Behavioural Brain Research 216 (2011) 525–530

Contents lists available at ScienceDirect

Behavioural Brain Research

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

Research report

The relationships between trait anxiety, place recognition memory, and learning strategy

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ARTICLE INFO

Article history:
Received 17 June 2010
Received in revised form 17 August 2010
Accepted 23 August 2010
Available online 27 August 2010

Keywords:
Learning strategy
Spatial memory
Anxiety
Y-maze
Visual platform water maze

ABSTRACT

Rodents learn to navigate mazes using various strategies that are governed by specific regions of the brain. The type of strategy used when learning to navigate a spatial environment is moderated by a number of factors including emotional states. Heightened anxiety states, induced by exposure to stressors or administration of anxiogenic agents, have been found to bias male rats toward the use of a striatum-based *stimulus–response* strategy rather than a hippocampus-based *place* strategy. However, no study has yet examined the relationship between natural anxiety levels, or trait anxiety, and the type of learning strategy used by rats on a dual-solution task. In the current experiment, levels of inherent anxiety were measured in an open field and compared to performance on two separate cognitive tasks, a Y-maze task that assessed place recognition memory, and a visible platform water maze task that assessed learning strategy. Results indicated that place recognition memory on the Y-maze correlated with the use of place learning strategy on the water maze. Furthermore, lower levels of trait anxiety correlated positively with better place recognition memory and with the preferred use of place learning strategy. Therefore, competency in place memory and bias in place strategy are linked to the levels of inherent anxiety in male rats.

1. Introduction

The hippocampus, striatum, and amygdala play key roles in the acquisition of information, its subsequent retrieval, and the resulting behavioral responses [1–5]. However, differences in the way that information is learned result from complex interactions of these memory systems in the brain. The hippocampus dominates when the demands of a task require the use of extra-maze cues to navigate to a goal, and subsequently results in the use of a *place learning* strategy [2,6,7]. Alternatively, when the task requires navigation by visual cues proximal to a goal or reflexive motor-based responses, such as always turning right to reach a goal, the striatum has greater influence, and thus results in the use of a *stimulus–response* learning strategy [2,6,7]. The amygdala has a dual role in multiple memory systems. On one hand, the amygdala modulates emotion-dependent learning and memory processes independent of the striatum and the hippocampus [2]. Additionally, the amygdala can act in concert with the hippocampus and striatum to influence learning strategy and bias rats away from a place strategy and toward a response strategy [4].

Use of a hippocampus-dependent place strategy is more complex, but also more flexible, than a striatum-dependent stimulus–response strategy and thus increases navigational efficiency when the demands of the task are changing frequently. Navigation to an escape platform in a water maze using a place strategy requires rats to develop a spatial relationship between extra-maze cues and the escape platform. Therefore, rats ignore intra-maze cues within the water tank, such as a marked or a visible platform, and implement an allocentric search strategy by assessing the environment outside the maze. Consequently, when the escape platform is visibly marked and relocated to a new quadrant following training, rats that have adopted a place strategy return to the previous quadrant, and exhibit prolonged escape latencies and increased path lengths [8].

A *stimulus–response* strategy uses one of two approaches based on the demands of the task. To navigate toward a goal in a maze without regard to intra-maze or extra-maze cues, rats can be guided by proprioceptive cues and adopt a reflexive motor response, learning to turn in the same direction on each trial [5,7]. Alternatively, if reaching the goal is dependent on salient intra-maze cues, such as a visible platform or a flag that signifies the location of the goal, the use of a stimulus–response strategy is also appropriate [6,8]. Whether using an egocentric 'turn this way'

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doi:10.1016/j.bbr.2010.08.028

approach or navigating via an intra-maze cue, the striatum dominates when a stimulus–response strategy is used, resulting in a behavioral response that is distinctly different from rats using a hippocampus-dependent place strategy. A stimulus–response strategy may increase efficiency on some tasks, however it is more rigid and less adaptable to changing conditions [8]. The efficiency of adopting either a place or a stimulus–response strategy is therefore task-dependent, but can be influenced by other components of multiple memory systems [4,9].

The amygdala appears to moderate the preferential use of a place or stimulus–response strategy [1,4,5,10–13]. Use of place or stimulus–response learning is modulated by changes in emotional arousal such as increases in anxiety or stress [1,5,11,14,15]. On a dual-solution water maze task, when anxiety was raised by injection of an anxiogenic drug either peripherally, or directly into the amygdala, male rats were more likely to adopt a response strategy [1]. Therefore, increased state anxiety biases male rats toward the use of the striatum and away from the use of the hippocampus on navigational tasks that can be solved by either strategy.

Interestingly, male rats with low levels of inherent, or trait, anxiety displayed faster acquisition and better retention on a standard water maze task that required a place strategy solution compared to rats high in anxiety [16–20]. What remains to be investigated is whether the preferential use of one learning strategy is associated with inherent trait anxiety in the same way increased state anxiety biases rats toward a stimulus–response strategy over a place strategy. To examine the relationship between learning strategies and naturally occurring trait anxiety, the current study assessed learning and memory on two cognitive tasks and correlated performance with anxiety levels measured in the open field. We hypothesized that rats with lower natural anxiety, as measured by the percentage of time spent in the center of the open field, would show better place recognition on a non-aversive test of spatial memory (Y-maze), and would be biased toward the use of a place strategy as indicated by increased escape latencies and path lengths on a test of learning strategy (visible platform water maze).

2. Materials and methods

2.1. Animals

All procedures were approved by the Tulane University Institutional Animal Care and Use Committee in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals (1996). Subjects were 14 unmanipulated male Long-Evans rats obtained from Harlan, Inc. (Indianapolis, IN) at 55 days of age. Rats were pair-housed with free access to food and water under a 12:12 light-dark cycle (lights on at 07:00). Rats were allowed 5 days of habituation to housing conditions and were handled daily during this time to acclimate to experimenters. For all testing procedures, rats were brought into the testing room 30 min prior to testing and left undisturbed to habituate to the change in environment.

2.2. Y-maze

At 60–65 days of age, spatial memory was assessed using a Y-maze constructed of grey Plexiglas consisting of three identical arms (51 cm × 10 cm × 30 cm; Stoelting ANY-maze, Wood Dale, IL) and surrounded by extra-maze cues of varying shapes and sizes. During the 15-min information trial, rats could explore the start arm and a second arm, but access to the third arm was blocked by a plastic partition [21–27]. The start, novel, and familiar arms were randomly assigned and counterbalanced across rats to account for preference of spatial cues [21]. Following the initial information trial, rats were returned to their home cages for a delay interval of 4 h. After the delay, each rat re-entered the Y-maze in the same start arm and was allowed to explore all three arms during a 5-min retention trial. Retention trials were video-recorded for later scoring where entry into an arm was defined as all four paws crossing into the arm proper. The maze was cleaned thoroughly with 70% ethanol and air-dried after each trial to remove olfactory cues. Spatial memory was indicated by increased entries into the novel arm during the retention trial [21–27].

2.3. Visible platform water maze

One week after Y-maze testing, a modified version of the water maze task was used to examine learning strategy [8]. A white circular pool, 180 cm in diameter, was filled to a depth of 26 cm with water made opaque by the addition of non-toxic white paint and surrounded by extra-maze cues of varying shapes and sizes. The temperature of the water was maintained between 22 and 24 °C. A visible black platform, 9.5 cm in diameter, projected 3 cm above the water surface and was located 30 cm from the wall of the pool. One day prior to testing, rats were placed in the water maze without a platform present for a 1-min habituation swim. Twenty-four hours later, rats were trained on the task.

During training trials, the escape platform was located in the southwest quadrant of the pool. Eight training trials were conducted with the rat entering from each

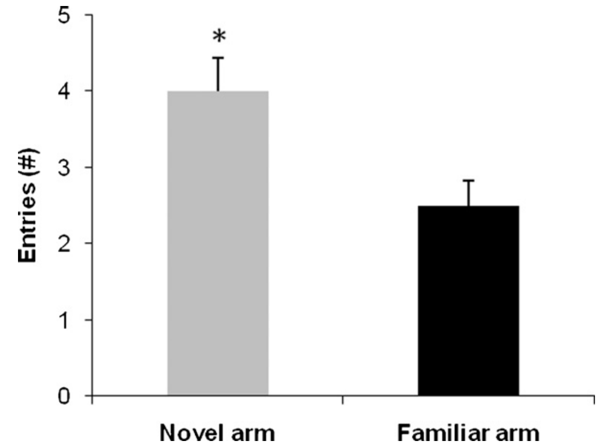

Fig. 1. A paired t-test indicated that male rats displayed significantly more entries into the novel arm of the Y-maze compared to entries into the familiar arm during a retention trial. n=13, *p<.05.

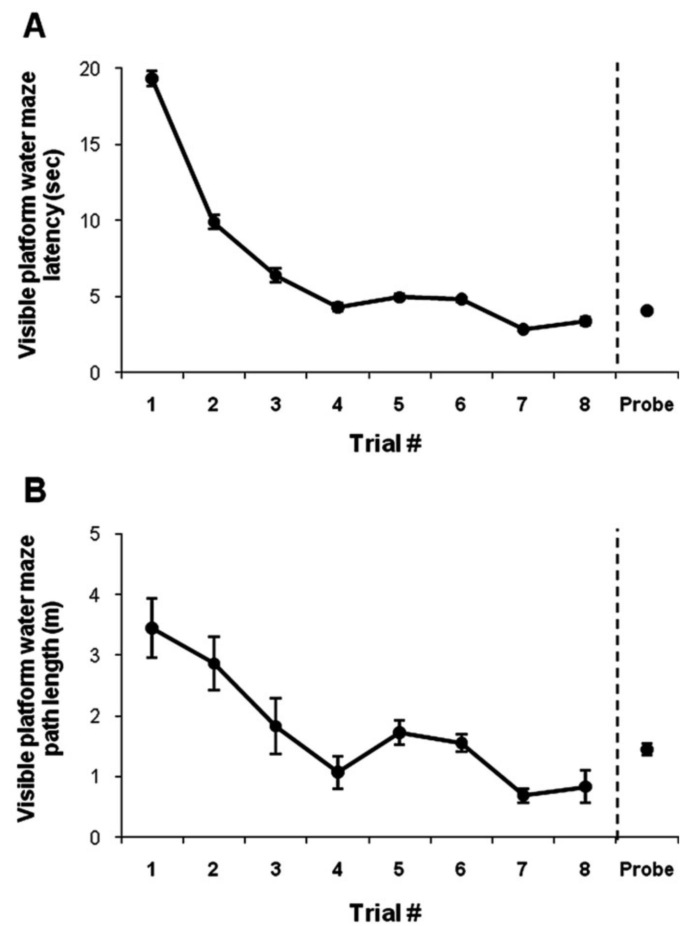

Fig. 2. Repeated measures analyses of variance indicated that male rats displayed significant decreases in (A) escape latency and (B) path length over 8 training trails on the visible platform water maze task, with performance maintained on probe trials when the visible platform was relocated to a different quadrant. n=14.