE. *Effects of sodium barbitone on learning and memory-storage of an appetitive and an aversive task.* PHARMAC. BIOCHEM. BEHAV. 17(5) 909–913, 1982.—In order to test the effects of sodium barbitone on the acquisition and retention of an appetitively and an aversively reinforced behavior, mice were trained in a spatial discrimination Y-maze task. Learning was observed in both situations, with acquisition unimpaired by the drug. Sodium barbitone did, however, affect retention of both tasks in all groups treated with the drug before training. Results are discussed in light of the various modes of action of this drug, i.e., as an inhibitor of protein synthesis, as a blocker of catecholamine biosynthesis, with regard to its effects on paradoxical sleep and on gamma-amino-butyric acid (GABA).

Sodium barbitone Learning Long-term memory Appetitive task Aversive task Y-maze

PROTEIN synthesis, according to several reports, is fundamental for the process of memory consolidation. It had been demonstrated that some protein inhibitors impair the acquisition and memory-storage of several behavioral tasks [1, 7, 8]. Recent data suggest that the blocking action of some drugs on retention may be specific to the task to be retained [15]. By far, the majority of the studies on memory consolidation have employed the well known situation of shuttle boxes and other active avoidance as well as passive avoidance situations. Very few studies have made use of appetitive control situations [2,9]. Some findings suggest that prevention of long-term memory by puromycin, as repeatedly reported in aversive studies, fails to occur in appetitive situations [12,15]. These data suggest the possibility that there is more than one neuronal mechanism for memory consolidation.

It has been demonstrated that chronic administration of sodium barbitone drastically inhibits the synthesis of proteins in brains of rats and mice [13,14]. With regard to the wide usage of barbiturates, as in the case of some epilepsies which are treated with long-term administration, it may also affect learning and memory. It has indeed been shown, with chronic administration, in a passive avoidance situation, that a memory deficit resulted from treatment with sodium barbitone [13]. This study proposes to test the effects of acute administration of sodium barbitone on the acquisition and retention of two different tasks in order to compare its effects on appetitively and aversively reinforced behaviors.

EXPERIMENT 1

METHOD

Subjects and Drug Administration. 75 mice (*Mus musculus*—13 females and 62 males) were placed in individual cages with food and water ad lib, and divided into five groups: T-R (task control group); S-T-R (saline control group); D-T-R (experimental group with drug administration 30 min before training); T-D-R (experimental group with drug administration 30 min after training); D-T-D-R (experimental group with drug administration 30 min before training and 30 min before retention test, drug-dissociation paradigm). The abbreviations used stand for: T=training; R=test of retention; D=drug; S=saline.

The drug used was sodium barbitone (5,5-dietilbarbiturate of sodium; Merck and Company, Darmstad, Germany). The drug, dissolved in distilled water at concentration of 6 mg/ml, was injected intraperitoneally in a volume of 60 mg/Kg of body weight. A Y-maze with a 32 cm runway and 30 cm arms was used in the Y-maze learning task. The goal boxes were

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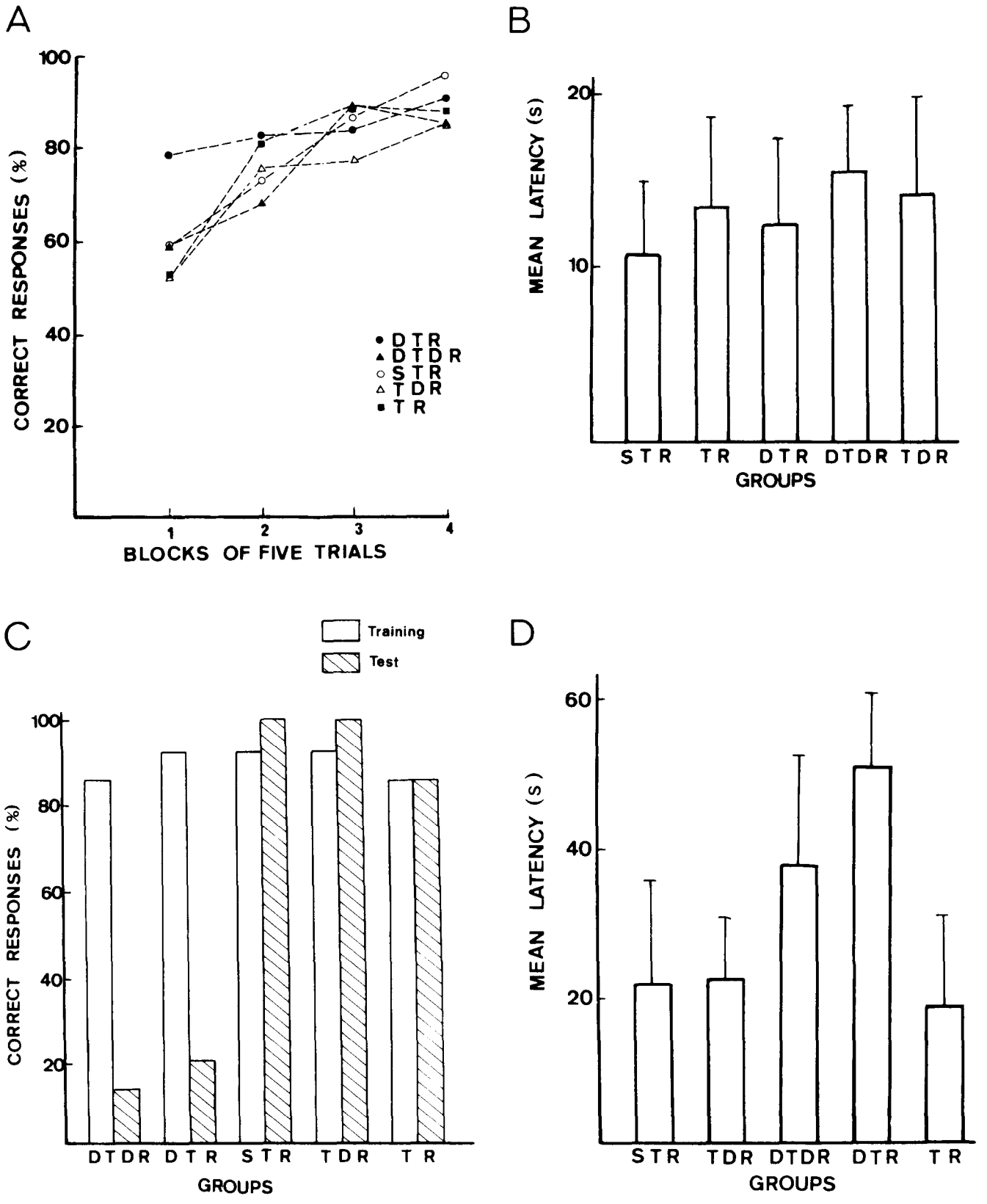

FIG. 1. (A) Subjects' performance during the last 20 training trials in each group. (B) Mean latency time required for the subjects to reach the goal box. Each bar represents the group mean for the 11th training trial. (C) Effects of sodium barbitone on retention of appetitive learning. Comparison of performances in the last learning trial and in the first trial of the retention test. (D) Mean latency time required for the subjects to reach the goal box in the first trial of the retention test.

separated from the end of the arms by sliding doors. The animals were water deprived for 22 hr before each experimental session during the three days of experimentation. To determine initial preference for one of the arms of the maze, each subject was placed in the start box and tested in five non-reinforced trials. For the first 10 test trials; the preferred arm was blocked. The final 20 trials were conducted with both arms opened. Entry in the correct goal box was rewarded with 0.05 ml of water, available on a small dish. On the third day the animals were submitted to a 20 trial retention session with both arms open.

RESULTS

Figure 1 A shows the percentage of correct responses during training (2nd day of experimentation), in blocks of five trials. Differences between group means were not significant ($F=1.5526$; $p>0.005$; Analysis of Variance test).

No differences among groups were noted when the mean time required for the animals to reach the goal box was considered (Fig. 1 B; $F=1.0447$; $p>0.005$; Analysis of Variance test).

The effects of administration of sodium barbitone on retention of appetitive learning are shown in Fig. 1 C. Comparison of differences between percentage of correct responses in the last trial of the acquisition training and in the first retention test response revealed significant differences for groups D-T-D-R and D-T-R ($\chi^2=49.5370$; $p<0.005$; Chi square test).

Figure 1 D shows the mean time required for the animals to reach the goal box in the first trial of the retention test. There were significant differences between D-T-R and the other groups ($F=3.9196$; $p>0.005$; Analysis of Variance test).

EXPERIMENT 2

METHOD

Subjects and Drug Administration. 30 mice (*Mus musculus*—10 females and 20 males), 3 months old, were divided into 3 groups (D-T-R; D-T-D-R; S-T-R) and were submitted to the same treatments as in Experiment 1.

Y-Maze Learning. The apparatus was the same as in Experiment 1. During training the animals were maintained on a schedule of water deprivation of 24 hours. Subjects were submitted to the experimental conditions for four days. On the first and second sessions the same procedure described in Experiment 1 was utilized, except for the injections. On the 3rd day, the animals received either drug or saline, and were submitted to four trials. On the second trial they received an electric shock (0.9 mA during 3 sec) through the cage floor whenever a correct response occurred.

These animals were retested 24 hr later. Latencies and correct responses were recorded.

RESULTS

Figure 2 A shows the acquisition of the Y-maze discrimination, in terms of percentage of correct response. All groups showed good learning scores. There were no significant differences between groups ($F=0.053$; $p>0.005$; Analysis of Variance test).

Table 1 shows the results obtained on the 3rd day. As can be seen, animals from the three groups showed good retention scores (data from 1st and 2nd trials). Data from the last 2 trials show that the animal could remember the shock pre-

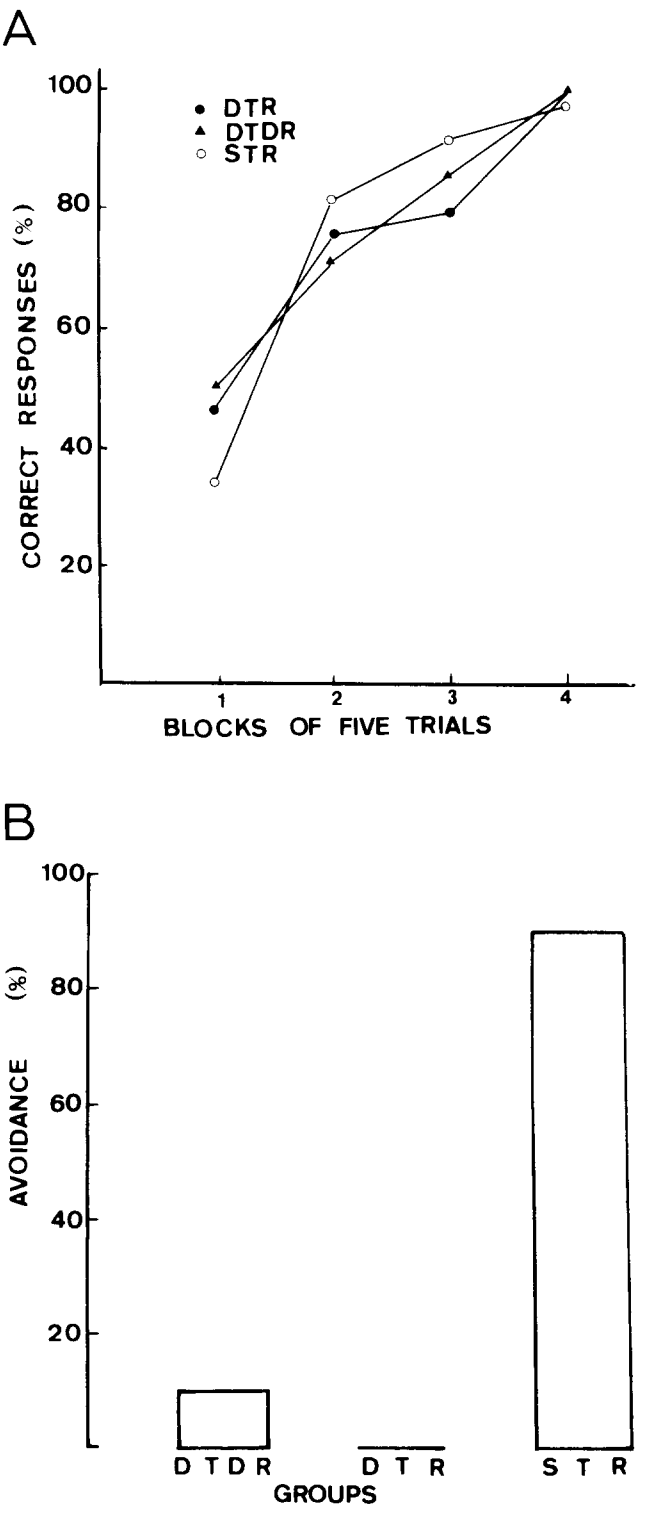

FIG. 2. (A) Subjects' performance during the last 20 training trials in each group. (B) Effects of acute administration of sodium barbitone and/or saline on retention of avoidance in the first trial of retention test.

TABLE 1

PERCENTAGE OF CORRECT RESPONSES AND MEAN TIME REQUIRED FOR THE ANIMALS TO REACH THE GOAL BOX FOR THE FOUR TRIALS ON THE 3rd DAY OF EXPERIMENT 2

	Trials							
	1		2		3		4	
Groups	%	Latency*	%	Latency	%	Latency	%	Latency
D-T-R	100	23.4	100	10.6	0	114.0	0	127.0
D-T-D-R	80	23.9	100	9.7	0	112.0	0	118.0
S-T-R	90	19.1	100	5.8	0	121.0	0	119.0

*Seconds.

sented after the 2nd trial (latencies significantly greater than for non-shock groups).

The effects of injected sodium barbitone on retention of the active avoidance response are shown in Figure 2 B. This figure shows the percentage of avoidance responses in the three groups during the first trial of retention test. Groups D-T-R and D-T-D-R did not show retention of the avoidance response.

DISCUSSION

The present study demonstrates that sodium barbitone, a drug which had previously been shown to be capable of preventing long-term memory in an aversive situation [1,2], is also capable of analogous effects in an appetitive situation. In addition, the present study further confirms the blocking effects of this substance in an aversive task introduced in the same experimental situation which was used for the appetitive task. Therefore, the differential blocking effects on aversive and appetitive situations found for puromycin [12,15], are not confirmed in the case of sodium barbitone.

The blocking action of sodium barbitone was observed when it was administered before training. Administration 30 minutes after training did not affect long-term memory.

Previous studies have indicated that a short period of cerebral protein synthesis inhibition, established during discrimination training, is sufficient to produce amnesia; but that a similar period of inhibition initiated 30 minutes or longer after training has no effect [3, 16, 17]. These results have suggested that cerebral protein synthesis occurring close to training may be sufficient for long-term memory of discrimination training. The present findings provide additional support to this conclusion.

An explanation of the present results in terms of the hypothesis of state-dependent learning was discarded, since animals in the group D-T-D-R also had their memory impaired by the drug, both in the aversive and the appetitive situations.

Previous work demonstrated that chronic ingestion of sodium barbitone decreased brain synthesis [13]. The present results showed impairment on memory consolidation with acute sodium barbitone administration for tasks controlled by positive and negative reinforcement. This could be explained by inhibition of protein synthesis if it comes to be shown that acute sodium barbitone treatment also produces such an effect. Other explanations for the memory deficit found should also be mentioned. One of these is related to the well known fact that barbiturates reduce the amount of time spent in the REM phase of sleep and in this respect at least, barbiturate-induced sleep differs from physiological sleep [10]. The demonstrations that REM sleep deprivation impairs the formation of long-term memory [4,6] make it possible that sodium barbitone could affect memory due to the associated REM sleep deprivation, rather than through protein synthesis inhibition.

Another alternative explanation may be found in the suggested involvement of catecholamine synthesis in memory consolidation (e.g., [4]). Since sodium barbitone is a proposed blocker of catecholamine biosynthesis [4] its inhibitory effects on memory consolidation could be due to catecholamine synthesis inhibition, as well.

Finally, a variety of reports have examined and suggested the involvement of gamma-amino-butyric acid (GABA) neurons in memory consolidation [5,11] and recent observations support the notion that depressant and anticonvulsant barbiturates could prolong GABA-mediated inhibition by increasing the lifetime of the activated ion channel, therefore affecting memory consolidation [18].

Thus, it is not possible to attribute the effects of sodium barbitone on retention solely to the inhibition of brain protein synthesis. It is possible that its effects on memory may be due to either one of the other mechanisms (i.e., REM sleep deprivation, catecholamine synthesis inhibition, and prolongation of GABA-mediated responses) or to any combination of the four influences.

Further research is needed to isolate the relative importance of the four influences on consolidation under action of sodium barbitone.

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