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Unstable CA1 place cell representation in rats with entorhinal cortex lesions

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Abstract

Recent studies emphasize the importance of the entorhinal cortex in spatial representation and navigation. Furthermore, evidence is accumulating to show that spatial processing depends on interactions between the entorhinal cortex and the hippocampus. To investigate these interactions, we examined the effects of entorhinal cortex lesions on the activity of hippocampal CA1 place cells. Rats received bilateral radiofrequency lesions of the entorhinal cortex or sham lesions before place cell recording. Place cells were recorded as the rats performed a pellet-chasing task in a cylinder containing three cue-objects. Entorhinal cortex lesions did not abolish place cell spatial firing but reduced noticeably discharge rate and field size. Most importantly, the lesions affected firing field stability when cells were recorded both in constant conditions and following cue manipulations (object rotation, object removal). These findings indicate that the entorhinal cortex is necessary for the stability of hippocampal representations across exposures to a familiar environment. Consistent with the recent discovery of grid cells in the medial entorhinal cortex, our results suggest that the entorhinal cortex contributes to providing a spatial framework that would enable the hippocampus to maintain stable environment-specific representations.

Introduction

Spatial navigation in mammals involves a large network of brain structures that are thought to play specific roles. Within this network, the hippocampus is considered to have a central role based on the existence, in rodents, of place cells in the CA1 and CA3 regions. The most conspicuous correlate of place cell firing is the animal's location in space. Each place cell is intensely active when a rat's head is in a cell-specific part of the environment called the firing field (e.g. O'Keefe & Dostrovsky, 1971; Muller & Kubie, 1987). Firing fields can be seen in all regions of the environment accessible to the rat so that place cells might provide the neural substrate of spatial representations or 'cognitive maps' (O'Keefe & Nadel, 1978).

Place cell firing has been shown to be modulated by a variety of sensory information including external (allothetic) and internal (idiothetic) information (O'Keefe & Conway, 1978; Muller & Kubie, 1987; Gothard *et al*., 1996; Save *et al*., 2000). Because sensory inputs to the hippocampus result from cortical processing, a crucial issue is to determine how cortical areas contribute to establish and/or modulate place cell firing properties. Among these areas, the entorhinal cortex may play a pivotal role because it is a relay for most cortical inputs and outputs of the hippocampal formation. Polymodal sensory inputs reach the entorhinal cortex via projections from the perirhinal, postrhinal, parietal, piriform and insular cortices (Burwell & Amaral, 1998; Sewards & Sewards, 2003). The entorhinal cortex sends projections to the hippocampus via the perforant pathways. It projects both directly and indirectly (via the trisynaptic loop) to the dentate gyrus, CA3, CA1 and subiculum. Return projections from the hippocampus to the entorhinal cortex originate in CA1 and the subiculum (Witter *et al*., 2000). This pattern of connectivity strongly suggests that the entorhinal cortex provides an essential input to the place cell system for establishing and maintaining location-specific firing. Supporting this hypothesis, it has been shown that direct entorhinal–CA1 circuitry can maintain place cell activity when the CA3–CA1 circuit is interrupted (Brun *et al*., 2002).

Electrophysiological evidence is also consistent with this hypothesis. Principal cells with location-specific firing have been recorded in the entorhinal cortex (Quirk *et al*., 1992; Fyhn *et al*., 2004). Recently, 'grid cells' were found in the medial entorhinal cortex that display location-specific firing in the form of a structure of firing fields repeating at regular intervals, i.e. a grid-like pattern, over the whole environment (Hafting *et al*., 2005). Although the function of grid cells and their relationships with place cells are still unclear, these results show that location-specific firing is already established upstream from the hippocampus.

There is little direct evidence of the interaction between the hippocampus and the entorhinal cortex. The objective of the present study was therefore to examine the contribution of the entorhinal cortex to place cell firing by recording place cells in rats with bilateral lesions of the entorhinal cortex. By making these lesions, it was critical not to damage neighboring regions, such as subiculum, perirhinal cortex or postrhinal cortex, that may also be involved in place cell firing. Because complete damage to the entorhinal cortex is difficult to accomplish without damaging the neighboring structures, we tried to produce extensive lesions that were limited as much as possible to the entorhinal cortex even if they were not complete. Our first question was whether such lesions would abolish location-specific

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firing in the hippocampus. Because we found preservation of place cell activity in entorhinal cortex-damaged rats, we then asked whether entorhinal lesions affect the ability of place cells to use environmental or idiothetic cues to maintain stable firing fields.

Materials and methods

Subjects

Eleven naive Long-Evans male rats, purchased from a commercial supplier (Janvier, Le Genest-St-Isles, France), were housed in individual cages (40 cm long × 26 cm wide × 16 cm high) with food and water available *ad libitum* and kept in a temperature-controlled room (20 ± 2 °C) with natural light/dark cycle. One week after arrival, animals were handled daily by the experimenter for 5 days. They were then submitted to a progressive food deprivation schedule until they reached 85% of their initial body weight and trained to forage for 20-mg food pellets (pellet-chasing task) in the recording cylinder. Following training, surgery for lesion and electrode implantation was made. Rats were assigned to entorhinal cortex lesion (ENTO, $n = 5$) or sham-lesioned (CONT, $n = 6$) groups.

Surgery

Lesion and electrode implantation were performed during the same surgery. Rats were first injected with atropine sulfate (0.25 mg/kg, i.p.) and then deeply anesthetized by injection of sodium pentobarbital (40 mg/kg i.p., Sanofi Santé Animal, Libourne, France). Additional injections of ketamine (50 mg/kg i.p., Imalgène, Merial, France) could be made to maintain appropriate anesthesia throughout surgery. The rats were placed in a Kopf stereotaxic apparatus (Kopf Instruments, Tujunga, CA, USA).

Entorhinal cortex lesions

We used a technique similar to that in previous studies (Parron & Save, 2004a,b; Parron *et al*., 2004). A midline incision of the scalp was made and the skin and muscles were carefully retracted to expose the skull. Holes were drilled above the target regions. Bilateral lesions of the entorhinal cortex were made by passing a radio-frequency current at the tip of an electrode (70 °C for 15 s; RFG 4 model, Radionics, Burlington, MA, USA) lowered in the brain at the following coordinates relative to bregma: AP: −6.8 mm, L: ±4.3 mm and ±5.4 mm; and AP: −8 mm, L: ±5 mm (Zilles, 1985). For each lesion point, the electrode was lowered very slowly until its tip touched the floor of the brain or calvarium. As soon as the electrode slipped slightly in the antero-posterior axis, lowering was stopped and the electrode was raised 1 mm (Cho & Kesner, 1996). This position was taken as the dorso-ventral coordinate of the lesion. Due to the antero-posterior curvature of the calvarium, this coordinate was different for each lesion point, thus allowing us to damage the entorhinal cortex along its entire extent. Sham-lesioned rats were treated in the same way as lesioned rats except that no current was passed through the electrode.

Electrode implantation

Electrodes were ten 25-μm nichrome wires forming a bundle that was inserted into a 30G stainless steel guide cannula secured to a pin of a 12-pin Mill-Max connector. Each wire was connected to a different pin on the connector. Three drive screws with nylon cuffs were attached to the connector with acrylic. When implanted, the electrodes and the cannula could be moved down in the brain by screwing the screws into the nylon cuffs. Implantation was made after entorhinal cortex lesions. Three miniature screws and a T-shaped screw were fixed in the skull to anchor the electrode carrier. A hole for the electrodes was then drilled above the target area. The tips of the electrodes were implanted above the dorsal hippocampus at the following coordinates relative to bregma: AP: −3.8 mm, L: −3.0 mm, DV: −1.5 mm (Paxinos & Watson, 2004). The bottom (nylon cuffs) of the three drive screws assembly was then cemented to the skull. The rats were sutured and received injections of an antibiotic (terramycine, 60 mg/kg, i.m., Pfizer, Paris, France) and an analgesic (tolfedine, 0.06 mg/kg, s.c., Vetoquinol, Lure, France). The animals were allowed to recover for 10 days before cell screening started.

At the completion of the experiment, lesioned and sham-lesioned rats received a lethal dose of sodium pentobarbital and were transcardially perfused with a 10% formalin solution. The brains were then removed and stored in a 4% formalin solution. Coronal 20-μm tissue sections were cut at −20 °C using a microtome cryostat (Leica CM3050) at the level of entorhinal cortex. Every second section was mounted and stained with cresyl violet. The slides were observed under the microscope to determine the lesion extent.

All treatments were performed in accordance with the NIH guide for the care and use of laboratory animals (NIH publication no. 86-23, revised 1987), European guidelines (European Community Council Directive, 24 November 1986, 86/609/EEC) and National guidelines (Council directive no. 87848 of the Direction des Services Vétérinaires de la Santé et de la Protection Animale permission no. 13.24 from the Ministère de l'Agriculture et de la Pêche to E.S.).

Recording methods

After 10 days of postsurgical recovery, the electrodes were lowered (by steps of 25–50 μm) over the course of several days, as the rats performed the pellet-chasing task. Recordings were performed with a Datawave Acquisition System (DataWave, Longmont, CO, USA). Signals were amplified 10 000 times, band-pass filtered between 0.3 and 10 kHz, and sent to a 250 kHz analog-to-digital (A/D) board in a PC computer. Waveforms of identified units were sampled at 32 kHz and stored. The headstage possessed an LED for tracking the rat's head position. The LED was tracked at 50 Hz with a digital spot follower that received RGB signals from a CCD colour camera fixed above the apparatus. The LED was detected in a grid of 256 × 256 square regions (pixels), which was reduced at analysis stage to a grid of 64 × 64 pixels, 25 mm on a side.

Apparatus

The recording apparatus was similar to that used in previous studies (Save *et al*., 1998, 2005; Paz-Villagrán *et al*., 2002). Briefly, it was a dark grey cylinder with a grey floor (50 cm high, 76 cm diameter), visually isolated from the rest of the laboratory by a cylindrical curtain (250 cm diameter). The apparatus was lit by indirect light provided by four 25-W bulbs placed on the ceiling in a 60 × 60-cm square arrangement. During all phases of the study, a radio tuned to an FM station was fixed to the ceiling in a central position relative to the cylinder, producing background music >70 dB to mask non-controlled directional sounds. Three objects differing from each other in colour, size, shape and texture were used as intramaze cues: a glass bottle (29 cm high), a white plastic cylinder (21 cm high) and a black wooden cone (20 cm high). Their locations were fixed relative to each other. Each object was placed against the wall of the cylinder, and their arrangement formed an isosceles triangle (Fig. 1).

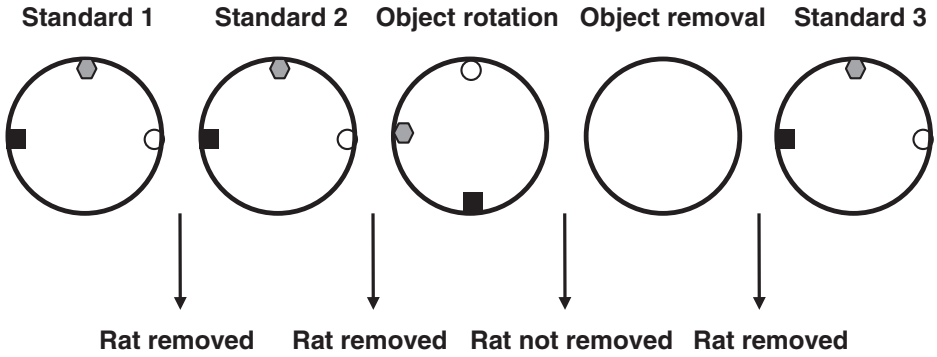


FIG. 1. Experimental protocol. Place cells were recorded in five successive 16-min sessions. The rat was removed between each session except between Object Rotation and Object Removal sessions.

The computer and the monitoring and recording equipment were located in a room adjacent to the room containing the cylinder.

Experimental procedure

Each day, the electrodes were screened as the rats performed the pellet-chasing task. Once unit waveforms of sufficient amplitude (> 100 μV, i.e. approximately three times the background noise) were isolated, a sequence of five 16-min recording sessions separated by 5–10-min intervals was run (Fig. 1). In the first standard session (STD1), the object cues were at the locations used during screening, allowing for the establishment of initial firing field location and characteristics. The second standard session (STD2) was similar to STD1 and was conducted to check for firing field stability in constant conditions. The third session (Object Rotation) was run with the three objects rotated 90° counterclockwise as a rigid set around the arena center, to examine whether they controlled firing field location. At the end of the Object Rotation session, the objects were removed while the animal remained in the apparatus. A fourth session (Object Removal) was then run to examine firing field stability in the absence of the objects. Previous work has shown that firing fields remain stable after the objects are removed, suggesting the contribution of olfactory and idiothetic cues (Save *et al*., 2000). Occasionally (see Results), a last standard session (STD3) was run to check for signal consistency.

Between sessions (except between Object Rotation and Object Removal), the rat was disconnected, placed back in its home cage and the floor was cleaned with a wet sponge to neutralize olfactory cues. The home cage was located in the adjacent room containing the recording equipment. At the end of the intersession interval, the experimenter took the rat in the hand and brought it to the curtained environment from one of four possible entrances. The animal was then connected to the recording system and introduced in the cylinder from different starting positions on each session.

Data analysis

Behavioral analysis

For each recording session, object exploration was estimated by measuring the overall time spent by the animal in a 10-cm-diameter area centered on the objects during STD1. The distance run during the whole session and the mean running speed were also measured.

Unit analysis

As our purpose was to study the effect of entorhinal cortex lesions on hippocampal place cells, only well-isolated cells with clear location-specific activity were included in the data set. Interneurons were not taken into account. The first step in off-line analyses was to refine boundaries for waveform clusters that were defined before recording. Candidate waveforms were discriminated using Datawave off-line sorting software, which allows waveform separation based on at least eight spike voltage parameters, including spike amplitude, spike duration, maximum and minimum spike voltage, and the time of occurrence of maximum and minimum spike voltages. It was also possible to discriminate according to the voltage at experimenter-defined times of the waveforms. Waveforms were then processed with a Plexon offline sorter (Dallas, TX, USA) to refine cluster boundaries, remove outlier waveforms, and calculate sorting statistics and autocorrelation functions. Inter-spike interval histograms were built for each unit and the whole unit was removed from analysis if the histogram revealed the existence of interspike intervals < 2 ms (refractory period), inconsistent with good isolation. Only waveforms of sufficient amplitude (> 100 μV) were further analysed. The cluster boundaries established for the first session were used for subsequent sessions.

Once single units were well separated, we calculated the positional firing rate distribution. The total time the light was detected (dwell time) and the total number of spikes in each pixel were accumulated for the session duration (16 min). Dividing the total number of spikes by the dwell time in each pixel allowed us to construct a firing rate map for the session (Muller *et al*., 1987). In such maps, pixels that were not visited by the rat are displayed as white and pixels that were visited, but in which no spike occurred during the session, are displayed as light grey. The firing rate is coded as levels of grey from low (light grey) to high (dark grey). The values used as boundaries between categories were determined for STD1 and applied to subsequent sessions of a recording sequence to allow for comparison between these sessions for a given cell.

A firing field was defined as a set of at least nine pixels contiguous on a side with a firing rate above the mean (i.e. above the total number of spikes divided by the session duration). Several numerical measures were used to describe the positional firing patterns: (i) the in-field mean firing rate was the total number of spikes emitted by the cell while the rat was in the firing field divided by the total time spent in the field; (ii) the in-field peak firing rate was defined as the mean between the pixel with the highest firing rate and the eight surrounding pixels; (iii) the field size in pixels; (iv) the spatial coherence was a computed autocorrelation between the rate for each pixel and the average rate of the eight neighboring pixels – it measures the local smoothness of firing rate contours and is a way to quantify the strength of spatial firing for a cell (Muller & Kubie, 1989); and (v) the information content measured the amount of information (in bits)

conveyed about spatial location by a single action potential emitted by a single cell (Markus *et al.*, 1994). This was calculated according to the formula:

$$I = \sum_i (\lambda_i/\lambda) \times \log_2(\lambda_i/\lambda) \times P_i$$

where λ_i is the mean firing rate in each pixel, λ is the overall mean firing rate, and P_i is the probability of the animal to be in pixel i (i.e. dwelling time in pixel/total dwelling time). The minimal value of positional information content is 0 for a cell which does not provide any information about location.

To estimate the stability of firing fields between sessions, pixel-by-pixel cross-correlations were made between pairs of firing rate arrays. First, the correlation between the two firing rate arrays (R_0) was used as a measure of similarity between firing fields (similarity score; see Paz-Villagràn *et al.*, 2004). A high similarity score indicated a great similarity between the two firing rate arrays. Second, we calculated a cross-correlation as the firing rate array of the first session was rotated in 6° steps relative to the firing rate array of the second session. The angle associated with the highest correlation (R_{max}) was taken as the rotation angle of the firing field between the two sessions or session halves. We considered firing fields to be spatially stable when R_{max} matched R_0, therefore corresponding to a 0° (± 20°) rotation angle.

For within-session comparisons, correlations were made between the firing rate arrays of the two session halves. To normalize the distribution, each R_{max} value was transformed into a Z_{max} score. These Z_{max} scores were used for calculation of the means and standard errors and for statistical analyses.

Results

Histology

Figure 2 displays a series of coronal planes (adapted from Paxinos & Watson, 2004) showing the extent of entorhinal cortical lesion in the five rats. In all rats, the medial regions of the entorhinal cortex including the caudomedial, medial and ventro-intermediate regions (Paxinos & Watson, 2004) were extensively damaged and the lateral regions including the dorsolateral, and lateral entorhinal cortex were partially damaged. Laterally, marginal damage to the perirhinal cortex was found in rats E1, E2 and E4 at the most dorsal levels. The lesion

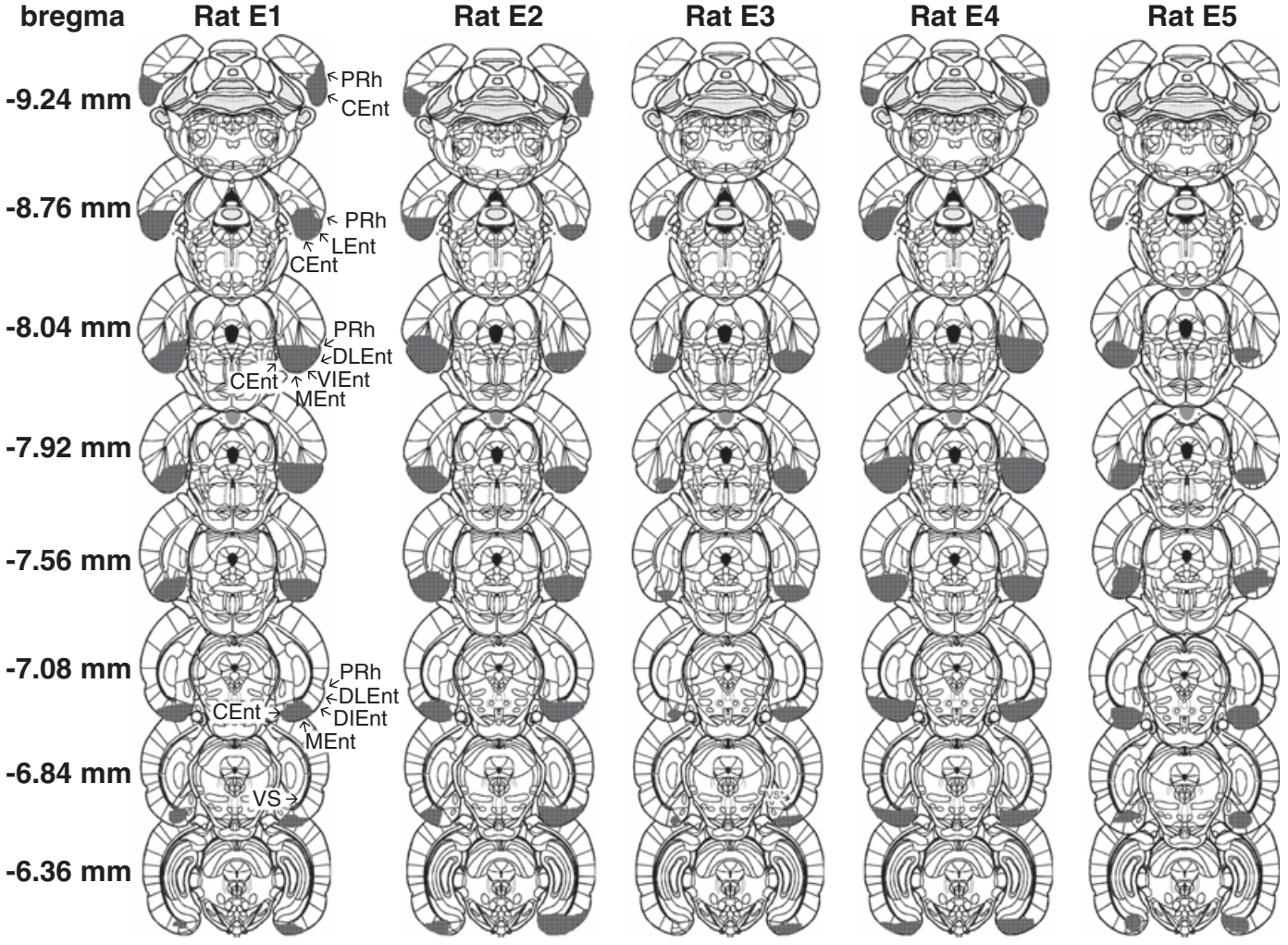


FIG. 2. Coronal planes (adapted from Paxinos & Watson, 2004) showing entorhinal cortex lesion extent for all five rats (in dark grey). Coordinates (in mm) are relative to Bregma. Abbreviations: PRh, perirhinal cortex; Cent, caudomedial entorhinal cortex; Lent, lateral entorhinal cortex; DLEnt, dorsalateral entorhinal cortex; VIEnt, ventral intermediate entorhinal cortex; Ment, medial entorhinal cortex; DIEnt, dorsal intermediate entorhinal cortex; VS, ventral subiculum (Paxinos & Watson, 2004).

slightly encroached on the ventral subiculum at the most ventral levels in rats E2, E4 and E5. No damage to the presubiculum and parasubiculum was found. In no case did the lesions invade the hippocampus. The magnitude of lesions was fairly similar in rats E1, E2, E4 and E5. Rat E3 had a smaller lesion of the entorhinal cortex contralateral to the recorded side. Note, however, that the lesion of the entorhinal cortex ipsilateral to the recorded hippocampus was as extended as in the other rats.

Behaviour

During STD1, ENTO rats explored more objects than CONT rats (mean object exploration: ENTO, 185.0 s; CONT, 145.4 s, $t_{70} = 2.56$, $P < 0.05$) although they did not exhibit greater locomotor activity (mean distance run: ENTO, 47.4 m; CONT, 37.2 m, $t_{70} = 1.39$, $P > 0.05$; Mean running speed: ENTO, 4.9 cm/s; CONT, 3.9 cm/s, $t_{70} = 1.34$, $P > 0.05$).

Place cell activity

A total of 125 putative place cells with complex burst firing were recorded from CA1 in 11 rats. Eighty-five cells were from five ENTO rats (E1: four cells, E2: 23 cells, E3: 45 cells, E4: four cells, E5: nine cells) and 40 cells were recorded from six CONT rats (C1: 17 cells, C2: three cells, C3: one cell, C4: 12 cells, C5: four cells, C6: three cells). The data from the controls confirmed our previous findings using the same protocol (Cressant *et al*., 1997; Save *et al*., 1998, 2005).

Basic firing characteristics (STD1)

Table 1 displays the basic firing characteristics in ENTO and CONT groups. Place cell discharge was weaker in ENTO rats than in CONT rats, as shown by several measures of firing, including mean firing rate ($t_{123} = 3.70$, $P < 0.001$), in-field firing rate ($t_{123} = 2.45$, $P < 0.05$) and peak firing rate ($t_{123} = 2.26$, $P < 0.05$). In addition, fields were noticeably smaller in ENTO rats (CONT: 51.1 pixels, ENTO: 33.0 pixels, $t_{123} = 3.44$, $P < 0.001$). In contrast, there was no difference in measures of information content ($t_{123} = 0.13$, $P > 0.05$) and coherence ($t_{123} = 1.10$, $P > 0.05$), suggesting that spatial selectivity was not affected by the lesion (Table 1).

We also calculated a parameter similar to that used by Miller & Best (1980), reflecting the saliency or robustness of spatial firing and corresponding to the difference between the field rate and the mean rate divided by the mean rate. However, unlike Miller and Best, we found no significant difference between the two groups ($t_{123} = 0.859$, $P = 0.39$).

Constant conditions (STD1–STD2)

Fifty pairs of standard sessions (STD1–STD2) were recorded in ENTO rats (85 cells, 1.7 cells per session) and 21 pairs were recorded in CONT rats (40 cells, 1.9 cells per session). We found that 17/85 cells ceased firing or started to fire in STD2 in ENTO rats. This outcome never occurred in CONT rats. For cells that still had a field in STD2, the distribution of the similarity scores (R_0) between STD1 and STD2 was clearly different in the two groups (Fig. 3).

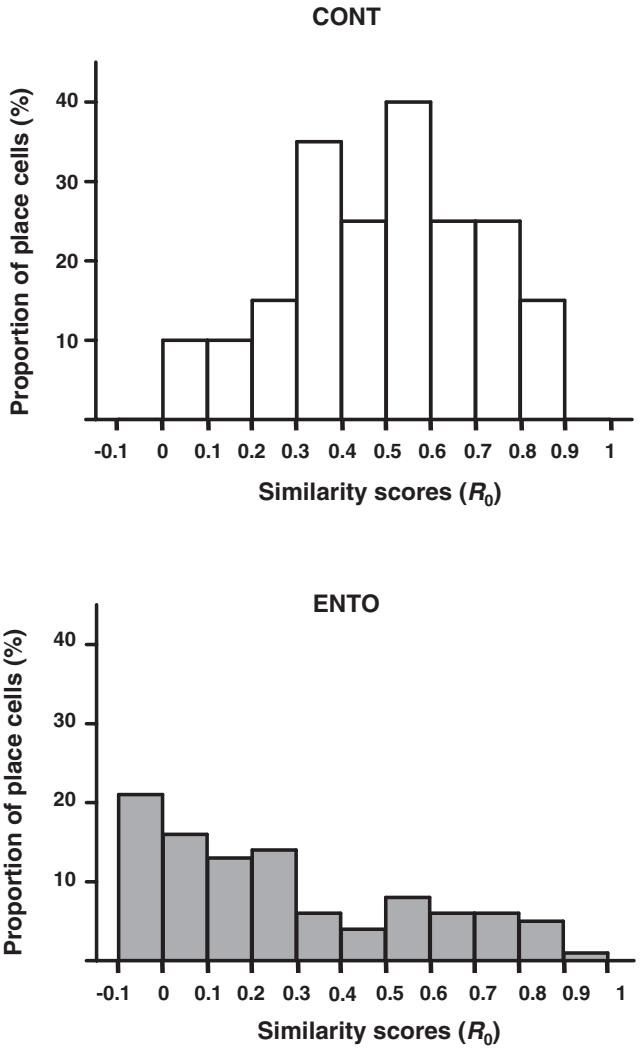


FIG. 3. Distribution of similarity scores between STD1 and STD2 in ENTO and CONT groups.

Although the high similarity scores in CONT rats (mean $R_0 = 0.590 \pm 0.05$) suggested that firing fields were stable in the two standard sessions, the distribution in ENTO rats was displaced to the left, suggesting that fields had changed between the two sessions (mean $R_0 = 0.280 \pm 0.03$; CONT vs. ENTO: $t_{123} = 3.08$, $P < 0.01$). This finding was further confirmed by rotational cross-correlation analyses (see Methods), which revealed that a substantial proportion of fields in ENTO rats had an R_{max} departing from R_0 (for a rotation

TABLE 1. Firing parameters of place cells in CONT and ENTO rats

	Number of cells	Firing rate (Hz)	In-field mean firing rate (Hz)	In-field peak firing rate (Hz)	Coherence	Information content	Field size (pixels)
CONT	40	0.46 ± 0.07**	2.64 ± 0.41*	5.84 ± 1.18*	0.62 ± 0.03	1.70 ± 0.10	49.70 ± 5.37*
ENTO	85	0.23 ± 0.02	1.70 ± 0.15	3.57 ± 0.38	0.63 ± 0.03	1.64 ±0.07	33.01 ± 2.33

Data are presented as mean ± SEM. Asterisks indicate significant differences: *$P < 0.01$ and **$P < 0.001$ (t-tests for independent samples).

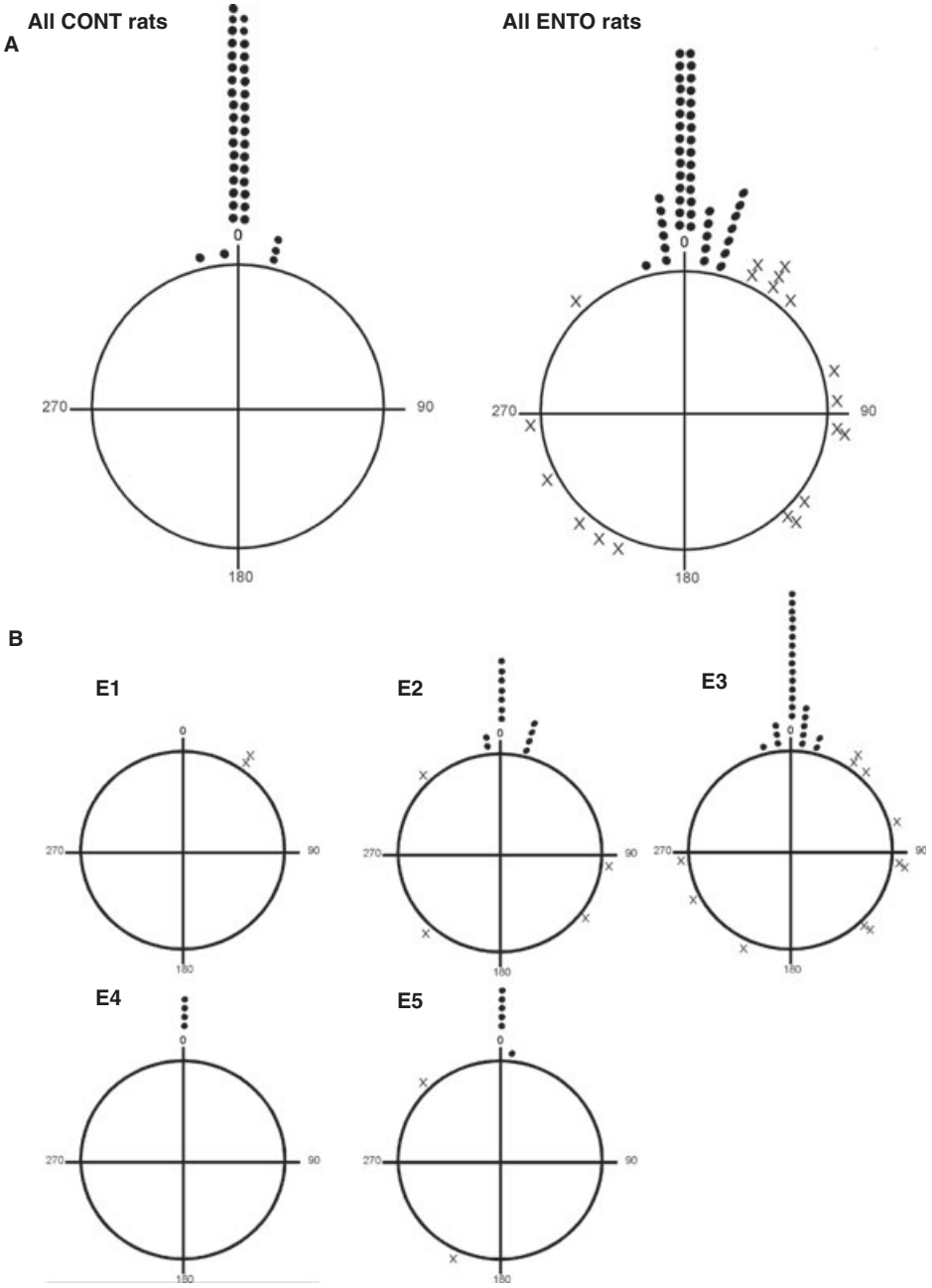


FIG. 4. (A) Circular distribution in 10° bins of the rotation angles of firing fields corresponding to R_{max} between STD1 and STD2 in ENTO and CONT rats. The black circles correspond to rotation angles of fields that were stable and the crosses fields that changed location. (B) Circular distribution in 10° bins of the rotation angles of firing fields corresponding to R_{max} between STD1 and STD2 for each ENTO rat.

angle greater than 20°). Visual inspection of the rate maps was consistent with this quantitative analysis. Figure 4 shows the distribution of field rotation angles between STD1 and STD2 in all ENTO and CONT rats and for each ENTO rat. As shown in Table 2, in ENTO rats 49/85 cells had stable fields (similarity score for these cells: $R_0 = 0.561 \pm 0.070$) and 19/85 cells had shifted fields. The remaining cells ceased firing. There was no difference in similarity scores for stable fields between ENTO and CONT groups ($t_{87} = 0.333$, $P > 0.05$). Figure 5 displays examples of firing rate maps in CONT and ENTO rats.

That many cells in ENTO rats had fields that changed between the two standard sessions or that stopped firing was indicative of a

TABLE 2. Number of cells in the three categories of response after the transition between STD1 and STD2 for each entorhinal rat

	Stable fields	Shifted fields	Stop firing
Rat E1	0	2	2
Rat E2	13	4	6
Rat E3	27	11	7
Rat E4	4	0	0
Rat E5	5	2	2
All rats ($n = 85$ cells)	49	19	17

A large proportion (42%) of fields remapped (shifted or disappeared). In contrast, all cells in CONT rats had stable fields.

remapping. Thus, we first asked if intrinsic properties could explain the propensity of some cells in ENTO rats to remap. We compared the firing characteristics (mean firing rate, information content, coherence, field size, in-field firing rate, in-field peak firing rate) of stable fields and fields that changed in the ENTO group in the two standard sessions, none of which revealed significant differences (*t*-tests, all P values >0.05), indicating that cells whose fields changed did not differ from cells with stable fields in terms of firing characteristics.

Remapping can be of different types: 'global', 'rotational' or 'rate' remapping (Leutgeb *et al*., 2005). In global remapping, fields shift to unpredictable angular and radial positions, and cells may stop firing after previously being active or become active after being previously inactive. In rotational remapping, fields rotate to unpredictable angular positions while their size, shape and radial positions are unchanged (Muller & Kubie, 1987). In rate remapping, the firing rate of place cells varies but field locations remain constant (Leutgeb *et al*., 2005). A number of cells stopped firing in the present study, suggesting a global remapping. However, because entorhinal cortex lesions may produce complex effects on place cell firing, we also looked at possible 'rotational' or 'rate' remapping in cells that had a field in both STD1 and STD2 ($n = 68$).

We compared R_{max} for cells that shifted field location to the similarity scores (R_0) for cells with stable fields, hypothesizing that if they are similar, this would indicate a rotational remapping. This hypothesis was not confirmed, however, as R_{max} was lower than the similarity score of stable fields (maximal angular correlation: $R_{max} = 0.25 \pm 0.03$, similarity scores: $R_0 = 0.56 \pm 0.07$; $t_{66} = 2.89$, $P < 0.01$). We also analysed the distribution of the rotation angles for these fields (shown as crosses in Fig. 4B) and found no evidence for a consistent bias, therefore suggesting that fields shifted to an unpredictable location (Watson U^2 test: $n = 19$; $U^2 = 0.069$; $P > 0.05$). Rate remapping was examined for fields whose location was stable in STD1 and STD2. A comparison of the firing rate in STD2 and STD1 failed to reveal a significant change (STD1: 0.22 ± 0.02 Hz, STD2: 0.21 ± 0.03 Hz; $t_{48} = 0.238$, $P > 0.05$). We also compared ENTO and CONT rats using the normalized rate difference between the two sessions (high − low/high + low, Leutgeb *et al*., 2005; McHugh *et al*., 2007) but no significant difference was found ($t_{87} = 0.73$, $P > 0.05$). In addition, no difference was found for the other firing parameters (information content, coherence, field size, in-field firing rate, in-field peak firing rate, all P values >0.05).

Thus, the results do not support the hypothesis of rotational or rate remapping but rather suggest that entorhinal cortex lesions produced a global remapping. In constant conditions, 42% of the cells in ENTO rats remapped, including 22% that had a field that shifted to an unpredictable location (Similarity score: $R_0 = 0.074 \pm 0.02$) and 20% that stopped firing. We tested the possibility that cells that stopped firing in STD2 needed more time to be reactivated. We thus extended the duration of STD2 session up to 64 min for a few (five) recordings. In no case, however, did cells resume firing.

It is also possible that ENTO rats would be merely slower than CONT rats to establish a stable representation of the environment. According to this hypothesis, ENTO rats would display more instability in the early recording sequences and more stability in the last sequences. Figure 6 shows the percentage of remapping as a function of training in ENTO rats and indicates that place cells displayed remapping responses throughout the experiment. Thus, instability does not appear to be related to delayed spatial learning abilities, at least within the range of study days used in this experiment.

Overall, while 100% of the fields were stable between STD1 and STD2 in CONT rats, only 58% (49/85) were stable in ENTO rats ($\chi^2_1 = 23.73$, $P < 0.001$). Interestingly, simultaneously recorded cells in ENTO rats displayed coherent responses in all sessions (see all simultaneously recorded cells in the Supplementary material, Fig. S1).

Finally, we investigated if the occurrence of field instability was linked to alterations of locomotion and object exploration. This was possible because all cells in a sequence produced coherent responses. We thus compared the behavior of ENTO rats (distance run, running speed, object exploration) in STD1 and STD2 separately for stable fields and for fields that changed between sessions. The results did not reveal any difference, however. In particular, no specific alteration of locomotion and exploratory activity was found for sessions in which fields were changed in STD2 relative to STD1 (all P values >0.05).

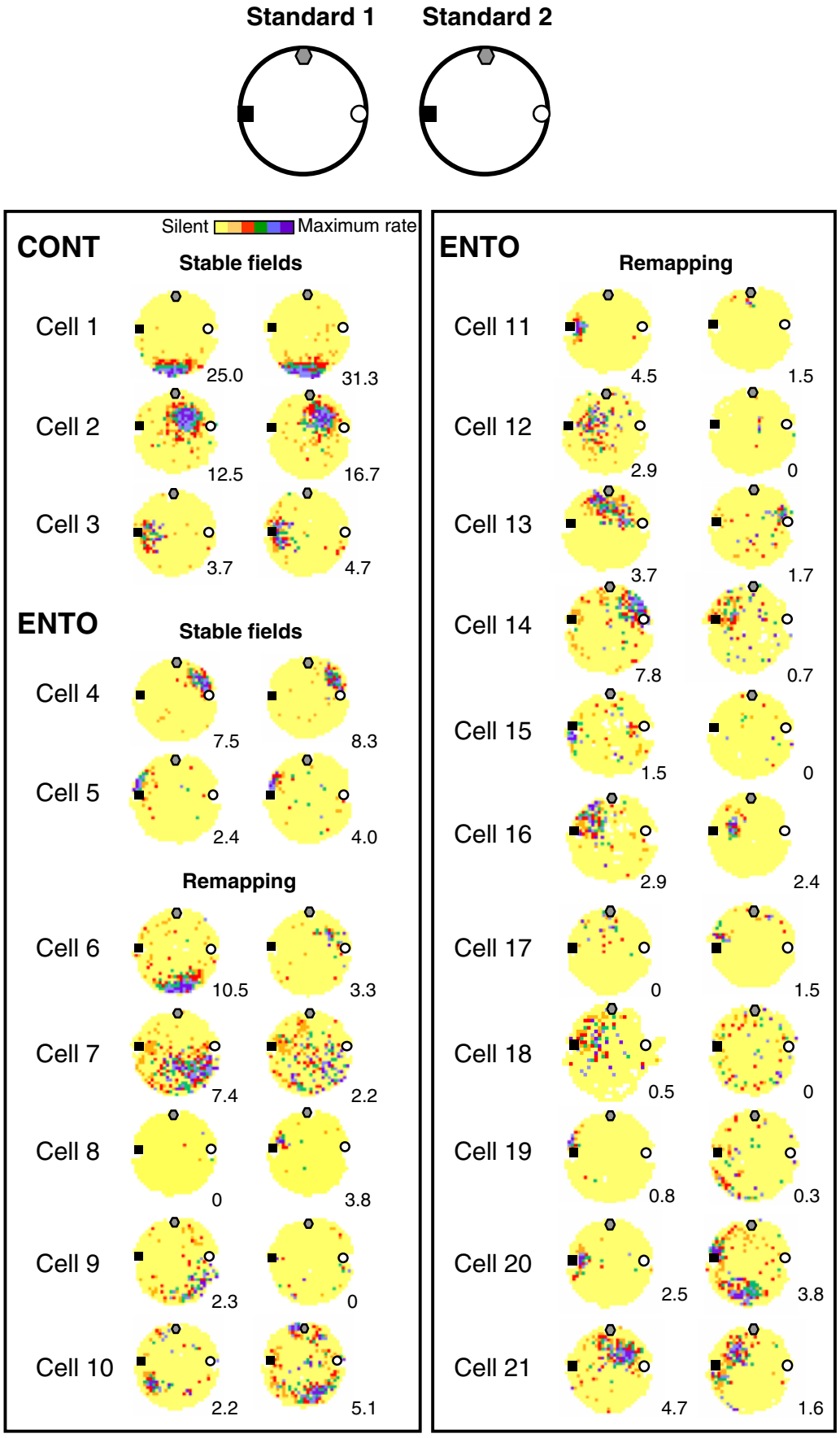
Cue manipulations (STD2–Object Rotation–Object Removal)

Only cells that had a stable field according to our criteria in STD1 and STD2 were further recorded in the sequence of three sessions involving object manipulations (STD2–Object Rotation–Object Removal) in ENTO and CONT rats. Forty-nine cells in ENTO rats that had stable fields in STD1 and STD2 and two cells that were silent in STD2, i.e. fired only a few spikes, but displayed a clear firing field in the object rotation session were further recorded in sessions that involved object manipulations, i.e. object rotation and object removal. Fifty-two sequences of three sessions were recorded, 31 in ENTO rats (51 cells, 1.6 cells per sequence) and 21 in CONT rats (40 cells, 1.9 cells per sequence). Thus, a total of 91 place cells were analyzed. The results of object rotation and removal are summarized in Table 3.

Object rotation was made to examine whether firing fields are controlled by the objects. Consistent with a large amount of data (e.g. Muller & Kubie, 1987; Cressant *et al*., 1997), 100% of the firing fields in CONT rats rotated an equivalent amount (90°), indicating that place cells relied on the objects to anchor their fields. In contrast, in ENTO rats, fields that were stable in constant conditions were not as well controlled by the objects. Object rotation produced field disappearance or appearance in four cells (4/51, 8%). The remaining 47 cells had fields in both STD2 and Object Rotation sessions. Figure 7A shows the distribution of field rotation angles for these cells in Object Rotation sessions and shows that many fields were not controlled by the objects (22/51, 43%). Twenty-nine per cent of fields (15/51) shifted and 14% (7/51) remained stable relative to the background cues. The remaining fields (49%) were controlled by the objects (25/51). Representative examples of the three response types are shown in Fig. 7B. Thus, object rotation induced a remapping in 37% of the cells in ENTO rats (field appearance, disappearance and shifted fields, 19/51) and 0% in CONT rats ($\chi^2_1 = 18.74$, $P < 0.001$). In addition, ENTO rats were impaired in using the objects to anchor place cell firing relative to CONT rats ($\chi^2_1 = 25.0$, $P < 0.001$).

We also looked at the coherence of the responses in small ensembles of simultaneously recorded cells. Of 11 sequences (32 cells), four resulted in incoherent responses to rotation, i.e. some fields rotated and some remapped. In CONT rats, all cells had a coherent response.

Object removal was made to examine whether place cells would maintain stable firing fields in the absence of the objects by using other sensory information such as olfactory and/or idiothetic information (Save *et al*., 2000). In a previous study, we showed that in the absence of objects most fields remained stable, therefore reflecting the complementary contribution of these two kinds of information (Save *et al*., 2005). Consistent with these results, we found that 65% (26/40) of cells in CONT rats had stable fields (R_{max}: 0.370 ± 0.04, angle: $-8.1 \pm 12.5°$). Twenty per cent of cells (8/40) stopped firing and 15% (6/40) had fields that shifted (R_{max}: 0.42 ± 0.09, angle: $-59 \pm 47.5°$).



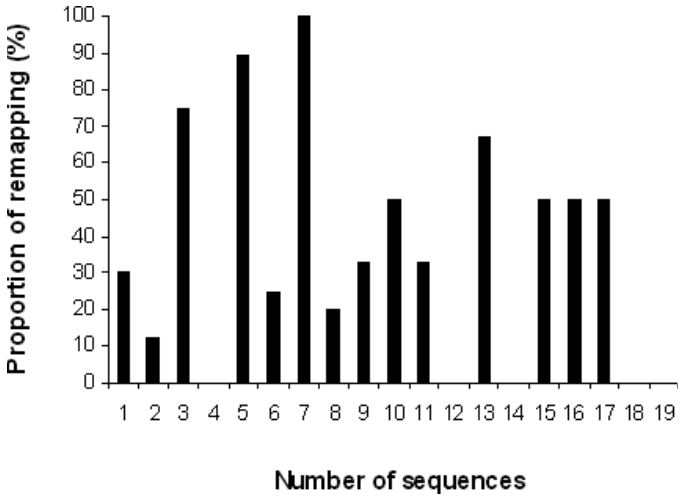


FIG. 6. Percentage of remapping as a function of training in ENTO rats.

TABLE 3. Effects of object manipulation on place cell firing after object rotation and object removal in ENTO and CONT rats

	Object rotation		Object removal	
Cells	Controlled	Non-controlled	Stable	Changed
ENTO	49%	51%	24%	76%
CONT	100%	0%	65%	35%

Figure 8A (left) displays the distribution of rotation angles for the cells that had a field in both Object Rotation and Object Removal sessions. Representative examples of rate maps are shown in Fig. 8B (left).

Figure 8A (right) displays the distribution of rotation angles for the cells that had a field in both Object Rotation and Object Removal sessions in ENTO rats. It shows that 39% of cells (20/51) had fields that shifted unpredictably (R_{max}: 0.300 ± 0.04, angle: 210.48 ± 25.37°) whereas only 24% of cells (12/51) had stable fields (R_{max}: 0.560 ± 0.09, angle: 5.44 ± 2.61°). Furthermore, 37% of cells (19/51) stopped firing. A global chi-squared analysis showed that the CONT and ENTO rats differed regarding the distribution of these three categories of responses ($\chi^2_1 = 15.68$, $P < 0.001$). More specifically, a difference between the two groups was found in the proportion of fields that remained stable vs. the fields that shifted ($\chi^2_1 = 12.8$, $P < 0.001$). In contrast, there was no difference in the proportion of cells that still fired vs. the cells that stopped firing ($\chi^2_1 = 3.26$, $P > 0.05$). Representative examples of rate maps are shown in Fig. 8B (right). In ENTO rats, a large majority of fields (76%) remapped following cue removal. Remapping occurred immediately following cue removal, therefore suggesting an exacerbated sensitivity to cue removal rather than a deficit in using idiothetic and olfactory information for path integration.

To examine whether damaging the entorhinal cortex would affect the use of idiothetic information during the cue removal session, correlations were made between the firing rate arrays of the two object removal session halves. It was hypothesized that because the use of idiothetic cues induces an accumulation of errors (Etienne & Jeffery, 2004), the firing fields would progressively drift during the course of the session. The correlation between the two session halves would therefore be smaller in ENTO rats (drifted fields) than in CONT rats (stable fields). In ENTO rats, cells with fields that had remained stable or had shifted after cue removal were included in the analysis ($n = 32$). No difference was found between the two session halves for shifted and stable fields in ENTO rats ($t_{30} = 0.4$, $P = 0.699$) and between ENTO and CONT rats (ENTO, within session $R_0 = 0.307 \pm 0.05$; CONT, within session $R_0 = 0.207 \pm 0.03$, $t_{62} = 0.3$, $P = 0.806$). In summary, in ENTO rats, object removal induced a large proportion of remapping but the newly established fields remained stable throughout the session.

Discussion

We recorded place cells in rats with bilateral lesions of the entorhinal cortex while they performed a pellet-chasing task in a cylinder containing three objects. We found that entorhinal lesions did not abolish place cell activity but affected place cell properties and impaired firing field stability in repeated constant conditions or following cue manipulations (object rotation, object removal). The results suggest that the entorhinal cortex is part of a system providing a stable spatial reference frame to the hippocampus that is crucial to maintain spatial representations.

Although damage to the entorhinal cortex removed a substantial amount of inputs to the hippocampus, it did not prevent the formation of firing fields, suggesting that some spatial information did reach the hippocampus. First, it is possible that information could still reach the hippocampus through spared entorhinal tissue even if it was limited. A small number of grid cells (see below) in the dorsolateral entorhinal cortex may provide enough input to allow place cell firing (Solstad *et al*., 2006). Second, there are indirect projections from the perirhinal cortex to the hippocampus via the lateral entorhinal cortex, a region that was partially damaged in our study as well as direct projections to CA1 (Naber *et al*., 1999). The postrhinal cortex is indirectly connected with the hippocampus via the perirhinal cortex, the medial entorhinal cortex but also directly projects to CA1 (Naber *et al*., 2001a, b). Firing fields could then be established on the basis of relatively limited and simple information. In spite of this, however, place cells were unable to anchor their fields relative to salient objects, suggesting that integration of sensory information was disrupted. A third non-exclusive possibility is that subcortical pathways could still convey spatial information to the hippocampus, by-passing the entorhinal cortex. Both the anterior thalamus and the medial septum may be part of such pathways as they send direct projections to the hippocampus and exert an influence on place cells (Mizumori *et al*., 1989; Calton *et al*., 2003). However, whether these subcortical pathways can, by

FIG. 5. Examples of STD1 and STD2 firing rate maps in ENTO and CONT rats. The corresponding peak rate (Hz) is indicated beside each map. In CONT rats all fields were stable (cell 1: R_0 0.81, R_{max} 0.81, angle 0°; cell 2: R_0 0.57, R_{max} 0.57, angle 0°; cell 3: R_0 0.90, R_{max} 0.90, angle 0°). In ENTO rats, a number of fields remained stable (cell 4: R_0 0.89, R_{max} 0.89, angle 0°; cell 5: R_0 0.56, R_{max} 0.58, angle 12°) but 42% remapped. These latter fields underwent a variety of alterations and modifications suggesting a global remapping. Changes included stop of firing (cell 7: R_0 0.19, no field; cell 9: R_0 0.05, no field; cell 11: R_0−0.03, no field; cell 12: R_0−0.02, no field; cell 15: R_0−0.05, no field; cell 18: R_0−0.01, no field), appearance of fields (cell 8: R_0−0.04, no field; cell 17: R_0 0.01, no field), modification in location, shape, size and field rate (cell 6: R_0−0.09, R_{max} 0.31, angle 72°; cell 10: R_0 0.01, R_{max} 0.13, angle 132°; cell 13: R_0−0.01, R_{max} 0.18, angle, 270°, cell 14: R_0−0.09, R_{max} 0.08, angle 90°; cell 16: R_0 0.36, R_{max} 0.36, angle 0°; cell 19: R_0 0.002, R_{max} 0.2, angle 46°; cell 20: R_0 0.12, R_{max} 0.34, angle 36°; cell 21: R_0−0.05, R_{max} 0.26, angle 246°). R_0 is the similarity score and R_{max} is the highest correlation between the two firing rate arrays, therefore allowing determination of field rotation between the two sessions.

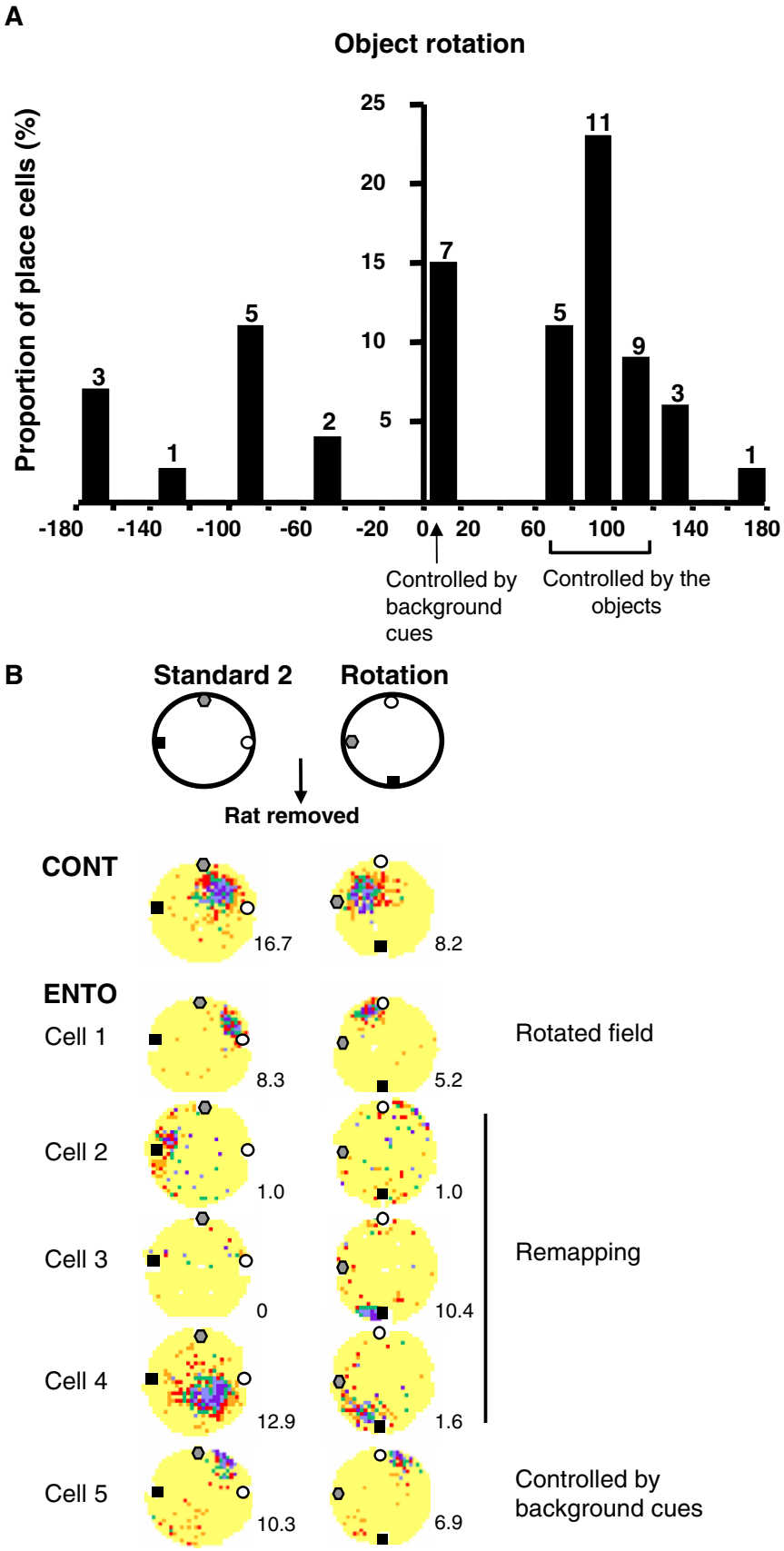


FIG. 7. (A) Distribution of field rotation angles (degrees) grouped in 20° bins, after object rotation in ENTO rats. The number of cells is displayed above each bar. (B) Examples of firing rate maps in CONT and ENTO rats for Object Rotation. The corresponding in-field peak rate (Hz) is indicated beside each map. In ENTO rats, cell 1 had a field controlled by the objects, cells 2–4 a field that remapped, and cell 5 a field that remained stable relative to background cues.

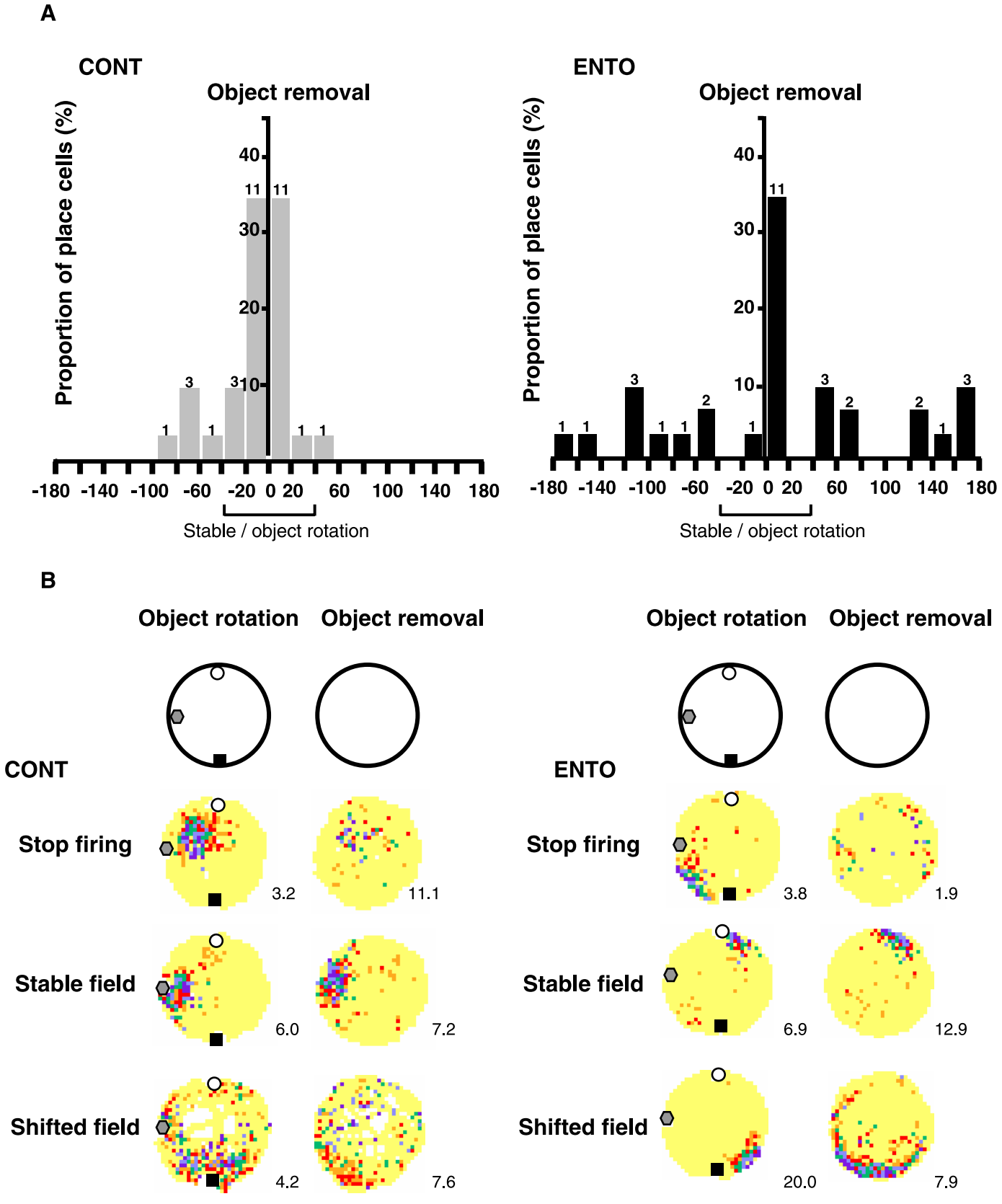


FIG. 8. (A) Distribution of field rotation angles (degrees) grouped in 20° bins, after object removal in CONT and ENTO rats. The number of cells is displayed above each bar. (B) Examples of firing rate maps in CONT and ENTO rats. The corresponding in-field peak rate (Hz) is indicated beside each map.

themselves, maintain CA1 place cell activity is uncertain. A last possibility relates to the fact that the rats were familiarized to the apparatus before surgery (see Materials and methods). This might have allowed the place cell system to generate and store spatial representations of the environment.

Location-specific firing was not disrupted in entorhinal-lesioned rats, and information content and spatial coherence of place cells were not diminished compared with control rats, indicating that spatial selectivity and coherence of place cell firing were not altered. In contrast, place cells recorded in entorhinal-lesioned rats discharged at a lower rate than place cells recorded in control rats. This deficit cannot be attributed to lower running speed (McNaughton *et al*., 1983) in entorhinal rats as both groups displayed similar locomotor activity. More plausibly, it may be attributed to a loss of sensory inputs to the hippocampus. Consistent with this idea, decreases of mean firing rate, in-field firing rate and peak firing rate were also found in visually deprived rats (Save *et al*., 1998). The field size was also diminished in entorhinal-lesioned rats. This effect has generally not been found in studies reporting the effects of cortical lesions on place cell activity (Muir & Bilkey, 2001; Paz-Villagràn *et al*., 2002; Kyd & Bilkey, 2003; Save *et al*., 2005) and seems therefore to be specific of entorhinal lesions. Although small fields may be at least partly a consequence of decreased discharge, previous results have shown that a lower firing rate is not systematically correlated with smaller field size (e.g. Save *et al*., 1998; Kyd & Bilkey, 2003).

Our results are not entirely consistent with those of Miller & Best (1980). These authors found that lesions of the entorhinal cortex altered the spatial robustness of place cells. Using this parameter, we did not find any difference between the two groups. In addition, information content and coherence were not modified by entorhinal cortex lesions. Miller and Best's results may be explained by the larger extent of their lesions that included damage to the subiculum, and pre- and para-subiculum. Supporting this hypothesis, lesions of the pre- and para-subiculum have been found to affect the information content in hippocampal place cells (Liu *et al*., 2004).

Another major finding of the study is that entorhinal cortex lesions disrupted the stability of firing fields both in constant conditions and following cue manipulations. While all fields in control rats were stable in constant conditions and followed object-cue rotation, many fields in entorhinal cortex-lesioned rats remapped (42% in constant conditions and 43% in Object Rotation). In spite of multiple exposures to the familiar environment, the environment-specific representation was not re-activated in entorhinal-lesioned rats as it was in control rats. A new representation was formed that was not re-activated in subsequent exposures to the same environment. Thus, the entorhinal cortex does not appear to be critically involved in the emergence of spatial representations in the CA1 subfield of the hippocampus but may be important for reactivation of existing representation. According to computational models of memory processes, re-activation of the appropriate representation of a familiar environment probably requires a comparison process between the current sensory inputs and the stored representations. This comparison allows the system to determine whether an input is familiar or novel. In the hippocampal system, the CA1 region would perform comparison of direct inputs from entorhinal cortex layer III with output from region CA3 (McClelland *et al*., 1995; Hasselmo *et al*., 2000; Lisman & Otmakhova, 2001). When exposed to a familiar environment, the CA3 output would match the entorhinal input and the corresponding representation would be re-activated in the CA1 neurons. If the entorhinal input is disrupted or degraded, the comparison process cannot operate properly in CA1 and our results show that the hippocampus generates unpredictable, new representations. The new representations may result from operation of the intrinsic hippocampal circuitry, therefore maintaining at all costs a representation of the environment that may be used for navigation.

The recently discovered grid cells provide another complementary interpretation, however (Hafting *et al*., 2005; Sargolini *et al*., 2006). Grid cells have multiple firing fields that constitute a tessellating structure extending to the entire environment. The field grids of neighboring cells are offset relative to each other but have identical spacing and orientation (Witter & Moser, 2006). A crucial property is that the grid cell firing pattern is not affected by re-arrangements or removal of environmental cues and persists in darkness (Hafting *et al*., 2005), suggesting that grid cells are involved in path integration-based localization. Thus, grid cells seem to implement a metric framework, i.e. a spatial coordinate system, upstream from the hippocampus (McNaughton *et al*., 2006). The grid cell system output is transmitted to CA1, which in turn projects back to the entorhinal cortex through the deep layers, suggesting that grid cell and CA1 place cell representations interact (Witter & Moser, 2006). A recent model predicted that entorhinal lesions would produce broader firing hippocampal place fields following lesion of the dorsal medial entorhinal cortex and smaller fields following lesion of the ventral medial entorhinal cortex (Solstad *et al*., 2006). We found a decrease in field size, possibly suggesting a greater impact of ventral entorhinal cortex lesions on place cell firing in our study. Calton *et al*. (2003) showed that lesions of the postsubiculum, a structure that projects to the superficial layers of the entorhinal cortex (Caballero-Bleda & Witter, 1993) induced unpredictable shifts of place field location similar to those observed in the present study. This suggests that grid cells may integrate information from the postsubiculum, i.e. from the head direction system to maintain stable fields. Interestingly, hippocampal lesions do not seem to dramatically affect spatial firing in the entorhinal cortex (Fyhn *et al*., 2004).

Our results are compatible with the idea that grid cell firing is necessary for stabilization and/or re-activation of spatial representations in CA1. In the absence of entorhinal cortex, the hippocampal representations are labile and vulnerable to interference. It follows that, when an animal with entorhinal lesions is re-exposed to a familiar environment, re-activation of the representation fails and the place cell system elaborates a new representation.

Another prediction resulting from the above considerations is that damaging the grid cell system should also disrupt the ability of place cells to rely on path integration to maintain stable fields in the absence of environmental cues. Thus, removing the objects as the rat forages in the cylinder does not affect the stability of firing fields in control rats (Save *et al*., 2005) but may produce a progressive drift of the firing fields in entorhinal-lesioned rats. In entorhinal-lesioned rats, many fields remapped shortly after object removal, indicating that they were very sensitive to this manipulation, but the newly established fields remained stable throughout the session. Thus, self motion-based stability of firing fields after cue removal does not depend on the entorhinal cortex, a result that departs from current hypotheses on the role of the entorhinal cortex (McNaughton *et al*., 2006). Other interpretations could account for this result, however. First, alternative sources of information could be used by place cells for path integration-based localization. The head direction cell system may be such a potential source as idiothetic cues are integrated by head direction cells (Knierim *et al*., 1998). Second, in the present task the animal was not required to use idiothetic information and thus may have used other information such as background cues to maintain stable fields. It is possible therefore that lesions of the entorhinal cortex would alter place cell stability if more demanding path integration processing is required,

i.e. in darkness or during specific path integration tasks (Save *et al*., 2000; Parron & Save, 2004b).

Overall, our results confirm the existence of a functional interaction between the entorhinal cortex and hippocampal place cell firing (e.g. Brun *et al*., 2002; Parron *et al*., 2006). Although the entorhinal cortex is not critical for the build-up of hippocampal spatial representations, it is crucial for the stability of such representations across sessions. Thus, the medial entorhinal cortex, via the grid cell system, may provide a spatial coordinate system that would serve to maintain stable hippocampal place fields. It cannot be ruled out, however, that the deficits found in the present study result from the combined lesion of the medial and lateral regions of the entorhinal cortex. Recent data suggest that these two regions mediate distinct processes (Knierim *et al*., 2006). Clearly, the respective contribution of these two regions remains to be determined.

In a more general vein, the framework provided by the entorhinal cortex may participate to a spatiotemporal coordinate system for conjointly encoding spatial and non-spatial information in episodic memory. Interestingly, our results show that the effects of entorhinal cortex lesions closely parallel the effect of aging on place cell stability (Barnes *et al*., 1997), thus suggesting that degeneration of the entorhinal cortex contributes to memory decline in normal and pathological aging.

Supplementary material

The following supplementary material may be found on
http://www.blackwell-synergy.com

Fig. S1. Smoothed rate maps of all the cells in ENT0 rats that displayed substantial modifications of their fields including stop of firing, appearance of fields, changes in location, shape, size or field rate.

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References

Barnes, C.A., Suster, M.S., Shen, J. & McNaughton, B.L. (1997) Multistability of cognitive maps in the hippocampus of old rats. *Nature*, **388**, 272–275.

Brun, V.H., Otnaess, M.K., Molden, S., Steffenach, H.A., Witter, M.P., Moser, M.B. & Moser, E.I. (2002) Place cells and place recognition maintained by direct entorhinal-hippocampal circuitry. *Science*, **296**, 2243–2246.

Burwell, R.D. & Amaral, D.G. (1998) Cortical afferents of the perirhinal, postrhinal, and entorhinal cortices of the rat. *J. Comp. Neurol.*, **398**, 179–205.

Caballero-Bleda, M. & Witter, M.P. (1993) Regional and laminar organization of projections from the presubiculum and parasubiculum to the entorhinal cortex: an anterograde tracing study in the rat. *J. Comp. Neurol.*, **328**, 115–129.

Calton, J.L., Stackman, R.W., Goodridge, J.P., Archey, W.B., Dudchenko, P.A. & Taube, J.S. (2003) Hippocampal place cell instability after lesions of the head direction cell network. *J. Neurosci.*, **23**, 9719–9731.

Cho, Y.H. & 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.

Cressant, A., Muller, R.U. & Poucet, B. (1997) Failure of centrally placed objects to control the firing fields of hippocampal place cells. *J. Neurosci.*, **17**, 2532–2542.

Etienne, A. & Jeffery, K. (2004) Path integration in mammals. *Hippocampus*, **14**, 180–192.

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

Gothard, K.M., Skaggs, W.E. & McNaughton, B.L. (1996) Dynamics of mismatch correction in the hippocampal ensemble code for space: interaction between path integration and environmental cues. *J. Neurosci.*, **16**, 8027–8040.

Hafting, T., Fyhn, M., Molden, S., Moser, M.B. & Moser, E.I. (2005) Microstructure of a spatial map in the entorhinal cortex. *Nature*, **436**, 801–806.

Hasselmo, M.E., Fransen, E., Dickson, C. & Alonso, A.A. (2000) Computational modeling of entorhinal cortex. *Ann. NY Acad. Sci.*, **911**, 418–446.

Knierim, J.J., Kudrimoti, H.S. & McNaughton, B.L. (1998) Interactions between idiothetic and external landmarks in the control of place cells and head direction cells. *J. Neurophysiol.*, **80**, 425–446.

Knierim, J.J., Lee, I. & Hargreaves, E.L. (2006) Hippocampal place cells: parallel input streams, subregional processing, and implications for episodic memory. *Hippocampus*, **16**, 755–764.

Kyd, R. & Bilkey, D.K. (2003) Prefrontal cortex lesions modify the spatial properties of hippocampal place cells. *Cereb. Cortex*, **13**, 444–451.

Leutgeb, S., Leutgeb, J.K., Barnes, C.A., Moser, E.I., McNaughton, B.L. & Moser, M.B. (2005) Independent codes for spatial and episodic memory in hippocampal neuronal ensemble. *Science*, **309**, 619–623.

Lisman, J.E. & Otmakhova, N.A. (2001) Storage, recall, and novelty detection of sequences by the hippocampus: elaborating on the SOCRATIC model to account for normal and aberrant effects of dopamine. *Hippocampus*, **11**, 551–568.

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

Markus, E.J., Barnes, C.A., McNaughton, B.L., Gladden, V.L. & Skaggs, W.E. (1994) Spatial information content and reliability of hippocampal neurons: effects of visual input. *Hippocampus*, **4**, 410–421.

McClelland, J.L., McNaughton, B.L. & 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.

McHugh, T.J., Jones, M.W., Quinn, J.J., Balthasar, N., Coppari, R., Elmquist, J.K., Lowell, B.B., Fanselow, M.S., Wilson, M.A. & Tonagawa, S. (2007) Dentate gyrus NMDA receptors mediate rapid pattern separation in the hippocampal network. *Science*, **317**, 94–99.

McNaughton, B.L., Barnes, C.A. & O'Keefe, J. (1983) The contributions of position, direction, and velocity to single unit activity in the hippocampus of freely-moving rats. *Exp. Brain Res.*, **52**, 41–49.

McNaughton, B.L., Battaglia, F.P., Jensen, O., Moser, E.I. & Moser, M.B. (2006) Path integration and the neural basis of the 'cognitive map'. *Nature Rev. Neurosci.*, **7**, 663–678.

Miller, V.M. & Best, P.J. (1980) Spatial correlates of hippocampal unit activity are altered by lesions of the fornix and entorhinal cortex. *Brain Res.*, **194**, 311–323.

Mizumori, S.J.Y., Barnes, C.A. & McNaughton, B.L. (1989) Reversible inactivation of the medial septum: selective effects on the spontaneous unit activity of different hippocampal cell types. *Brain Res.*, **500**, 99–106.

Muir, G.M. & Bilkey, D.K. (2001) Instability in the place field location of hippocampal place cells after lesions centered on the perirhinal cortex. *J. Neurosci.*, **21**, 4016–4025.

Muller, R.U. & Kubie, J.L. (1987) The effects of changes in the environment on the spatial firing of hippocampal complex-spike cells. *J. Neurosci.*, **7**, 1951–1968.

Muller, R.U. & Kubie, J.L. (1989) The firing of hippocampal place cells predicts the future position of freely moving rat. *J. Neurosci.*, **9**, 4101–4110.

Muller, R.U., Kubie, J.L. & Ranck, J.B. (1987) Spatial firing pattern of hippocampal complex-spike cells in a fixed environment. *J. Neurosci.*, **7**, 1935–1950.

Naber, P.A., Lopes da Silva, F.H. & Witter, M.P. (2001a) Reciprocal connections between the entorhinal cortex and hippocampal fields CA1 and the subiculum are in register with the projections form CA1 to the subiculum. *Hippocampus*, **11**, 99–104.

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

Naber, P.A., Witter, M.P. & 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.
O'Keefe, J. & Conway, D.H. (1978) Hippocampal place units in the freely moving rat: why they fire where they fire. *Exp. Brain Res.*, **31**, 573–590.
O'Keefe, J. & Dostrovsky, J. (1971) The hippocampus as a spatial map. Preliminary evidence from unit activity in the freely moving rat. *Brain Res.*, **34**, 171–175.
O'Keefe, J. & Nadel, L. (1978) *The Hippocampus as a Cognitive Map*. Oxford University Press, London.
Parron, C., Poucet, B. & Save, E. (2004) Entorhinal cortex lesions impairs the use of distal but not proximal landmarks during navigation in the rat. *Behav. Brain Res.*, **154**, 345–352.
Parron, C., Poucet, B. & Save, E. (2006) Cooperation between the hippocampus and the entorhinal cortex in spatial memory: a disconnection study. *Behav. Brain Res.*, **170**, 99–109.
Parron, C. & Save, E. (2004a) Comparison of the effects of entorhinal or retrosplenial cortical lesions on habituation, reaction to spatial and non spatial changes during object exploration in the rat. *Neurobiol. Learn. Mem.*, **82**, 1–11.
Parron, C. & Save, E. (2004b) Evidence for entorhinal and parietal cortices involvement in path integration in the rat. *Exp. Brain Res.*, **159**, 349–359.
Paxinos, G. & Watson, C. (2004) *The Rat Brain in Stereotaxic Coordinates*, 5th edn. Academic Press, New York.
Paz-Villagràn, V., Lenck-Santini, P.P., Save, E. & Poucet, B. (2002) Properties of place cell firing after damage to the visual cortex. *Eur. J. Neurosci.*, **16**, 771–776.
Paz-Villagràn, V., Save, E. & Poucet, B. (2004) Independent coding of connected environments by place cells. *Eur. J. Neurosci.*, **20**, 1379–1390.
Quirk, G.J., Muller, R.U., Kubie, J.L. & Ranck, J.B. (1992) The positional firing properties of medial entorhinal neurons: description and comparison with hippocampal place cells. *J. Neurosci.*, **12**, 1945–1963.
Sargolini, F., Fyhn, M., Hafting, T., McNaughton, B.L., Witter, M.P., Moser, M.B. & Moser, E.I. (2006) Conjunctive representation of position, direction, and velocity in entorhinal cortex. *Science*, **312**, 758–762.
Save, E., Cressant, A., Thinus-Blanc, C. & Poucet, B. (1998) Spatial firing of hippocampal place cells in blind rats. *J. Neurosci.*, **18**, 1818–1826.
Save, E., Nerad, L. & Poucet, B. (2000) Contribution of multiple sensory information to place field stability in hippocampal place cells. *Hippocampus*, **10**, 64–76.
Save, E., Paz-Villagràn, V., Alexinsky, T. & Poucet, B. (2005) Functional interaction between the associative parietal cortex and hippocampal place cell firing. *Eur. J. Neurosci.*, **21**, 522–530.
Sewards, T.V. & Sewards, M.A. (2003) Input and output stations of the entorhinal cortex: superficial vs. deep layers or lateral vs. medial divisions ? *Brain Res. Rev.*, **42**, 243–251.
Solstad, T., Moser, E.I. & Einevoll, G.T. (2006) From grid cells to place cells: a mathematical model. *Hippocampus*, **16**, 1026–1031.
Witter, M.P. & Moser, E.I. (2006) Spatial representation and the architecture of the entorhinal cortex. *Trends Neurosci.*, **29**, 671–678.
Witter, M.P., Wouterlood, F.G., Naber, P.A. & Van Haeften, T. (2000) Anatomical organization of the parahippocampal-hippocampal network. *Ann. NY Acad. Sci.*, **911**, 1–24.
Zilles, K. (1985) *The Cortex of the Rat, a Stereotaxic Atlas*. Springer Verlag, Berlin.