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Differential Effects of Intrahippocampally or Systemically Applied Picrotoxin on Memory Consolidation in Rats[1]

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GRECKSCH, G. AND H. MATTHIES. *Differential effects of intrahippocampally or systemically applied picrotoxin on memory consolidation in rats.* PHARMAC. BIOCHEM. BEHAV. **14**(5) 613–616, 1981.—The effect of picrotoxin on retention of a brightness discrimination was investigated using hippocampal or systemical injections. Using intraperitoneal injections an improvement of retention was found. If picrotoxin was hippocampally injected a retention impairment was seen. A role of the GABA-ergic systems in the hippocampus of the rat for memory consolidation was suggested.

Memory consolidation Hippocampus Picrotoxin Rat GABA

THE role of neurotransmitters for learning and memory has been intensively investigated in the last years. It has become evident that modulation of several neurotransmitter systems influences memory consolidation processes in various learning tasks and in various animal species. Since the hippocampus is a region found to be important in the consolidation process [1, 3, 11, 13, 14, 17, 18], we have investigated the involvement of hippocampal transmitter systems in memory consolidation by means of hippocampal topical application of transmitter agonists and antagonists. After hippocampal injection of the cholinomimetic drug, oxotremorine, and the dopaminergic agonist, apomorphine, we found an improvement of consolidation, and after the cholinolytic, scopolamine, and the dopamine receptor blocker, haloperidol, an impairment [7,8]. The GABA-level enhancing substance, n-dipropylacetate (n-DPA), also produced an improvement in consolidation [9]. To verify the role of hippocampal GABA-ergic systems in memory processes, experiments using GABA antagonists were carried out. In the present experiment we investigated picrotoxin as a GABA-antagonist [16,21] using the learning task employed previously, a brightness discrimination in a Y-chamber. Picrotoxin is known to be a central nervous system stimulant. Numerous studies have shown that posttraining systemical administration of stimulants causes improvements of memory consolidation [2, 4, 5, 6, 10, 20]. For this reason we have investigated the effect of hippocampally or systemically injected picrotoxin on consolidation under the same experimental conditions.

METHOD

The experiments were performed on 41 male Wistar rats of our own breeding stock. Twenty-one animals received chronically implanted microcannulas in each dorsal hippocampus (coordinates according to Skinner [22]: AP=3.1, lateral=3.1, vertical=3.1 mm) one week before the learning experiment.

Training Procedure

The training task used was that of brightness discrimination in a semiautomatic Y-chamber. The animals were motivated by electrical footshocks [19]. After application of a starting stimulus (1.0 mA) to the grid floor, each animal escaped from the starting compartment of the Y-chamber. In order to avoid the stimulus, the rat had to run into the illuminated alley of the chamber; entering the dark alley of the chamber was punished by footshock. A trial was evaluated as correct only when the animal ran directly into the illuminated alley after application of the starting stimulus. To avoid position training the illuminated alley was changed after every three trials. Training was terminated after 31 trials. The intertrial interval was 1 min (30–90 sec).

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Retention Test

Twenty-four hours after training retention was tested in a relearning procedure, which was carried out in the same manner as the training procedure.

Evaluation

In each case the incorrect trials (errors) during training and relearning were tallied; these values served as a basis for calculating the saving scores:

$$\text{Percentage savings} = \frac{\text{training errors} - \text{relearning errors}}{\text{training errors}} \times 100$$

Injections

All injections were performed immediately after training. The animals with intrahippocampal cannulas received 1.0 μg picrotoxin/hippocampus or 1 μl of artificial cerebrospinal fluid (ACF)/hippocampus. Picrotoxin was dissolved in ACF. In each case 1 μl of solution per hippocampus was injected for approximately 30 sec.

The other 20 animals received systemical injections of 1 mg/kg picrotoxin IP or saline.

Statistics

Statistical evaluation was performed using the two-tailed Mann-Whitney U test.

Control of the position of the intrahippocampal cannulas. Upon completion of the experiment the brains of the rats with chronic microcannulas were stereomicroscopically checked for correct position of the cannulas. Only animals showing exact cannula position in the region CA1 of the dorsal hippocampus were used for evaluation.

RESULTS

The mean values of the training errors were approximately the same in control and experimental groups.

Intrahippocampal Injections

As depicted in Fig. 1 the bilateral hippocampal injection of 0.1 μg picrotoxin/hippocampus immediately after training caused a pronounced impairment of retention. The animals treated with picrotoxin showed significantly more errors in the relearning test 24 hours after training and significantly lower saving scores compared to the controls.

Systemic Injections

Contrary to hippocampal application, peripheral injection of picrotoxin improved the retention performance (Fig. 2). We found that the picrotoxin treated animals showed significantly higher saving scores and fewer errors in the relearning test than the saline controls.

DISCUSSION

The experiments showed that picrotoxin causes different effects depending on how it is administered. Using intraperitoneal injections we found an improvement of the retention performance. This is in accordance with the findings from the literature [2, 6, 10, 15]. Picrotoxin applied in posttraining also antagonizes the amnestic effect of the protein synthesis inhibitor anisomycin [5]. If picrotoxin was hippocampally injected, a retention impairment was seen. This finding corresponds with the results of the experiment with hip-

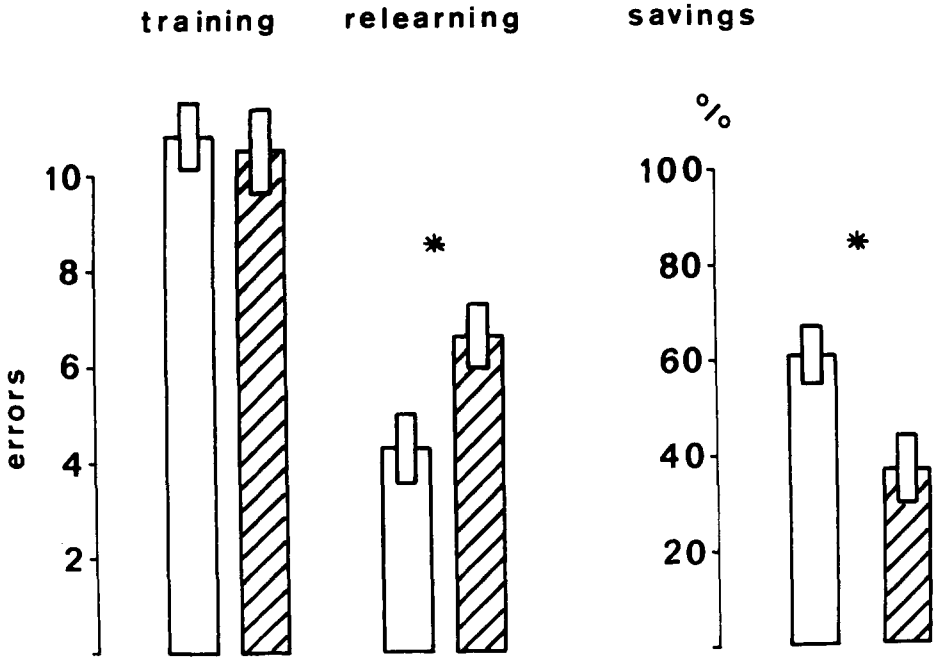

FIG. 1. Effect of posttraining hippocampal injection of picrotoxin on the retention of a brightness discrimination. Errors in training and relearning and saving scores (±SEM). Open bars=control group; n=10. Shaded bars=picrotoxin group; n=11. *$p<0.05$.

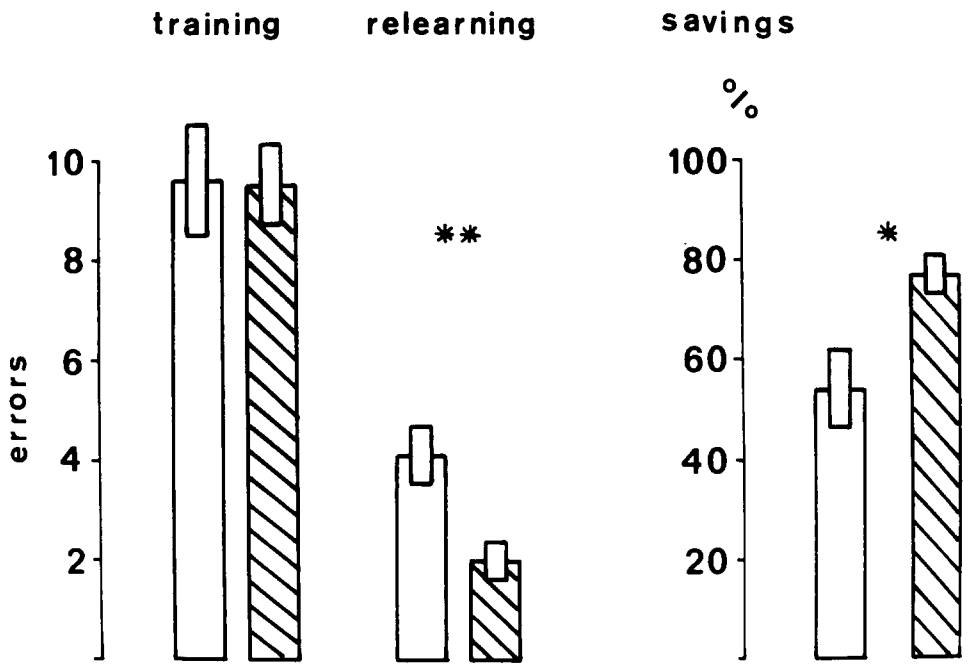

FIG. 2. Effect of posttraining systemical injection of picrotoxin on the retention of a brightness discrimination. Errors in training and relearning and saving scores (±SEM). Open bars=control group; n=10. Shaded bars=picrotoxin group; n=10. *$p<0.05$; **$p<0.02$.

pocampal injections of the GABA-level enhancing substance, n-dipropylacetate.

The opposite results in our investigation of systemic versus hippocampal application of picrotoxin can not be a dose dependent effect, since both dosages were chosen in such a way that no behavioral convulsions and no seizures in the hippocampal EEG were found. But these findings using picrotoxin are not necessarily inconsistent. Kim and Routtenberg [12] found a retention disruption following topical picrotoxin injection into the substantia nigra. Following peripheral injection numerous brain structures would be affected by the picrotoxin. In conclusion, following hippocampal picrotoxin injection, GABA-ergic systems in the hippocampus were blocked resulting in an impairment of consolidation processes. The results obtained with n-DPA and picrotoxin suggest a role of the GABA-ergic systems in the hippocampus of the rat for memory consolidation.

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