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Hippocampal Place Cells Encode Intended Destination, and Not a Discriminative Stimulus, in a Conditional T-Maze Task

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ABSTRACT: The firing of hippocampal place cells encodes instantaneous location but can also reflect where the animal is heading (prospective firing), or where it has just come from (retrospective firing). The current experiment sought to explicitly control the prospective firing of place cells with a visual discriminada in a T-maze. Rats were trained to associate a specific visual stimulus (e.g., a flashing light) with the occurrence of reward in a specific location (e.g., the left arm of the T). A different visual stimulus (e.g., a constant light) signaled the availability of reward in the opposite arm of the T. After this discrimination had been acquired, rats were implanted with electrodes in the CA1 layer of the hippocampus. Place cells were then identified and recorded as the animals performed the discrimination task, and the presentation of the visual stimulus was manipulated. A subset of CA1 place cells fired at different rates on the central stem of the T depending on the animal's intended destination, but this conditional or prospective firing was independent of the visual discriminative stimulus. The firing rate of some place cells was, however, modulated by changes in the timing of presentation of the visual stimulus. Thus, place cells fired prospectively, but this firing did not appear to be controlled, directly, by a salient visual stimulus that controlled behavior. © **2011 Wiley Periodicals, Inc.**

KEY WORDS: spatial cognition; goals; trajectory encoding; memory; place fields

INTRODUCTION

Place cells in the hippocampus encode both an animal's location and aspects of its ongoing behavior. For example, individual place cells fire at different rates within their place fields when the rat is traveling to or from different locations (Markus et al., 1995; Frank et al., 2000; Wood et al., 2000; Ferbiteneau and Shapiro, 2003; Bower et al., 2005; Dayawansa et al., 2006; Lee et al., 2006; Smith and Mizumori, 2006; Ainge et al., 2007a; Lipton et al., 2007; Ji and Wilson, 2008) or when it is under different motivational states (Kennedy and Shapiro, 2009). Thus, the activity of some place cells may be described as conditional; whether a cell fires at a high rate or not within its place field depends on aspects of the animal's experience. More recently, studies have shown that place cells anticipate an animal's future destination (Ainge et al., 2007a; Johnson and Redish, 2007; Pastalkova et al., 2008).

How does conditional place field activity arise? One possibility is that place cells are driven by both spatial information—the physical location of the animal—and contextual inputs (Jeffery et al., 2004; Kennedy and Shapiro, 2009; c.f. Oler et al., 2008). These contextual inputs range from the presence of a specific odor (Anderson and Jeffery, 2003) to the prospective choice of a specific destination from several alternatives (Ainge et al., 2007a). If a given place cell is driven by both location and contextual inputs, then the removal of the latter should cause place cells to lose any contextual modulation. Thus, the same place cell may exhibit conditional activity in one situation, and nonconditional activity in another.

Earlier work has shown that, in the absence of polarising cues, the location of place fields on a maze with equivalent arms may reflect the rat's perspective on the location of reward (O'Keefe and Speakman, 1987). In the current experiment, we examined a separate, but related issue: Can the prospective activity of place cells be controlled by a stimulus that indicates where food is located?

To address this question, we devised a conditional discrimination task in which rats traveled through a specific location either in the presence or absence of information about the reward location provided by a light cue. Critically, on some trials, this light would be active before the rat traveled through a given place field; in other trials, the light came on after the animal passed through a place field. We hypothesized that place cells on the central stem of the maze would fire prospectively—with respect to the animal's intended destination—when information about the reward location was available, but the same cells would exhibit traditional fields when the animal traversed the central stem without knowing which arm contained reward. Our results show that conditional place cell activity in CA1 is not controlled by a conditional stimulus, but instead reflects the intended trajectory of the animal.

MATERIALS AND METHODS

Subjects

Six male Lister hooded rats served as subjects in this experiment. The rats were housed individually

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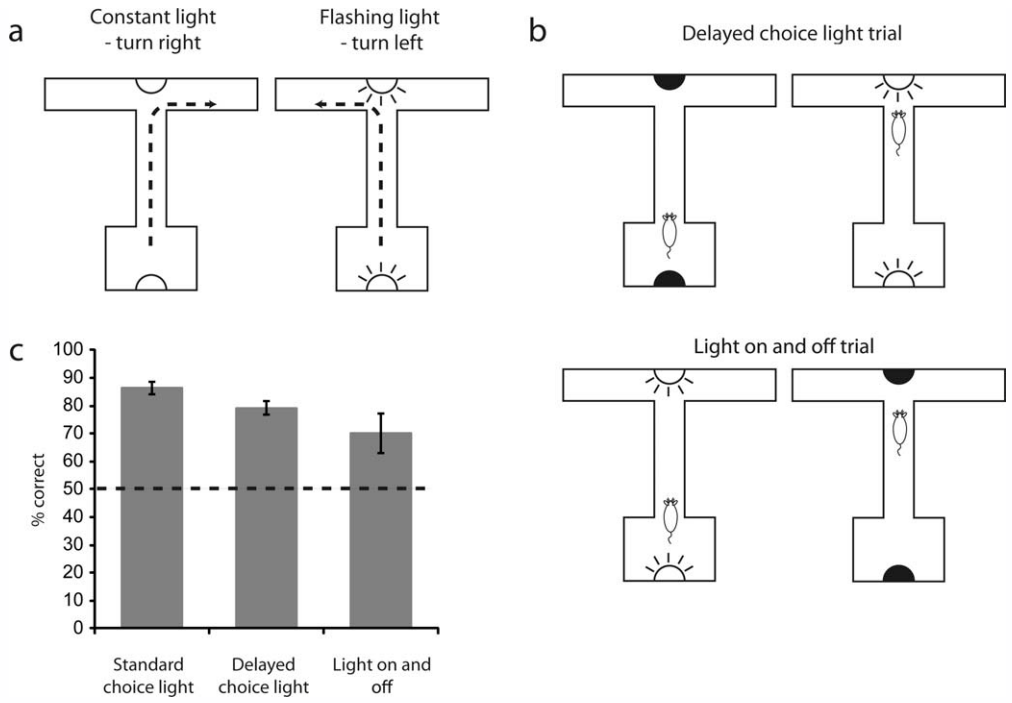

FIGURE 1. Conditional T-maze task controlled by visual discriminative stimulus. (a) Standard choice light trials where the cue is present for the whole trial. A constant light means that the right arm is rewarded. A flashing light means that the left arm is rewarded. (b) Delayed choice light trials (top); the light cue is not present at the beginning of the trial and is only turned on when the rat approaches the choice point. Light on and off trials (bottom); the light cue is initially present but is turned off as the rat approaches the choice point. (c) Performance on the three types of trial during recording [mean ± standard error of mean (SEM)].

and kept on a 12-h light/dark cycle, with training and testing occurring during the light portion of the cycle. To motivate the rats on the behavioral task, their food access was controlled to maintain their weights at ∼90% of their free-feeding weight. Water was freely available to the rats in their cages. At the time of electrode implantation, rats weighed ∼315–415 g.

Apparatus

The modified T-shaped maze was built from wood, painted black, and rested on a plus-shaped stand 76 cm from the ground. At the base of the T, there was a start box (30 × 30 cm^2). From the start box, the central stem of the T extended 110 cm to the T junction. The central stem and arms of the T were 10 cm wide, with walls of 9.5 cm height. The left and right arms of the T were 38.8 cm long. At the end of each T arm was a small bowl in which chocolate Weetos rewards (1/4 Loops; Weetabix, Kettering, UK) could be placed by the experimenter. The disproportionate length of the central stem was deliberate, as our interest was in place fields on this region of the maze. White light emitting diodes (LEDs) were placed in the center of the back wall of the start box, at the center of the T junction, and at the end of each T arm, all 9 cm from the floor of the maze. A small switch at the base of the T-maze allowed the experimenter to control the onset of each trial, and on certain trials, the onset of the discriminative light. The switch was connected to an input/output board on the recording system which allowed the experimenter to control the sequence of constant and flashing light stimuli occurring on the maze via custom written programs. The maze was centered within a square enclosure (1.85 m^2) created by black curtains. A false roof was created by stretching a white sheet over the curtain rails. Centered in the false roof was a commutator (Dragonfly Research and Development, Ridgeley, WV), which allowed free movement of the rat when attached to a recording cable. To increase the salience of the constant and flashing discriminative stimuli, the task was run without illumination within the curtained enclosure.

Behavioral Training

Our goal was to record from place cells with place fields on the central stem of the T-maze as rats performed a conditional visual discrimination. Prior to electrode implantation, rats were handled for 5 days, and then introduced to the conditional visual discrimination task.

Trials in the task proceeded in the following way. First the experimenter brought the animal into the curtained enclosure, and placed it in the start box. The experimenter stood behind the start box, and pressed the switch to turn on the maze lights. The lights on the maze would then either flash (10 Hz), or be illuminated constantly. If a flashing light was presented, entries to the left arm of the T were rewarded; if the constant light was presented, entries to the right arm of the T were rewarded (Fig. 1a and Supplementary Movies 1 and 2). This was a reference memory task, and the contingencies of the task

were the same throughout training and recording. On different days, however, the sequence of flashing and constant light trials was changed. The order of flashing and constant light trials was pseudorandomized such that a maximum of three trials of the same type were presented consecutively. Rats were trained on the lights-on trials until they reached a criterion of 2 consecutive days with $\geq$80% correct responses over 40 trials.

Recording Electrodes

For each electrode array, four tetrodes were constructed from 25 μm nichrome wire (California Fine Wire, Grover City, CA), threaded through 27-gauge thin-wall steel cannula (Small Parts, Miami Lakes, FL), and affixed to an 18-pin socket (Millmax, Oyster Bay, NY). Individual wires from each tetrode were wrapped around a pin of the socket, and then coated with silver conductive paint. An animal ground wire was soldered to one of the remaining pins of the socket, and the other pin was affixed to the cannula holding the electrodes. The socket and three advancing screws were affixed to one another in a tripod formation (Kubie, 1984).

Surgery

The recording electrodes were implanted in a standard stereotaxic surgery. Rats were anesthetized with isoflourane and placed in a stereotaxic frame (David Kopf Instruments, Tujunga, CA). Rats were given an intraperitoneal injection of Hartman's solution (5 ml) to maintain hydration, and were given subcutaneous injections of carprofen and buprenorphine. The procedure was conducted under isoflourane anesthesia.

The skull was exposed, and two electrode arrays were implanted 3.5 mm posterior to bregma, $\pm$2.5 mm lateral to the midline (one on each side), and 1.8 mm ventral to the dura. The arrays were affixed to the skull via small skull screws and dental cement. The ground wire from each array was attached to a different skull screw.

Following surgery, rats were placed on a warming pad until they recovered from the anesthesia. They were given an additional 5 ml of Hartman's solution at this time, and an additional dose of carprofen the next day.

Screening and Recording

Screening for cells began after a 1 week recovery period. Individual rats were brought into the recording room and the recording cable was plugged into the electrode assembly while the rat consumed a Weeto on the experimenter's lap. The rat was then placed in a large cylindrical container where it could move about freely.

The signals from each tetrode wire were amplified with a unity-gain operational amplifier at the end of the recording cable, and then passed through a pre-amplifier to the recording system (Axona, Herts, UK). The signal was bandpass filtered (600–6,000 hz) and amplified (5,000–20,000 times). Individual electrodes were recorded differentially with respect to a quiet wire on another tetrode. The rat's position was tracked by a video tracker (Axona) that recorded the position of the LEDs at the end of the recording cable. If no units were encountered in a screening session, the electrodes were advanced by turning the screws on the electrode assembly.

Recording Task

If place cells were identified in the screening environment, a 10 min session within the circular environment was recorded prior to the conditional T-maze testing. The animal was subsequently assessed during performance of the conditional T-maze task. Recording sessions in the conditional T-maze task were comprised of the following types of trials:

1. Fully-cued trials: The discriminative lights were illuminated in the start box, at the choice point, and at the end of the reinforced arm. They continued to be illuminated until the rat reached either the rewarded or the nonrewarded container.
2. Choice-light-trials: The discriminative lights were illuminated in the start box and at the choice point of the T, but not at the end of the correct choice arm. Thus, the rat had to use the choice-point light to select the maze arm that contained reward.
3. Delayed choice-light trials: On these trials, the discriminative lights at the choice point were only illuminated when the rat reached the T junction (Fig. 1b, top).
4. Light-on-and-off trials: For these trials, the lights in the start box and at the choice point were illuminated as the rat was placed in the start box, but were extinguished when the rat was halfway up the central stem. Thus the rats had to remember which type of light (constant or flashing) they had seen in the start box to correctly identify to arm of the T that was to be rewarded on a given trial (Fig. 1b, bottom).
5. No-light trials: On these trials the lights were not illuminated. Neither arm choice was reinforced. These were control trials, run at the end of the recording session, which allowed us to determine whether conditional firing was due to the discriminada (the constant or flashing light) or the destination of the animal (the left or right arm of the T).

The trials were run in blocks. Ten fully-cued trials were run at the start of the day to reinforce the previously learned behavior. The trials were then run in blocks of 8 or 10 trials in the following sequence: choice-light, delayed choice-light, light-on-and-off. This sequence was then repeated and at the end of the session a block of no-light trials was run. (Our preliminary training with the task showed that mixing the trial-types, as opposed to giving blocks of specific trial-types, resulted in substantially poorer performance.) At the end of the T-maze training a second 10 min session in the circular environment was recorded.

Perfusion and Histology

At the end of the experiment, rats were given an overdose of sodium pentobarbital and the electrode position was marked by passing current through the tetrodes, making small electrolytic

lesions. Rats were perfused transcardially with saline, followed by 4% formalin, and the brains removed. Brains were kept in 4% formalin mixed with 4% potassium ferrocyanide for at least 48 h which elicited a Prussian blue reaction marking the end of the tetrode. Brains were sectioned on a freezing microtome. Fifty micrometer sections were cut with every section around the cannula on each side of the brain being taken for analysis.

Place Cell Identification and Analysis

Initial data analysis was performed using Klusters analysis software (Hazan et al., 2006) on the data from all of the trials combined. Spikes were sorted into clusters using comparisons of peak amplitude, energy, and first principal component. Autocorrelograms were generated for each cluster to ensure that no spikes fired within 1.5 μs of any other spikes from the same cluster. Cluster quality was assessed using the L-ratio and isolation distance (i.d.), as described by Schmitzer-Torbert et al. (2005). Firing rate maps were generated by dividing the maze into a grid of 56 × 56 pixels (each pixel being 2.5 × 2.5 cm^2). The firing rate for each pixel was calculated by dividing the number of spikes fired in that pixel by the number of seconds that the rat spent there. Cells were deemed to have place fields on the maze if there were at least six adjacent pixels with a firing rate of at least three times the session mean firing rate. Only cells with well defined place fields on the T-maze were used for further analysis. Recordings from consecutive days were closely examined and cells that reappeared on multiple days were only counted once. This was done conservatively such that any cell that was similar to those from a previous day was excluded. This will have resulted in new cells being excluded but guards against the possibility of effects being magnified by counting cells in the analysis more than once.

Analysis

Our interest was in the properties of place fields that occurred in the start box, the stem of the maze, and the choice point. In these locations, fields could be modulated by the discriminative stimulus or by memory. Fields after the choice-point (in the goal arms of the T-maze) were not considered further. The following analysis dealt only with trials on which the rat chose the correct arm.

The region of interest on the maze (central stem and start box) was divided into eight equally sized bins. In our initial analyses, we considered the firing rate of each place cell in terms of the journey made by the animal (left vs. right choice at the T), the type of trial (fully-cued, choice-light, delayed choice-light, light-on-and-off, no light), and the location of the place field (bins 1–8). The journey and type of trial were treated as between-subjects conditions within an analysis of variance (ANOVA); bin was treated as a within-subject condition. Statistical significance was defined as an F-ratio for a main effect or interaction with a $P < 0.05$.

This three-way ANOVA was run on each cell. It allowed us to identify three classes of place cells. The first were traditional place cells, those with a place field (and therefore a significant difference in firing rates between the bins) but no modulation of firing on different journeys or trial types. The second class of cells were those whose firing was significantly different depending on whether the rat was headed for the left or right arm of the T. In our initial categorization of cells, we placed cells in this category if they showed a significant main effect of journey, or any significant interaction with journey. Our third category was comprised of cells that fired at significantly different rates depending on the type of trial, or showed a significant interaction with trial type.

Planned Comparisons

Our interest was in the modulation of place cell firing, particularly prospective firing, as a function of the information available to the animal as it traversed a given cell's firing field. To address this, we conducted comparisons of different trial types for cells exhibiting significant journey or trial type effects in the initial analyses. Cells without significant journey or trial-type effects—traditional place cells—were not considered in this analysis, as their firing was not modulated by the responses or the contingencies of the environment.

Our first planned comparison was designed to address the following question: Do place cells exhibit conditional firing when the animal "knows" where it is going? To test this, we compared the firing of place cells on trials where the animal could determine which arm of the T was rewarded before it passed through the cell's firing field to trials where this information was not available to the animal when it passed through the cell's field.

The trials that make up this comparison differed depending on the location of the recorded cell's firing field. Therefore, we analyzed each place field separately. For place fields in the start box or beginning of the stem (bins 1–4) the discriminative light was on as the animal traversed the field in the fully-cued, the choice-light, and the light-on-and-off trials. The light was not on for the delayed choice-light and the no-light trials as the animal traversed this portion of the maze. Thus, our comparison for place fields at the beginning of the T-maze was: fully-cued, choice-light and light-on-and-off trials vs. delayed choice-light and no-light trials.

For place fields near the choice point of the T (bins 6–8), a different comparison was made; fully-cued, choice-light, and delayed-choice-light trials vs. light-on-and-off and no-light trials. As in the previous comparison, the rationale behind this test was to see if firing differed when information on the location of the rewarded arm was available to the animal when it traversed the place field as opposed to when it was not. It should be noted that this comparison may underestimate any differences due to the lights, as in the light-on-and-off trials, the animal's choice may be guided by its memory from the previously illuminated stimulus.

RESULTS

Behavior

The rats required, on average, 12 days of training on the conditional T-maze task to reach a >80% performance level

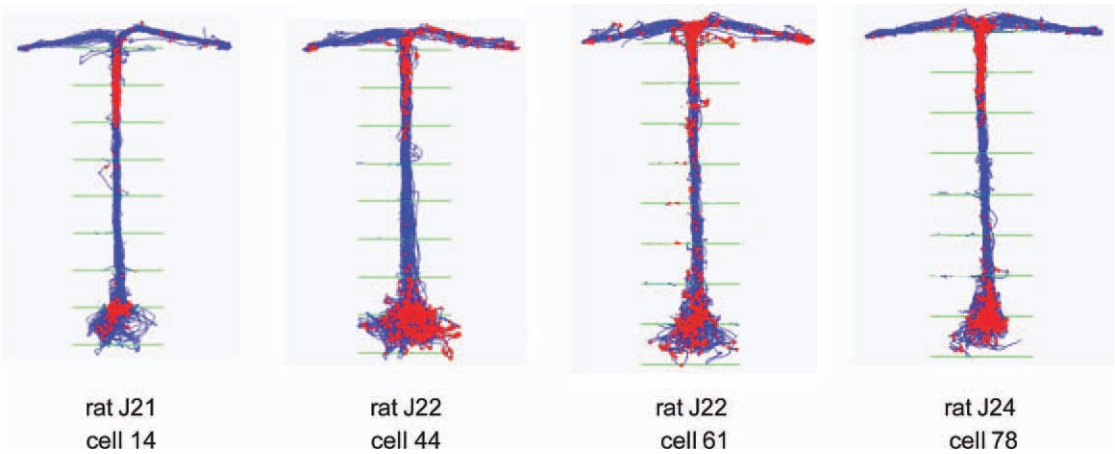

FIGURE 2. Four examples of place cells that possessed fields in both the start box of the T-maze, and the choice point. For cells with multiple fields, this was the most common pattern observed. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

prior to surgery. Following recovery from surgery and upon the encounter of place cells, rats were returned to the T-maze task. In these recording sessions, rats experienced both the light-on trials that they had originally been trained on and trials in which the light onset was manipulated. In the final form of the task, rats were typically given 10 fully-cued, 20 choice-light, 16 delayed choice-light, 16 light-on-and-off, and 10 no-light trials.

The average performance of the rats on these different trials is shown in Figure 1c (performance on the no-light trials is not shown as neither arm was rewarded). When the discriminative lights were on for the entire trial (fully-cued and choice-light trials), performance was >80% correct. A somewhat lower mean level of performance was observed on trials where the discriminative lights were turned off before the animal reached the choice point, but there was no overall significant difference between the three trial types [$F_{(2,8)} = 3.2$, $P = 0.095$].

Place Field Location on the T-Maze

126 cells from CA1 were recorded during performance of the conditional T-maze task. Twenty-five were classed as interneurons based on waveform characteristics interspike interval and firing rate. The remaining cells were well isolated with a mean i.d. of 53.07 ± 7.24 (SEM) and mean L-ratio of 0.05 ± 0.01 (SEM). Example waveforms from cells with good and average/poor isolation can be seen in Supplementary Figure 1. Some place cells fired in more than one location and thus 127 place fields were identified. The majority of the fields, 57, were observed in the start box of the maze. Twenty-eight fields were observed near the middle of the central stem of the T, 27 fields occurred at the choice point, and 16 fields fired on the goal arms. Each place field was analyzed individually.

For cells that possessed more than one place field, the incidence of cells with fields both in the start box and in the choice point was striking. About 58% of the cells with fields at the choice point also had fields in the start box region (Fig. 2). The incidence of choice-point and start-box fields was higher than choice-point and fields elsewhere on the maze, as only 8% of the choice point cells exhibited a second firing field on the central stem of the maze, and only 14% of choice point cells also fired on the goal arm (although the proximity of the two locations likely means that some of this activity reflects a single field).

Initial Classification of Cell Correlates

To determine whether place fields were modulated by the discriminative stimulus or by the different conditions in which the stimulus was presented, we compared the firing of each cell on the appropriate trials of different journey types (left or right T choices) and trial-types (cued, light-on, delayed light-on, light-on-and-off, no-light). The initial classification of the cells from this analysis is shown in Figure 3. Of the 93 cells

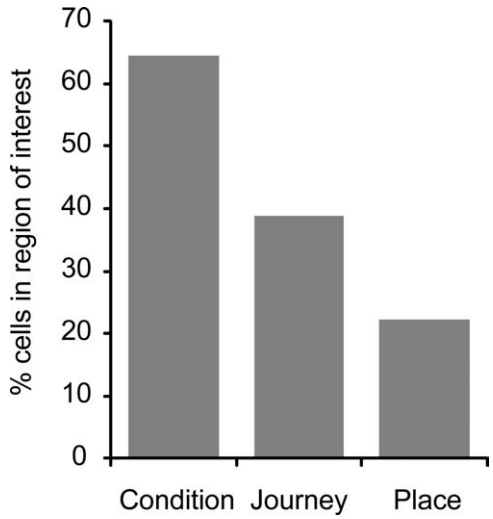

FIGURE 3. Percentage of place cells in the region of interest (central stem or start box) showing significant patterns of firing in response to condition (presence or absence of the cue light), journey (left vs. right turns) or place (traditional place cells that fire in their place field but not in response to contextual features of the trial).

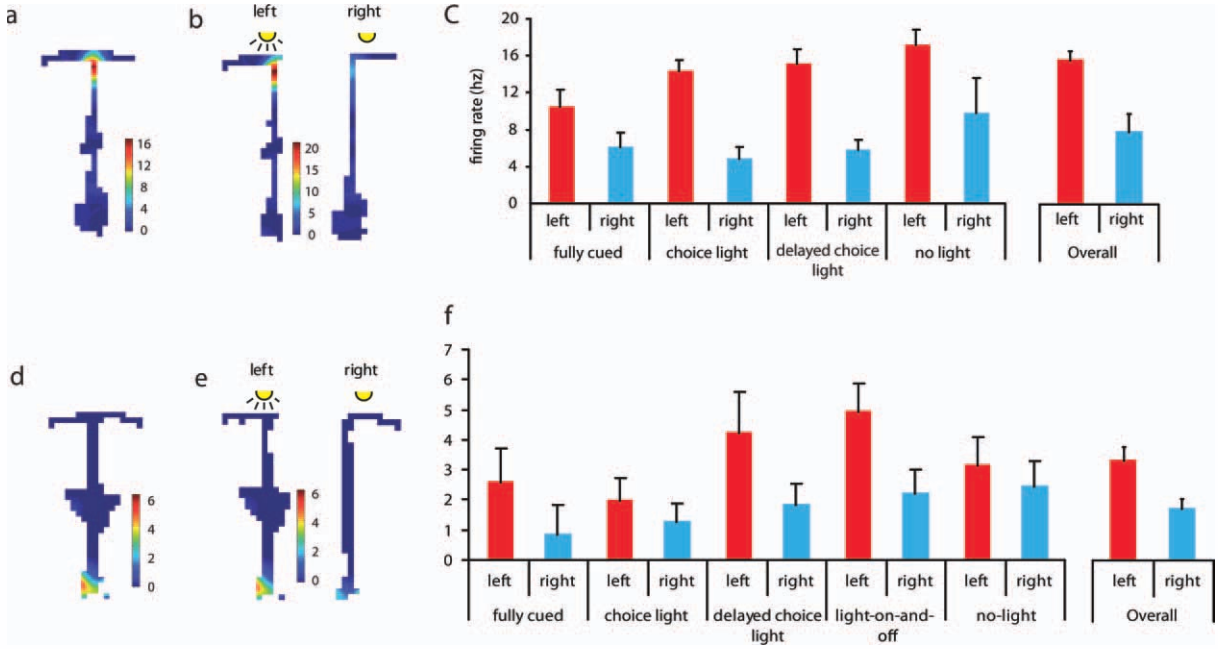

FIGURE 4. Two examples of journey dependent place cells. (a,d) Firing rate map for an example place cell with a field on the maze. Areas of the maze where the cell had low firing rates are represented in blue and those with high firing rates in red (see scale bar for details). (b,e) Rate maps for the same cell split up into left and right trials. Note the dramatic change in firing rate on the left and right trials. (c,f) Histogram illustrating the firing rates on left and right turn trials during all of the different conditions. Note that the cells have higher firing rates on left turn trials than right turn trials in all conditions. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

recorded on the T-maze with fields on the central stem or in the start box, 38 (40.8%) showed significant modulation of their field depending on whether the rat made a left or right journey. These were cells that had a significant main effect of journey type, or a significant interaction between journey type and trial-type, bin, or both. 64 cells (68.8%) showed a significant difference in firing depending on the task conditions. It should be noted that cells with significant journey and trial type effects counted in both categories. Finally, 20 cells (21.5%) showed no difference in firing with respect to the journey- or trial-type. These we refer to as traditional place cells.

Place Cell Firing is Modulated by the Journey Type

Examples of cells whose firing was modulated by the rat's destination are shown in Figure 4. In the top example, the cell fired more at the choice point on trials where the rat made a left turn, as opposed to trials on which it made a right turn. A similar pattern of firing was exhibited by a cell with a place field in the start box in the bottom example of Figure 4.

In both examples, although there were significant differences in firing rates on right- as opposed to left-trials, firing was seen on both. A comparable pattern was seen in the other cells that showed a significant main effect of journey type. This is likely an example of rate remapping between journey types.

Planned Comparisons

Cells showing significant differences in firing rate on different journeys may do so in response to the conditional stimulus (the constant or flashing light) or because the animal is about to make a specific turn (to the left or right arm of the T). To distinguish between these possibilities, we examined the firing rate differences between left and right journeys occurring in the presence or the absence of the light. If the differences in normalized firing rates between journeys are driven by this discriminative stimulus, then large differences should occur on trials in which the discriminative light is present, and little difference should occur on trials where the light is not illuminated. Conversely, if differences in firing on left and right journeys reflect the intended destination of the animals, then they should be found regardless of the discriminative stimulus's presence.

As shown in Figure 5a, the difference in firing rates on left and right trials for place fields exhibiting significant journey effects was similar when the discriminative stimulus, the constant or flashing light, was present (lights on) or absent (lights off) ($t_{(41)} = -0.49$, $P = 0.62$). These cells maintained their prospective firing when the discriminative stimulus was not present, and the differences in firing rates between left and right journeys in the presence of the discriminative stimulus were significantly correlated with the differences in the absence of the discriminative stimulus ($r = 0.66$; $P < 0.001$; Fig. 5b). This suggests that the prospective firing of journey cells reflects

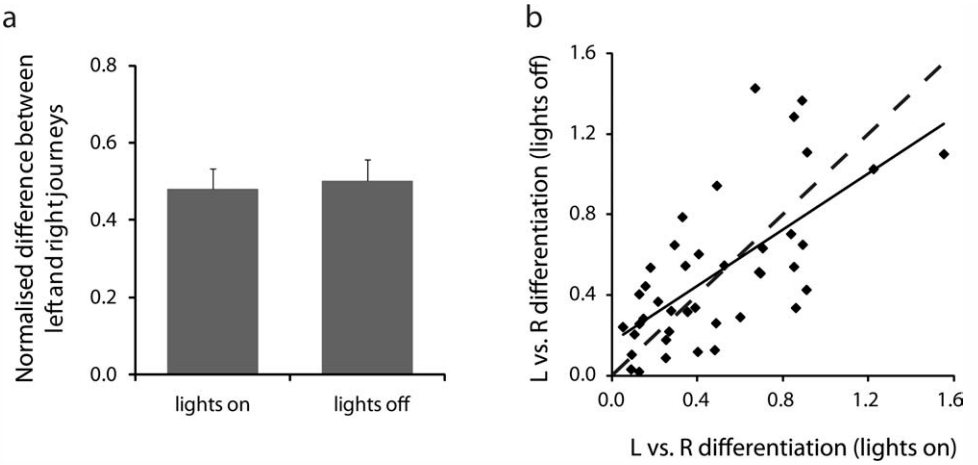

FIGURE 5. Modulation of conditional firing by the cue light. (a) Normalized firing rates of left vs. right trials for cells showing a significant effect of journey type during trials when the light was present and when it was absent. A score of zero represents equal firing rates on left and right turn trials. Note that on average cells that differentiate between left and right turn trials do so even when the cue is not present. (b) Normalized firing rate for left vs. right turn trials when the lights were present plotted against the same measure when the lights were absent. Note that the two measures are very close to being perfectly correlated (dashed line).

the destination of the animal, and not the discriminative stimulus.

Task-Modulated Place Fields

Although place cell activity did not directly discriminate between the constant and flashing lights, many cells (68.8%) showed significant differences in firing rate across the different task conditions or significant interactions between task conditions and firing locations or journey-types. Different patterns of firing were observed in different place fields. For example, for some fields, firing was highest during the initial cued trials (Fig. 6a). For others, firing was highest on trials where the conditional lights were extinguished before the animal reached the choice point (Fig. 6b).

Different conditions elicited the highest firing rate across the population of place fields sampled. This is illustrated in Supplementary Figure 2. The modal trial type for the highest firing rate was the no light trial. This raises the possibility that place cell firing rate increased throughout the session. To examine this possibility the firing rates in the two sessions in the circular environment before and after the T-maze testing were compared (see place maps for pre- and post-testing sessions, Fig. 6). On average the firing rate was 1.4× lower in the second circle session than in the first for the cells. This is in marked contrast to the 4.6× average change between conditions on the T-maze, suggesting that a simple increase in firing rate across the session cannot account for the change in firing rates within the different types of trial.

Interneurons

The firing rates of interneurons were also assessed for task and journey correlates. For the majority (19/20) of interneurons, no differences in firing rates were observed on the central stem of the maze for journeys to the left or right goal arms. Differences in firing rates for different types of light conditions were observed in a small number of interneurons (5/20). However, these differences did not reflect dramatic modulation of firing rates across condition. Even though interneurons fired in all locations on the maze, their firing rates were different in different locations; a significant difference in firing rate across locations on the central stem was observed in 19 of the 20 interneurons recorded.

Histology

Histological assessment of the brains following completion of the experiment confirmed the placement of the electrodes in the CA1 region of the hippocampus (Supplementary Fig. 3).

DISCUSSION

The current study sought to test whether the firing of place cells in the CA1 region of the hippocampus could be controlled by a visual stimulus in a conditional visual discrimination task. The main novel finding of this study was that many place cells fired at different rates on the central stem of the maze when the rat was headed to different destinations, but that this differential firing was independent of the visual discriminada signaling reward location. We also observed that the majority of place cells showed a significant difference in firing rate or shift in location across different types of trials. Finally, we found that place fields were distributed unequally on the maze, with several cells firing both at the beginning of the maze and at the choice point. These findings are considered below.

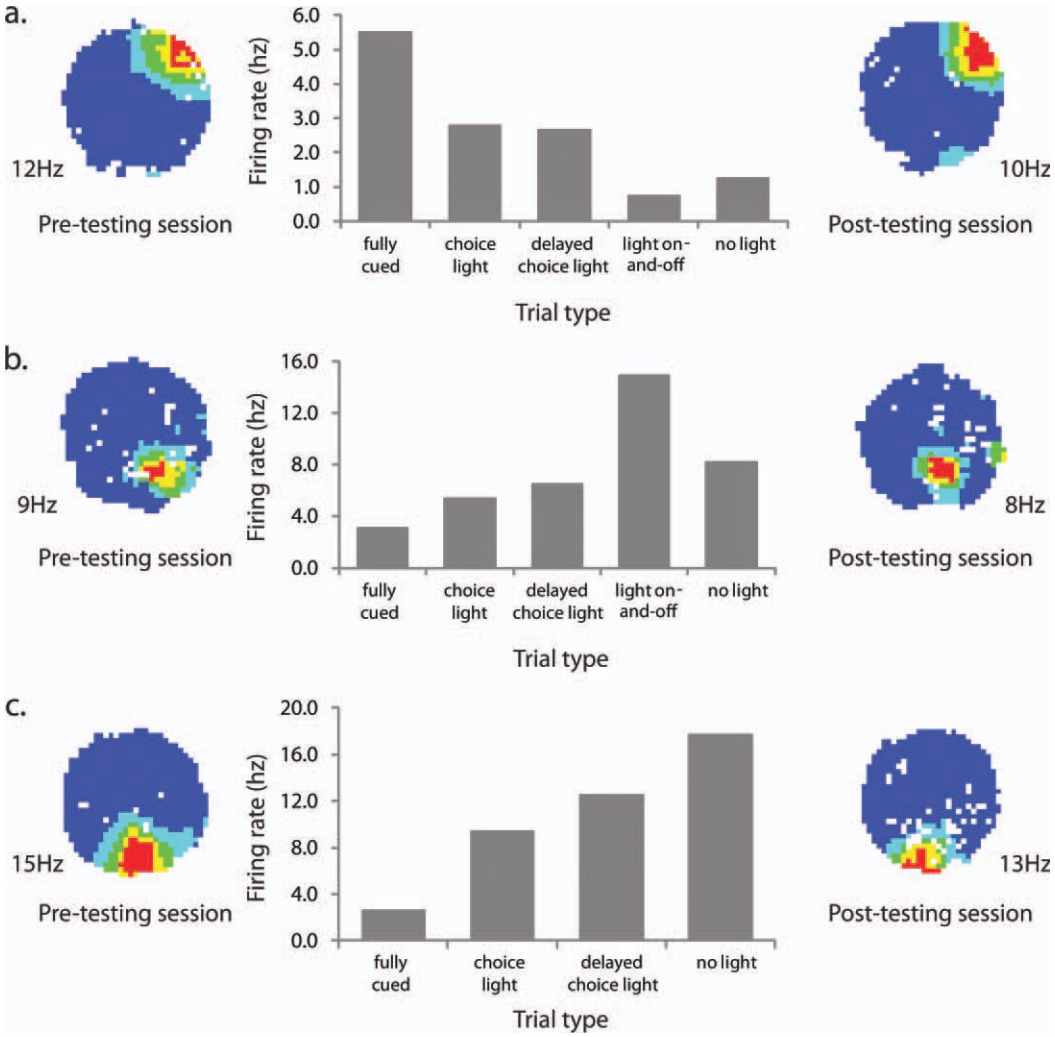

FIGURE 6. Three examples of place cells that change their firing rate in response to different conditions. Note in all three examples that firing rates in the session in the circular environment before and after the T-maze session did not differ (see inset rate maps). These pairs of maps are plotted with a consistent scale for the firing rate colors. (a) An example of a place cell that has its highest firing rate in the cued trials. (b) An example of a place cell that has it highest firing rate in the light on and off trials. (c) An example of a place cell that has its highest firing rate in the no light trials. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

Conditional Place Fields Reflect the Animal's Destination, Not the Discriminative Stimulus

A primary aim of this experiment was to test whether place cell activity could be controlled by a visual discriminative stimulus. Although differences in firing were observed on the central stem of the T-maze for trials in which the animal chose the right goal arm as opposed to the left, these differences persisted on trials in which no discriminative stimulus was presented. Thus, this differential firing likely reflects the animal's intended destination, and not an encoding of contextual information signaling location of reward provided by the discriminative stimulus.

Our findings may complement those from a recent study by Kennedy and Shapiro (2009). They found that some place cells encoded the animal's motivational state (hunger or thirst) on a maze where food or water could be obtained at the end of a trident-shaped maze. In their task, however, the location of the food or water reward was moved from trial to trial between the three terminal arms of the maze. Thus, in contrast to the task we employed, rats had no way of knowing which turn they would make at the choice point while they traversed the central stem. Unsurprisingly, in the absence of a known trajectory, place cells failed to represent trajectory, but did represent the animal's motivational state. It's possible that such motivational states are quite salient to the animal, and are therefore represented more robustly than an arbitrary association between a visual discrimada and the location of a reward.

The lack of control by the light over place cell firing also implies that the encoding of a visual discriminative stimulus occurs outside the hippocampus. It suggests that not all elements of attended experience are evident in the activity of

hippocampal principal neurons (Morris and Frey, 1997), at least in CA1.

Recent theories have suggested that the hippocampus may be involved in combining spatial information from grid cells in the medial entorhinal cortex (EC) with contextual information, possibly from the lateral EC. The present data suggest that the convergence of contextual and spatial information needed to solve the current task may occur earlier in the hippocampal–entorhinal circuitry. The CA1 region of the hippocampus may then be responsible for relaying the intended behavioral choice to regions involved in action selection, such as the ventral striatum (van der Meer and Redish, 2009).

Task-Related Differences in Firing Rates

Although the prospective firing of place cells was not controlled by the lights, differences in overall firing rates were observed in nearly 2/3 of the cells for different types of trials. This suggests that place cells were sensitive to manipulations of a behaviorally relevant stimulus. Previous studies have demonstrated that place cells in the hippocampus encode nongeometric changes in environmental stimuli (Anderson and Jeffery, 2003; Jeffery et al., 2004; Leutgeb et al., 2005). In particular, one study demonstrated that place cells change their firing rate in response to changes of color of the environment while retaining their spatial consistency (Leutgeb et al., 2005). This suggests that place cells are sensitive to contextual features of the surrounding environment. The current study extends this by showing that the majority of place cells in CA1 respond to the presence or absence of behaviorally relevant stimuli. Within-session changes in place cell firing rates have been previously reported in portions of a radial arm maze and in a Y-maze (Frank et al., 2004, 2006; Ainge et al., 2007a). The mechanism for these changes is not fully understood, and an open question is whether such changes are specific to tasks with learning and memory demands.

Place Fields Firing at the Start Box and Choice Point

The place fields we identified on this T-maze task were not distributed evenly throughout the environment. Approximately 45% of the fields recorded were located in the start box. This finding is consistent with recent observations of an over-representation of the start location on a spatial maze (Ainge et al., 2007a, b). In other studies, place field over-representation has been observed at goal locations (O'Keefe and Conway, 1978; Hollup et al., 2001; Hok et al., 2007). A possible account for these findings is that the hippocampus over-represents locations where the animal spend more time, such as the start box in our experiments, or the goal location the Hollup and Hok studies. In our studies, it's not entirely clear why over-representation of the start box is warranted. However, one possibility is that rodents have a predisposition to form a representation of a base location, from which potential journeys can originate (Wallace and Whishaw, 2003).

A second finding of note is that more than half of the place cells with fields in the start box of the maze also exhibited firing at the choice point of the maze. This additional activity was not observed elsewhere on the maze. One possibility is that this activity is akin to the look-ahead place cell activity demonstrated by Johnson and Redish (2007). In the current task, as in the task used by Johnson and Redish, rats did not appear to achieve the automaticity at the choice point observed in a previous win-stay Y-maze task (Ainge et al., 2007a). Thus, such look-ahead activity may be a feature of tasks which entail more deliberative processing at locations of consequence.

SUMMARY

The novel finding from this study was that CA1 place cell firing is modulated by the intended destination of the rat and not by a salient visual stimulus that controls behavior. This suggests that CA1 is responsible for the output of the behavioral response or trajectory that has been selected for a given trial. The association of contextual features within the environment with specific behavioral responses must happen upstream of CA1. Whether this is within the CA3/DG network, the entorhinal cortex or elsewhere remains to be determined. However, CA1 place cells do respond to the presence or absence of a behaviorally relevant stimulus, suggesting that aspects of on-going behavior and/or changes in the environment can have an impact on information output from the hippocampus.

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