Place Memory Is Intact in Rats With Perirhinal Cortex Lesions

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Two experiments compared the effects of bilateral lesions of the hippocampal formation (HPC) or perirhinal cortex (PRh) on rats’ performance of an allocentric spatial working memory task—delayed matching-to-place (DMTP) in a water maze. DMTP trials consisted of paired swims, and the hidden platform was moved to a new location on each trial. Performance was assessed with intervals between the first and second swim (i.e., retention delays) of 4, 30, 120, and 300 s. The rats received extensive presurgery training in Experiment 1 and no presurgery training in Experiment 2. In both experiments, rats with HPC lesions displayed DMTP deficits at all delays, taking longer and swimming farther to find the platform on the second swims than did sham-operated controls. By contrast, rats with PRh lesions displayed normal DMTP acquisition and performance. The results suggest that, unlike the functions of HPC, those of PRh are not critical for allocentric spatial working memory.

Most models or theories of how memory functions are organized within the temporal lobes assume that interactions between the hippocampal formation (hippocampus proper, dentate gyrus, and subicular complex) and the rhinal cortex (perirhinal and entorhinal cortices) are critical for normal memory abilities. The anatomical findings are consistent with this view because there are extensive reciprocal connections between these areas and the rhinal cortex is a major site of convergence for neocortical inputs to the hippocampal formation (Burwell, Witter, & Amaral, 1995; Deacon, Eichenbaum, Rosenberg, & Eckmann, 1983; Witter, Groenewegen, Lopes da Silva, & Lohman, 1989).

There is much debate about how to characterize the representational properties of various temporal lobe structures and the nature of their interactions. Some propose that the hippocampal formation is important for memory of only certain kinds of information, such as places (Jarrard, 1993; M. Morris, Garrud, Rawlins, & O’Keefe, 1982; O’Keefe & Nadel, 1978) or contexts (Hirsh, 1974; Jarrard, 1993), whereas others suggest a more general role in representing the relations among individual items in memory (Eichenbaum, Otto, & Cohen, 1994). It has been proposed that the rhinal cortex has a special role in recognition memory (Mishkin & Murray, 1994; Murray, 1996), or memory for objects (Eacott, Gaffan, & Murray, 1994; Mumby & Pinel, 1994; Murray, 1996), or intermediate-term representation of individual (i.e., nonrelational) items involving potentially any type of stimulus (Eichenbaum et al., 1994).

Functional dissociations have been observed after selective lesions of the hippocampal formation or rhinal cortex in rats and monkeys (reviewed in Eichenbaum et al., 1994; Murray, 1996). One well-documented dissociation involves impairments of object-recognition memory after perirhinal cortex lesions (rats: Eichenbaum, Neave, & Aggleton, 1996; Mumby & Pinel, 1994; monkeys: Gaffan & Murray, 1992; Meunier, Bachevalier, Mishkin, & Murray, 1993; Zola-Morgan, Squire, Amaral, & Suzuki, 1989) but not after lesions restricted to the hippocampal formation (rats: Mumby, Wood, & Pinel, 1992; Rothblat & Kromer, 1991; monkeys: O’Boyle, Murray, & Mishkin, 1993).

The present experiment assessed the effects of perirhinal cortex lesions on a discrete-trial allocentric spatial memory task, which is widely believed to require the functions of the hippocampal formation (see review by Barnes, 1988; O’Keefe & Nadel, 1978). The perirhinal cortex receives input from the temporal, parietal, occipital, cingulate, and insular cortices and shares reciprocal connections with the subiculum and the CA1 subfield of the hippocampus (Deacon et al., 1983; Kosel, Van Hoesen, & Rosene, 1983; Suzuki & Amaral, 1990) and with the adjacent entorhinal cortex, which in turn provides a major input to the hippocampal formation through the perforant path (Insausti, Amaral, & Cowan, 1987; Witter et al., 1989). The perirhinal cortex is thus considered an important link in the pathway through which highly processed information from polymodal association cortices reaches the hippocampal formation.

It is much less clear which aspects of hippocampal function are dependent upon input from the perirhinal cortex. Given the strong projections linking these two structures, it is logical to predict that spatial memory abilities would be impaired after perirhinal cortex lesions. However, the findings from previous studies that examined the effects of perirhinal cortex damage on spatial memory are inconsistent and therefore inconclusive. Some report that performance of place-memory tasks in a water maze is normal after perirhinal cortex lesions (e.g., Kolb, Buhrmann, McDonald, & Sutherland, 1994; Wiig & Bilkey, 1994b), whereas others report impairments (Nagahara, Otto, & Gallagher, 1995; Wiig & Bilkey, 1994a). In a more recent
study, rats with excitotoxic lesions of perirhinal cortex performed normally on a T-maze spatial-alternation task and on spatial (position) discrimination and nonmatching-to-position tasks in an operant chamber (Ennaceur et al., 1996).

In the two experiments of the present study, rats with bilateral lesions of either the hippocampal formation or perirhinal cortex were tested on a delayed matching-to-place (DMTP) task in a water maze. The DMTP task required rats to find an escape platform in a new target location on each trial and to remember this location across a variable retention delay. The rats could not see the platform, so they had to learn its location relative to extramaze cues. The use of a different hidden-platform location on each DMTP trial and the employment of a range of retention delays emphasized the importance of intermediate-term storage of the platform location on individual trials. The DMTP task thus required allocentric spatial working memory.

Previous studies indicate that presurgery training can attenuate deficits on memory tasks after temporal-lobe lesions (see Mishkin & Murray, 1994). In the present study, therefore, the rats received presurgery DMTP training in Experiment 1 and no presurgery training in Experiment 2.

**Experiment 1**

This experiment compared the effects of bilateral lesions of the hippocampal formation or perirhinal cortex on rats’ DMTP performance. In a previous study, rats with lesions of the entorhinal cortex that also included parts of perirhinal cortex displayed delay-dependent deficits on a DMTP task similar to the one used in the present experiment (Nagahara et al., 1995). The lesions in the Nagahara et al. study included much of the entorhinal cortex, and therefore it is not clear whether perirhinal cortex damage contributed to the deficits. Moreover, the rats in the Nagahara et al. study and in another study that reported impaired spatial memory after perirhinal cortex lesions (Wiig & Bilkey, 1994a) did not receive any training in the water maze before surgery. In the present experiment, we gave our rats extensive DMTP training to facilitate the interpretation of the results by ruling out any deficits caused by impaired learning of the procedural aspects of the task.

**Method**

**Subjects**

Nineteen adult male Long-Evans hooded rats served as subjects. They weighed between 300 and 350 g at the start of the experiment and were housed individually in opaque plastic cages in a colony room that was maintained at 21°C with a 12-hr light–dark cycle (lights on at 8 a.m.). The rats had continuous access to water in their home cages and were fed approximately 25 g of standard laboratory rat food once per day.

**Apparatus**

The DMTP task was conducted in a water maze (R. G. M. Morris, 1981), 137 cm in diameter and 46 cm high, and filled with water (23°C) to a depth of approximately 30 cm. The water was made opaque by adding instant skim milk powder. A movable Plexiglas platform (10 cm × 10 cm × 28 cm) was hidden approximately 2 cm below the surface of the water. The rats could not see the platform, but several extramaze cues (e.g., posters, shelves, a computer, etc.) were visible from within the pool, and the rats could learn the location of the platform relative to these distal cues. Swim paths and latencies were recorded using a VP118 Super Tracker with HVSwater software (HVS Image Ltd., Hampton, United Kingdom), and these raw data were stored on an IBM-compatible personal computer for later analysis.

**Procedure**

All training, testing, and surgical procedures were conducted during the light phase. Rats were transported to the testing room in groups of six or seven and were singly housed there on top of a cart inside covered, wire mesh cages. A black curtain separated the cart from the rest of the room so that the rats could not see the extramaze cues that were visible from within the pool.

The DMTP task. Four equally spaced points along the perimeter of the pool were designated as cardinal compass points (north, south, east, and west) and served as release points for placing the rats into the pool. Each DMTP trial consisted of paired swims. During the first swim of a trial (i.e., the acquisition swim), the rat was placed into the water, facing the pool wall, at one of the release points and allowed to search for the hidden platform. After finding the platform and climbing onto it, the rat was allowed to remain there for 10 s before the experimenter removed it. If a rat failed to find the platform and climb onto it within 60 s, the experimenter guided it there by hand. During the second swim of a trial (i.e., the retention test), the rat was again placed into the pool at the same release point and allowed to search for the platform, which remained in the same location. The main dependent measures used to gauge retention of the hidden platform location on each trial were the escape latency and the swim-path length on both the first and second swims.

The platform was moved to a new location on successive trials, within and across sessions, cycling repeatedly through the sequence of 10 locations shown in Figure 1. This sequence included a similar number of platform locations near the edge of the pool, near the center of a quadrant, and near the center of the pool, which were chosen to prevent the rats from adopting successful strategies that obviated the need to use allocentric spatial relations, such as preferentially searching in a particular area of the pool, or along the pool wall, or at some fixed distance from the wall. Each release point was used approximately the same number of times for each of the 10 platform locations.

There were four DMTP trials per session, with an intertrial interval of approximately 12 min. Rats spent the intertrial intervals in the stainless steel, wire mesh cages and the retention delays in an opaque plastic, shoebox cage atop the transport cart. It was not possible to return the shoebox cage to the cart during trials that used a 4-s retention delay. On those trials, the rats were removed from the platform and carried to the release point inside the shoebox cage.

Preliminary training. The rats were first trained on a conventional water maze task with a fixed hidden platform location. On each trial, the rat was placed into the pool, facing the wall, at one of the release points. A pseudorandom schedule determined the release point for each trial, with the constraint that each of the four release points was used once before the next random sequence of four release points was determined. The rat was allowed a maximum of 60 s to find the hidden platform, which was located in the center of the northeast quadrant on every trial. If a rat failed to find the platform and climb onto it within 60 s, the experimenter
Figure 1. The sequence of 10 platform locations that were used for the delayed matching-to-place task. The platform was moved to the next location in the sequence on successive trials within and between sessions. N = north; E = east; S = south; W = west.

guided it there by hand. Each rat received eight trials per day for 4 days, with an intertrial interval of approximately 6 min. A moving-platform procedure was then gradually introduced: The platform was moved to a new location for the fifth session and to another new location for the sixth session. On the seventh and eighth sessions, the platform was in a new location for the first four trials, and then moved to a new, previously unused, location for the remaining four trials.

Presurgery DMTP training. After completion of preliminary training, the rats received 10 DMTP sessions with a 4-s retention delay between the first and second swim of each trial. The delay was subsequently increased to 30, 120, and 300 s, and each rat received four sessions (i.e., 16 trials) at each delay before moving on to the next longest delay.

Retention curves were obtained during a final phase of presurgery training. Each rat received 15 mixed-delay sessions, which consisted of four trials, one at each of the four delays (i.e., 4, 30, 120, and 300 s). The order of delays used within a mixed-delay session was randomly determined.

Surgery. Rats received either perirhinal cortex lesions (Group PRh, n = 7), hippocampal lesions (Group HPC, n = 7), or sham surgery (Group SHAM, n = 5). Surgery was performed under pentobarbital anesthesia (65 mg/kg), between 24 and 72 hr after a rat's last presurgery mixed-delay session. Hippocampal lesions were made with intrahippocampal microinjections of ibotenic acid (5 μg/μl; Sigma Chemical, St. Louis, MO) in 0.1 M phosphate-buffered saline (PBS, pH 7.4). The rats were placed in a stereotaxic apparatus (Kopf Instruments, Tujunga, CA), their scalps were incised and retracted, and small holes (1.5 mm in diameter) were drilled in their skulls to enable bilateral placement of 30-gauge cannulas at the stereotaxic coordinates shown in Table 1. A 10-μl Hamilton syringe was mounted in an infusion pump (KD Scientific, Boston, MA) and connected to the cannulas with polyethylene tubing. Ibotenic acid was infused at each site at a flow rate of 0.1 μl/min over a period of 2.5 min for a total injection volume of 0.25 μl per site. The cannulas were left in place for 2 min after each infusion to allow diffusion of the neurotoxin. After surgery, the incision was closed with stainless steel wound clips, and 1 mg diazepam (Hoffmann-La Roche, Mississauga, Ontario, Canada) was administered intramuscularly as a prophylaxis against seizures.

Table 1

<p>| Cannula Coordinates Relative to Bregma (in Millimeters) for Neurotoxic Lesions of the Hippocampal Formation |
|-------------------------------------------------|---------------------------------|-------------------|</p>
<table>
<thead>
<tr>
<th>Anteroposterior</th>
<th>Mediolateral</th>
<th>Dorsoventral</th>
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<tbody>
<tr>
<td>−3.1</td>
<td>±1.0</td>
<td>3.6</td>
</tr>
<tr>
<td>−3.1</td>
<td>±2.0</td>
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<td>±5.2</td>
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<td>−5.0</td>
<td>±5.2</td>
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<td>−5.8</td>
<td>±4.4</td>
<td>4.4</td>
</tr>
<tr>
<td>−5.8</td>
<td>±5.1</td>
<td>6.2</td>
</tr>
<tr>
<td>−5.8</td>
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<td>7.5</td>
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</table>
Rats in Group SHAM received the same general treatment as those in Group HPC, with the exceptions being that no ibotenic acid was infused, the cannulas were lowered to a depth of only 2 mm below bregma, thus sparing the dorsal hippocampus, and no postsurgery diazepam was administered.

In preparation for making the perirhhinal cortex lesions, a coronal scalp incision was made and the skull overlying the perirhhinal cortex was exposed. A hole was cut into the skull with a dental drill, the dura overlying the rhinal fissure was incised, and portions of the perirhhinal cortex and lateral entorhinal cortex were aspirated with a vacuum pump and a glass Pasteur pipette. The cavity was filled with Gelfoam (Upjohn, Don Mills, Ontario, Canada), and the incision was closed with wound clips.

All rats received antibiotic following surgery (penicillin G, 15,000 units im; G. C. Hanford, Syracuse, NY). One rat died following hippocampal surgery, leaving 6 rats in Group HPC. The remaining rats were allowed to recover for 10–14 days before behavioral testing recommenced.

Postsurgery DMTP testing. After recovery, all rats were retrained on the DMTP task with a 4-s retention delay until their second-swim escape latencies reached a stable level. The criterion of stability was that the mean latency per session could not differ by more than 5 s over any three consecutive sessions, with the additional constraint that each rat had to receive a minimum of four sessions. After rats reached the criterion of stable postsurgery performance with a 4-s delay, they received 15 mixed-delay sessions, using procedures that were identical to those of the presurgery mixed-delay sessions.

Visible-platform test. Within 48 hr of the final mixed-delay session, a visible-platform test was administered during a single session. The top 3 cm of the platform was wrapped with black tape and the platform was raised so that it extended approximately 2 cm above the surface of the water. The remaining procedures were identical to the DMTP task with a 4-s delay: There were 4 paired swims, and the platform was moved to a new location for every second swim.

Histology. Upon completion of behavioral testing, the rats were given an overdose of sodium pentobarbital and were perfused transcardially with saline, followed by 10% neutral buffered formalin. The brains were removed and stored in 10% formalin solution before being frozen and sectioned along the coronal plane at a thickness of 30 μm. Every 10th section through the lesioned area was mounted on a glass slide and stained with cresyl violet.

Statistical analyses. Data from the preliminary training trials, from the presurgery DMTP acquisition sessions, and from the postsurgery DMTP reacquisition sessions were analyzed with repeated measures analyses of variance (ANOVA), with group (lesion type) as a between-subjects factor and session as a repeated measures factor. Data from the mixed-delay sessions were analyzed with repeated measures ANOVA, with group as a between-subjects factor and time-of-testing (presurgery vs. postsurgery) and delay as repeated measures factors.

Results

Histological Findings

Hippocampal lesions. Figure 2 shows the location and extent of the largest and smallest of the hippocampal lesions. The ibotenic acid injections produced extensive loss of cells in all principle subfields of the hippocampus and dentate gyrus. The fimbria and fornix were largely spared in each rat, and in each rat there was also some minor sparing of dentate granule cells and CA1 pyramidal neurons in the most temporal portions of the hippocampal formation. There was some minor sparing of neurons in the medial aspect of the most anterior sections of the dentate gyrus in 2 rats. The extent of damage to the subiculum was variable, but there was some bilateral loss of subicular cells in all rats, which was incomplete in every case. There was no evidence of damage to the thalamus or rhinal cortex in any of the rats with hippocampal lesions. There was, however, some thinning of parietal cortex near the sites where the injection cannulae were inserted.

Rhinal cortex lesions. Most of the rats in Group PRh sustained rhinal cortex damage that was roughly symmetrical in the two hemispheres (see Figure 3, top panel). Damage to areas outside of the rhinal cortex, however, was more variable. In all rats, the area that sustained the most bilateral damage was perirhinal cortex. All rats also sustained some bilateral damage to lateral entorhinal cortex, but most of this region was spared in every case, and the medial entorhinal cortex was completely spared in both hemispheres of all rats. There was slight unilateral damage to ventro-posterior portions of temporal association cortex (area Te2; Burwell et al., 1995) in 3 PRh rats.

Figure 3 shows the depth of the rhinal cortex lesions. A portion of the ventral subiculum was damaged unilaterally in 2 PRh rats and bilaterally in 2 others. Slight damage occurred in the most temporal portion of the CA1 field, which was unilateral in 4 PRh rats and bilateral in 2 others.

Behavioral Findings

Presurgery training. The three groups were well matched for presurgery performance. There were no significant differences among the groups in their escape latencies on the preliminary training trials, F(2, 15) < 1. Mean latency for each group reached an asymptote during the second session with a fixed platform location. Each group displayed an increase in mean latency each time the platform was moved.

Figure 2. Location and extent of the largest (grey) and smallest (black) hippocampal formation lesions in Experiment 1 at three coronal planes (distance from bregma shown in millimeters). Coronal drawings based on the rat brain atlas of Paxinos and Watson (1986).
Figure 3. The top panel shows the approximate boundaries of perirhinal, postrhinal, and entorhinal cortex on lateral surface of rat brain, according to Burwell et al. (1995), and the location and extent of the largest (grey) and smallest (black) perirhinal cortex lesions in the present experiment. The bottom panel shows the depth of largest (grey) and smallest (black) perirhinal cortex lesions at three coronal planes. Coronal drawings are based on the rat brain atlas of Paxinos and Watson (1986). Arrowheads indicate the approximate dorsal and ventral boundaries of the perirhinal cortex, based on Burwell et al.

to a new location during preliminary training. This increase was always temporary, lasting from one to three trials before returning to the previous asymptote (data not shown).

The three groups also mastered the DMTP task with a 4-s delay at similar rates. The mean second-swim escape latencies reached an asymptote in each group between the fourth and sixth sessions (data not shown). The effects of increasing the retention delay to 30, 120, and 300 s were
Figure 4. The mean second-swim escape latencies (±SEM) on the first five postoperative sessions of delayed matching-to-place reacquisition in Experiment 1. HPC = hippocampal formation lesions; PRh = perirhinal cortex lesions; SHAM = sham surgery.

negligible, as none of these increases caused a significant change in mean second-swim latency in any of the groups. There were no significant differences among the groups in second-swim latencies on the presurgery mixed-delay sessions, F(2, 15) < 1. The main effect of delay duration was also not significant, F(3, 45) = 1.80, p = .16.

Post-surgery DMTP testing. Most rats reached the criterion of stable performance on the fifth session of DMTP reacquisition with a 4-s delay, with the exception of 1 HPC rat that reached criterion on the seventh session, 1 PRh rat that reached criterion on the eighth session, and 2 PRh rats that reached criterion on the seventh session. Figure 4 shows the mean second-swim latencies for the first five sessions. The mean latency was significantly longer in group HPC than in Group SHAM (p < .05) or group PRh (p < .05), whereas Groups SHAM and PRh did not differ significantly.

Figure 5 shows the mean second-swim latencies for each group on the presurgery and post-surgery mixed-delay ses-

Figure 5. The mean presurgery and post-surgery second-swim latencies (±SEM) that were determined on mixed-delay sessions in Experiment 1. HPC = hippocampal formation lesions; PRh = perirhinal cortex lesions; SHAM = sham surgery.

sions. Between-group comparisons of post-surgery performance indicated that HPC rats had significantly longer second-swim latencies than did SHAM rats (p < .05) or PRh rats (p < .05), but the SHAM rats and PRh rats did not differ significantly. Within-group comparisons indicated that the HPC rats' post-surgery second-swim latencies were significantly longer than their presurgery second-swim latencies, F(1, 15) = 11.91, p < .01; in contrast, presurgery and post-surgery second-swim latencies were not significantly different in Groups SHAM and PRh.

Although the foregoing analysis of second-swim latencies would appear to indicate that HPC rats were impaired in their ability to remember the platform location, the same general pattern of results was observed for first-swim latencies. That is, there were no differences among the groups on first-swim latencies before surgery, but post-surgery first-swim latencies were significantly longer in Group HPC than in Groups SHAM (p < .05) and PRh (p < .05). Therefore, an analysis that considers the difference between first-swim and second-swim latencies should provide a more meaningful index of retention than second-swim latencies alone. Accordingly, we calculated a savings ratio for each trial—second-swim latency as a proportion of total swim time for the trial (i.e., second-swim latency/first-swim latency + second-swim latency)—with the assumption that smaller savings ratios indicate better retention of the platform location.

Figure 6 shows post-surgery savings ratios for the groups at each delay. There was a significant effect of group, F(2, 15) = 12.42, p < .001, and nonsignificant effects of delay, F(3, 45) = 2.11, p > .10, and Group × Delay interaction, F(6, 45) < 1. Group HPC had significantly higher savings ratios at all delays than did Group SHAM (ps < .05) or Group PRh (ps < .05). There were no significant differ-

Figure 6. The mean savings ratios (±SEM) at each retention delay, as determined on post-surgery mixed-delay sessions in Experiment 1. The savings ratio represents second-swim latency as a proportion of total swim time for the trial (i.e., second-swim latency/first-swim latency + second-swim latency). Smaller ratios are assumed to reflect better retention of the platform location. HPC = hippocampal formation lesions; PRh = perirhinal cortex lesions; SHAM = sham surgery.
PERIRHINAL CORTEX AND SPATIAL MEMORY

Figure 7. Mean path length (±SEM) of first swims and second swims on trials of the post-surgery mixed-delay sessions in Experiment 1. The data are collapsed across all retention delays. HPC = hippocampal formation lesions; PRh = perirhinal cortex lesions; SHAM = sham surgery.

Figure 8. Mean swimming speed (±SEM) of first swims and second swims on trials of the post-surgery mixed-delay sessions in Experiment 1. The data are collapsed across all retention delays. HPC = hippocampal formation lesions; PRh = perirhinal cortex lesions; SHAM = sham surgery.

Figure 9. Mean first- and second-swim escape latencies (±SEM) on the visible-platform trials in Experiment 1. HPC = hippocampal formation lesions; PRh = perirhinal cortex lesions; SHAM = sham surgery.

ences between Group PRh and Group SHAM at any delay (p > .05).

As shown in Figure 7, the difference in mean path length from the first swim to the second swim on post-surgery trials also indicated an impairment in Group HPC and normal performance in Group PRh. Rats in Groups SHAM and PRh swam significantly shorter distances on second swims than on first swims: SHAM, F(1, 15) = 7.92, p < .02; PRh, F(1, 15) = 15.35, p < .001. Path lengths did not differ significantly between groups SHAM and PRh (p > .05). In contrast, although HPC rats also swam shorter distances on second swims than on first swims, this difference was not statistically significant, F(1, 15) = 4.16, p = .06. Moreover, first- and second-swim path lengths of HPC rats were significantly longer than those of SHAM rats (p < .05) or PRh rats (p < .05).

As shown in Figure 8, the rats in all three groups swam significantly faster on second swims than on first swims (p < .05), but there were no significant differences among the groups in overall swimming speed, F(2, 12) = 1.65, p > .20.

Visible-platform test. Figure 9 shows the mean first- and second-swim escape latencies on the visible-platform trials. Second-swim latencies were significantly shorter than first-swim latencies, F(1, 15) = 5.33, p < .05, but there was no significant difference among the groups, F(2, 15) < 1, and a nonsignificant Group × Swim interaction, F(2, 15) = 1.10, p > .05.

Discussion

Pretrained rats with perirhinal cortex lesions did not differ significantly from the controls on any of the DMTP performance measures, even when the retention delay was as long as 300 s. In contrast, rats with lesions of the hippocampal formation were severely impaired on the DMTP task at all retention delays, performing significantly worse than the sham-lesioned controls in terms of (a) second-swim escape latencies, (b) second-swim path lengths, and (c) savings ratios. There were no significant differences among the groups in terms of swimming speed on DMTP trials or in escape latencies on the visible-platform test, which suggests that the DMTP deficits of the HPC rats were not due to a general deficiency in swimming ability. The use of a different hidden-platform location on each DMTP trial and the employment of a range of retention delays should have emphasized the importance of intermediate-term storage of the platform location on individual trials. Together the results suggest that the HPC lesions impaired the rats’ ability to remember the location of the hidden platform and that this ability was unaffected by the PRh lesions.

The finding of DMTP deficits in our HPC rats is consistent with several previous demonstrations of impaired performance on tasks that require allocentric spatial memory after damage to the hippocampal formation (Barnes, 1988; O’Keefe & Nadel, 1978). Therefore, this finding was entirely expected. However, previous studies of spatial memory abilities after PRh lesions have produced conflicting findings. Thus, the normal DMTP performance of our PRh rats is
consistent with reports that rats with perirhinal cortex lesions were unimpaired on a spatial reference-memory task in a water maze (Wig & Bilkey, 1994b), on a T-maze spatial alternation task (Ennaceur et al., 1996), and on spatial (position) discrimination and nonmatching-to-position tasks in an operant chamber (Ennaceur et al., 1996), and inconsistent with reports of impaired acquisition of a reference-memory water maze task (Wig & Bilkey, 1994a) and delay-dependent deficits on a DMTP task similar to the one used in the present experiment in rats with lesions of the entorhinal and perirhinal cortex (Nagahara et al., 1995).

Our rats received extensive presurgery DMTP training, whereas the rats in the Wig and Bilkey (1994a) and Nagahara et al. (1995) studies did not receive any training in the water maze before surgery. It is possible, therefore, that presurgery training in our experiment obscured a significant place-memory impairment in our PRh rats. According to this interpretation, the functions of the perirhinal cortex may play an important role in the early stages of developing a representation of the spatial layout of the testing room but not in the long-term maintenance or storage of this representation, nor in its subsequent use in place navigation. Another possibility is that the perirhinal cortex is involved in the acquisition of the procedural aspects of water maze tasks. Both possibilities suggest that it would be premature to conclude from the present findings that place-memory abilities are entirely unaffected by PRh damage.

Experiment 2

This experiment was conducted to determine whether the extensive presurgery training administered in Experiment 1 may have obscured a place-learning deficit in rats with PRh lesions. The hypothesis that presurgery training can attenuate performance deficits on memory tasks after PRh lesions predicts that DMTP performance would be impaired in PRh-lesioned rats that did not receive presurgery training. As in Experiment 1, we compared the effects of bilateral lesions of the hippocampal formation or perirhinal cortex on rats’ DMTP performance, but this time the rats received no water maze training before surgery.

Method

Subjects

Nineteen adult male Long-Evans hooded rats served as subjects. They weighed between 300 and 350 g at the start of the experiment and were housed individually in opaque plastic cages in a colony room that was maintained at 21 °C with a 12-hr light–dark cycle (lights on at 8 a.m.). The rats had continuous access to water in their home cages and were fed approximately 25 g of standard laboratory rat food once per day.

Procedure

Surgery. Rats received either perirhinal cortex lesions (Group PRh, n = 7), hippocampal lesions (Group HPC, n = 6), or sham surgery (Group SHAM, n = 6). Surgery was performed under pentobarbital anesthesia (65 mg/kg). The hippocampal lesions were made with N-methyl-D-aspartate (NMDA; 5.1 M, dissolved in 0.1 M PBS, pH 7.2); NMDA was infused at a flow rate of 0.15 μl/min over a period of 160 s for a total injection volume of 0.4 μl per site. The cannulae were left in place for 2 min after each infusion. All other surgical procedures were identical to those of Experiment 1, including the injection coordinates (Table 1).

DMTP training. Two weeks after surgery, the rats were trained on the DMTP task using general procedures similar to those of Experiment 1. They received four trials (i.e., four pairs of swims) per daily session, with an intertrial interval of approximately 5 min.

The moving-platform procedure was gradually introduced during a preliminary training phase that lasted four sessions. The interval between swims on each trial was approximately 4 s. The platform was in a fixed location for all swims of the first session and was moved to a new location for all swims of the second session. Two new platform locations were used on the third session, each one for two trials (i.e., four swims), and another two new locations were used in the same manner on the fourth session.

After completion of the preliminary training phase, the rats received 10 DMTP sessions, with a 4-s retention delay between the first and second swim of each trial. The delay was subsequently increased to 30, 120, and 300 s, and each rat received four sessions (i.e., 16 trials) at each delay before moving on to the next longest delay.

Next, each rat received 10 mixed-delay sessions, each of which consisted of four trials, one at each of the four delays (i.e., 4, 30, 120, and 300 s). The order of delays used within each mixed-delay session was randomly determined.

Finally, a visible-platform test was administered in a single session, using procedures identical to those in Experiment 1.

Results

Histological Findings

The lesions were similar to and within the range of lesion sizes depicted in Figure 2. The NMDA injections produced extensive cell loss bilaterally throughout the hippocampus and dentate gyrus, with variable damage to the subiculum in every case. There was also minor damage to the parietal cortex where the injection cannula had been inserted, but this cortical damage was less than that which accompanied the ibotenate lesions in Experiment 1. There was no evidence of damage to the thalamus or rhinal cortex in any of the rats with hippocampal lesions.

The PRh lesions were roughly symmetrical in the two hemispheres (see Figure 3, top panel). Most of the perirhinal cortex was removed, and each rat sustained minor damage to the lateral entorhinal cortex; the medial entorhinal cortex was completely spared in both hemispheres of all rats. There was slight unilateral damage to ventro-posterior portions of area Te2 in 2 PRh rats and bilateral damage to this region in another PRh rat. Each PRh lesion also included minor bilateral damage to anterior portions of the postrhinal cortex, and unilateral damage to the ventral subiculum and the temporal CA1 field.

Behavioral Findings

The overall results were the same as in Experiment 1. To summarize, the PRh rats did not differ significantly from the
SHAM rats at any stage of DMTP training and testing, whereas the HPC rats were impaired at every stage.

Figure 10 shows the escape latencies during the preliminary training phase. Each time the platform was moved to a new location, there was a transient increase in escape latencies in Groups SHAM and PRh, followed by a rapid decrease in escape latencies over successive swims to the same platform location. Moving the platform had less effect on the escape latencies of the HPC rats, and they showed little improvement in escape latencies over successive swims to a particular location.

Figure 11 shows the second-swim escape latencies during each phase of formal DMTP testing—acquisition with a 4-s retention delay, testing at longer delays, and mixed-delay testing. One rat in Group PRh became ill and did not receive mixed-delay testing, which left data from 6 rats in Group PRh for this stage of the experiment. The HPC rats displayed deficits during all phases, whereas the PRh rats did not differ significantly from controls. Repeated measures ANOVA revealed that second-swim escape latencies during DMTP acquisition with a 4-s retention delay were significantly longer in HPC rats than in SHAM rats, $F(1, 10) = 9.89, p =$

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**Figure 10.** The mean second-swim escape latencies ($\pm SEM$) during preliminary training in Experiment 2. HPC = hippocampal formation lesions; PRh = perirhinal cortex lesions; Sham = sham surgery.
HPC versus SHAM, $F(1, 10) = 27.28, p < .001$; HPC versus PRh, $F(1, 10) = 66.64, p < .0001$; and PRh versus SHAM, $F(1, 10) = 4.15, p > .05$.

As in Experiment 1, the HPC rats’ first-swim latencies were significantly longer than those of the SHAM and PRh rats (not shown). Therefore, we calculated the savings ratios for mixed-delay sessions in the same manner as in Experiment 1. The general results were the same as for second-swim latencies: There were no significant differences in the savings ratios for Groups PRh and SHAM, $F(1, 10) < 1$, whereas the savings ratios in Group HPC were significantly higher (i.e., performance was poorer) than in Group SHAM, $F(1, 10) = 30.96, p < .001$, or in Group PRh, $F(1, 10) = 49.87, p < .0001$.

General Discussion

The main finding was that extensive bilateral lesions of the perirhinal cortex failed to disrupt DMTP acquisition or performance in either pretrained or un pretrained rats. Rats with perirhinal cortex lesions did not differ significantly from sham-lesioned rats on any of the DMTP performance measures, even when the retention delay was as long as 300 s. These results suggest that the perirhinal cortex does not play a critical role in allocentric spatial working memory. In contrast, rats with lesions of the hippocampal formation were severely impaired on the DMTP task at all retention delays, regardless of whether or not they received presurgery training. The latter results merely confirm the already well-established view that the hippocampal formation makes an essential contribution to allocentric spatial memory. However, the inclusion of the HPC group in these experiments was important for at least two reasons: First, it enabled a direct comparison of the effects of PRh lesions with those of HPC lesions; most other studies examining the effects of rhinal cortex damage on memory have included only the rhinal-lesion group and are, therefore, not as well controlled. Second, the demonstration of performance deficits in HPC rats indicates that the lack of impairment in PRh rats was not due to the task’s being insensitive to disruption of spatial memory.

Previous studies of spatial memory abilities after PRh lesions have produced conflicting findings. The normal DMTP performance of our PRh rats is consistent with reports that rats with perirhinal cortex lesions were unimpaired on a spatial reference-memory task in a water maze (Wiig & Bilkey, 1994b), on a T-maze spatial alternation task (Ennaceur et al., 1996), and on spatial (position) discrimination and nonmatching-to-position tasks in an operant chamber (Ennaceur et al., 1996). In contrast, the present findings are inconsistent with studies that found impaired acquisition of a spatial reference-memory task in a water maze after perirhinal cortex lesions (Wiig & Bilkey, 1994a) and delay-dependent deficits on a DMTP task similar to the one used in the present experiment after extensive entorhinal cortex lesions that also included parts of perirhinal cortex (Nagahara et al., 1995). Rats with combined lesions of the perirhinal and entorhinal cortices were also impaired on two spatially guided radial-arm maze tasks, one of which was a
delayed nonmatching-to-sample (DNMS) task that explicitly required the use of working memory for allocentric spatial information (Otto, Wolf, & Walsh, 1997).

The rats in the Nagahara et al. (1995) and Otto et al. (1997) studies sustained substantially more damage to entorhinal cortex than did the rats in our experiment, which may account for the differences between their findings and ours. Such an account is consistent with the results of other reports of spatial memory deficits following lesions of the entorhinal cortex (e.g., Goodlet, Nichols, Halloran, & West, 1987; Ramirez & Stein, 1984; Schenk & Morris, 1985), and it suggests that there are functional subdivisions within different regions of the rhinal cortex. The functions of the entorhinal cortex may be more important for spatial learning than those of the perirhinal cortex. From this viewpoint, perirhinal cortex damage may not have contributed to the place-learning deficits of the rats that received entorhinal-perirhinal cortex lesions in the Nagahara et al. (1995) study, and the extent of the entorhinal cortex damage that was sustained by the PRh rats of our experiment may have been insufficient to disrupt place learning.

It is more difficult, however, to provide a straightforward account of the inconsistencies between our findings and those of the Wig and Bilkey (1994a) study based on differences in the size or location of the rhinal cortex lesions. The electrolytic lesions in the Wig and Bilkey (1994a) study spared more of the perirhinal cortex and caused less collateral damage to the entorhinal cortex than did the aspiration lesions in our study. Lesioned rats in the former study took longer than control rats to locate a fixed hidden platform during acquisition trials, which suggests that the perirhinal cortex can make a significant contribution to place learning in some situations. One factor that can accentuate performance deficits on memory tasks after brain damage is a lack of presurgery training (see Mishkin & Murray, 1994). Wig & Bilkey’s rats received no presurgery training. However, the presence or absence of presurgery training made little difference in our study. The presurgery training in Experiment 1 was not sufficient to preserve normal DMT performance in rats with hippocampal lesions, nor was the absence of presurgery training in Experiment 2 sufficient to produce deficits in rats with perirhinal cortex lesions. It is more likely that specific cognitive or behavioral demands determine whether a particular place-memory task will be sensitive to perirhinal cortex damage. The present DMT task and the fixed hidden-platform task used in the Wig and Bilkey (1994a) study both required allocentric spatial memory, but in the latter task a representation of the platform location would presumably build gradually over several trials (i.e., a reference memory task), whereas the DMT task relies on a representation of the platform location after only a single trial (i.e., a working memory task).

Although the present experiment sought to determine whether damage to the perirhinal cortex would disrupt allocentric spatial working memory, an associated goal was to assess the nature of functional interactions between perirhinal cortex and the hippocampal formation—namely, to test the hypothesis that these structures are serial compo-

ments of a single memory system and that the memory functions of hippocampal formation depend upon input from perirhinal cortex. The findings from studies of anterograde amnesia in human and nonhuman primates with medial temporal-lobe damage and a vast literature on the effects of hippocampal ablations on memory abilities in rats have led many investigators to view the hippocampal formation and perirhinal cortex as two components of a temporal-lobe memory system (Eichenbaum et al., 1994; Squire, 1992; Zola-Morgan, Squire, & Ramus, 1994). This system is thought to support the representation of relational information about individual items or events that the subject experiences (i.e., declarative memory). Recent proposals suggest that the memory functions of the hippocampal formation critically depend upon information that it receives from the parahippocampal region—which in primates includes the entorhinal and perirhinal cortices and the parahippocampal cortex, and in rats the entorhinal, perirhinal, and postrhinal cortices (Eichenbaum et al., 1994). This predicts that any memory deficits that are caused by lesions restricted to the hippocampal formation should also be observed following lesions of the parahippocampal region, provided the latter sufficiently disrupt the flow of critical information to the hippocampus. Our results suggest that the spatial memory functions of the hippocampal formation do not depend upon inputs from the perirhinal cortex.

It has been suggested that, based on the nature of the parallel routes by which neocortical information may reach the hippocampal formation through the perirhinal and postrhinal cortices, the postrhinal cortex is likely to be more important for spatial information processing than is the perirhinal cortex (Burwell & Amaral, 1996; Suzuki, 1996). Given that our lesions spared most of the postrhinal cortex bilaterally but damaged most of the perirhinal cortex (see Figure 3), our results are consistent with what has been suggested by the anatomical findings.

It is difficult to argue that the present PRh lesions were not complete enough to disrupt the output from perirhinal cortex to the hippocampal formation, because most or all of the perirhinal cortex was removed bilaterally in each PRh rat (see Figure 3). Moreover, the extent of the PRh lesions was similar to those of two previous studies that found impaired object-recognition abilities after rhinal cortex lesions in rats (Glen & Mumby, 1996; Mumby & Pinel, 1994) and larger than perirhinal lesions that produced deficits on a water maze reference-memory task (Wiig & Bilkey, 1994a). Still, it is entirely possible that the above mentioned model is basically correct, and all that is needed to make it more accurate is to distinguish the contributions of the perirhinal cortex from those of the other components of the parahippocampal region, such as the entorhinal and postrhinal cortices. In other words, lesioning different portions of the parahippocampal region may produce different disconnection effects, but lesioning all of them together (i.e., a complete parahippocampal lesion) may, in fact, reproduce the effects of a hippocampal lesion. We did not aim to make complete lesions of the parahippocampal region in our rats and instead were interested in whether the perirhinal cortex makes a critical contribution to memory functions of the
hippocampal formation. In summary, the findings suggest that it does not.

Our findings fit well with other recent evidence of a double dissociation of function between the hippocampal formation and the perirhinal cortex. Neither rats (Kesner, Bolland, & Dakis, 1993; Mumbay, Pinel, Kornecek, Shen, & Redila, 1995; Mumbay et al., 1992; Rothblat & Kromer, 1991) nor monkeys (O'Boyle et al., 1993) with hippocampal lesions were impaired on the nonrecurrent-items DNMS task, which is used to assess object-recognition memory. Lesions of the rhinal cortex that spared the hippocampal formation produced DNMS deficits both in rats (Mumbay & Pinel, 1994) and in monkeys (Meunier et al., 1993; Zola-Morgan et al., 1989). The opposite pattern of results has been observed on tests of place memory, including various water maze tasks (e.g., Bolhuis, Stewart, & Forrest, 1994; M. Morris et al., 1982; Sutherland, Whishaw, & Kolb, 1983): Hippocampal lesions in rats impaired performance on such tasks (see Barnes, 1988, for a review), whereas perirhinal cortex lesions did not (Kolb et al., 1994; Wiig & Bilkey, 1994b; but see Wiig & Bilkey, 1994a). Evidence supporting a critical role for perirhinal cortex in object recognition also comes from electrophysiological studies (e.g., Brown, 1996) and from a recent study that compared the location and extent of immediate early-gene (c-fos) expression in rats presented with familiar and novel objects (Zhu, McCabe, Aggleton, & Brown, 1996). This double dissociation suggests that the contribution made by the perirhinal cortex to object-recognition memory is not dependent on information from the hippocampal formation and that the contribution made by the hippocampal formation to place memory is not dependent on information from the perirhinal cortex.

The notion that the perirhinal cortex and the hippocampal formation can function independently in some situations still allows for interactions between them in solving problems or performing tasks that require the primary functions of both structures. For example, Gaffan and Parker (1996) found evidence that interactions of the perirhinal cortex and fornix are required for performance of a task that requires monkeys to remember spatial arrangements of individual objects (i.e., object-in-place memory).

In summary, we draw two main conclusions from this experiment: (a) The functions of the perirhinal cortex are not essential for allocentric spatial working memory in rats, and (b) the contributions of the hippocampal formation to spatial memory do not rely on either direct or indirect inputs from the perirhinal cortex. These findings are consistent with the idea that the perirhinal cortex and hippocampal formation can function independently to serve certain memory abilities, a view previously expressed by others (e.g., Ennaceur et al., 1996; Gaffan, 1994; Meunier et al., 1996) and one that would receive valuable support from demonstrations of a double dissociation following PRH and HPC lesions within subjects and within the same experiment. Such a demonstration would mitigate arguments that the apparent double dissociation that appears to exist within the literature is an artifact of different lesion sizes and locations, and different memory tasks, across different experiments.

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