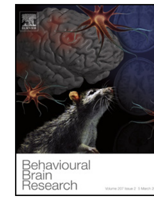
Behavioural Brain Research 267 (2014) 126–132

Contents lists available at ScienceDirect

Behavioural Brain Research

journal homepage: www.elsevier.com/locate/bbr



Research report

Can rats solve the active place avoidance task without the room-bound cues?

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HIGHLIGHTS

- Rats are able to solve the active place avoidance task without the room-bound cues.
- Their performance without the room-bound cues is not as efficient as with them.
- Pretraining with the room-bound cues is needed for learning the task without them.

ARTICLE INFO

Article history:
Received 11 July 2013
Received in revised form 7 March 2014
Accepted 10 March 2014
Available online 25 March 2014

Keywords:
Spatial behavior
Avoidance behavior
Cognitive coordination
Idiothetic navigation

ABSTRACT

The active place avoidance task is used in the research of spatial cognition. Rats are trained on a rotating arena to avoid an aversive stimulus delivered in a part of the room while being transported toward it by the arena rotation. The task tests the ability of rats to navigate with respect to distal cues in the room and to ignore confusing cues on the arena. The demand for cue segregation makes the task suitable for studying neural mechanisms responsible for cognitive coordination. An incidental observation made in our laboratory implied that overtrained rats may be able to solve the task without the room-bound cues. The aim of this study was to test this observation. The room-bound cues were hidden by switching off the lights. Rats trained only in darkness did not learn the task at all. Rats that were initially pre-trained in light performed considerably better. In a few exceptional dark sessions they even reached the level of performance observed in light. The rats needed the aversive stimuli to keep off the to-be-avoided sector. Without them, they continued their behavior, but with no spatial relationship to the to-be-avoided sector. We conclude that rats are able to solve the place avoidance task without the room-bound cues, but not as efficiently as in their presence.

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1. Introduction

The active place avoidance task was designed more than 10 years ago [1] to study spatial cognition in rodents. Today, it is recognized as a well-established cognitive test in behavioral, lesional, electrophysiological and pharmacological studies, e.g. [2–12].

The active place avoidance task is carried out on a circular arena, whose rotation continuously dissociates orientation cues into arena-bound and room-bound subsets. Each of the subsets of cues forms a reference frame in which rats are able to navigate and form independent spatial memories [13].

In the standard version of the active place avoidance task, a rat is supposed to avoid a 60° wide sector defined in the room frame. If the rat enters the sector, then an aversive stimulus, a mild electrical current, is delivered. Since the to-be-avoided sector is defined solely with respect to the room-bound cues, the arena-bound cues seem to be irrelevant for the solution of the task. However, Wesierska et al. [4] showed that rats with unilateral hippocampal lesions are only able to learn the task when the arena-bound cues are covered by shallow water. This experiment confirmed that rats process the room-bound and arena-bound cues simultaneously. It is now accepted that the task requires the ability to segregate orientation cues into distinct room-bound and arena-bound subsets. The rats must use the room-bound cues for navigation and ignore the arena-bound cues to solve the task successfully [4,5]. The demand for segregation of the continuously dissociated cues makes the active place avoidance task unique among spatial tasks.

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http://dx.doi.org/10.1016/j.bbr.2014.03.028
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The active place avoidance task has been adapted for mice [14], and, for humans in real maze [15] and in virtual reality [16].

Similar to the active place avoidance task, some tasks designed for humans, such as the Wisconsin card sorting test or the Stroop task, also require segregation of cues into subsets, as well as evaluation of their relevance for the task solution. Patients with various neuropsychiatric disorders are commonly impaired in these tasks [17]. For this reason, the active place avoidance task has been used in the research of human neuropsychiatric disorders in animal models, e.g. [8,12,18–20]. The main conclusions were based on the assumption that rats capable to solve the task are also capable to segregate the cues into coherent subsets.

An incidental observation made in our laboratory, however, suggested, that overtrained rats might be able to solve the task in darkness, i.e. without the room-bound cues. Such rats would then not need to solve the conflict between the two groups of cues, possibly solving the task without cue segregation. If this were true, then alternative strategies would need to be considered for interpretation of the results.

The purpose of the present study was to confirm or rule out the existence of alternative strategies that do not involve the room-bound cues. Rats were trained to solve the active place avoidance task either in light or in darkness; i.e. with or without the room-bound cues, respectively. Some of the rats trained in darkness had been pretrained in light. Test sessions were carried out after each block of five training sessions. The purpose of the test sessions was to study the avoidance strategies employed by the rats and to rule out the presence of any remaining uncontrolled room-bound cues in darkness. The results showed that some rats are able to solve the active place avoidance task without the room-bound cues relatively well, but only if they had been pretrained in their presence.

2. Materials and methods

2.1. Subjects

The subjects ($n = 22$) were naive male Long-Evans rats obtained from the breeding colony of the Institute of Physiology, Academy of Sciences of the Czech Republic. The rats were 3–5 months old and weighed 350–540 g at the beginning of the experiment. They were housed in a temperature-controlled environment at 21 °C. Food and water were freely available. All manipulations were performed during the light phase of a 12/12 h light/dark cycle. The procedures were in accordance with guidelines of the Animal Protection Act of the Czech Republic, National Institutes of Health (NIH) and with the directive of the European Communities Council (2010/63/EC).

2.2. Apparatus

The active place avoidance apparatus (Fig. 1A) was a modification of that used originally by Cimadevilla and his colleagues [1]. It consisted of a rotatable metallic circular arena (82 cm in diameter), enclosed with a 30 cm high transparent perspex wall. The perspex wall did not allow the animal to escape from the arena, while it provided good visibility of the room-bound environment. The apparatus was elevated 70 cm above the floor of a 4 m × 5 m room containing numerous room-bound cues.

The animal was tracked via an overhead TV camera registering an infrared light-emitting diode (LED) that the rat carried on its back. The TV camera was connected to a computerized tracking system (iTrack, Bio-Signal Group, DE, USA) located in an adjacent room. The system recorded the rat's position in the room as well as the angular displacement of the rotating arena every 40 ms, and it calculated the position of the rat on the arena. Based on the actual room position, the computer delivered electrical stimuli to the rat via a shock-delivering cable attached to the animal.

2.3. Habituation

The animals were handled by the experimenter for several minutes every day during 1 week. On the last day of handling, the rats' skin was pierced between the shoulders by a hypodermic needle. The sharp end of the needle was cut off and a small loop was made to prevent the needle from slipping out. The loop served as an anchor for attaching an alligator clip, which was connected to the shock-delivering cable. Several hours after the hypodermic needle was implanted, a 10-min habituation session on the arena was carried out. The animals were allowed to freely explore the rotating arena without receiving aversive stimuli. The behavioral training started 1–3 days after the habituation session.

2.4. Behavioral training

The rats were trained on the rotating arena to avoid a directly imperceptible 60° wide sector defined on the arena surface (Fig. 1A). The rotation speed had been set to 1 revolution per minute (rpm). The sector did not rotate with the arena, but was stable in the room reference frame. The location of the sector remained constant throughout the whole experiment. Whenever the rat entered the to-be-avoided sector for more than 0.2 s, the tracking system delivered an aversive stimulus and counted an entrance. The aversive stimulus was a mild foot-shock (50 Hz, 0.5 s). The exact value of the shock current was determined for each rat individually (between 0.4 and 0.8 mA) in order to elicit a rapid escape reaction but to prevent freezing. If the rat did not leave the sector after the first aversive stimulus, additional stimuli were given every 1.5 s, but no more entrances were counted. A new entrance into the to-be-avoided sector was counted if the animal had left the sector and returned back after a longer period than 1.5 s.

In total, 22 rats were trained in the avoidance task. Some rats were trained exclusively in light ($n = 8$), some in darkness ($n = 4$) and some in both conditions ($n = 10$; seven out of these 10 rats were initially trained in light and three in darkness). The training in each condition proceeded for either 15 or 25 sessions, except for four rats that had been trained in dim light for only 10 sessions before they were trained in darkness for another 15 sessions. These rats were originally intended for training only in darkness, however, some light from the adjacent corridor and from the ventilation system penetrated into the room. When it became apparent from the performance of the rats in test sessions (see below) that they were probably still able to use the room-bound cues under these light conditions, we identified these sources of light and removed them. Since the conditions were not completely controlled, we did not analyze them. The training sessions lasted 20 min. They were labeled by a number denoting the order of the session followed either by the letter L or D, indicating the light and dark conditions, respectively.

2.5. Test sessions

Test sessions were used to verify the absence of any remaining room-bound cues in darkness and to test whether the rats that had mastered the task in light could solve it in darkness. They were carried out after each block of five training sessions in all rats. The test sessions consisted of three to four phases and are described as follows: Phase 1 (10 min): the aversive stimuli were switched on and the arena rotated at the standard speed of 1 rpm. Phase 2 (10 min): the aversive stimuli were switched off. Phase 3 (5 or 10 min): the aversive stimuli remained switched off and the speed of the arena rotation was altered. It was decreased to 70 s

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