European Journal of Neuroscience, Vol. 20, pp. 1379–1390, 2004 © Federation of European Neuroscience Societies

Independent coding of connected environments by place cells

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Keywords: hippocampus, place cells, rat, spatial context, unit recordings

Abstract

Place cells are hippocampal neurons that have a strong location-specific firing activity in the rat's current environment. Collectively, place cells also provide a signature of the rat's environment as their ensemble activity is markedly different when recorded in distinct apparatuses. This phenomenon, referred to as 'remapping', suggests that each environment activates a different hippocampal map. In this study, we sought to determine the independence of such maps. In Experiment 1, we used a cylinder apparatus that was divided into two equal halves by a central barrier with an aperture allowing the rat to freely commute between the two sides. A local change in one side failed to induce field remapping in the changed side, thus precluding any significant conclusion to be drawn. We therefore designed Experiment 2 in which place cells were first recorded while rats explored three distinct high-walled boxes. Most cells had distinctive firing fields in each box. A runway was then added to connect two initially unrelated boxes. This manipulation altered the firing of some cells but the fields in each box were still clearly distinguishable. The final manipulation consisted of changing one box and allowing the rat to commute freely between the changed and unchanged boxes. While the firing fields remapped in the changed box, they were most usually unaltered in the unchanged box. These results suggest that the hippocampus holds a set of independent maps for each box, and that each specific map is activated mainly according to the rat's current sensory environment.

Introduction

For the last two decades, the hippocampus of the rat has been thought to host a spatial representation of the animal's environment (O'Keefe & Nadel, 1978). The main evidence in support of this theory is the existence of place cells, which are pyramidal neurons located in the CA1 and CA3 regions of the hippocampus and whose firing is strongly correlated with the location of a freely moving rat in its environment (O'Keefe & Dostrovsky, 1971). The cell-specific region of intense discharge is called the 'firing field'. The firing fields of place cells can be seen in all regions of the environment accessible to the rat, so that they collectively provide the units of the putative environmental map.

In addition to continuously providing the rat with information about its current position, place cells also signal the identity of the rat's current environment. For example, Muller & Kubie (1987) recorded the activity of place cells in rats that explored either a circular or a square apparatus and found that the location-specific firing pattern of each place cell was markedly different in each apparatus. Some cells active in one apparatus were silent in the other apparatus and the fields of cells active in both apparatuses were unrelated. This phenomenon, referred to as 'remapping', suggests that the identity of an environment is encoded by a unique selection of active place cells (or 'active subset') and furthermore by a unique spatial arrangement of the firing fields of the selected place cells. The circumstances that trigger a remapping include changing the shape of the apparatus (Muller & Kubie, 1987; Lever *et al*., 2002), changing the colour of the cue card within the apparatus (Bostock *et al*., 1991; Kentros *et al*., 1998), or changing the orientation of the apparatus relative to the background cues (Cressant *et al*., 2002). Remapping can occur immediately after the change in conditions (Kentros *et al*., 1998) or take place after a moderate (Bostock *et al*., 1991; Sharp *et al*., 1995; Shapiro *et al*., 1997) or substantial delay (Lever *et al*., 2002). Finally, remapping is generally reversible: restoring the initial conditions usually restores the initial firing fields (Muller & Kubie, 1987; Kentros *et al*., 1998; Cressant *et al*., 2002; Lever *et al*., 2002).

The remapping phenomenon is of considerable interest as it suggests that the hippocampus learns and holds distinct maps for distinct environments. However, it also raises several questions. A first question has to do with the conditions in which remapping is usually studied. Typically, the design of a remapping experiment consists of exposing the rat to the initial apparatus, then removing the rat from this apparatus and finally, after the appropriate change is made, exposing the rat to the modified apparatus (Muller & Kubie, 1987; Bostock *et al*., 1991; Kentros *et al*., 1998; Cressant *et al*., 2002; Lever *et al*., 2002). In this design therefore the rat is exposed sequentially to different environments. This procedure does not demonstrate if and how the hippocampus is capable of activating two separate maps when the rat is moving between two distinct environments.

Another, more fundamental issue raised by the remapping phenomenon concerns the nature of the putative hippocampal map. At one extreme, the hippocampal representation is purely local. That is, the hippocampus holds a set of completely independent maps for each apparatus, and each specific map is activated only according to the rat's current environment. At the other extreme, the nature of the hippocampal map can be thought to be holistic. In that view, activation of a specific map also evokes activation of a more general

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Received 3 March 2004, revised 17 May 2004, accepted 23 June 2004

doi:10.1111/j.1460-9568.2004.03570.x

representation that includes other regions of space with which the rat's current environment is connected. One way to dissociate these two possibilities is to study the remapping properties of place cells when the rat is commuting between two connected environments. The view that the hippocampal map is only local predicts that a change resulting in remapping of cell firing in one environment should not influence cell firing in the other, unchanged environment. Conversely, the holistic view predicts that a remapping in the changed environment should be reflected in major changes in the other unchanged environment. The results demonstrate that, at least in our conditions, the hippocampal map is mainly driven by the sensory data from the rat's current environment, therefore supporting the local encoding hypothesis.

Materials and methods

Subjects

Naïve Long–Evans black hooded male rats (R. Janvier, St.-Berthevin, France) weighing 300–350 g were used. The rats were housed one per cage on a natural light-dark cycle in a temperature-controlled room (20° ± 2°) with *ad-libitum* access to water. On arrival, they were handled daily for two weeks. Next, the rats were food-deprived to 85% of *ad-libitum* body weight and trained to chase pellets in either a large cylinder (Experiment 1) or in each of three distinct boxes (Experiment 2) prior to electrode implantation. Lighting of the experimental room was provided by four 25 W bulbs fixed to the ceiling at symmetrical positions. A food dispenser attached to the ceiling dropped food pellets at random locations on the floor of the apparatus. A radio tuned to an FM station fixed to the ceiling provided background noise > 70 dB to mask uncontrolled directional sounds. The experimenter stood in an adjacent room that contained the unit recording system, the computer and a TV monitor that displayed the overhead view from the camera. All procedures used in this study were performed in compliance with institutional guidelines (council directives #87848, October 19, 1987, Ministère de l'Agriculture et de la Forêt, Service Vétérinaire de la Santé et de la Protection Animale; permission #13.76 to BP; NIH publication N°86–23, revised 1985).

Surgery

At the end of training, surgery to implant an array of ten microwire electrodes was performed under sterile conditions and general anaesthesia. The electrodes were made of 25 μm nichrome wire and formed a bundle threaded through a piece of stainless steel tubing (Kubie, 1984). Each wire was attached to a pin on the outside of a circular connector. The tubing was attached to the centre pin of the connector and served as the animal ground as well as a guide for the microwires. The connector, tubing, and wires could be moved down in the brain by turning screws attached to the connector into nylon cuffs that were attached to the rat's skull.

The tips of the electrode bundle were implanted above the dorsal CA1 pyramidal cell layer. The rat was anaesthetized with pentobarbital (40 mg/kg i.p.), injected with atropine sulphate (0.25 mg/kg i.p.) to prevent respiratory distress and put in a Kopf stereotaxic apparatus. The skull surface was exposed and holes for the electrodes and to anchor the electrode carrier were drilled at appropriate locations. Three miniature screws and a T-shaped screw were placed in the skull to anchor the recording-electrode array. The tips of the recording electrodes were implanted at stereotaxic coordinates: 3.8 mm posterior and 3.0 mm lateral to bregma and 1.5 mm below the dura (Paxinos & Watson, 1986). Once electrodes were in place, sterile petroleum jelly was applied to the exposed brain surface and around the guide tube for the electrodes. Next, dental acrylic was put over the jelly and around the tube to cover the skull hole. Finally, the bottoms of the three drive-screw assemblies were attached to the anchor screws. As a postoperative treatment, the rats received an i.m. injection of antibiotic (terramycine, 60 mg/kg). The animals were given one week to recover from surgery before recordings were made.

At the completion of the experiment, each rat was killed with a lethal dose of pentobarbital and perfused intracardially with 0.9% saline followed by 4% formalin. Just before death, positive current (15 μA for 30 s) was passed through one of the microwires to deposit iron that could be visualized after reaction with potassium ferrocyanide (Prussian blue). The brain was removed and stored for 1 day in 3% ferrocyanide. Later, 40 μm coronal sections were made. Every fifth section was stained with cresyl violet for verification of electrode placements.

Recording methods

Beginning one week after surgery, activity from each microwire was screened daily while the rat underwent additional pellet chasing sessions in the boxes. A set of tubes was attached to the food dispenser in the ceiling in such a way that food pellets could land in either box. Screening and recording were performed with a cable attached at one end to a commutator that allowed the rat to turn freely. The other end of the cable was connected to a light emitting diode (LED) for tracking the rat's head position, a headstage with a unity gain preamplifier for each wire field and finally a connector that mated with the electrode connector cemented to the rat's skull. The fixed side of the commutator was connected to a distribution panel. From the panel, the desired signals were amplified 10 000-fold with low-noise differential amplifiers, band-pass filtered from 0.3 to 10 kHz, and sent to a 250 kHz analogue-to-digital (A/D) board in a Pentium computer. The data acquisition system (DataWave, Longmont, CO) recorded a 1 ms burst of 32 samples at 32 kHz each time the voltage exceeded an experimenter-defined threshold. Before the initial recording session, spike discharges of single units were separated using on-line clustering software (DataWave Discovery, Longmont, CO) to simplify later off-line separation. Briefly, scatterplots of the most characteristic waveform parameters (e.g. peak voltage and waveform duration) were generated from the signals emanating from putative units recorded on each channel.

The rat's head position was tracked by locating the LED set on the midline 1 cm above the head and 1 cm behind the headstage. Tracking was carried out using a TV-based digital spot follower that received RGB signals from a CCD colour camera fixed to the ceiling above the apparatus. The LED was detected at 50 HZ in a grid of 256 × 256 square regions (pixels) that was reduced at analysis stage to a 64 × 64 grid of pixels 30 mm on a side.

Unit discrimination

Cells selected for analysis had to be well-discriminated complex-spike cells with clear location-specific firing in at least one region of the environment. Thus, only waveforms of sufficient amplitude (> 100 μV with a background noise level < 30 μV) were kept for analysis. Similarly, cells that fired too slowly (i.e. with peak rate < 1 Hz or with mean firing rate < 0.2 Hz) were not analysed. Lastly, because our purpose was to measure changes in the same cell across different manipulations, cells that were lost before the session series

was completed, or whose waveforms changed too much between two sessions, were discarded from further analysis.

The first step in off-line analyses was to refine boundaries for waveform clusters that were defined before recording. Candidate waveforms were discriminated based on at most eight characteristic features including maximum and minimum spike voltage, spike amplitude (from peak to trough), time of occurrence of maximum and minimum spike voltages, spike duration, and voltage at two experimenter-defined points of the waveforms. The settings established for a given session were generally used for subsequent sessions. Once single units were well separated, positional firing rate distributions were calculated. The total time the light was detected in each pixel (dwell time) and the total number of spikes in each pixel were accumulated for the session duration. The rate in each pixel was the number of spikes divided by the dwell time. For each session, a firing rate map was constructed using the method described by Muller *et al.* (1987) to visualize the positional firing rate distribution. The rate in each pixel (i.e. the number of spikes divided by the dwell time) was then assigned a grey shade from a scale of four shades. Thus, white pixels represented locations where the firing rate was exactly 0.0 Hz for the whole session. Firing rates were shown as light grey, dark grey and black pixels from low to high. The values used as boundaries between categories were determined for the first session recorded for a given cell. To permit comparisons among positional firing distributions across several sessions for a cell, the rate categories used for subsequent sessions were the same as for the first session.

Experiment 1

In this preliminary experiment, we sought to measure changes in place cell firing in a cylinder apparatus that was divided into two halves between which the rat could freely commute. After exploration of the standard cylinder, one side of the cylinder was changed and modifications of firing were assessed in both changed and unchanged sides.

Apparatus

A cylinder 1 m in diameter with walls 50 cm in height was used. The cylinder was visually isolated from the rest of the laboratory by a concentrically placed cylindrical curtain 250 cm in diameter and height. Additional walls were set in the cylinder so that, seen from above, it looked like an inverted U-shaped apparatus. In particular, a barrier 50 cm in height and 10 cm in width divided the cylinder into two equal halves. When in one half of the cylinder, the rat could not see anything in the other side (except when it was at the border between the two sides). The barrier was 90 cm in length so that it provided a 10-cm wide aperture between its top end (as seen from the TV monitor) and the cylinder wall. This aperture allowed the rat to freely explore the whole cylinder. Each half of the cylinder was made visually different by adding flat black (left side) and white cards (right side) that covered the whole half of the cylinder wall. When necessary, a salient change was brought to the right side of the cylinder by replacing the white walls with light grey walls with black stripes and setting an additional barrier that also modified the geometry.

Presurgery behavioural training

Rats were trained to chase 20-mg food pellets in the apparatus for one week before electrode implantation. Each rat was thus placed daily in the apparatus for 15 min during which time 20-mg food pellets were delivered from the automatic food dispenser in the ceiling. The rat learned to run almost constantly and covered the accessible area several times during a recording session, thus allowing us to sample place cell activity everywhere in the apparatus. Rats were not exposed to the altered environment before place cells were actually recorded.

Testing protocol

Each electrode in a rat was checked twice a day during performance of the pellet-chasing task in the apparatus. If no recordable cell could be isolated on a screening day, the electrode bundle was advanced 25–50 μm. Once a cell, or set of cells, was judged suitable for recording, it was recorded for two successive sessions, both 25 min in duration. Session 1 was conducted while the rat explored the standard apparatus. The rat was then disconnected, removed from the apparatus, and returned to its home cage in the adjacent room. The right side of the cylinder was modified as described above (see section 'apparatus') and a second recording session was conducted after the rat was connected and placed in the left (unchanged) side. This protocol was repeated for each rat whenever a new cell or set of cells were isolated. The repetition allowed for the possibility that cell activity would vary in a sequence-dependent fashion.

Data presentation and analysis

A firing field was defined as a set of at least nine contiguous pixels that shared at least one edge and with firing rate above the grand mean rate. Visual assessments of field positions were complemented by numerical analyses. To measure similarity of firing fields in the unchanged (left) and changed sides of the cylinder, a pixel-by-pixel correlation was calculated for each cell when its positional rate distribution for each side in the first (standard) session was superimposed on its positional rate distribution for the same side in the second session. Thus, we numerically estimated the similarity of firing fields for the unchanged and changed sides by calculating, for each cell, the pixel-by-pixel correlation $R_{left1\text{-}left2}$ and $R_{right1\text{-}right2}$, respectively. A high correlation was taken to reflect similar firing fields in the corresponding side of the apparatus.

Results: experiment 1

From an initial pool of 35 CA1 cells recorded from two rats, 32 satisfied our acceptance criteria and were kept for further analysis. In rat 1, 20 place cells were recorded in four sequences over 4 weeks, while 12 place cells were recorded in three sequences over 2 weeks in rat 2.

All cells fired very differently in the two sides of the cylinder, with 9 out of 32 having a distinctive field in both sides, and 23 out of 32 having a field in only one side (left 10; right 13). Thus, firing fields were selective of the specific side of the cylinder explored by the rat. Measurements of field similarity before and after the change brought to the right side of the cylinder yielded scores of 0.36 ± 0.03 and 0.32 ± 0.04 in the unchanged and changed sides, respectively ($t_{31} = 0.86$, n.s.). These similarity scores are extremely significant ($P << 0.0001$ with d.f. ≈500 in each side) and show that fields were mostly unaltered in both sides (see Fig. 1 for representative examples). The failure of fields to be altered in the changed side of the apparatus was observed regardless of the rat's experience with the change; fields were as likely to be unaltered on the first rat's exposure to the change as on the last exposure (Fig. 1).

To measure the rat's reaction to the change brought to the right side of the cylinder, the dwell times in each side were compared between sessions 1 and 2. Dwell times were normalized to account for differences in the slightly reduced area of the right side after the change. The two rats distributed their time almost evenly between

the two sides during both session 1 (left 47 ± 3%; right 53 ± 3%) and session 2 (left 46 ± 4%; right 54 ± 4%). Similarly, the number of transitions between the two sides of the apparatus was marginally, but not significantly, increased by the change in the right side (session 1, 1.17 ± 0.11 transition/min; session 2, 1.38 ± 0.08, n.s.).

Discussion: experiment 1

To examine the local vs. holistic nature of place cell firing, it is first necessary to show that a change in one region results in a change in the fields observed in that region. Only under these circumstances is it possible to determine if place cell firing is also affected in the other, unchanged region. Our results show that fields were unaltered in the changed side of the apparatus, thus precluding any significant conclusion to be drawn.

Several reasons might account for the failure of hippocampal place cells to detect the change. First, the modifications brought to the apparatus may have not been sufficient to entail that a new environment was created. Thus, even though the wall colour and shape of the right side of the cylinder were changed, the overall aspect of this region was unchanged in several aspects such as its gross geometry and topology. Second, exposure of the rats to the changed side may have not been extensive enough, as suggested by the recent demonstration that a lengthy period of experience is necessary for place cells to have distinctive firing fields in geometrically different environment (Lever *et al*., 2002). Third, it is possible that starting the rat on the unchanged side allowed it to maintain the previously established hippocampal representation even after walking into a contextually very different part of the environment. This effect might even be amplified by the extensive rat's experience of a permanent connection area between the two sides of the apparatus. Each of these factors might have contributed, singly or in combination, to the absence of remapping in the changed side of the apparatus. It is therefore impossible to draw any significant conclusion about local vs. holistic encoding of space. Accordingly, we conducted Experiment 2 with the purpose of ensuring, from the outset, that exposing the rat to different boxes would induce an immediate remapping of place cell firing.

Experiment 2

In this experiment, rats received extensive presurgery exposure to three separate boxes, a protocol demonstrated to induce reliable remapping (Lever *et al*., 2002). The rats were then exposed to a composite apparatus in which two familiar boxes were connected through a runway. Finally one box of the complex apparatus was replaced with a third, familiar box so as to measure changes in place cell firing in both changed and unchanged boxes.

Apparatus

Three boxes, chosen to be as different as possible from each other, were used. They were: (i) a white plastic circular box 48 cm in diameter and 44 cm in height; (ii) a light red square-shaped plastic box 45 cm on each side and 31 cm in height, and (iii) a brown wooden hexagon-shaped box with each side being 28 cm in length (distance between two opposite sides: 45 cm) and 33 cm in height. All boxes had open top and bottom and lay on a large piece of grey plastic paper. The three boxes had a sliding aperture 15 cm in width, which provided a salient visual and tactile polarizing cue to the animal. This aperture could be opened simply by removing the sliding portion of the box, so that it was possible, when necessary, to connect two boxes with one another. When present, the connection between two boxes was made with a U-shaped corridor runway 15 cm in width with walls 32 cm in height. The floor and wall of the runway were made of white plastic. The two ascending branches of the U-runway were 30 cm long while the horizontal branch was 50 cm long. During presurgery behavioural training, only one box was used at a time. It was positioned at the centre of a circular area delimited by a 2.5 m high opaque curtain 2.5 m in diameter. During postsurgery testing, several boxes could be used simultaneously. Therefore, the circular box was slightly moved to the left relative to the centre of the curtained area so as to leave room for either the square or the hexagonal box, which appeared slightly

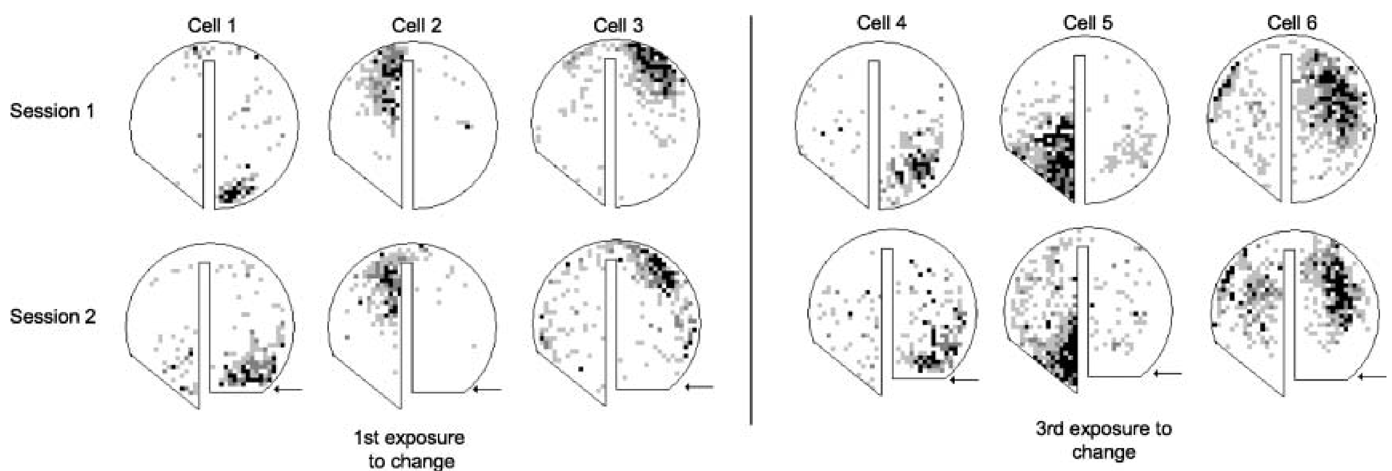

FIG. 1. Rate maps of six cells recorded in Experiment 1. Three cells were simultaneously recorded during the first and third (two weeks later) exposure of rat 2 to a change in the right side of the cylinder. Following session 1 in the familiar apparatus, the cues of the right part were changed during session 2 and a barrier was added in the right end (arrow). The firing fields in the unchanged part were recognizable across sessions. Similarly, there was almost no change in firing in the right part of the apparatus. These outcomes were observed on both exposures ($R_{left1\text{-}left2}$ values for cells 1–6: 0.49, 0.28, 0.47, 0.50, 0.36, 0.16; $R_{right1\text{-}right2}$ values for cells 1–6: 0.20, 0.45, 0.46, 0.22, 0.43, 0.31). In all maps, pixels in which no firing was observed are white. Pixels in which firing did occur are shown as grey-shaded squares. The highest firing is coded as black, and intermediate rates are shown as light grey and dark grey from low to high. Median firing rates for black (peak firing) pixels: cell 1, 10.3 AP/s; cell 2, 4.9 AP/s; cell 3, 4.9 AP/s; cell 4, 3.0 AP/s; cell 5, 9.6 AP/s; cell 6, 5.2 AP/s.

offset to the right in the TV view of the apparatus (see Fig. 2 for relative positions of boxes).

Presurgery behavioural training

Extensive behavioural training in each apparatus was carried out before electrode implantation. For three weeks before surgery, each rat was thus placed daily in each box for 15 min. This extensive period of familiarization ensured that the rat had learned the boxes and thus increased the likelihood that place cells would produce distinctive firing fields in each box (i.e. would remap). During each exposure, rats were trained to retrieve 20-mg food pellets that were delivered from the automatic food dispenser in the ceiling. As there were three boxes, the rats underwent three 15 min session per day, one session in each box in a pseudo-randomized order. At no time during presurgery training did the rats experience the connecting runway between the boxes.

Testing protocol

Each electrode in a rat was checked twice a day during performance of the pellet-chasing task in two of the three boxes. Each box was used an equal number of times to balance the rat's experience. The order in which the rat was exposed to each box during screening sessions was pseudo-random. When a cell, or set of cells, was judged suitable for recording, it was recorded for six successive sessions (Fig. 2). After each session, the rat was disconnected, removed from the apparatus, and returned to its home cage in the adjacent room. After the appropriate modification was made, the rat was brought back into the recording room, reconnected, and put into the modified apparatus facing the sliding aperture. The first four recording sessions were 10 min in duration and were conducted while the rat explored one of the individual boxes in the following order: session 1 in the circle, session 2 in the square, session 3 in the circle and session 4 in the hexagon. The purpose of these four sessions was to document the remapping phenomenon under traditional conditions.

Before session 5, the sliding openings of the circular and hexagonal boxes were removed and the connection between the two boxes was established by adding the U-shaped runway corridor. The rat was returned to the recording room, reconnected, and put into the circle. The recording session was started immediately while the rat could commute between the circle and the hexagon.

For session 6, the hexagonal box was removed and replaced with the square box while the U-shaped runway remained in place. The rat was returned to the recording room, reconnected, and put into the circular box for the last recording session. The rat could now commute between the circle and the square rather than between the circle and the hexagon. Both sessions 5 and 6 were 20 min in duration. The complete 6-session protocol was repeated for each rat whenever a new cell or set of cells were isolated.

Data presentation and analysis

As in Experiment 1, a firing field was defined as a set of at least nine contiguous pixels that shared at least one edge and with firing rate above the grand mean rate. Visual assessments of field positions were complemented by numerical analyses. To measure similarity of firing fields in the different conditions, a pixel-by-pixel correlation was calculated for each cell when its positional rate distribution for a given session/box was superimposed on its positional rate distribution for a different session/box. Thus, we numerically estimated the similarity of firing fields for the circular box by calculating, for each cell, the pixel-by-pixel correlation $R_{cir1\text{-}cir3}$ between the positional firing distributions observed in sessions 1 and 3. A high correlation $R_{cir1\text{-}cir3}$ was taken to reflect similar cell's positional firing patterns in the circle. In the same way, to numerically estimate the similarity of firing between two different boxes, we calculated the pixel-by-pixel correlations $R_{cir1\text{-}sq2}$ between the positional firing distributions for the circle (session 1) and the translated square (session 2), $R_{cir3\text{-}hex4}$ between the positional firing distributions for the circle (session 3) and the hexagon (session 4), and finally $R_{sq2\text{-}hex4}$ between the positional firing distributions for the square (session 2) and the translated hexagon (session 4). Non-overlapping pixels were excluded from the analysis. Although this method does not allow measuring subtle topological transforms of firing fields, it is sufficient to assess the gross changes expected if remapping occurs. Thus, if the cells remapped their activity when the rat was transferred from one box to another, the difference in associated positional firing patterns had to yield low correlations.

The same type of analysis was applied to the composite apparatus used in both sessions 5 and 6. For these sessions, however, the analysis was broken down into specific regions of interest. To measure the overall effect of adding the connection between the circle and the hexagon (session 5), two regions of interest were considered. On one hand, we cross-correlated, for each cell, the firing fields in the circle during sessions 3 and 5 ($R_{cir3\text{-}cir5}$). On the other hand, we cross-correlated the cell firing fields in the hexagon during sessions 4 and 5 ($R_{hex4\text{-}hex5}$). If adding the runway corridor between the two boxes had no effect on the firing fields in each individual box, the two correlations $R_{cir3\text{-}cir5}$ and $R_{hex4\text{-}hex5}$ should be high.

The effects of substituting the hexagonal box with the square box during session 6 were measured in two ways. First, we compared firing activity in the region that was unchanged across sessions 5 and 6 (i.e. the area combining the circular box and the runway, $R_{unchanged}$). We made the same comparison for the region that was changed across sessions 5 and 6 (i.e. $R_{hex5\text{-}sq6}$). This latter correlation was expected to be weak if remapping had indeed occurred. In contrast, the former correlation, $R_{unchanged}$, was expected to be high if box substitution only affected the local properties of cell firing, but low if box substitution had a more general effect that spread across regions distant from the change. Finally, to document in more detail possible changes within the unchanged region of the apparatus, we also calculated the cross-correlations between the firing fields observed in the circular box ($R_{cir5\text{-}cir6}$) and in the runway ($R_{run5\text{-}run6}$) during sessions 5 and 6. The major reason for conducting the latter analysis is because the runway was in the direct vicinity of the changed box whereas the circular box was some distance away

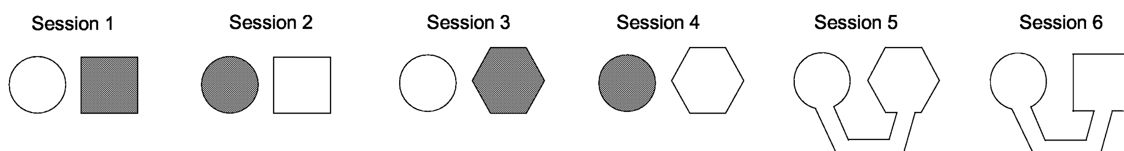

FIG. 2. Recording protocol (Experiment 2). Two boxes were present during the first four sessions, but only one box (shown in white) was used at a time (the area not accessible to the rat on each recording session is shown in dark grey). The boxes were at the centre of a curtained area (not shown). The rat was disconnected and returned to its home cage between each pair of recording sessions. The first four sessions were 10 min in duration; the last two sessions were 20 min in duration.

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from it. Thus, it could be expected that the effects of box substitution might affect cell firing in a more obvious way for the runway than for the circle.

Results: experiment 2

From an initial pool of 115 cells recorded from five rats, only 85 cells (69 from CA1, 16 from CA3) satisfied our acceptance criteria (see section on unit discrimination) and were kept for further analysis. As no obvious difference was observed among cells in these two regions, they were all processed in the same manner. The number of completed sequences of six recording sessions and recorded place cells varied considerably between rats (rat 1, five cells recorded in three sequences over 4 weeks; rat 2, 11 cells, five sequences, 12 weeks; rat 3, 33 cells, nine sequences, 10 weeks; rat 4, 32 cells, eight sequences, seven weeks; rat 5, four cells, two sequences, two weeks).

Cell firing in individual boxes

Of the 85 place cells recorded in the three boxes, 14 had a firing field in one box only, 32 had a field in two boxes, and 39 had a field in all three boxes. Table 1 summarizes the number of cells that fall into these different categories for each box or combination of boxes.

Figure 3 shows firing rate maps obtained from a representative subset of six different place cells in the three boxes. For a vast majority of cells, the firing fields observed in different boxes were clearly distinguishable. Cell firing fields were stable in the two sessions performed with the circle (mean similarity score, $R_{cir1\text{-}cir3} = 0.41 \pm 0.04$ calculated on an average of 200 pixels). In contrast, firing fields in the different boxes were strikingly different as shown by the very low mean similarity scores ($R_{cir1\text{-}sq2} = -0.01 \pm 0.01$; $R_{cir3\text{-}hex4} = 0.02 \pm 0.02$; $R_{sq2\text{-}hex4} = -0.02 \pm 0.01$). Because correlation coefficients are not normally distributed, paired *t*-tests were conducted on their *z*-transforms. These analyses confirmed that similarity scores for the sessions conducted with the circle were significantly greater than 0.0 ($z = 0.55 \pm 0.08$, $P < 0.001$). On the contrary, none of the similarity scores for the different boxes was significantly different from 0.0 ($-0.01 < z < 0.02$, n.s.). Only in three cells did the fields look similar in the square and the hexagon (i.e. similarity scores were > 0.139, $P < 0.05$), an occurrence so rare that it can be expected to happen just by chance. Finally, a *t*-test confirmed that field similarity was much greater for sessions run with the circle than for sessions run in different boxes ($P < 0.001$). Thus, as expected, firing fields were clearly stable in the circle and distinguishable in different boxes.

As almost half of the cells had a field in all three boxes, the possibility exists that fields in one box were simply rotational equivalents of fields in other boxes. This hypothesis of rotational remapping (Bostock *et al.*, 1991) states that place fields maintain the same distance and angular relationships to each other, but the orientation of the entire representation rotates to a new bearing in the environment (Knierim, 2003). In the present study, rotational remapping is possible, for example, if the rat fails to identify polarizing cues in the box, in spite of the salient visual and tactile cues provided by the sliding aperture (note, however, that field stability in the circle argues against random field rotation across sessions). According to the rotational remapping hypothesis, if field ensembles in one box are rotational equivalents of field ensembles in another box, the rotation angles of the two cells should be similar (and therefore the difference of rotation angles close to zero). To test this hypothesis, we analysed pairs of simultaneously recorded cells that happened to have a field in at least two boxes. Pixel-by-pixel cross-correlations between the firing rate maps of each cell were calculated as its positional firing pattern in one box was rotated against its positional firing pattern in the other box in 1° steps. The rotation associated with the highest correlation (R_{max}) was taken as the rotation of the field between the two boxes. The same analysis was carried out for the second, simultaneously recorded cell. We conducted this analysis on seven cell pairs with a field in two boxes and 22 cell pairs with a field in three boxes for a total of 73 comparisons. Figure 4A shows representative maps of two simultaneously recorded cells. Both had a field in the

TABLE 1. Number of firing fields in each box, or combination of boxes (Experiment 2)

Box(es)	Number of firng fields
One box	
Circle	8
Square	6
Hexagon	0
Two boxes	
Circle + Square	18
Circle + Hexagon	6
Square + Hexagon	8
Three boxes	
Circle + Square + Hexagon	39

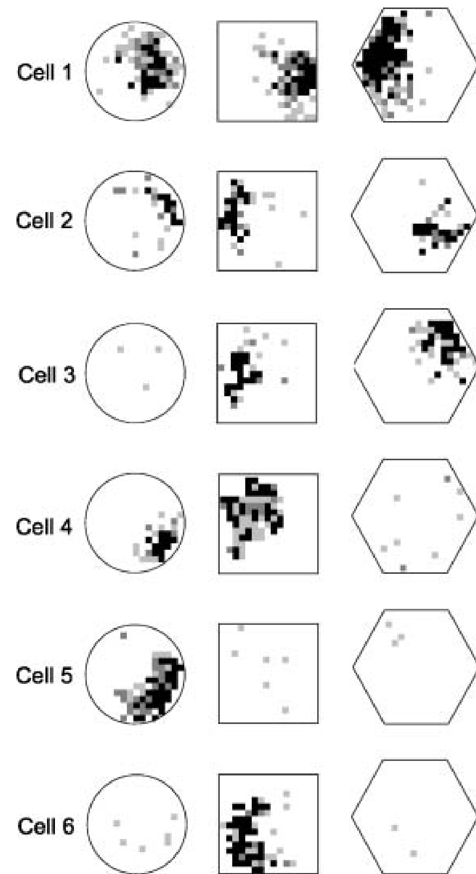

FIG. 3. Rate maps of six cells in the different boxes used in Experiment 2. Cells 1 and 2 had a firing field in all three boxes; cells 3 and 4 had a field in two boxes; cells 5 and 6 had a field in one box only. Median firing rates for black (peak firing) pixels: cell 1, 10.3 AP/s; cell 2, 4.9 AP/s; cell 3, 4.9 AP/s; cell 4, 10.8 AP/s; cell 5, 4.2 AP/s; cell 6, 1.2 AP/s.

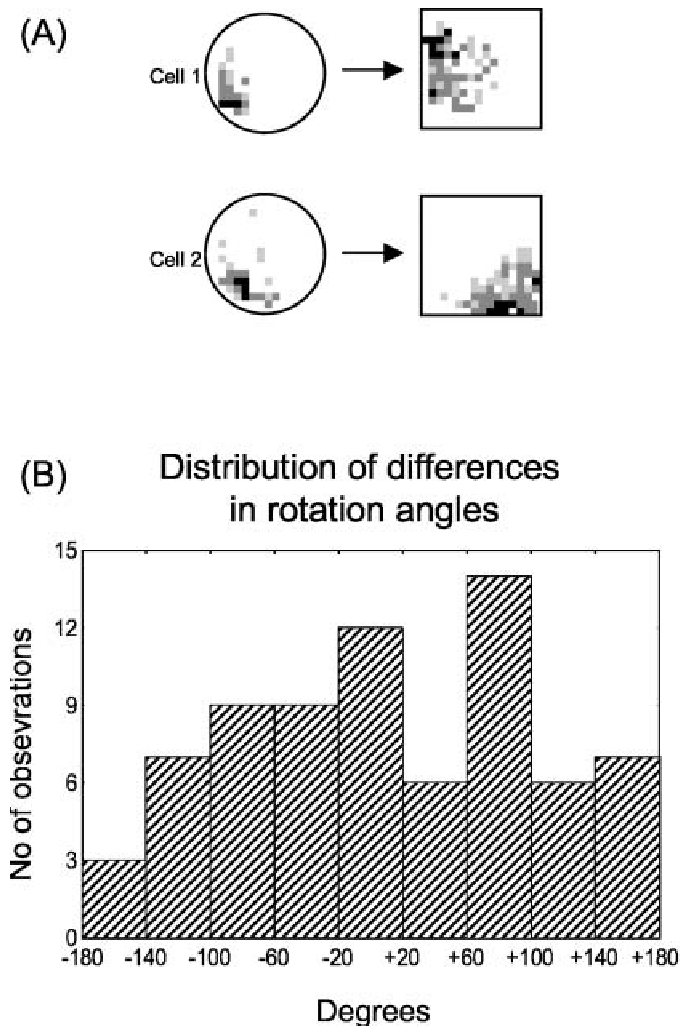

FIG. 4. Testing the rotational remapping hypothesis (Experiment 2). (A) Rate maps of two simultaneously recorded cells in the circle and the square. Both cells had a field in each box. The rotations needed to obtain the best match between the fields in circle and square were 39° ($R_{max} = 0.412$) for cell 1 and 291° ($R_{max} = 0.491$) for cell 2. The difference between the two rotation angles (252°, i.e. −108°) makes it unlikely that fields in one box were simple rotational equivalents of fields in the other box (such rotational remapping would predict a difference close to 0°). Median firing rates for black (peak firing) pixels: cell 1, circle 4.1 AP/s, square 7.7 AP/s; cell 2, circle 3.6 AP/s, square 11.2 AP/s. (B) Distribution of the differences in field rotation of simultaneously recorded cells. The rotation needed to obtain the best match between the fields in two boxes was calculated for each pair of simultaneously recorded cells that had a field in at least two boxes. The difference between the rotation angles for each pair of cell/box was then plotted in 40° bins. As there was no strong bias for the 0°-centred bin (from −20° to +20°), it is unlikely that field ensembles in one box were rotational equivalents of field ensembles in the other box, thus allowing us to discard the rotational remapping hypothesis.

circle and the square, but it is clear that different rotations are needed to superimpose the field of cell 1 and the field of cell 2 in the two boxes. The distribution of differences in rotation angles (Fig. 4B) failed to reveal a significant bias for 0° ($\chi^2 = 10.9$, d.f. = 8, n.s.), thus making it unlikely that field ensembles in one box were rotational equivalents of field ensembles in other boxes. Additionally, eight of the 12 near-0° values in the histogram came from triplets of simultaneously recorded fields that did not rotate congruently. To sum up, no support was found for the hypothesis that the existence of cells with fields in several boxes was caused by rotational remapping.

Effects of adding the connecting runway on cell firing

Adding the runway between the circular and hexagonal boxes induced complex effects that for the most part are illustrated in the rate maps shown in Fig. 5. Each row shows the firing fields for a different cell; the first three columns show firing fields in individual boxes, and the fourth column is for the session after connection of the circle and hexagon. Remapping can include quite different categories of firing modifications such as modification of field location and shape, cessation of firing or conversely, newly formed fields by previously silent cells (Muller & Kubie, 1987). In addition, such effects can be specific to a box or occur in both boxes. Thus, a first measure of the effects of runway addition is simply the number of cells whose activity was changed one way or another in either the circle or the hexagon. This count revealed that 31 out of 85 (36%) cells had altered activity as a result of runway addition. Activity of the remaining 54 out of 85 (64%) cells was unchanged.

Considering only cells that had a field in either the circle or the hexagon (see Table 1), the most common outcome, seen in 52 out of 79 cells (66%), was that fields were unchanged (cells 1–3, Fig. 5). These effects were reflected in the large similarity scores, which were significantly above zero (hexagon, mean $R_{hex4\text{-}hex5} = 0.40 \pm 0.03$; $z = 0.48 \pm 0.05$, $P < 0.001$; circle, $R_{cir3\text{-}cir5} = 0.27 \pm 0.04$; $z = 0.40 \pm 0.08$, $P < 0.001$). With regard to the hexagon, 12 out of 53 (19%) fields had low similarity scores (< 0.139), thereby indicating a significant change in firing. Nine of these 12 fields were located in the area next to the newly available opening (cell 5, Fig. 5). With regard to the circle, 26 out of 71 (37%) fields underwent significant changes after runway addition (cell 4, Fig. 5). Contrary to the hexagon, there was no relation between the location of the field in the circle and the likelihood it would remap. That is, even fields that were away from the runway were likely to change after it was added. Two additional observations are that four out of six cells initially silent developed a new field in either the circle or hexagon after runway addition, and that many cells (67 out of 85, 79%) developed a new field in the runway (cell 2, Fig. 5).

Effects of box substitution on cell firing

The crucial manipulation made before conducting session 6 consisted of substituting the hexagonal box with the square box while leaving unchanged the circular box and runway. As for the effects of runway addition, we first counted the number of cells whose activity was changed by box substitution. Changing the hexagon to a square in session 6 resulted in altered firing for a vast majority of cells (71 out of 85, 83%) in the substituted box (i.e. hexagon vs. square), but only for a small number in the unchanged circle and runway (13 out of 85, 15%). One cell previously silent in the circle developed a new field after box substitution (cell 5 in Fig. 5). Ten of the 71 fields (16%) in the circle and 2 out of 67 (3%) fields in the runway had low similarity scores indicative of remapping. Thus, for the most part fields previously established in the unchanged region were stable (see representative examples in the right-most two columns of Fig. 5).

These effects were confirmed by the similarity scores which were significantly above 0.0 for the unchanged region (i.e. circle and runway; $R_{unchanged} = 0.43 \pm 0.03$, $z = 0.63 \pm 0.08$, $P >> 0.001$) but not distinguishable from 0.0 for the changed box ($R_{changed} = 0.03 \pm 0.01$, $z = 0.03 \pm 0.01$, n.s.). The fields seen in the square during session 6 were generally similar to those seen during session 2 ($R_{sq2\text{-}sq6} = 0.33 \pm 0.03$, $z = 0.45 \pm 0.06$, $P < 0.001$) although similarity scores were low for 18 out of 71 fields (25%), indicative of some remapping (cell 3, Fig. 5). The distribution of correlation coefficients

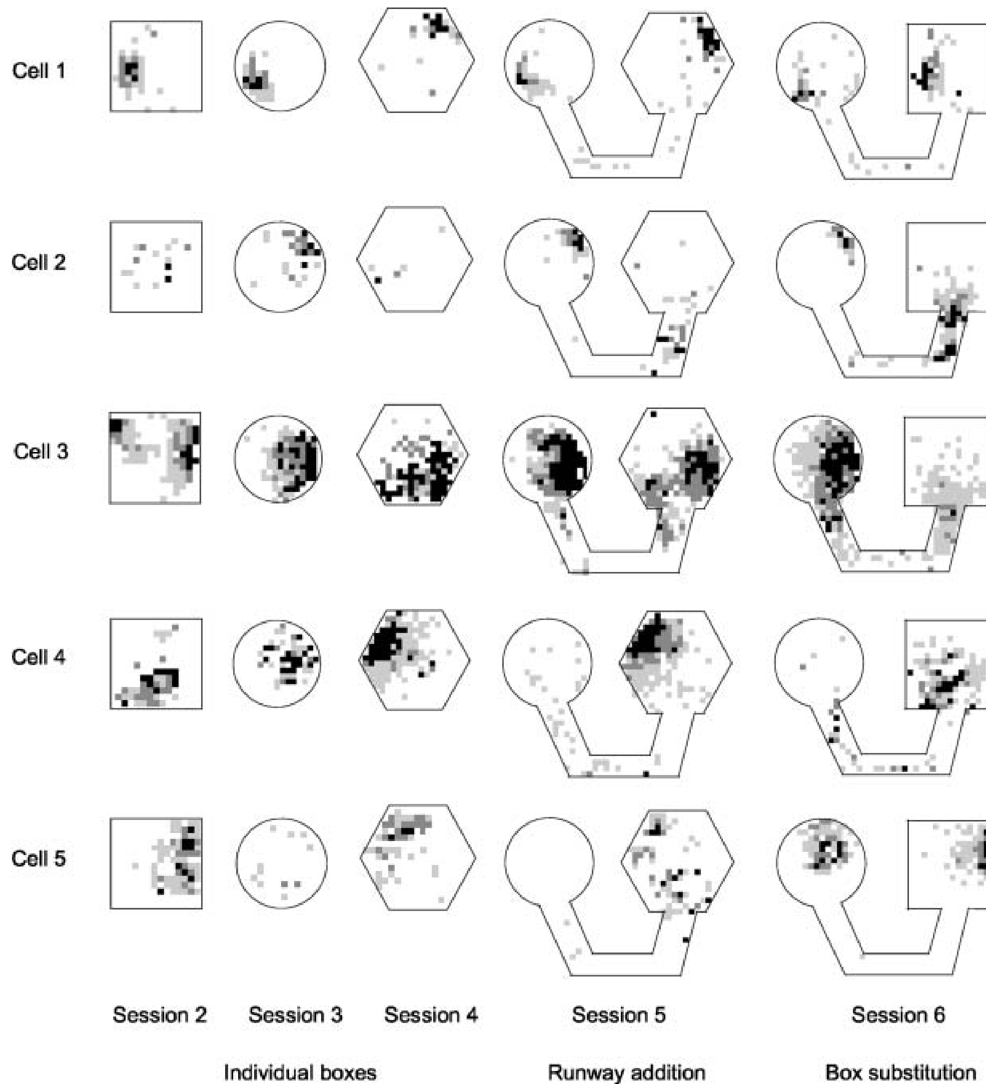

FIG. 5. Effects of runway addition and box substitution (Experiment 2). The rate maps of five cells are shown for sessions 2–6. Runway addition had only minimal effects on cells 1–3. Cell 4 ceased firing in the circle when the runway connected the two boxes, whereas cell 5 developed a weak field at the boundary between the hexagon and the runway. After box substitution, all fields underwent some remapping in the changed box ($R_{changed}$ values for cells 1–5: −0.07, 0.18, 0.05, −0.10, −0.14). Nevertheless, the firing of cells 1–4 in the unchanged region (circle + runway) was hardly affected by box substitution ($R_{unchanged}$ values for cells 1–4: 0.74, 0.44, 0.64, 0.35). In contrast, cell 5, which was initially silent, developed a new field ($R_{unchanged}$ −0.04; such remapping in the circle was observed for only two cells). Lastly, cell 3 had a clearly different field in the square after box substitution. Median firing rates for black (peak firing) pixels: cell 1, 29.2 AP/s; cell 2, 12.0 AP/s; cell 3, 4.0 AP/s; cell 4, 25.7 AP/s; cell 5, 6.2 AP/s.

for the changed and unchanged regions of the apparatus is shown in a histogram (see Fig. 6) from which it is evident that most fields remapped in the changed region but remain essentially the same in the unchanged region ($t_{84} = 7.52$, $P >> 0.001$).

Measurements of field similarity in more restricted regions confirmed this impression. Fields were generally unaffected by box substitution in either the circle ($R_{cir5\text{-}cir6} = 0.43 \pm 0.03$, $z = 0.59 \pm 0.06$, $P >> 0.001$) or the runway ($R_{run5\text{-}run6} = 0.40 \pm 0.03$, $z = 0.63 \pm 0.08$, $P < 0.001$) though similarity was slightly but not significantly decreased in the runway compared to the circle ($t_{84} = 1.12$, n.s.). This was caused by two runway fields near the changed box that were altered by box substitution (cell 2, Fig. 5).

A detailed inspection of the temporal firing patterns for the fields near the changed box revealed that these alterations tended to occur abruptly from the first exposure to the square. For three cells that ceased firing after box substitution, no firing was ever observed at the boundary between the runway and the square, thus suggesting that cessation of firing was immediate. For cells that instead developed a new field at the border between the runway and the square, the rat's path was replayed until firing occurred in the newly observed field. Of three cells so analysed, one fired in the new field during the rat's first pass through the field location while the other two cells fired during the second pass (time of firing is not relevant for this analysis as it depends on the rat's exact path). The same analysis, performed for remapped fields in the changed box, revealed that for all cells the modifications in firing occurred during the first pass through the field, again suggesting immediate adaptation of cell firing to environmental changes.

Consistency of effects induced by box substitution

Table 2 provides an overall summary of the effects of box substitution. A cell was considered to remap its activity if it met any of the following three conditions: (i) its field similarity score

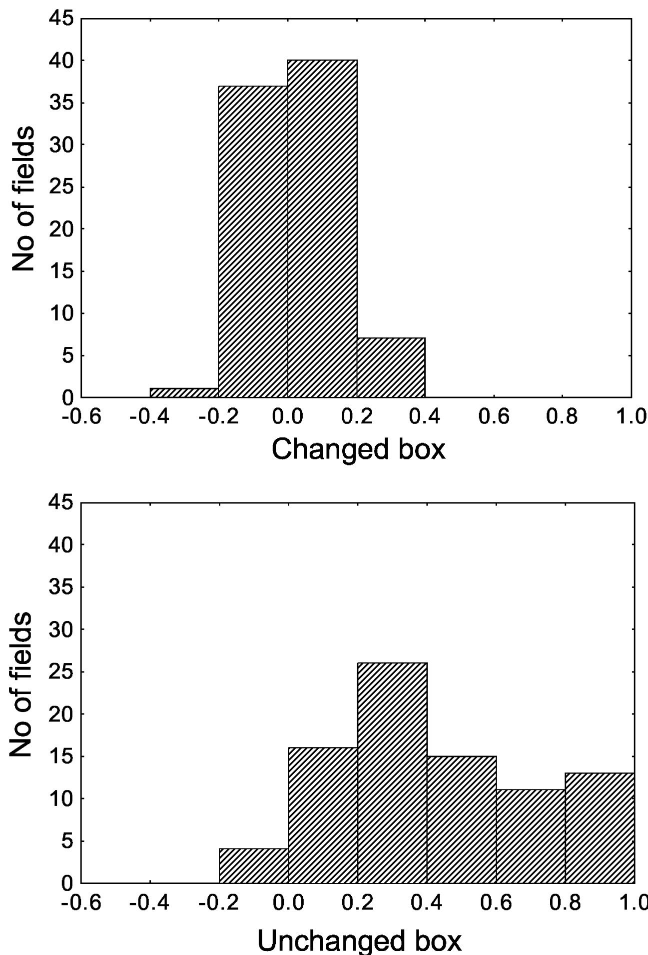

FIG. 6. Distribution of correlation coefficients for the changed and unchanged regions of the apparatus (Experiment 2). Most fields in the unchanged region were essentially unaffected by box substitution whereas fields in the changed region remapped, as shown by the leftwards-shifted distribution of the correlation coefficients.

TABLE 2. Across rat distribution of remapping cells in the changed and unchanged region (Experiment 2)

	Number of remapping cells after box substitution	
	Changed region (hexagon vs. square)	Unchanged region (circle + runway)
Rat 1	4/5	0/5
Rat 2	10/11	2/11
Rat 3	28/33	3/33
Rat 4	25/32	8/32
Rat 5	4/4	0/4
Total	71/85	13/85

A cell was considered to remap if (i) field similarity was low (< 0.139); (ii) from silent it became active after box substitution, or (iii) it stopped firing.

was less than 0.139 [i.e. the probability that its field was unchanged was > 0.05 with d.f. ≈200 (number of pixels used for computing correlations)]; (ii) from previously silent, it became active, or (iii) from previously active, it ceased firing. The data of Table 2 confirm that the patterns of changed and unchanged cell activity were markedly different in the substituted box and the stable circle ($\chi^2 = 10.3$, d.f. = 4, $P < 0.035$) and additionally reveal that this result was consistent across all five rats. We also asked if there was any effect of repeated exposure to box substitution. Figure 7 shows the percentage of remapped fields for both unchanged (i.e. circle and runway) and changed regions of the apparatus over successive recording days. Again, the effects were very consistent across recording days and there was no reliable effect of successive exposures on the occurrence of remapping in either the changed or unchanged region (both *F*s < 1). Thus, box substitution induced fields to remap in the changed region and to remain unaltered in the unchanged region in all five rats. This pattern was observed from the first to the last exposure to box substitution.

Behavioural effects of box substitution

To see if and how the rats reacted to box substitution, the dwell times in each region of the apparatus were compared between session 5 and session 6. Dwell times were normalized to account for differences in the area of the boxes and runway. While rats distributed their time almost evenly in the three regions during session 5 (circle, 28%; hexagon, 33%; runway, 39%), a significant bias for the square was observed during session 6 (circle, 24%; square, 45%; runway, 31%; $P < 0.001$ for square vs. other boxes). In contrast, the number of visits to each region was unchanged after box substitution (session 5, 16.4 ± 1.2; session 6, 17.4 ± 1.5, n.s.). Thus, on average rats commuted back and forth as frequently during both sessions 5 and 6, but tended to make longer visits to the changed box during session 6. Although we cannot discard a spontaneous preference for the square box, this pattern may reflect detection of the change by the rat.

Discussion: experiment 2

The principle of Experiment 2 was to ensure, from the outset, that the rat would activate a different hippocampal map, specific to each box so that changing one box within the connected apparatus would reliably result in field remapping in that box. The boxes and connecting runway were made as different as possible so that each

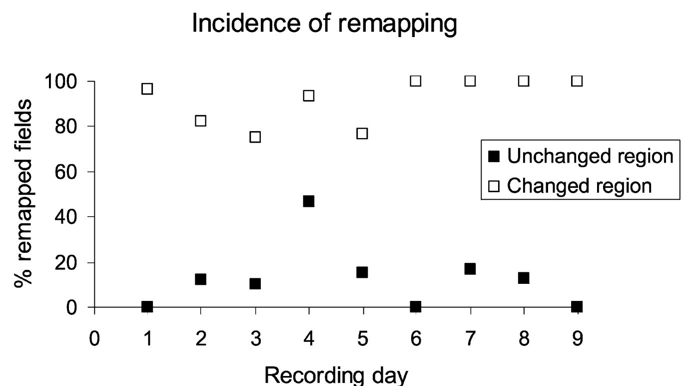

FIG. 7. Time-course of the incidence of remapping across successive exposure to box substitution (Experiment 2). There was no clear-cut experience-dependent change in the incidence of remapping in either the changed or unchanged regions.

part of the apparatus differed in texture, shape, colour, and size. Furthermore, the boxes had walls high enough to provide independent environments that were out of view of each other, as in previous studies of remapping (Muller & Kubie, 1987; Kentros *et al*., 1998; Tanila, 1999; Oler & Markus, 2000; Lever *et al*., 2002). Rats were made familiar with each box prior to unit recordings. As suggested by Lever *et al*. (2002), this increased the likelihood that cells would fire in a distinctive way in each box. The consistent remapping observed when recordings were made with different boxes indicates that these conditions were sufficient to convince the rat's hippocampal place cells that the circular, square, and hexagonal boxes were different.

Under these circumstances, we showed that (i) adding a runway connection between two separate boxes preserves a significant fraction of the firing fields observed in each box even though some local changes are observed; (ii) substituting one of the two connected boxes with a third box results, as expected, in field remapping in the changed box, and (iii) fields in the unchanged, connected box are unaltered. Overall, this result suggests that the hippocampus holds a set of independent local maps.

The addition of a runway between two previously unconnected boxes induced complex effects. Although a substantial proportion of fields in the boxes was unaffected by the newly available runway, evidence for remapping was found in approximately 36% of the cells. Furthermore, remapping was more prevalent in the circular box than in the hexagonal box. Lastly, the few fields that were seen to change in the hexagonal box after runway addition were located near the box opening. It is therefore likely that the change in those fields was caused by local modifications of visual and topological properties induced by opening the hexagonal box. It is clear that, if place cells are part of a navigation system, the topology of space must be an important determinant of their activity (Muller *et al*., 1996; Gaussier *et al*., 2002) and topological changes must result in changes in place cell activity. In contrast, field changes in the circular box were usually unrelated to the field location in that box. One possibility for the deeper alteration of fields in the circular box following runway addition is that the rat was consistently introduced in that box on each recording session. As the first task for the rat to accomplish when introduced into the apparatus is to locate itself, it is arguable that attending to details of the environment may result in increased detection of the change. Whatever the explanation, the effects observed when a new connection is established between two familiar environments suggest that the connectivity of space influences hippocampal representations. Rather than inducing a complete remapping, however, new connections alter activity only of a subset of place cells. This effect is similar in many respects to the partial remapping phenomenon described by Tanila *et al*. (1997) and Knierim (2002) and suggests that at least in some conditions, previous representations can be modified, but not replaced, by new information. This result confirms that hippocampal activity may reflect regularities in an environment in a very flexible way, such that incoming information can readily be incorporated into existing representations (Eichenbaum *et al*., 1999).

The critical result of Experiment 2 is that when one box was changed, therefore resulting in a clear remapping of firing fields in that box, place cell activity was usually unaffected in the other, unchanged, box. That is, even though the rat demonstrably experienced a change in a region of the environment, this did not affect its recognition of the unchanged region. This result supports the local encoding hypothesis in which place cells are primarily driven by the rat's current environment. We address the significance of this finding in the general discussion.

General discussion

By virtue of their location-specific activity, place cells provide information about the rat's current position in the environment (O'Keefe & Dostrovsky, 1971). As a collective, however, the subset of active place cells also provides a signature of the rat's current environment. That is, the ensemble of place cells sustains a pattern of activity specific to the spatial environment currently experienced by the rat (Muller & Kubie, 1987). In this study, we questioned the nature of the spatial representation encoded by place cells. Specifically, we asked whether the hippocampus holds a set of independent maps, with place cells being primarily sensory-driven by the local environment, or if it holds a more general representation, in which each environment-specific local map is related to other local maps. This alternative possibility entails place cells receiving information not only from sensory data, but also from other place cells, some of which convey information about remote parts of the environment. By this view, alteration to one part of the environment should propagate across the whole network and affect even those cells representing perceptually unchanged regions.

Experiment 1 revealed that some precautions must be taken to address this issue. More specifically, the recording protocol must be designed in such a way that exposing the rat to a new box will reliably result in field remapping in that box. Experiment 2 therefore comprised three distinct stages. First, rats were exposed to three distinctive boxes in which place cells yielded distinguishable firing fields. Then, a runway connection was added between two separate boxes. Finally, substituting one of the two connected boxes with a third box resulted in field remapping in the changed box but unchanged firing fields in the unchanged box. Our main conclusion is that, under these circumstances, the hippocampus holds a set of independent local maps.

Interestingly, this result closely parallels those of a previous study of head direction cells (Taube & Burton, 1995). In this study, rats commuted between two connected environments. Rotation of cell's preferred directions was induced in one of the environments by rotating a prominent visual cue. Firing in the other environment was nevertheless unchanged, thus demonstrating independent representation of the two environments by head direction cells, much as for place cells in the present work. The parallel between the two studies also strengthens the idea that both head direction and place cells are part of a tightly coupled network whose activity is triggered by similar sensory inputs (Knierim *et al*., 1995; Jeffery *et al*., 1997).

Several studies have demonstrated that remapping often occurs in an all-or-none fashion. For example, Cressant *et al*. (2002) recorded place cells while rat explored a T-maze at the centre of a room that contained a large number of available cues. They found that rotating the maze by 45° produces a complete remapping, suggestive of the replacement of the entire map. Therefore, the present results appear to conflict with the cohesive hippocampal map revealed by the study of Cressant *et al*. (2002). A cohesive map would suggest rather that a remapping should be observed everywhere in the environment, including the unchanged regions. Such an outcome was very unusual in the present study. The most plausible explanation for this discrepancy is that while the rat had full visual access to the external environment in the study of Cressant *et al*. (2002), in the present design it was prevented from seeing the contents of a box when it was in the other box. Thus, the two boxes constituted visually independent environments, making it very likely that the rat would activate a distinct map for each box rather than a single map as in the T-maze study.

The process by which distinct maps were activated in time as the rat moved from one box to another is also of interest, as the shift from one activity pattern to another was immediate. That is, place cells were firing in the appropriate location in a given box on the very first exploration episode of that box as if mere exposure to that box was sufficient to re-activate the corresponding map. This observation confirms the findings of previous experiments that studied changes in place cell activity as rats ran back and forth between two separate environments (Wilson & McNaughton, 1993; Skaggs & McNaughton, 1998; Tanila, 1999). For example, Skaggs & McNaughton (1998) gave rats access to two visually identical square boxes connected by a narrow runway. To an extent that varied across rats, some cells had distinct firing fields in each box whereas other cells had apparently identical fields in the two boxes. For each class of cell, each exploratory episode of a specific box evoked the activity pattern to that box. A related finding was reported by Tanila (1999) who demonstrated that a transition from a familiar environment into a novel but visually identical environment was associated with strong changes in place fields. Both Skaggs & McNaughton (1998) and Tanila (1999), however, found that a fraction of the place cells were likely to adopt the same firing patterns in the two visually similar environments. In contrast, when the connected boxes are visually different (present study), place cell firing patterns are consistently distinguishable and specific to each box. Thus, there is a body of converging data showing that place cells can switch between several activity states in accordance with both the local environment and the rat's location within that environment. The new finding here is that each of these activity states appears independent of the others even when the animal is allowed to move freely between separate environments. The dynamics of such activity states is an open question, however. Because sampling place cell activity requires time to get an accurate view of firing fields, it is difficult from the present study to understand how the switch process operates. Based on the relative independence of the fields in one box with respect to what happens in the other box, it may be argued that the time-scale of the switch is short, therefore suggesting the operation of a strong attractor (Wilson & McNaughton, 1993). This conclusion, however, is at odds with the recently reported weak nature of the attractor that triggers the shifts in place cell activity (Knierim, 2002). Thus, our finding that the hippocampus holds relatively independent maps for each environment also raises the issue of the temporal dynamics of activity states in the hippocampus, an issue that will require simultaneous recording of many place cells.

Although Experiment 2 yielded clear-cut results suggesting that place cells can encode parts of an extended connected space independently, we must acknowledge that using a different protocol might have provided a different outcome. In other words, it is possible that, under certain circumstances, place cells show more global encoding of space. For example, the existence of distinct boundaries between the boxes might promote local encoding. Thus, the lack of such clear boundaries in Experiment 1 might be the reason why fields did not remap in the changed part. Among the other critical factors to manipulate, the order in which the rats experienced the changed and unchanged box might be important. In this view, firing in the circular box might be more stable after changing the other box because it was experienced first. Whether the same outcome is observed when the rat is exposed to the changed box first is an open question. Finally, it will be important to test the effects of pretraining. In fact, pretraining in separate environments may have biased place cells to favour a local representation, and it is unclear if a more holistic representation might have occurred if the environments were connected from the beginning. Thus, different factors may have contributed to the observed pattern of independent coding of space. Even though further studies will be necessary to disentangle the role of each factor, it remains that the phenomenon of local coding observed in the present study can be useful for addressing many questions about the functional properties and the temporal dynamics of place cell firing. In a more general perspective, the observation of both partial remapping (after connection of two separate boxes) and independent coding of each environment (following box substitution) may reflect the capability of hippocampal representations to encode the salient features of a situation and integrate new and familiar information in overlapping memory representations (Eichenbaum *et al*., 1999).

Acknowledgements

Support for this work was provided by the French Centre National de la Recherche Scientifique and Ministère de la Recherche (ACI Neurosciences Intégratives et Computationnelles) and the Mexican Consejo Nacional de la Ciencia y la Tecnologia (CONACYT grant # 149872 to VPV). We thank J.-P. Banquet, R. Chatila, P. Gaussier, M. Quoy and all members of the ACI Group for stimulating discussions and helpful suggestions.

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