Available online at www.sciencedirect.com
SCIENCE DIRECT
Cognitive Brain Research 25 (2005) 741 – 746
COGNITIVE BRAIN RESEARCH
www.elsevier.com/locate/cogbrainres

Research Report

Partial unilateral inactivation of the dorsal hippocampus impairs spatial memory in the MWM

José M. Cimadevilla [a,*], Ruben Miranda [b], Laudino López [b], Jorge L. Arias [b]

[a] *Department of Neuroscience, University of Almeria, 04120 Almeria, Spain*
[b] *Laboratory of Psychobiology, University of Oviedo, 33003 Principado de Asturias, Spain*

Accepted 3 September 2005
Available online 10 October 2005

Abstract

The hippocampus is one of the more widely studied structures related with spatial memory. In this study, we assessed the effect of unilateral inactivation of the dorsal hippocampus with tetrodotoxin (TTX) on the performance displayed by Wistar rats in the spatial version of the Morris water maze. In experiment 1, we injected into the dorsal hippocampus in two different groups of rats 1 μl of saline solution or 5 ng of TTX in 1 μl of saline each day immediately after the training during four consecutive days. This procedure blocked consolidation and impaired spatial memory in the TTX group. In experiment 2, a new group of subjects was trained in the Morris water maze for 8 days and was administered 1 μl of saline on day 7 (saline session) and TTX on day 8 (TTX session) into the dorsal hippocampus 40 min before the training. Only the treatment with TTX altered the retrieval of memories. These experiments showed that unilateral interventions on the dorsal hippocampus can affect consolidation as well as retrieval of well-established spatial memories.

Theme: Neural basis of behaviour
Topic: Learning and memory: systems and functions

Keywords: Consolidation; Rat; Retrieval; Reversible lesion; TTX

1. Introduction

Much neuroscience research is concerned with establishing the substrate of our mental processes. In this respect, memory is one of the intellectual proprieties more deeply studied and the hippocampus, a brain structure directly related with it, since its lesion disrupts memory processes [8,23–25] and because long-term potentiation (LTP), a physiological property of the hippocampus, is considered necessary for memory formation in the brain [4,18].

For many years, the Morris water maze (MWM) has been widely used in rodents to study the role of the hippocampus [9,10,14,15,18]. Under normal conditions, allothetic as well as idiothetic information is available in the MWM for orientation. As many studies have demonstrated, hippocampal lesion or temporal inactivation causes serious impairments in spatial memory based on allothetic stimuli [7,15,16,22]. Hence, reversible neural inactivation has revealed hippocampal participation in encoding and retrieval, as well as long-term storage of spatial information [6,22].

An interesting issue about the hippocampus concerns how cells responsible for encoding and retrieval are distributed. It was elegantly demonstrated that, when the dorsal part of both hippocampi lost more than 30% of tissue, retrieval of previously acquired information failed [5,17]. Nevertheless, several studies in rats have revealed that unilateral hippocampal inactivation [9,10] that presumably involves more than 30% of the tissue, or complete lesion of the hippocampus [18], does not interfere with encoding processes of new spatial information in the MWM or with its retrieval. It is important to point out that subjects were

* Corresponding author. Fax: +34 950015473.
E-mail address: jcimadev@ual.es (J.M. Cimadevilla).

0926-6410/$ - see front matter © 2005 Elsevier B.V. All rights reserved.
doi:10.1016/j.cogbrainres.2005.09.001

trained after the lesion or under the effect of TTX in all these studies.

However, the temporal dimensions of administration of a treatment into the hippocampus must be considered. Therefore, when inactivation occurred after animals had been trained under normal conditions (no treatment applied), tetrodotoxin (TTX) injection aimed unilaterally at the dorsal hippocampus has been reported to impair consolidation as well as to retrieve allothetic memories in an active place avoidance task [7].

It is not clear if spatial memories in the MWM can be disturbed by unilateral inactivation of the dorsal hippocampus if the treatment is applied after training. This could help to disclose the role of hippocampus in navigation and memory.

To explore this hypothesis, we trained rats in the MWM. In experiment 1, the dorsal hippocampus was inactivated each day immediately after the training in order to assess the role of this treatment in short-term consolidation processes. In experiment 2, in a different group of animals, after they had reached an asymptotic level in the MWM, unilateral injections into the dorsal hippocampus were applied 40 min before retrieval. Results showed that, in both experiments, unilateral hippocampal inactivation impaired performance.

2. Materials and methods

2.1. Animals

The experimental animals were 4-month-old male Wistar rats (315–430 g), obtained from the breeding colony of the University of Oviedo, Spain. The animals were housed in pairs in plastic cages with food and water available ad libitum in a room with artificial light (lights on: 08:00–20:00) and with a constant temperature of 20–21 °C. Nineteen animals were used for experiment 1 (control group $n = 9$ – one discarded after the histology – and experimental group $n = 10$) and ten for experiment 2 (two of them discarded after the histology). Subjects were distributed randomly into the groups.

The work was conducted in accordance with the European Communities Council Directive of 24 November 1986 (86/609/EEC) for the care and use of laboratory animals.

2.2. Surgery

The animals were unilaterally implanted with stainless-steel cannulae (0.5 mm inner diameter) aimed at the right dorsal hippocampus. The rats were mounted on a stereotaxic frame under ketamine (50 mg/kg i.p.) and xilazynum (20 mg/kg i.m.) anesthesia. The skull was exposed by a sagittal incision, and one hole for the guide cannula (1.5 mm in diameter) was made according to the atlas of Paxinos and Watson [20], 4 mm caudal from bregma and ±2.5 mm lateral from midline. The 10-mm-long guiding cannula was introduced dorso-ventrally 2 mm below the skull surface to a position above the dorsal hippocampus. Two holes were drilled to permit the positioning of anchoring bolts. The cannula and bolts were cemented to each other and the bone with dental acrylate. Finally, the skin was sutured around the implant, and the training started 1 week later.

2.3. Intrahippocampal injections

Control injections of 1 μl saline, or inactivating injections of 5 ng of TTX in 1 μl saline, were made through a guide cannula. The animal was gently restrained by hand, the guide cannula cleaned and a 30-gauge injection needle was inserted so that the needle protruded 2 mm from the guide into the dorsal hippocampus. The injection solution was delivered for 60 s using a Hamilton syringe connected to the injection needle with a short piece of polyethylene tubing. The needle was left in place for another 60 s before it was slowly removed. The spread of TTX in the neural tissue was estimated to cover a region approximately 1.4 mm in diameter [30].

2.4. Histology

Animals were anesthetised with thiopental (100 mg/kg) and perfused transcardially with saline followed by formalin (10%) for 20 min. After removing the brains, they were embedded in paraffin, and 30 μm histological slices were extracted, stained with cresyl violet and verified to correspond to the dorsal hippocampus.

2.5. Apparatus

The circular pool (150 cm diameter × 40 cm deep) made of fibre glass painted black was placed on a metal platform 35 cm high, in the centre of a room (4 m × 4 m) with several landmarks (in the North, a white wall on which eight patterned plates hung, in the South, a shelf and a map, in the West, a computer and a table and in the East, a door and a covered window). Two lamps located near the floor in the South and West indirectly illuminated the whole room. A black round platform (10 cm in diameter) occupied the centre of one of the four virtual quadrants in which the pool was divided conceptually to assess the animals' behaviour. This was located in the Northern quadrant, 30 cm from the wall of the pool. The water level was kept 1.5 cm above the platform, and the temperature of the water was maintained between 22 °C and 23 °C. The experiment was recorded using a tracking system (Noldus, Netherlands) that provided information about the tracks displayed by the animals. The latency to the platform, the total distance covered, the mean swimming speed, the time spent in each virtual quadrant of the pool as well as the time the animals swam in a 15 cm ring at the periphery were measured.