Spatial Memory in the Rat Requires the Dorsolateral Band of the Entorhinal Cortex

Hill-Aina Steffenach,[1] Menno Witter,[1,2]
May-Britt Moser,[1] and Edvard I. Moser[1,*]
[1]Centre for the Biology of Memory
Norwegian University of Science and Technology
NO-7489 Trondheim
Norway
[2]Research Institute Neuroscience
Department of Anatomy
VU University Medical Center
Amsterdam 1007 MB
The Netherlands

Summary

The extensive connections of the entorhinal cortex with the hippocampus and the neocortex point to this region as a major interface in the hippocampal-neocortical interactions underlying memory. We asked whether hippocampal-dependent recall of spatial memory depends on the entorhinal cortex, and, if so, which parts are critical. After training in a Morris water maze, rats received fiber-sparing lesions in the dorsolateral band of the entorhinal cortex, which mediates much of the visuospatial input to the dorsal hippocampus. These lesions entirely disrupted retention and retarded new learning. Spatial memory was spared by lesions in the ventromedial band, which connects primarily with ventral hippocampus, but these lesions reduced defensive behavior on an elevated plus maze, mirroring the effects of damage to ventral hippocampus. The results suggest that the functional differences between dorsal and ventral hippocampus reflect their connectivity with modules of the entorhinal cortex that are differently linked to the rest of the cortex.

Introduction

Converging evidence suggests that the hippocampus is essential for fast encoding and storage of new episodic memories. The involvement of the hippocampus in memory is particularly evident in tasks where subjects must recall spatial relations between landmarks in order to locate a hidden goal object (O'Keefe and Nadel, 1978; Morris et al., 1982; Maguire et al., 1998; Teng and Squire, 1999; Ekstrom et al., 2003), although it is clear from studies in both humans and animals that the role of the hippocampus extends to other types of declarative memory as well (Scoville and Milner, 1957; Squire, 1992; Eichenbaum, 2000).

The encoding and consolidation of episodic memories is thought to rely on interactions between the hippocampus and other cortical structures (McClelland et al., 1995; Squire and Alvarez, 1995). The exact routes by which information about location and other elements of episodes reach the hippocampus are not known, nor are

the neocortical output structures in which hippocampal memories are implemented for long-term retention. However, in terms of connectivity, the entorhinal cortex stands out as the primary interface between the hippocampus and neocortex, linking the hippocampus with nearly all other association cortices (Witter et al., 1989; Burwell, 2000; Lavenex and Amaral, 2000; Witter and Amaral, 2004). This pattern of organization suggests that the entorhinal cortex plays a fundamental role in the computations that take place both before and after cortical information enters the hippocampus.

If hippocampal memories depend on interactions with the neocortex, one would expect lesions of the entorhinal cortex to severely impair hippocampal-dependent memory, with the profile of impairment mirroring that observed after damage to the hippocampus. Surprisingly, this prediction has not been upheld in studies of spatial memory after selective fiber-sparing lesions of the entorhinal cortex. While large mechanical or electrolytic lesions in the parahippocampal cortex impaired performance in a number of spatial tasks (Olton et al., 1978; Jarrard et al., 1984; Schenk and Morris, 1985; Rasmussen et al., 1989; Cho and Kesner, 1996), selective excitotoxic damage to the entorhinal cortex generally failed to mirror the pronounced spatial learning impairment normally observed after lesions of the hippocampus (Bouffard and Jarrard, 1988; Pouzet et al., 1999; Bannerman et al., 2001a, 2001b; Galani et al., 2002; Oswald et al., 2003; Burwell et al., 2004; Jarrard et al., 2004; but see Holscher and Schmidt, 1994 and Cho and Jaffard, 1994, 1995; for review, see Aggleton et al., 2000). The simplest interpretation of the spared spatial learning in animals with selective entorhinal lesions is that the visuospatial information that is required to solve hippocampal-dependent spatial learning tasks can reach the hippocampus via sparser routes. Such routes include the direct connections with the presubiculum and parasubiculum (Witter et al., 1988) and the perirhinal and postrhinal cortices (Naber et al., 1999, 2001), and the direct connections with subcortical structures such as the midline nuclei of the thalamus (Herkenham, 1978; Wouterlood et al., 1990; Dolleman-van der Weel and Witter, 1996). These inputs and their reciprocal outputs may be sufficient for the retention of spatial memories (see Aggleton et al., 2000). On the other hand, several studies have provided evidence for a modular organization of the entorhinal cortex, such that it is organized into recurrently connected bands that run parallel to the rhinal sulcus and cut across the medial and lateral subdivisions of the area (Witter et al., 1989; Dolorfo and Amaral, 1998a, 1998b). Spatially modulated neurons have been observed only in the most caudal portion of the dorsolateral band, i.e., the part made up by the medial entorhinal cortex near the postrhinal cortex and the rhinal fissure (Fyhn et al., 2004). This area receives most of the cortical visuospatial input to the entorhinal cortex (Burwell, 2000). More ventromedially located neurons exhibit little or no spatial modulation (Fyhn et al., 2004). Based on the extreme dorsocaudal position of the spatially modulated neurons, we hypothesized that
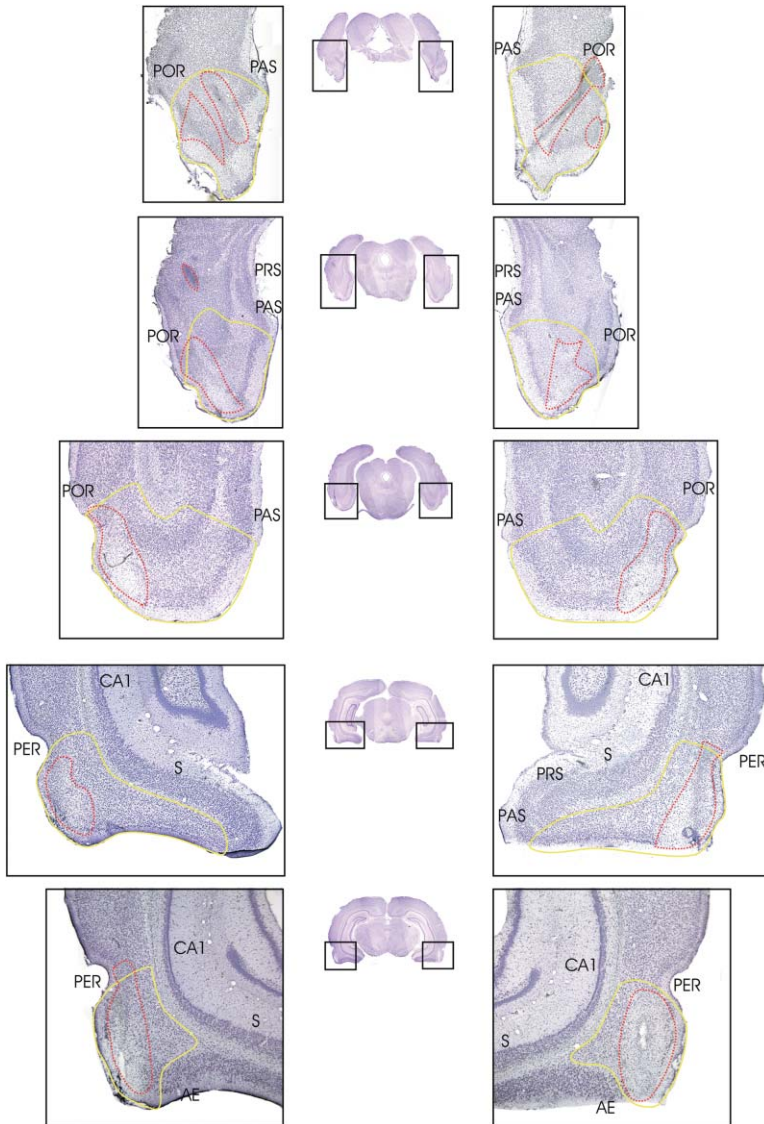
*Correspondence: edvard.moser@cbm.ntnu.no

Figure 1. Coronal Sections Stained with Cresyl Violet, Indicative for Neuronal Cell Bodies, after a Representative Cytotoxic Lesion of the Dorsolateral Band of the Entorhinal Cortex

Sections are shown for rat 10647 at five levels between the anterior and posterior poles of the entorhinal cortex (middle columns: low magnification; left and right columns: high magnification). Yellow traces show outlines of the entorhinal cortex at each coronal level. Red traces indicate borders of the lesion (including areas with partial cell loss). Adjacent cortical areas are indicated (PER, perirhinal cortex; POR, postrhinal cortex; PAS, parasubiculum; PRS, presubiculum; S, subiculum; AE, amygdaloentorhinal transition area). Note that the lesion covers nearly the entire anteroposterior extent of the entorhinal cortex. The dorsolateral band (closest to the perirhinal and postrhinal cortices) is extensively lesioned. The damage stretches into the intermediate band in some places, and there is minor damage to the perirhinal and postrhinal cortices.

previous lesion studies may have missed that particular part of the entorhinal cortex. The present study thus examined the ability of rats to recall spatial information after lesions that specifically targeted the dorsolateral band and compared their performance to that of animals with sham lesions or lesions in the ventromedial band.

Results

Lesions Aimed at the Dorsolateral Band

The first aim of the study was to determine whether spatial memory requires the dorsolateral band of the entorhinal cortex. A total of 124 rats were trained to asymptote in a conventional reference memory task in the Morris water maze. Within 36 hr after the last training session, the rats received N-methyl-D-aspartate (NMDA)-induced excitotoxic lesions in the dorsolateral band or sham lesions. Recall was tested on a probe trial 7 days later. This part of the study was conducted in five replications, each containing approximately five lesioned animals and five sham animals.

The lesions in the dorsolateral band were generally limited to the target area, forming a bilateral strip of damage beneath the rhinal fissure and the postrhinal border (Figures 1 and 2). The lesions always extended into the intermediate band, but the ventromedial band was spared. The lesions generally involved both superficial layers II and III and deep layers V and VI, although the damage to layers II and III was usually larger. The damage was generally more extensive in the dorsocaudal part of the band than in the ventrorostral part. The lesions covered 38.8% ± 3.5% of the area of the entorhinal cortex as measured in the unfolded maps (mean ± SEM; range, 9.8%–56.1%). The border between damaged and healthy tissue was sharp, implying that the density of intact neurons within the spared parts of entorhinal cortex was within the normal range.

Most dorsolateral lesions caused some additional damage to the adjacent parts of the perirhinal or postrhinal cortices, and in the overlying temporal cortex around the cannula tracks (Figure 1). The amount of such damage varied between animals. Three rats were excluded
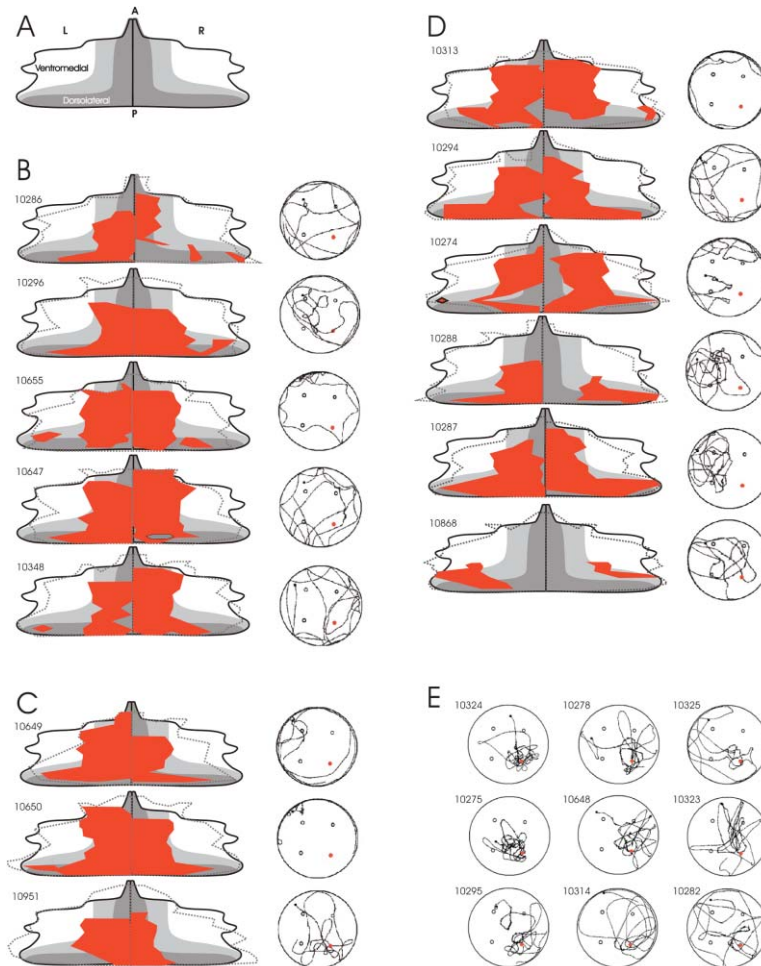
Figure 2. Unfolded Maps Showing Extent of Lesions in the Entorhinal Cortex and Performance on the Retention Test for Each Animal in the Dorsolateral Band Group

(A) Average unfolded map based on the entire sample of individual maps. To construct each map, the lateral border of the entorhinal cortex in each brain section was mapped onto the vertical line in the center of each diagram, and sections were unfolded onto straight lines perpendicular to the line that represents the lateral border. L, left hemisphere; R, right hemisphere; A, anterior; P, posterior. A schematic representation of the three parallel bands in entorhinal cortex is indicated. The dark gray area represents the dorsolateral band, light gray represents the intermediate band, and white represents the ventromedial band.

(B–D) Outline of lesions (left) and swim paths on the postsurgical retention test (right) for individual animals with lesions in the dorsolateral band. Animals are grouped according to the amount of inadvertent damage to adjacent structures ([B], perirhinal damage only; [C], perirhinal and postrhinal damage; [D], damage in both hippocampal and perirhinal or postrhinal areas). Individual maps are indicated with stippled gray contours. Lesions are indicated in red. The solid black contour shows the average unfolding. In the swim path diagrams, the position of the unavailable platform is marked by a red circle (southeast quadrant). Note that none of the three groups performed above chance level on the retention test.

(E) Swim paths on the postoperative retention test for 9 of the 28 sham-operated dorsolateral control animals (ranked as number 1, 4, 6, 9, 13, 17, 20, 21, and 25, with time in the target zone as the ranking variable; rat 10324 was number 1, rat 10275 was number 4, rat 10295 was number 6, etc.).

from the dorsolateral group because of extensive hippocampal damage. Two were excluded because the entorhinal cortex was essentially spared on one side, and five rats were taken out because the most posterior brain sections (containing the dorsocaudal portion of the medial entorhinal cortex) were lost when the brains were cut. Among the remaining animals in the dorsolateral group, five had additional damage only in the perirhinal cortex. Three animals had some damage in the postrhinal cortex as well, mostly in the anterior half. Six animals had minor lesions in the subiculum or CA1 of the ventral hippocampus, in addition to some damage to the perirhinal or postrhinal cortices. Behavioral results were first analyzed separately for these three subgroups, but in the absence of any detectable behavioral differences, the subgroups were subsequently combined.

All rats learned to approach the platform within ~10 s during the 6 days of preoperative training (Supplemental Figure S1A at http://www.neuron.org/cgi/content/full/45/2/301/DC1/). The escape latencies decreased quickly to ~20 s during the first 2 days and then decreased slowly during the remainder of the pretraining phase. On day 7, retention was tested on a probe trial during which the platform remained unavailable for 60 s (Figure 3A). Most rats showed a strong bias toward the platform position during the search period, spending an average of 43.6% ± 2.0% of the search time within a circular zone around the platform (mean ± SEM). The zone covered 12% of the pool surface. The rats were ranked, matched, and assigned to surgery groups according to the proportion of time they spent in the platform zone.

The effect of the lesions was tested on a second probe trial 7 days after the surgery (Figure 3B). While the sham-operated animals continued to search at the training location (38.0% ± 3.0% in the platform zone), those with lesions in the dorsolateral band displayed no memory of the target location, spending only 8.7% ± 2.9% of total search time in the circular zone, which was no more than the time they spent in the corresponding zones of the three other quadrants. Thirteen of the 14 rats with lesions in the dorsolateral band had swim times near chance level (range, 0%–16.1% in the target zone; the expected time with a nonbiased search pattern would be 12%). One outlier spent 41.2% in the target zone (rat 10951; 26.0% lesion). This rat had less damage to the dorsocaudal pole than other rats in the dorsolateral band group (Figure 2B; see posterior end of the unfolded
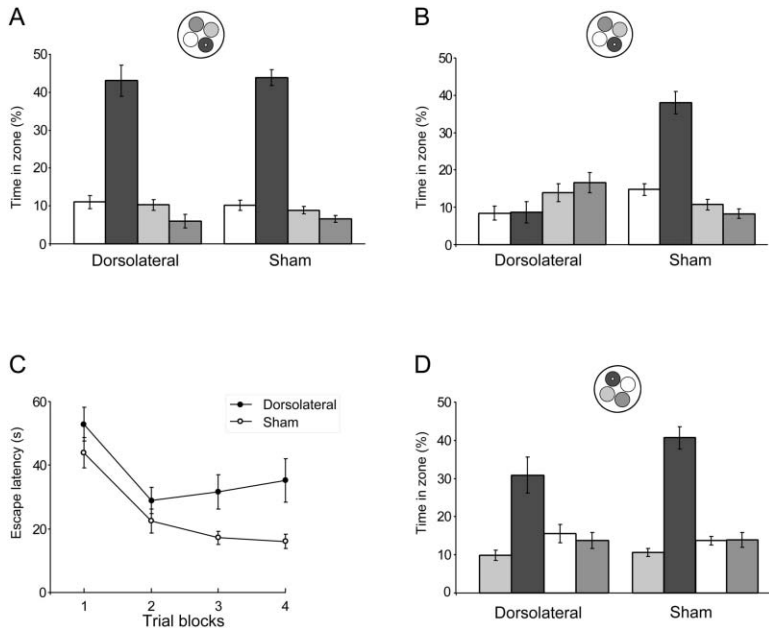
Figure 3. Spatial Memory in the Water Maze after Lesions in the Dorsolateral Band of the Entorhinal Cortex

(A and B) Probe trials at the end of pretraining (A) and 7 days after pretraining and surgery (B). Bars indicate time spent in a circular zone around the platform position (black) and in corresponding zones of the three other quadrants (means ± SEM; see inset). Chance level is 12%.
(C) Latency to locate the hidden platform during reversal training after surgery (means ± SEM).
(D) Probe trial after postoperative training with the platform at a new location.

map). There was no significant correlation within the dorsolateral group between lesion size and time spent in the target zone (r = −0.43; n = 14; p = 0.13). A repeated-measures analysis of variance of time spent in the platform zone and in corresponding zones of the three other pool quadrants showed a significant group × quadrant effect [F(3, 120) = 23.9; p < 0.001] with a significant group difference in the target zone [t(40) = 6.2; p < 0.001].

The search times of animals in the dorsolateral group were not related to the amount of accompanying damage in adjacent structures. The five rats with additional damage only in the perirhinal cortex spent between 4.5% and 16.1% of the trial in the platform zone (Figure 2B). The zone times of the three rats with inadvertent lesions in both the perirhinal and postrhinal cortices were 3.0%, 0%, and, in the case of the previously mentioned outlier (rat 10951), 41.2% (Figure 2C). The six rats with additional minor damage in the subiculum or hippocampus spent between 0% and 13.5% in the target circle (Figure 2D).

The ability of the rats with entorhinal lesions to learn a new, modified version of the task was tested by presenting the platform in the opposite quadrant at the end of the probe trial and using this position as the goal on subsequent trials. The rats received four blocks of training over the course of 8 hr (one block of four trials and three blocks of two trials). Escape latencies decreased in both groups [block effect: F(3, 132) = 19.1; p < 0.001] but were longer in the animals with dorsolateral lesions [Figure 3C; group effect: F(1, 44) = 8.1; p < 0.01]. When the new memory was probed at the end of training, both groups searched in the platform zone, and the group × quadrant interaction effect was no longer significant [F(3, 108) = 2.1; p = 0.10; Figure 3D]. The main effect of quadrant was significant [F(3, 108) = 42.5; p < 0.001]. There was no correlation between lesion size and time in the target zone in the dorsolateral group (r = 0.01; n = 14).

Lesions Aimed at the Perirhinal and Postrhinal Cortices

The lesions in the dorsolateral band were accompanied by minor to moderate damage in adjacent parts of the perirhinal or anterior postrhinal cortices (Figure 1). To determine whether the retention impairment in the dorsolateral group was caused by this additional damage, we prepared a separate group of animals with lesions that specifically targeted the affected areas of perirhinal and postrhinal cortex. The damage to the perirhinal cortex in these animals was fairly complete and selective (Figure 4). The lesion also included postrhinal cortex, but primarily the anterior part of the area. There was only very minor damage to the entorhinal cortex, mostly at the adjacent dorsolateral tip of layer II. The size of the perirhinal and postrhinal lesions was generally larger than that in the animals with entorhinal lesions.

Lesions in the perirhinal and postrhinal cortices did not impair retention on the postoperative probe trial (Figure 5). Rats with such lesions spent as much time in the platform zone as simultaneously trained sham-operated animals [Figure 5B; quadrant: F(3, 30) = 9.4, p < 0.001; group × quadrant interaction: F < 1]. The rate of learning in the reversal task was unimpaired (Figure 5C; group difference in escape latencies: F < 1), and the lesioned animals searched extensively in the correct quadrant on the final probe trial [Figure 5D; quadrant: F(3, 30) = 16.9, p < 0.001; group × quadrant interaction: F(3, 30) = 2.1, p > 0.10]. These results suggest that damage to the perirhinal and postrhinal cortices did not contribute detectably to the retention impairment of the dorsolateral entorhinal group, consistent with the fact that the impairment in the latter group was independent of the amount of accompanying damage to the perirhinal and postrhinal cortices.

Finally, we checked whether the impairment after lesions of the entorhinal cortex was due to needle-induced mechanical damage to overlying areas. An additional sham group was prepared in which the needle was low-
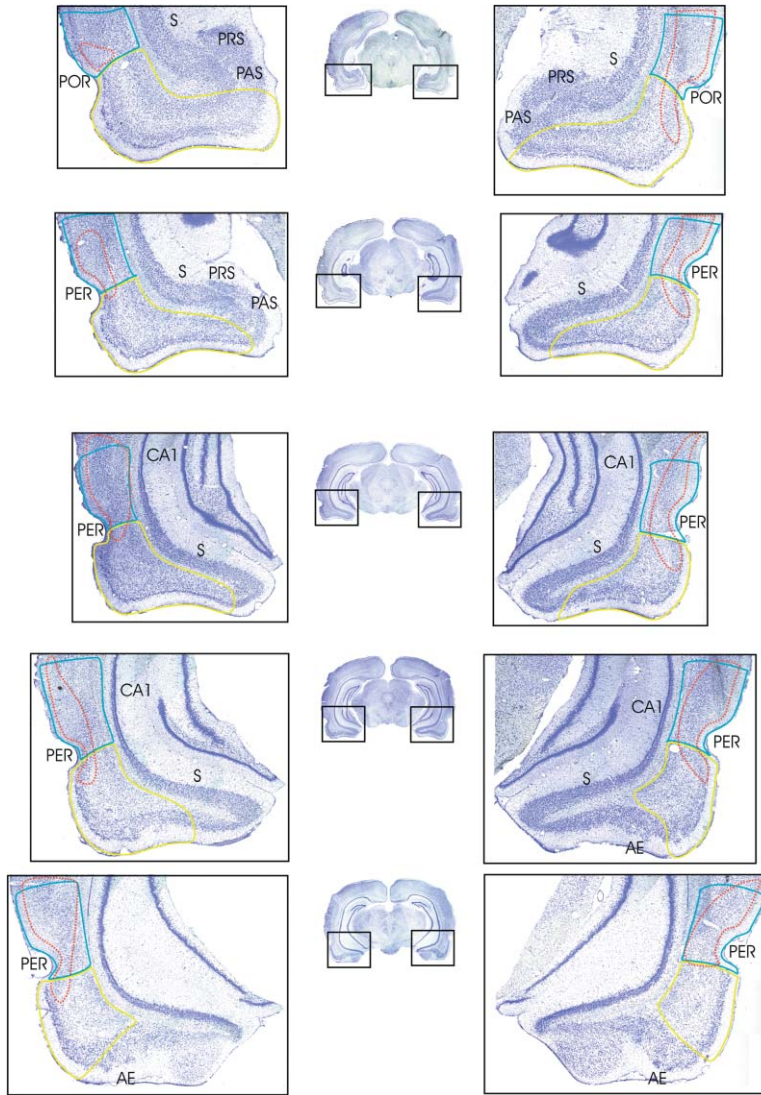
Figure 4. Coronal Sections Showing Cresyl Violet Stains of Neuronal Cell Bodies after a Representative Cytotoxic Control Lesion in the Perirhinal and Anterior Postrhinal Cortices

Sections are taken slightly more anterior than those in Figure 1. Yellow traces indicate borders of the entorhinal cortex, blue traces indicate borders of the perirhinal and postrhinal cortices, and red indicates the lesion. Abbreviations as in Figure 1. Note that both the perirhinal and anterior postrhinal cortices were extensively lesioned, yet this animal exhibited good retention (29.4% in the platform zone on the postoperative retention test).

ered into the dorsolateral band of entorhinal cortex without infusion of the excitotoxin. The needle tracks caused similar damage in the perirhinal, postrhinal, and temporal cortices as in the group with entorhinal lesions (Supplemental Figure S2 at http://www.neuron.org/cgi/content/full/45/2/301/DC1/). Rats with such mechanical lesions spent as much time in the platform zone on the postoperative retention test as a group of simultaneously trained sham-operated animals in which the needle was not inserted [Supplemental Figure S3B; quadrant: $F_{(3, 30)} = 21.6$, $p < 0.001$; group × quadrant interaction: $F_{(3, 42)} = 1.1$, $p > 0.3$]. The two groups performed similarly also after postoperative reversal training [Supplemental Figure S3C; quadrant: $F_{(3, 18)} = 5.6$, $p < 0.01$; group × quadrant interaction: $F < 1$].

Lesions Aimed at the Ventromedial Band

We next asked if spatial memory specifically depends on the dorsolateral band of the entorhinal cortex, or if more ventromedial parts are also required. Ten rats received bilateral lesions in the ventromedial band, using an oblique injection track that bypassed most of the hippocampus. These rats were tested together with eight sham-operated rats in two replications.

The lesions were generally limited to the ventromedial band, forming a bilateral strip of damage along the border with the parasubiculum (Figures 6 and 7). The lesions generally extended into the intermediate band, but the dorsolateral band was spared. Both deep and superficial layers were involved. The damage was more extensive in the caudal part of the ventromedial band than in the rostral part. On average, the ventromedial lesions covered 12.3% ± 1.1% of the area of the entorhinal cortex as measured in the unfolded maps (mean ± SEM). The range was 7.8%–20.1%. Ventromedial band lesions were thus generally smaller than the lesions in the dorsolateral band, although the largest ventromedial lesions caused a larger percentage of damage in the target band than the smallest dorsolateral lesions. The border between damaged and healthy tissue was sharp, similar to what was observed for the dorsolateral band lesions.

Lesions in the ventromedial band caused some additional damage in adjacent parts of the parasubiculum and presubiculum, as well as occasional minor damage
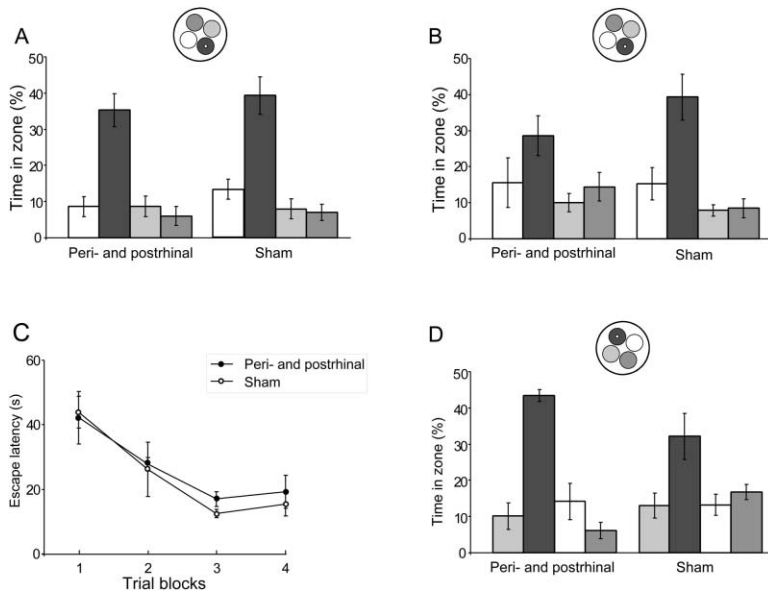
Figure 5. Recall after Excitotoxic Control Lesions in the Perirhinal and Anterior Postrhinal Cortices

(A and B) Probe trials at the end of pretraining (A) and 7 days after pretraining and surgery (B).

(C) Latency to locate the hidden platform during reversal training after surgery (means ± SEM).

(D) Probe trial after postoperative training with the platform at a new location. Symbols as in Figure 3.

in the subiculum and dentate gyrus around the cannula tracks. The amount of such damage was highly variable between animals. Three rats were excluded from the ventromedial group because of more extensive hippocampal damage.

Lesions in the ventromedial band had no significant effect on retention of the spatial task (Figure 8). The lesioned rats continued to search near the platform in the same way as the simultaneously trained sham-operated group (means of 32.7% and 35.5%, respectively, in the target zone; Figure 8B). Seven rats had damage almost exclusively in the ventromedial band. These animals showed consistently good retention (zone times from 20.6% to 48.9%; Figure 7). Three rats had additional damage to the intermediate band (zone times of 12.4%, 26.0%, and 55.0%; Figure 7). Within the ventromedial group, there was no significant correlation between lesion size and time spent in the target zone (r = −0.46; n = 8; p = 0.18). A repeated-measures analysis of variance of time spent in the four quadrant zones showed a significant effect of quadrant [$F_{(3, 48)}$ = 20.1; p < 0.001] but no group × quadrant interaction (F < 1). During subsequent reversal training, the escape latencies decreased equally in the two groups [Figure 8C; group and group × block effects: F < 1; block: $F_{(3, 48)}$ = 28.4, p < 0.001], and there was no difference in their search pattern on the final probe trial [Figure 8D; group × quadrant interaction: F < 1; quadrant: $F_{(3, 48)}$ = 15.3, p < 0.001].

Lesions in the ventromedial band were generally smaller than those in the dorsolateral band. Although the relative amount of damage overlapped, the ventromedial lesions may have been too small to impair any behavior. To determine whether the lesions were large enough to functionally perturb the circuitry of the ventromedial band, we tested a subset of the animals with ventromedial lesions in a task known to depend on the ventral hippocampus, to which the ventromedial band is strongly connected (Dolorfo and Amaral, 1998a). The elevated plus maze is such a task (Kjelstrup et al., 2002).

The plus maze has two open and two enclosed arms. During their first exposure to the maze, rats typically make few entries into the open arms. Clinically effective anxiolytics reduce this avoidance (Pellow et al., 1985; Kjelstrup et al., 2002). Six of the rats with lesions in the ventromedial band were tested on the plus maze. Whereas the sham-operated animals avoided the open arms, rats with lesions in the ventromedial band visited the open arms more frequently [Figures 8E and 8F; group difference: $t_{(8)}$ = 2.6; p < 0.05; two-tailed Student's t test], although the total time spent on these arms was not significantly enhanced [$t_{(8)}$ = 1.3; p > 0.2]. The results show a dissociation of function between the dorsolateral and ventromedial bands of the entorhinal cortex and suggest that the lesions were large enough to disrupt one function of the ventromedial band of entorhinal cortex.

Discussion

The present study aimed to determine whether the entorhinal cortex is necessary for the acquisition and retention of a hippocampal-dependent spatial reference memory task. We showed first that lesions that included the dorsolateral band of the entorhinal cortex entirely and consistently disrupted recall of a spatial navigation task acquired before the lesion. Second, lesions in the ventromedial band failed to affect spatial memory but reduced avoidance of the open arms on a plus maze test of defensive behavior. The results suggest a modular organization of the entorhinal cortex with an essential role for the dorsolateral band in spatial memory and the ventromedial band in control of defensive behavior.

The Entorhinal Cortex Is Necessary for Spatial Memory

The results point to a key role for the dorsolateral band of the entorhinal cortex in spatial computation or spatial representation. They are consistent with the existence of neurons with sharp and coherent place fields caudally
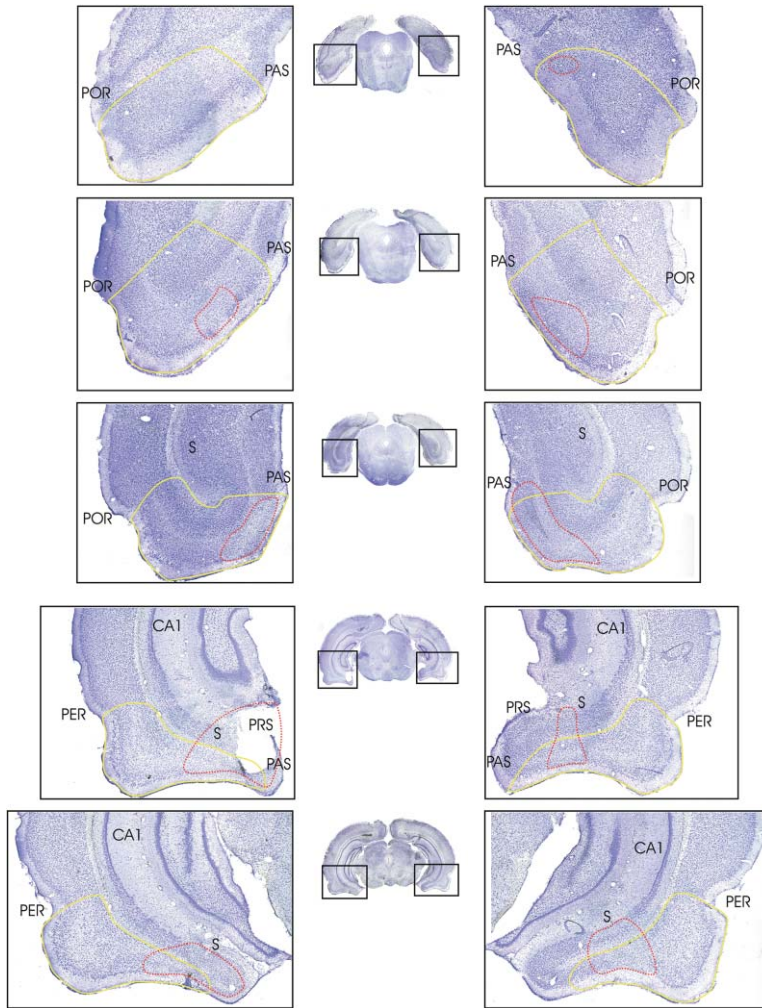
Figure 6. Coronal Sections Stained with Cresyl Violet after a Representative Cytotoxic Lesion of the Ventromedial Band of the Entorhinal Cortex

Sections are shown for rat 10948 at approximately the same five anteroposterior levels as in Figure 1. Abbreviations as in Figure 1. Note that the ventromedial parts of the entorhinal cortex are lesioned on both sides. The lesion stretches into parts of the intermediate band on the right side but leaves the dorsolateral band intact. Note additional damage to the subiculum, presubiculum, and parasubiculum.

in layers II and III of this area (Fyhn et al., 2004). Although such neurons have multiple fields dispersed over the entire recording arena, their information rate is comparable to that of place cells in CA1, and collectively they signal the rat's position as accurately as place cells in the hippocampus. Cells with similarly sharp spatial modulation have not so far been observed further upstream of the hippocampus, such as in the presubiculum (Taube et al., 1990; Cacucci et al., 2004) and parasubiculum (Taube, 1995) or the perirhinal and postrhinal cortices (Taube et al., 1990; Burwell et al., 1998; Burwell and Hafeman, 2003; Fyhn et al., 2004), implying that the dorsocaudal portion of the dorsolateral band may contribute directly to the computation of spatial information. The present study shows that the dorsolateral band not only expresses spatial information but is also necessary for storage, consolidation, or retrieval of such information.

The complete disruption of retention in the water maze is at odds with the relatively preserved spatial learning ability of rats with excitotoxic lesions of the entorhinal cortex reported in previous studies (Bouffard and Jarrard, 1988; Pouzet et al., 1999; Bannerman et al., 2001a, 2001b; Galani et al., 2002; Oswald et al., 2003; Burwell et al., 2004; Jarrard et al., 2004). At initial glance, the ability of rats to solve spatial memory tasks after fiber-sparing lesions in the entorhinal cortex may suggest that cortical inputs to the hippocampus other than those from the entorhinal cortex must be sufficient for spatial learning (see Aggleton et al., 2000 and Jarrard et al., 2004 for discussion). The only significant cortical inputs that could mediate such a function are those originating in the perirhinal and postrhinal cortices (Naber et al., 1999, 2001) and the pre- and parasubiculum (Witter et al., 1988). However, except perhaps for the parasubicular projection to the dentate gyrus (Witter et al., 1988), direct inputs from these areas are sparse and reach only limited parts of the transverse axis of the hippocampus (Naber et al., 1999, 2001). An alternative explanation of why previous studies with excitotoxic lesions failed to impair spatial memory is that the entorhinal lesions may have been incomplete. While the ventral part of the entorhinal cortex was damaged successfully in most of the studies, the extent of damage to more dorsal and caudal regions is uncertain, as sections including this part were generally not included in figures showing outlines of the damage. The reported injection sites and the reconstructions of three studies that aimed to include the more dorsal areas (Bannerman et al., 2001a; Galani et al., 2002; Oswald et al., 2003) suggest that the lesions
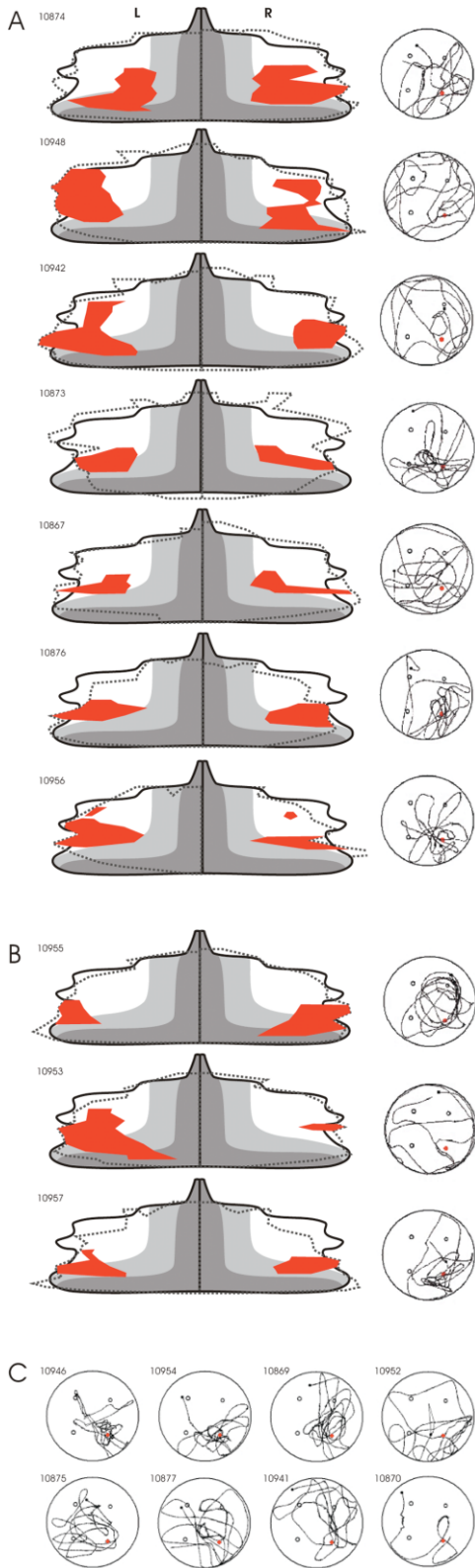
Figure 7. Unfolded Maps Showing Extent of Lesions in the Entorhinal Cortex and Performance on the Postoperative Retention Test for Each Animal with Damage in the Ventromedial Band

Animals are grouped according to the selectivity of the lesions ([A], damage primarily in the ventromedial band; [B], significant damage

spared the dorsocaudal area in which the strongest spatial modulation has been reported (Fyhn et al., 2004). The sparing may account for the lack of significant effects on spatial memory. This suggestion is in line with our findings in one animal (rat 10951) where the lesion largely missed the dorsocaudal pole of the entorhinal cortex and no spatial impairment was detected. Although the lesions of the remaining rats were far from complete with respect to removing the entire entorhinal cortex, or even removing the entire dorsolateral band, they all included the dorsocaudal pole. The complete and consistent disruption of retention in these animals suggests that the dorsocaudal pole of the medial entorhinal cortex plays an essential role in spatial memory. This does not rule out an additional role for sparser inputs to the hippocampus like the moderately dense projection from the parasubiculum (Kesner and Giles, 1998; Liu et al., 2001, 2004; Jarrard et al., 2004), but our results suggest that these alternative connections are not sufficient for maintaining and expressing spatial memory.

While the present study clearly points to a necessary role for the dorsolateral band of the entorhinal cortex in spatial memory, the exact functions performed by neurons in this region remain elusive. One outstanding issue is whether the entorhinal cortex is differently involved during encoding, storage, and retrieval of spatial memory. Lesions of the dorsolateral band abolished retention when the spatial task was learned before surgery but had a weaker effect on new learning. Escape latencies were longer during reversal learning, but there was no significant disruption of the search pattern on the final probe trial. At least two factors may account for the weaker impairment of new learning. First, the entorhinal lesions were not complete. Remaining tissue in the dorsolateral band, or in the intermediate and ventromedial bands, may have been sufficient to support new learning. Studies in the hippocampus have shown that retention of spatial memory in the water maze requires the integrity of large parts of the hippocampus, whereas less is required for new spatial learning (Moser and Moser, 1998a). It is possible that encoding in the intact animal engages a wide entorhinal and hippocampal network and that this distributed network must be activated in its entirety for successful retrieval of the stored information, rendering retention more vulnerable to lesions than new learning. Second, the effects on new learning may depend on the exact requirements of the behavioral task. In the present reversal task, only the platform was moved; other landmarks remained stationary. Although no memory was detected on the postoperative retention trial, further training may facilitate the retrieval of memories stored before surgery that were not accessible on the initial test (de Hoz et al., 2004). Such memories may speed up learning when the majority of sensory cues remain unaltered. Performance may also have benefited

also in the intermediate band). Symbols as in Figure 2. (C) Swim paths on the postoperative retention test for all eight sham-operated ventromedial control animals, ranked according to time spent in the target zone (top row: rank 1–4; bottom row: rank 5–8). Rat 10870 spent a significant part of the trial floating between northwest and southwest and in southeast.
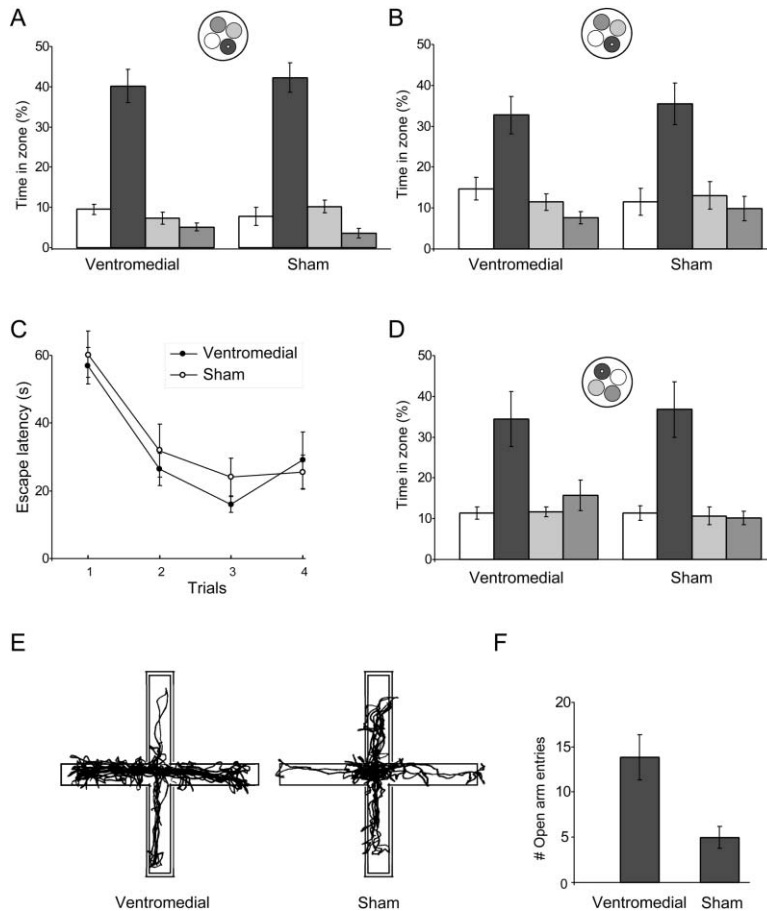
Figure 8. Behavioral Effects of Lesions in the Ventromedial Band of the Entorhinal Cortex

(A and B) Recall of spatial memory assessed by probe trials at the end of pretraining (A) and 7 days after pretraining and surgery (B). Symbols as in Figure 3.

(C) Latency to locate the hidden platform during reversal training after surgery (means ± SEM).

(D) Probe trial after postoperative training with the platform at a new location.

(E and F) Reduced avoidance of open arms in an elevated plus maze after lesions in the ventromedial band. (E) Paths of representative animals from the ventromedial and sham groups. West and east arms were open; north and south were closed. (F) Number of entries into the open arms.

from the use of short retention intervals during postoperative learning. Tests using different training environments and longer retention intervals may reveal a more permanent disruption of both encoding and maintenance processes.

A second outstanding issue is whether the dorsolateral band of the entorhinal cortex participates in computing allocentric location, stores memories of location, or does both. In principle, impaired performance in the water maze task may reflect perturbation of either of these functions. The fact that rats with dorsolateral lesions were eventually able to learn a new spatial task suggests that the dorsolateral band is not critical for spatial perception, but it does not exclude a role for the entorhinal cortex in perceptual functions in normal animals. Moreover, any role of the dorsolateral entorhinal cortex in spatial memory is likely to depend closely on interactions with its projection areas in the dorsal hippocampus, which are critical for encoding, consolidation, and retrieval of recent spatial memory (Moser and Moser, 1998a, 1998b; Riedel et al., 1999). While converging evidence implies a role for the hippocampus in storage of recent spatial information and certain neocortical areas in storage of remote spatial information (Teng and Squire, 1999; Frankland et al., 2004; Maviel et al., 2004), the exact contribution of the entorhinal cortex in spatial computation and information storage remains a target for future study.

Modular Organization of the Entorhinal Cortex

In addition to reestablishing an essential role for the entorhinal cortex in interfacing spatial information between the neocortex and the hippocampus, the present study points to the dorsolateral band as the critical region within the entorhinal cortex for recall of spatial memory. With the exception of one animal with a small lesion, retention was blocked in all animals with lesions in the dorsolateral band. In contrast, rats with lesions in the ventromedial band consistently recalled the target position. Although a direct comparison is complicated by the generally smaller size of the ventromedial lesions, these lesions were dispersed more or less evenly within the ventromedial band (Figure 7). Although they missed the anterior tip of the band, the individual lesions jointly covered a significant share of the area of origin of the entorhinal projections to the ventral third of the hippocampal formation (Witter, 1989; Dolorfo and Amaral, 1998a). The distributed location of the lesions does not rule out the possibility that spatial memory would be disrupted by larger ventromedial lesions, regardless of their exact location, although this seems less likely in view of the observation that retention was completely abolished by lesions in the dorsolateral band that were smaller than the largest lesions in the ventromedial band. The impairment of defensive behavior in the elevated plus maze further implies that the lesion was large enough to disrupt essential elements of the circuitry of

Table 1. Stereotaxic Coordinates and Injection Volume of NMDA

Group	Anteroposterior	Mediolateral	Dorsoventral	Angle (°)	Volume (μl)
Dorsolateral	−5.2[a]	±6.8	bottom + 0.5	0	0.04
	−5.8	±6.8	bottom + 0.5	0	0.03
	−6.4	±6.7	bottom + 0.5	0	0.04
	−7.0	±6.4	bottom + 0.5	0	0.04
	−7.6	±6.4	bottom + 0.5	0	0.03
	−8.2	±5.8	7.3[b]	0	0.04
		±4.7	6.7	0	0.04
	−8.6	±5.6	7.0	0	0.04
		±4.5	6.5	0	0.04
Perirhinal/postrhinal	−5.2[a]	±6.8	7.3[b]	0	0.04
	−5.8	±6.8	7.2	0	0.04
	−6.4	±6.7	6.9	0	0.04
	−7.0	±6.4	6.9	0	0.04
	−7.6	±6.4	6.5	0	0.04
Ventromedial	+1.1[c]	±1.0	bottom + 0.3	22[d]	0.04
	+0.7	±1.0	bottom + 0.3	22	0.04
	+0.4	±1.0	bottom + 0.3	22	0.04
	+0.1	±1.0	bottom + 0.3	22	0.04
	−0.4	±1.0	bottom + 0.3	26	0.04
	−0.9	±1.0	bottom + 0.3	26	0.04
	−1.4	±1.0	bottom + 0.3	26	0.04

For the deepest injection sites, the needle was lowered to the bottom of the brain and retracted 0.5 or 0.3 mm before infusion.
[a] Relative to bregma (+ is anterior).
[b] Relative to dura at anteroposterior 1.0, mediolateral 1.0.
[c] Relative to lambda (+ is anterior).
[d] Angled in coronal plane with tip pointing laterally and ventrally.

the ventromedial band. Together, these observations suggest that the dorsolateral and ventromedial bands have different roles, as predicted from the intrinsic and extrinsic organization of the entorhinal cortex (Witter et al., 1989; Dolorfo and Amaral, 1998a, 1998b). Further studies may determine whether spatial memory requires the entire extent of the dorsolateral band, including the more ventral areas targeted in previous excitotoxic lesion studies, or only the dorsocaudal pole in which sharp position-related firing was recorded (Fyhn et al., 2004). Cutting across the medial and lateral subdivisions of the entorhinal cortex, the dorsolateral band may comprise subdivisions mediating input of very different origin to the hippocampus (Naber et al., 1997; Burwell and Amaral, 1998a, 1998b; Burwell, 2000). Recent data showing place-modulated activity in medial but not lateral entorhinal cortex (E.L. Hargreaves et al., 2002, Soc. Neurosci., abstract) suggest that spatial memory may involve mainly the medial entorhinal cortex component of the dorsolateral band.

The modular organization of the entorhinal cortex provides a rationale for the functional differentiation between dorsal and ventral parts of the hippocampus (Moser and Moser, 1998b). The effects of lesions in the dorsal and ventral hippocampus mirror those observed here after lesions in the dorsolateral and ventromedial bands of the entorhinal cortex, respectively. Spatial memory is impaired in a number of tasks by lesions of the dorsal hippocampus but not by equally sized lesions in the ventral hippocampus (Moser et al., 1993, 1995; Bannerman et al., 2002; Pothuizen et al., 2004; but see de Hoz et al., 2003 and Ferbinteanu et al., 2003). The stronger involvement of the dorsal hippocampus in spatial tasks is matched by the presence of strong location-specific firing in most pyramidal neurons in the dorsal

hippocampus as compared to the weak spatial modulation of neurons at intermediate levels (Jung et al., 1994) and the near absence of spatial modulation at the ventral pole (K.G. Kjelstrup et al., 2003, Soc. Neurosci., abstract). Lesions of the ventral pole of the hippocampus inhibit food neophobia (Bannerman et al., 2002) and attenuate avoidance of open arms on an elevated plus maze (Kjelstrup et al., 2002), whereas dorsal lesions do not, pointing to a specific role for the ventral hippocampus in avoidance of fear-associated stimuli and possibly other emotion-dependent behaviors. Together with the anatomical tracing studies, these findings suggest that differentiation along the septotemporal axis of the hippocampus is a direct consequence of the different cortical connections of these areas mediated through topographically organized modules in the entorhinal cortex (Witter et al., 1989; Dolorfo and Amaral, 1998a; Burwell, 2000).

Experimental Procedures

Subjects
A total of 124 naive male Long-Evans rats (300–450 g) were housed in groups of four to six in large transparent polycarbonate cages (59 × 38 × 20 cm) with food and water available ad libitum. They were kept on a 12 hr:12 hr light/dark schedule and tested in the dark phase.

Behavioral Training
All rats were trained in a white Morris water maze (diameter, 198 cm; height, 50 cm; water depth, 40 cm; water temperature, 23°C ± 2°C; see Moser and Moser, 1998a). One quadrant contained a remotely controlled escape platform (11 cm diameter) that could be moved between an available level (submerged 1.5 cm) and an unavailable level (submerged 21 cm). The rats received 6 days of training, each day comprising two blocks of four consecutive trials (maximal trial length 120 s; time on platform 30 s; four start positions

varied in a predetermined and pseudorandom order). Position data were collected at 50 Hz (Axona Ltd., Herts, UK). A probe trial with the platform unavailable for 60 s was conducted at the beginning of day 7. The rats were released from the pool side opposite to the platform. All rats were allowed to escape on the platform at the end of the trial to prevent extinction. Time spent in a circular zone around the platform was compared with time in corresponding zones of the other pool quadrants (zones of 35 cm radius, each covering 12% of the pool surface). Only rats that spent >20% of the search time in this zone were operated and tested further (106/124 rats). These rats were ranked, matched, and assigned to the different surgery groups according to the proportion of time they spent around the platform.

Surgery

Within 36 hr after the probe trial on day 7, the rats were anesthetized with Equithesin (pentobarbital and chloral hydrate; 1.0 ml/250 g body weight), and lesions of the entorhinal cortex were made by bilateral injection of NMDA (Sigma). NMDA was dissolved in phosphate-buffered saline (pH 7.4; 0.1 M) and injected with a 1 μl Hamilton syringe mounted to the stereotaxic frame. Lesions in the dorsolateral band of entorhinal cortex were made by infusing 0.03–0.04 μl NMDA over 10–20 s at nine stereotaxic positions in each hemisphere, using bregma, midline, and dura at anteroposterior 1.0 and mediolateral 1.0 as references for the anteroposterior, mediolateral, and dorsoventral injection coordinates, respectively (Table 1). A subset of these coordinates was used also for perirhinal and postrhinal control lesions, except that NMDA was injected more dorsally than for the entorhinal lesions (Table 1). Lesions in the ventromedial band were made by using an angled approach (to avoid hippocampal damage), with lambda and the midline as references for the anteroposterior and mediolateral positions. The syringe was angled either 22° or 26° in the coronal plane, with the needle pointing laterally and ventrally (Table 1). For both types of lesions, the syringe was lowered to each target position 2 min before the injection started. After the injection, the syringe was left in place for 7 min before it was retracted. In sham-operated rats, the syringe was lowered into the dorsal neocortex, but no NMDA was infused. A separate sham-operated control group was operated to control for mechanical damage at deeper levels caused by the needle penetrations. The coordinates used in these animals were similar to those used for dorsolateral band lesions in the entorhinal cortex. Four of the 106 rats were lost after surgery.

Retention Test and New Learning

Seven days after completion of pretraining and surgery, a second retention test was conducted. Again, the platform was kept in its lower position for the first 60 s, and the swim pattern was recorded. The platform was raised at the opposite quadrant of the pool, and the rats were then retrained with this new goal position. This phase of training consisted of one block of four trials and three subsequent blocks of two trials. The blocks were separated by 2 hr intervals. Two hours after the last block, another probe test was conducted.

Elevated Plus Maze

The maze consisted of four equally illuminated white steel arms (12 × 50 cm) radiating at square angles from a central platform (12 × 12 cm) 50 cm above the floor. The maze was placed in a silent and dimly lit room (3 × 4 min; background noise, <55 dB at 50 kHz; light intensity on open arms, 30 lux). Two opposite arms were enclosed by 40 cm high walls of white steel. The other two arms were open but had transparent plastic ledges (0.3 cm) to prevent the rats from falling. Rats were released from the central platform, with their face pointing toward an enclosed arm. An observer watching the animal's behavior on a monitor behind a curtain counted entries into open and closed arms. An entry was scored when the rat moved into the arm with all four paws. The rat had to leave the arm entirely before another entry was scored. Position was tracked at 50 Hz (Axona Ltd., Herts, UK), and time spent in each arm and in the center were calculated. After each trial, the maze was cleaned with water.

Histology

The rats were killed with an overdose of Equithesin and perfused intracardially with saline and 4% formaldehyde, and the brains were stored in formaldehyde. Frozen sections were cut coronally (30 μm) and stained with cresyl violet. Every second section from the posterior end of the cerebrum to the anterior end of the entorhinal cortex was placed under a microscope attached to a Leica DC200 camera. Images were grabbed and merged by Adobe Photoshop (version 9), and outlines of the entorhinal cortex and the lesion were traced in Photoshop or, for the illustrations, in Corel Draw (version 11).

Unfolded Maps

Unfoldings of the entorhinal cortex were prepared according to procedures described previously (Insausti et al., 1997). For convenience, the lateral border of the entorhinal cortex in each section was mapped onto a straight line, and sections were unfolded onto straight lines perpendicular to the line that represents the lateral border. Lesions were mapped onto the surface of layer II and mapped only when they included layer II or III. Lesions in layers V and VI are described in the text but were not included in the unfolding. In case the borders of the entorhinal cortex were not obvious because of lesions, we estimated them on the basis of corresponding sections in nonlesioned animals. We did not differentiate between cytoarchitectonically defined subdivisions within the entorhinal cortex.

All unfoldings were represented as individual files in Corel Draw (version 11). The outlines of all complete maps were grouped, and an average unfolding was created. On this unfolding, a schematic representation of the three parallel bands in entorhinal cortex was indicated (Figures 2 and 7). All individual maps were subsequently scaled to best fit on top of this average map by linear scaling in the x and/or y direction in Corel Draw. For each animal, the area of the lesion was expressed as a proportion of the total area of the entorhinal cortex in the rat's individual map.

References

Aggleton, J.P., Vann, S.D., Oswald, C.J., and Good, M. (2000). Identifying cortical inputs to the rat hippocampus that subserve allocentric spatial processes: a simple problem with a complex answer. Hippocampus *10*, 466–474.

Bannerman, D.M., Yee, B.K., Lemaire, M., Wilbrecht, L., Jarrard, L., Iversen, S.D., Rawlins, J.N., and Good, M.A. (2001a). The role of the entorhinal cortex in two forms of spatial learning and memory. Exp. Brain Res. *141*, 281–303.

Bannerman, D.M., Yee, B.K., Lemaire, M., Jarrard, L., Iversen, S.D., Rawlins, J.N., and Good, M.A. (2001b). Contextual fear conditioning is disrupted by lesions of the subcortical, but not entorhinal, connections to the hippocampus. Exp. Brain Res. *141*, 304–311.

Bannerman, D.M., Deacon, R.M., Offen, S., Friswell, J., Grubb, M., and Rawlins, J.N. (2002). Double dissociation of function within the hippocampus: spatial memory and hyponeophagia. Behav. Neurosci. *116*, 884–901.

Bouffard, J.P., and Jarrard, L.E. (1988). Acquisition of a complex place task in rats with selective ibotenate lesions of hippocampal formation: combined lesions of subiculum and entorhinal cortex versus hippocampus. Behav. Neurosci. *102*, 828–834.

Burwell, R.D. (2000). The parahippocampal region: corticocortical connectivity. Ann. N Y Acad. Sci. *911*, 25–42.

Burwell, R.D., and Amaral, D.G. (1998a). Cortical afferents of the

perirhinal, postrhinal, and entorhinal cortices of the rat. J. Comp. Neurol. *398*, 179–205.

Burwell, R.D., and Amaral, D.G. (1998b). Perirhinal and postrhinal cortices of the rat: interconnectivity and connections with the entorhinal cortex. J. Comp. Neurol. *391*, 293–321.

Burwell, R.D., and Hafeman, D.M. (2003). Positional firing properties of postrhinal cortex neurons. Neuroscience *119*, 577–588.

Burwell, R.D., Shapiro, M.L., O'Malley, M.T., and Eichenbaum, H. (1998). Positional firing properties of perirhinal cortex neurons. Neuroreport *9*, 3013–3018.

Burwell, R.D., Saddoris, M.P., Bucci, D.J., and Wiig, K.A. (2004). Corticohippocampal contributions to spatial and contextual learning. J. Neurosci. *24*, 3826–3836.

Cacucci, F., Lever, C., Wills, T.J., Burgess, N., and O'Keefe, J. (2004). Theta-modulated place-by-direction cells in the hippocampal formation in the rat. J. Neurosci. *24*, 8265–8277.

Cho, Y.H., and Jaffard, R. (1994). The entorhinal cortex and a delayed non-matching-to-place task in mice: emphasis on preoperative training and presentation procedure. Eur. J. Neurosci. *6*, 1265–1274.

Cho, Y.H., and Jaffard, R. (1995). Spatial location learning in mice with ibotenate lesions of entorhinal cortex or subiculum. Neurobiol. Learn. Mem. *64*, 285–290.

Cho, Y.H., and Kesner, R.P. (1996). Involvement of entorhinal cortex or parietal cortex in long-term spatial discrimination memory in rats: Retrograde amnesia. Behav. Neurosci. *110*, 436–442.

de Hoz, L., Knox, J., and Morris, R.G.M. (2003). Longitudinal axis of the hippocampus: both septal and temporal poles of the hippocampus support water maze spatial learning depending on the training protocol. Hippocampus *13*, 587–603.

de Hoz, L., Martin, S.J., and Morris, R.G.M. (2004). Forgetting, reminding, and remembering: the retrieval of lost spatial memory. PLoS Biol. 2(8): e225 DOI: 10.1371/journal.pbio.0020225.

Dolleman-Van Der Weel, M.J., and Witter, M.P. (1996). Projections from the nucleus reuniens thalami to the entorhinal cortex, hippocampal field CA1, and the subiculum in the rat arise from different populations of neurons. J. Comp. Neurol. *364*, 637–650.

Dolorfo, C.L., and Amaral, D.G. (1998a). Entorhinal cortex of the rat: topographic organization of the cells of origin of the perforant path projection to the dentate gyrus. J. Comp. Neurol. *398*, 25–48.

Dolorfo, C.L., and Amaral, D.G. (1998b). Entorhinal cortex of the rat: organization of intrinsic connections. J. Comp. Neurol. *398*, 49–82.

Eichenbaum, H. (2000). A cortical-hippocampal system for declarative memory. Nat. Rev. Neurosci. *1*, 41–50.

Ekstrom, A.D., Kahana, M.J., Caplan, J.B., Fields, T.A., Isham, E.A., Newman, E.L., and Fried, I. (2003). Cellular networks underlying human spatial navigation. Nature *425*, 184–188.

Ferbinteanu, J., Ray, C., and McDonald, R.J. (2003). Both dorsal and ventral hippocampus contribute to spatial learning in Long-Evans rats. Neurosci. Lett. *345*, 131–135.

Frankland, P.W., Bontempi, B., Talton, L.E., Kaczmarek, L., and Silva, A.J. (2004). The involvement of the anterior cingulate cortex in remote contextual fear memory. Science *304*, 881–883.

Fyhn, M., Molden, S., Witter, M.P., Moser, E.I., and Moser, M.B. (2004). Spatial representation in the entorhinal cortex. Science *305*, 1258–1264.

Galani, R., Obis, S., Coutureau, E., Jarrard, L., and Cassel, J.C. (2002). A comparison of the effects of fimbria-fornix, hippocampal, or entorhinal cortex lesions on spatial reference and working memory in rats: short versus long postsurgical recovery period. Neurobiol. Learn. Mem. *77*, 1–16.

Herkenham, M. (1978). The connections of the nucleus reuniens thalami: evidence for a direct thalamo-hippocampal pathway in the rat. J. Comp. Neurol. *177*, 589–610.

Holscher, C., and Schmidt, W.J. (1994). Quinolinic acid lesion of the rat entorhinal cortex pars medialis produces selective amnesia in allocentric working memory (WM), but not in egocentric WM. Behav. Brain Res. *63*, 187–194.

Insausti, R., Herrero, M.T., and Witter, M.P. (1997). Entorhinal cortex of the rat: cytoarchitectonic subdivisions and the origin and distribution of cortical efferents. Hippocampus *7*, 146–183.

Jarrard, L.E., Okaichi, H., Steward, O., and Goldschmidt, R.B. (1984). On the role of hippocampal connections in the performance of place and cue tasks: comparisons with damage to hippocampus. Behav. Neurosci. *98*, 946–954.

Jarrard, L.E., Davidson, T.L., and Bowring, B. (2004). Functional differentiation within the medial temporal lobe in the rat. Hippocampus *14*, 434–449.

Jung, M.W., Wiener, S.I., and McNaughton, B.L. (1994). Comparison of spatial firing characteristics of units in dorsal and ventral hippocampus of the rat. J. Neurosci. *14*, 7347–7356.

Kesner, R.P., and Giles, R. (1998). Neural circuit analysis of spatial working memory: role of pre- and parasubiculum, medial and lateral entorhinal cortex. Hippocampus *8*, 416–423.

Kjelstrup, K.G., Tuvnes, F.A., Steffenach, H.-A., Murison, R., Moser, E.I., and Moser, M.-B. (2002). Reduced fear expression after lesions of the ventral hippocampus. Proc. Natl. Acad. Sci. USA *99*, 10825–10830.

Lavenex, P., and Amaral, D.G. (2000). Hippocampal-neocortical interaction: a hierarchy of associativity. Hippocampus *10*, 420–430.

Liu, P., Jarrard, L.E., and Bilkey, D.K. (2001). Excitotoxic lesions of the pre- and parasubiculum disrupt object recognition and spatial memory processes. Behav. Neurosci. *115*, 112–124.

Liu, P., Jarrard, L.E., and Bilkey, D.K. (2004). Excitotoxic lesions of the pre- and parasubiculum disrupt the place fields of hippocampal pyramidal cells. Hippocampus *14*, 107–116.

Maguire, E.A., Burgess, N., Donnett, J.G., Frackowiak, R.S., Frith, C.D., and O'Keefe, J. (1998). Knowing where and getting there: a human navigation network. Science *280*, 921–924.

Maviel, T., Durkin, T.P., Menzaghi, F., and Bontempi, B. (2004). Sites of neocortical reorganization critical for remote spatial memory. Science *305*, 96–99.

McClelland, J.L., McNaughton, B.L., and O'Reilly, R.C. (1995). Why there are complementary learning systems in the hippocampus and neocortex: insights from the successes and failures of connectionist models of learning and memory. Psychol. Rev. *102*, 419–457.

Morris, R.G.M., Garrud, P., Rawlins, J.N.P., and O'Keefe, J. (1982). Place navigation impaired in rats with hippocampal lesions. Nature *297*, 681–683.

Moser, M.-B., and Moser, E.I. (1998a). Distributed encoding and retrieval of spatial memory in the hippocampus. J. Neurosci. *18*, 7535–7542.

Moser, M.-B., and Moser, E.I. (1998b). Functional differentiation in the hippocampus. Hippocampus *8*, 608–619.

Moser, E.I., Moser, M.-B., and Andersen, P. (1993). Spatial learning impairment parallels the magnitude of dorsal hippocampal lesions, but is hardly present following ventral lesions. J. Neurosci. *13*, 3916–3925.

Moser, M.-B., Moser, E.I., Forrest, E., Andersen, P., and Morris, R.G.M. (1995). Spatial learning with a minislab in the dorsal hippocampus. Proc. Natl. Acad. Sci. USA *92*, 9697–9701.

Naber, P.A., Caballero-Bleda, M., Jorritsma-Byham, B., and Witter, M.P. (1997). Parallel input to the hippocampal memory system through peri- and postrhinal cortices. Neuroreport *8*, 2617–2621.

Naber, P.A., Witter, M.P., and Lopez da Silva, F.H. (1999). Perirhinal cortex input to the hippocampus in the rat: evidence for parallel pathways, both direct and indirect. A combined physiological and anatomical study. Eur. J. Neurosci. *11*, 4119–4133.

Naber, P.A., Witter, M.P., and Lopes da Silva, F.H. (2001). Evidence for a direct projection from the postrhinal cortex to the subiculum in the rat. Hippocampus *11*, 105–117.

O'Keefe, J., and Nadel, L. (1978). The Hippocampus as a Cognitive Map (Oxford: Clarendon Press).

Olton, D.S., Walker, J.A., and Gage, F.H. (1978). Hippocampal connections and spatial discrimination. Brain Res. *139*, 295–308.

Oswald, C.J., Bannerman, D.M., Yee, B.K., Rawlins, J.N.P., Honey, R.C., and Good, M. (2003). Entorhinal cortex lesions disrupt the

transition between the use of intra- and extramaze cues for navigation in the water maze. Behav. Neurosci. *117*, 588–595.

Pellow, S., Chopin, P., File, S.E., and Briley, M. (1985). Validation of open:closed arm entries in an elevated plus-maze as a measure of anxiety in the rat. J. Neurosci. Methods *14*, 149–167.

Pothuizen, H.H., Zhang, W.N., Jongen-Relo, A.L., Feldon, J., and Yee, B.K. (2004). Dissociation of function between the dorsal and the ventral hippocampus in spatial learning abilities of the rat: a within-subject, within-task comparison of reference and working spatial memory. Eur. J. Neurosci. *19*, 705–712.

Pouzet, B., Welzl, H., Gubler, M.K., Broersen, L., Veenman, C.L., Feldon, J., Rawlins, J.N.P., and Yee, B.K. (1999). The effects of NMDA-induced retrohippocampal lesions on performance of four spatial memory tasks known to be sensitive to hippocampal damage in the rat. Eur. J. Neurosci. *11*, 123–140.

Rasmussen, M., Barnes, C.A., and McNaughton, B.L. (1989). A systematic test of cognitive mapping, working-memory, and temporal discontiguity theories of hippocampal function. Psychobiol. *17*, 335–348.

Riedel, G., Micheau, J., Lam, A.G., Roloff, E.L., Martin, S.J., Bridge, H., de Hoz, L., Poeschel, B., McCulloch, J., and Morris, R.G.M. (1999). Reversible neural inactivation reveals hippocampal participation in several memory processes. Nat. Neurosci. *2*, 898–905.

Schenk, F., and Morris, R.G.M. (1985). Dissociation between components of spatial memory in rats after recovery from the effects of retrohippocampal lesions. Exp. Brain Res. *58*, 11–28.

Scoville, W.B., and Milner, B. (1957). Loss of recent memory after bilateral hippocampal lesions. J. Neurol. Neurosurg. Psychiatry *20*, 11–21.

Squire, L.R. (1992). Memory and the hippocampus: a synthesis from findings with rats, monkeys, and humans. Psychol. Rev. *99*, 195–231.

Squire, L.R., and Alvarez, P. (1995). Retrograde amnesia and memory consolidation: a neurobiological perspective. Curr. Opin. Neurobiol. *5*, 169–177.

Taube, J.S. (1995). Place cells recorded in the parasubiculum of freely moving rats. Hippocampus *5*, 569–583.

Taube, J.S., Muller, R.U., and Ranck, J.B., Jr. (1990). Head-direction cells recorded from the postsubiculum in freely moving rats. I. Description and quantitative analysis. J. Neurosci. *10*, 420–435.

Teng, E., and Squire, L.R. (1999). Memory for places learned long ago is intact after hippocampal damage. Nature *400*, 675–677.

Witter, M.P. (1989). Connectivity of the rat hippocampus. In Neurology and Neurobiology, Volume 52: The Hippocampus—New Vistas, V. Chan-Palay and C. Köhler, eds. (New York: Alan Liss, Inc.), pp. 53–69.

Witter, M.P., and Amaral, D.G. (2004). The hippocampal formation. In The Rat Nervous System, Third Edition, G. Paxinos, ed. (San Diego, CA: Elsevier Academic Press), pp. 637–703.

Witter, M.P., Holtrop, R., and van de Loosdrecht, A.A. (1988). Direct projections from the periallocortical subicular complex to the fascia dentata in the rat: An anatomical tracing study using phaseolus vulgaris leucoagglutinin. Neurosci. Res. Commun. *2*, 61–68.

Witter, M.P., Groenewegen, H.J., Lopes da Silva, F.H., and Lohman, A.H.M. (1989). Functional organization of the extrinsic and intrinsic circuitry of the parahippocampal region. Prog. Neurobiol. *33*, 161–254.

Wouterlood, F.G., Saldana, E., and Witter, M.P. (1990). Projection from the nucleus reuniens thalami to the hippocampal region: light and electron microscopic tracing study in the rat with the anterograde tracer Phaseolus vulgaris-leucoagglutinin. J. Comp. Neurol. *296*, 179–203.