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Modification of the Responses of Hippocampal Neurons in the Monkey During the Learning of a Conditional Spatial Response Task

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ABSTRACT

In order to analyze the function of the hippocampus in learning, the activity of single neurons was recorded while monkeys learned a task of the type known to be impaired by damage to the hippocampus. In the conditional response task, the monkey had to learn to make one response when one stimulus was shown, and a different response when a different stimulus was shown. It had previously been shown that there are neurons in the hippocampal formation that respond in this task, to, for example, a combination of a particular visual stimulus that had been associated in previous learning with a particular behavioral response. In the present study, it was found that during such conditional response learning, the activity of 22% of the neurons in the hippocampus and parahippocampal gyrus with activity specifically related to the task altered their responses so that their activity, which was initially equal to the two new stimuli, became progressively differential to the two stimuli when the monkey learned to make different responses to the two stimuli. These changes occurred for different neurons just before, at, or just after the time when the monkey learned the correct response to make to the stimuli. In addition to these neurons, which had differential responses that were sustained for as long as the recordings continued, another population of neurons (45% of those with activity specifically related to the task) developed differential activity to the two new stimuli, yet showed such differential responses transiently for only a small number of trials at about the time when the monkey learned. These findings are consistent with the hypothesis that some synapses on hippocampal neurons modify during this type of learning so that some neurons come to respond to particular stimulus–response associations that are being learned. Further, the finding that many hippocampal neurons started to reflect the new learning, but then stopped responding differentially (the transient neurons), is consistent with the hypothesis that the hippocampal neurons with large sustained changes in their activity inhibited the transient neurons, which then underwent reverse learning, thus providing a competitive mechanism by which not all neurons are allocated to any one learned association or event.

Key words: hippocampus, learning, memory, conditional response, association memory, amnesia, competition, synaptic modification, neuronal network

Bilateral damage to the temporal lobe in humans can cause anterograde amnesia (Scoville and Milner, 1957; Milner, 1972; Squire, 1986; Squire et al., 1989; Rolls, 1990a). A number of structures are damaged, and these include the hippocampus and the amygdala. Experimental investigations have been performed to determine which structures are crucial in producing the amnesia, and to analyze the neural bases of the different types of amnesia (Mishkin, 1982; Squire and Zola-Morgan, 1983; Zola-Morgan et al., 1986; Gaffan, 1987; Rolls, 1990a). In analyses of the way in which the hippocampus could contribute to a memory deficit, it has been shown that tasks that are particularly affected by hippocampal (or fornix) damage in the monkey include tasks in which both an object, and the place in which it was seen, must be remembered (Gaffan and Saunders, 1985; Gaffan, 1987; Parkinson, Murray and Mishkin, 1988). It is thus of interest that H.M., and humans with right temporal lobe damage, were impaired in remembering the positions in which objects had been placed on a board (Smith and Milner, 1981). We have recently reported that

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9.3% of neurons in the hippocampal formation respond to the spatial position of stimuli presented in a multiple object–place memory task (Rolls et al., 1989; Feigenbaum and Rolls, 1991).

Another task in which monkeys with fornix lesions are impaired is a conditional spatial response task in which one response must be made to one stimulus, and a different response to another stimulus (Gaffan and Harrison, 1984; Rupniak and Gaffan, 1987). In this task, the monkey had to press a panel a fixed number of times (e.g., four) in order to obtain a reward when one visual stimulus was shown. When a different stimulus was shown, the monkey had to withhold pressing the panel for 3 seconds in order to obtain reward. (This task is referred to below as an FR-DRO stimulus–response task.) After learning this task with one pair of stimuli, the monkeys had to learn the correct responses to a new pair of stimuli. It was found that monkeys with fornix damage showed slower rates of learning this task than control monkeys (Rupniak and Gaffan, 1987). It is important that this task cannot be solved by reward association learning, as each of the responses is equally associated with reward. (Indeed, reward association learning of the type normally involved in visual pattern discrimination learning in which the monkey must learn to approach one stimulus and avoid another is not impaired by damage to the hippocampus, see Gaffan, 1987; Rolls, 1990a. It is also worth noting that the deficits in some types of spatial learning produced by fornix damage in the monkey are not just transient deficits, see Gaffan and Harrison, 1989, where in Experiment 5 run months after the fornix section a learning deficit is still apparent.) Instead, the task requires the monkey to learn which response to make to one visual stimulus in order to get reward, and which different response to make to a different stimulus also in order to obtain reward. It is also important that this conditional response task requires new stimulus–motor response associations to be learned rapidly and flexibly (in the study by Rupniak and Gaffan, 1987, a new association was introduced once the previous one had been learned to criterion), and so is different from habit learning, in which relatively fixed motor responses are learned gradually (Mishkin and Petri, 1984). It is of interest in relation to the study of mechanisms of human memory that humans with right temporal lobe (including the hippocampus) damage also have deficits in learning conditional spatial response tasks (Petrides, 1985).

In order to analyze the functions of the hippocampus in the primate, and to advance the understanding of amnesia, we recorded the activity of single hippocampal neurons in the monkey during the performance of tasks known to be affected by hippocampal damage and in related tasks (see Rolls, 1989a,b, 1990a–c, 1991; Rolls and O'Mara, 1992). In this paper we describe the activity of single neurons in monkeys during the learning of conditional spatial response (e.g., the FR-DRO stimulus–response task described above) and related tasks. It has previously been shown that some hippocampal neurons (14% of the sample) have activity related to the performance of the conditional spatial response task, and that this activity is not for the majority related unconditionally to the stimuli used or to the movements made, but is instead related to the particular stimulus to motor response mappings made in the task (Miyashita et al., 1989). In particular, the firing of this population of neurons was shown to reflect a combination of the particular stimulus and the particular response associated by learning in the conditional response association task, and could not be accounted for by the motor requirements of the task, nor wholly by the stimulus aspects of the task, as demonstrated by testing their firing in related visual discrimination tasks (Miyashita et al., 1989).

Given these findings, it was of interest to determine how neuronal responses in the hippocampal formation and the parahippocampal gyrus might alter during this type of learning, for which the hippocampus is needed, to determine whether the neuronal responses might alter rapidly enough to provide a basis for the learning, or whether they reflected some more complex aspect of the learning process. In this study, in addition to analyzing the activity of neurons during the learning of the FR-DRO conditional spatial response task, their activity was measured in simpler visual discrimination tasks in order to provide controls to show that the activity of the neurons being analyzed was not related to movements. In 38 experiments the activity of hippocampal neurons was analyzed during the learning of these simpler visual discrimination tasks to investigate whether hippocampal neurons in the monkey might alter their activity not only during the learning of tasks such as conditional spatial response tasks for which the hippocampus is needed, but also in simpler learning tasks.

MATERIALS AND METHODS

The monkeys were all trained in the following tasks to criteria of 95% correct, and could switch between the different tasks without difficulty when familiar stimuli were used within 2–3 trials.

Conditional spatial response memory task

The task may be described as a conditional spatial response task with symmetrical reinforcement, nonspatial stimuli, and arm movement responses. The response requirement was for three arm reach responses to touch a screen when one stimulus was shown (FR3), and for response omission (DRO) when the other stimulus was shown. The task is therefore abbreviated to FR-DRO. The task was designed to be very similar to that used by Rupniak and Gaffan (1987), so that the neuronal responses recorded might be related to the deficit in the performance of the task shown to be produced by damage to the fornix. The task was identical to that used in our earlier study of hippocampal neuronal activity in an FR-DRO conditional response task (Miyashita et al., 1989).

The monkey initiated each trial by pressing a central key placed 25 cm in front of it. This sounded a 0.5 second tone (400 Hz) to give the monkey time to fixate the video monitor placed 30 cm in front of it, just above the key. At the end of the tone, a discriminative visual stimulus appeared on the screen. To one of the stimuli (the FR stimulus), the monkey had to touch the screen three times within 3 seconds in order to obtain reward. These trials thus required a fixed ratio (FR3) type of response within 3 seconds. The reward was fruit juice, which could be obtained by licking a tube in front of the mouth, and its availability was indicated on the video monitor by a white circle that appeared for one second immediately after the FR3 requirement was met. If the monkey failed to

touch the screen three times within 3 seconds, the first touch after this period resulted in a white rectangle appearing on the video monitor, which indicated that reward would not be obtained by licking. If the monkey licked, it obtained aversive saline solution. To the other stimulus of a pair (the DRO stimulus), the monkey had to withhold touching the screen for 3 seconds. If this response was performed correctly, the circle appeared on the screen, and the monkey could lick to obtain fruit juice reward. This was thus a differential reinforcement of omission (DRO) contingency. If the monkey pressed the response key incorrectly, then the white rectangle appeared at the end of the 3-second period, saline was obtained if it licked, and the next trial was delayed by an extra 5 seconds beyond the normal intertrial interval of 5 seconds. A PDP11 computer sequenced the trials randomly. The task was completely computer-controlled to ensure that no influence by the experimenters on the monkeys' behavior or on the neuronal activity was possible. The computer switched the stimuli on and off for each trial, and synchronized its data collection so that the stimulus was turned on at the start of the 21st bin of a peristimulus time histogram. The stimuli were displayed 30 cm from the monkey on a video monitor that subtended 12° at the retina. The stimuli for this and the other tasks were images digitized from television broadcasts. The resolution of these images was 256 pixels wide by 256 pixels high with 256 gray levels. The stimuli were digitized by the computer and loaded into the AED512 (Advanced Electronic Design, Inc.) video framestore from the computer disc for each trial.

The tasks could be run with different pairs of stimuli as the discriminative stimuli, but once a pair had been chosen, that pair was used for at least 50 and for up to 265 trials. Once a neuron had been isolated (see below) with differential activity in the task and sufficient data had been obtained with the then current pair of discriminative stimuli, the task was switched to use a completely new pair of stimuli that the monkey had never previously seen. The monkey had to learn how to respond to the new pair, e.g., had to learn which was the new FR stimulus and which the new DRO stimulus. In all tasks, the neuronal responses were measured from before the introduction of the new pair of stimuli and continued throughout this learning, and for at least 40 trials after the behavioral learning. The magnitude and latency of the neuronal responses to the FR and DRO stimuli were measured and printed out on every trial, and the monkey's performance was simultaneously monitored. It was possible to perform the analysis in 43 different experiments during the learning of this task.

Visual discrimination task with an arm movement response

In order to obtain evidence on whether neuronal activity in the hippocampus was also altered during the learning of a simpler, visual discrimination task, neuronal responses were also measured during the learning of the following task. This task was a Go/NoGo visual discrimination task with asymmetrical reinforcement using the same movement response as that required in the task described above; it is abbreviated to OA (operant task with arm movement response.) The monkey initiated each trial by pressing the central key placed 25 cm in front of it. This sounded a 0.5 second tone to give the monkey time to fixate the video monitor placed 30 cm in front of it, just above the response keys. At the end of the tone, a discriminative visual stimulus appeared on the screen. To one of the stimuli (the "Go" stimulus, usually a white circle, and usually different from the discriminative stimulus used in the FR-DRO task described above), the monkey could touch the monitor screen to obtain a delivery of the fruit juice reward into its mouth. To the other of the stimuli (the "NoGo" stimulus, usually a white square, and different from that used in the FR-DRO task described above), the monkey had to withhold touching the screen in order to avoid the delivery of aversive saline solution. In 36 separate experiments, the activity of a single neuron was analyzed while the monkey learned whether to make an arm movement response to a pair of completely new stimuli in this task (OA task).

The monkeys were also trained to perform a visual discrimination task with a lick response, which was run as a control to ensure that the neuronal responses were not due to obtaining reinforcement, licking for fruit juice, etc. This task is abbreviated to OP (operant task). If a circle, the positive discriminative stimulus (S+), appeared on the monitor, the monkeys could lick to obtain a fruit juice reward, and if a square of the same area and luminance, the negative discriminative stimulus (S−), appeared the monkeys had to withhold licking in order to avoid aversive hypertonic saline. A 0.5 second signal tone (400 Hz) preceded the presentation of the stimulus, and if the monkey was fixating correctly before the stimulus appeared, it had sufficient time to perform the discrimination and obtain multiple licks of the fruit juice tube in the short (1.0 second) period in which the stimulus was on. This procedure was designed to ensure fixation of the stimuli (Rolls et al., 1979) and was also used in the OA memory task. The order of presentation of the stimuli was randomized. The EOG recordings confirmed that this procedure resulted in consistent fixation of the stimuli.

Conditional spatial arm movement response task

The conditional spatial arm movement response task was also a conditional spatial response task with symmetrical reinforcement using nonspatial stimuli, but required the monkey to make a spatial arm movement response. Following one of the stimuli, a reach response to touch the right side of the monitor was required, and to the other stimulus, a reach to touch the left side was required. (The touch areas were each approximately 7 × 7 cm.) The stimuli consisted of black and white sine wave gratings of different spatial frequencies and orientations, and were not spatially restricted to any position on the screen. This is thus a conceptually simpler example than the FR-DRO task in which one spatial response must be learned to one stimulus, and a different spatial response must be learned to another visual stimulus. It was possible to analyze the activity of neurons in six experiments during the learning of this (MSMS) task.

Recording techniques

The activity of single neurons was recorded with glass-insulated tungsten microelectrodes (after Merrill and Ainsworth, 1972, but without the platinum plating) in three rhesus macaque monkeys (Macaca mulatta) (weight 3.0–6.5 kg) seated

in a primate chair using techniques that have been described previously (Rolls et al., 1976). The monkeys had been implanted under thiopentone sodium anesthesia with stainless-steel holders on which an adaptor could be fitted for the later daily recording sessions. The action potentials of single cells were amplified using techniques described previously (Rolls et al., 1979), converted into digital pulses using the trigger circuit of an oscilloscope, and analyzed on-line using a PDP11 computer. The computer collected peristimulus rastergrams of neuronal activity for each trial and displayed, printed, and stored each trial, as well as computing the peristimulus time histogram by summing trials of a given type. To facilitate latency measurements, the cumulative sum distribution was calculated from the sum peristimulus time histogram. For each trial the number of action potentials occurring in a 500 ms period (and a 250 ms period) starting 100 ms after the stimulus onset was printed. This period was chosen because the neurons studied responded to visual stimuli with latencies that were typically 100 ms or more, and the monkeys consistently fixated the stimuli for more than 500 ms. Fixation of the stimuli was confirmed using permanently implanted silver/silver chloride electrodes for electro-oculogram recording. The electro-oculogram recordings provided eye position with an accuracy of 1°–2° and were sampled by the computer every 10 ms and saved with the action potentials for each trial.

Radiographs were used to locate the position of the microelectrode on each recording track relative to permanently implanted reference electrodes and bony landmarks such as the posterior tip of the sphenoid bone (Aggleton and Passingham, 1981). The position of cells was reconstructed from the x-ray coordinates taken together with serial 50μ histological sections stained with cresyl violet that showed the reference electrodes and microlesions made at the end of some of the microelectrode tracks (see Feigenbaum and Rolls, 1991).

Treatment of results

For each cell, the rate of firing was measured in the poststimulus period and compared for the two stimuli being used. The poststimulus period in which the firing was compared typically started 100 ms after the stimulus onset (as the typical response latency of the neurons recorded was of this order), and typically lasted for 380 ms (as the typical latency for the monkey's responses was 400 ms), but the exact period chosen was determined on the basis of the poststimulus time within which each neuron differentiated most between the two stimuli as shown by cumulative sum and running randomization tests (Siegel and Castellan, 1989). Once the time window within which the neuron showed differential responses had been determined in these ways, a *t*-test was performed using all the data within this time window and for the relevant consecutive trials to determine the significance of the response difference of the neuron to the different visual stimuli. A criterion of $P < .01$ for statistical significance was used, but for the majority of neurons with differential activity the difference was significant at $P < .001$. (The running randomization tests that indicated the relevant trials during which the neuron was responding differentially were found to be most useful when they were set to analyze differences of neuronal firing over 3–7 trials with significance levels set at 0.05–0.01. The statistical validity of this procedure was confirmed using control data from the prestimulus period.)

The latency of neuronal responses, or the differential latency of the neuronal response, i.e., the latency at which it fired significantly differently for the different trial types, was determined using cumulative sum and running mean statistics. The cumulative sum (Woodward and Goldsmith, 1964) was calculated on-line, using 18 prestimulus bins as the reference. The point at which the slope of the cumulative sum changed was taken as the latency.

A performance index was used to determine when correct behavioral performance to the new pair of stimuli started. For each trial, a value was calculated as the value from the previous trial, +1 for a correct trial or −2 for an incorrect trial, with limits set at 0 and 10, and an initial value of 0. This statistic remained close to 0 while the monkey was performing at chance, but showed a steady increase when the monkey's performance increased above chance. This inflection point provided a clear indication of when the monkey learned the task with a new pair of stimuli.

RESULTS

It was possible to perform 87 experiments of analyzing the activity of a neuron throughout the learning of a task, with 70 different neurons with activity related to the FR-DRO task in the hippocampus or parahippocampal gyrus of three macaque monkeys (18 neurons from the first monkey, 41 from the next, and 11 from the last). In 43 experiments the FR-DRO conditional spatial response task was learned, in 6 experiments the conditional spatial arm movement response task (MSMS), in 36 experiments the visual discrimination task with an arm reach response (OA), and in 2 experiments the visual discrimination task with a lick response (OP). Two types of alteration of the responses of neurons were found during this learning. In the first type, the differential response that developed to the two stimuli as a result of the learning began at some stage during learning and then persisted relatively unchanged throughout the duration of the experiment, which was at least a further 40 trials. This type of neuronal response modification was termed *sustained*. The second type of alteration of neuronal responsiveness consisted of a differential neuronal response to the two stimuli that occurred for only a limited time during the experiment, typically at about the time when the monkey learned the task. This type of response alteration was termed *transient*. These two classes of response are considered separately below. A few neurons showed both types of response modification when tested with different pairs of new stimuli. None of the neurons in which the effects of learning were analyzed had movement-related activity, as shown by their lack of activity in at least one of the tests such as the visual discrimination task with an arm movement response (OA) or the conditional spatial tasks (FR-DRO and MSMS) when these tasks were performed with a different pair of stimuli. The evidence that these neurons did not have simple movement-related activity is described more fully by Miyashita et al. (1989). Further evidence for this becomes evident below, when it is shown that some of the neurons show only transient discrimination between the stimuli while the task is being learned, yet the differential movements continue to be made after the task is fully learned. The experiments were

performed only with neurons shown to be specifically related to the performance of these tasks as shown by these criteria, and in previous work such neurons were found to comprise 14% of those recorded in the hippocampus and parahippocampal gyrus (Miyashita et al., 1989).

Sustained changes in responsiveness

The responses of a neuron that showed a sustained alteration in its relative responsiveness to the stimuli during learning are illustrated in Figure 1 in raster and peristimulus firing rate histogram form. At the start of the set of trials in which the new pair of stimuli were presented (examples from which are shown as the first few trials in the top two Go/NoGo rastergrams), the neuron responded similarly (very little relative to the prestimulus period) to the two stimuli used in the task, which the monkey had not seen before. During the second block of trials, the neuron showed consistent differential responses to the two stimuli in the post-stimulus time period of 100–400 ms in which the monkey made the discrimination, with differential responses becoming evident between the stimulus to which the monkey had to touch the screen (left part of Fig. 1) and the other stimulus (right). During the same block, the monkey learned to consistently touch the screen (as shown by the T responses) when one stimulus was shown, and to withhold touching the screen when the other stimulus was shown. The first stages in the generation of differential responses by the neuron, and in the manifestation of the learning in behavior, are seen in the lower trials in the first (top) block rastergrams. Once the activity became differential, it remained differential until the end of the experiment, i.e., its differential responses were sustained. This particular neuron was recorded while the monkey learned the operant task with an arm movement responses, i.e., a Go/NoGo visual discrimination (OA) task. (Data for a neuron recorded during learning the conditional spatial response (FR-DRO) task are shown later in Fig. 4.)

The activity of another hippocampal neuron analyzed during learning is presented in a different way in Fig. 2. Again, it is clear that the neuronal activity was modified during the learning, and that the modification in the neuron's responses preceded the monkey's learning.

Sustained differential changes in firing became evident during learning in 19 of the 87 experiments. The changes were shown in each case to be significant at $P < .01$ using a t-test. (In most experiments, the difference was significant at $P \ll .001$.) The onset of these sustained differential changes relative to the introduction of the new pair of stimuli is shown in Figure 3a, and the onset relative to the monkey learning the new association is plotted in Figure 3b. The differential responses of these neurons to the stimuli typically became evident at or about the time of learning, with the neuronal response in a number of experiments clearly occurring just before the monkey learned, as shown in Figure 3b. (In five experiments, there was a transient period of differential responding before the sustained differential responses became finally established.) Because modifications of neuronal activity were found in learning both the conditional spatial response task (12/19 experiments) and the visual discrimination task (7/19 experiments), the neurons are shown combined in Figure 3. (In each task, the important aspect of the neuronal activity change was that it occurred at about the time of behavioral learning, as shown in Fig. 3b. The behavioral learning of the visual discrimination task took in the range of 2–10 trials, and the conditional spatial response task in the range of 10–60 trials.) The involvement of hippocampal neurons in both tasks is considered below.

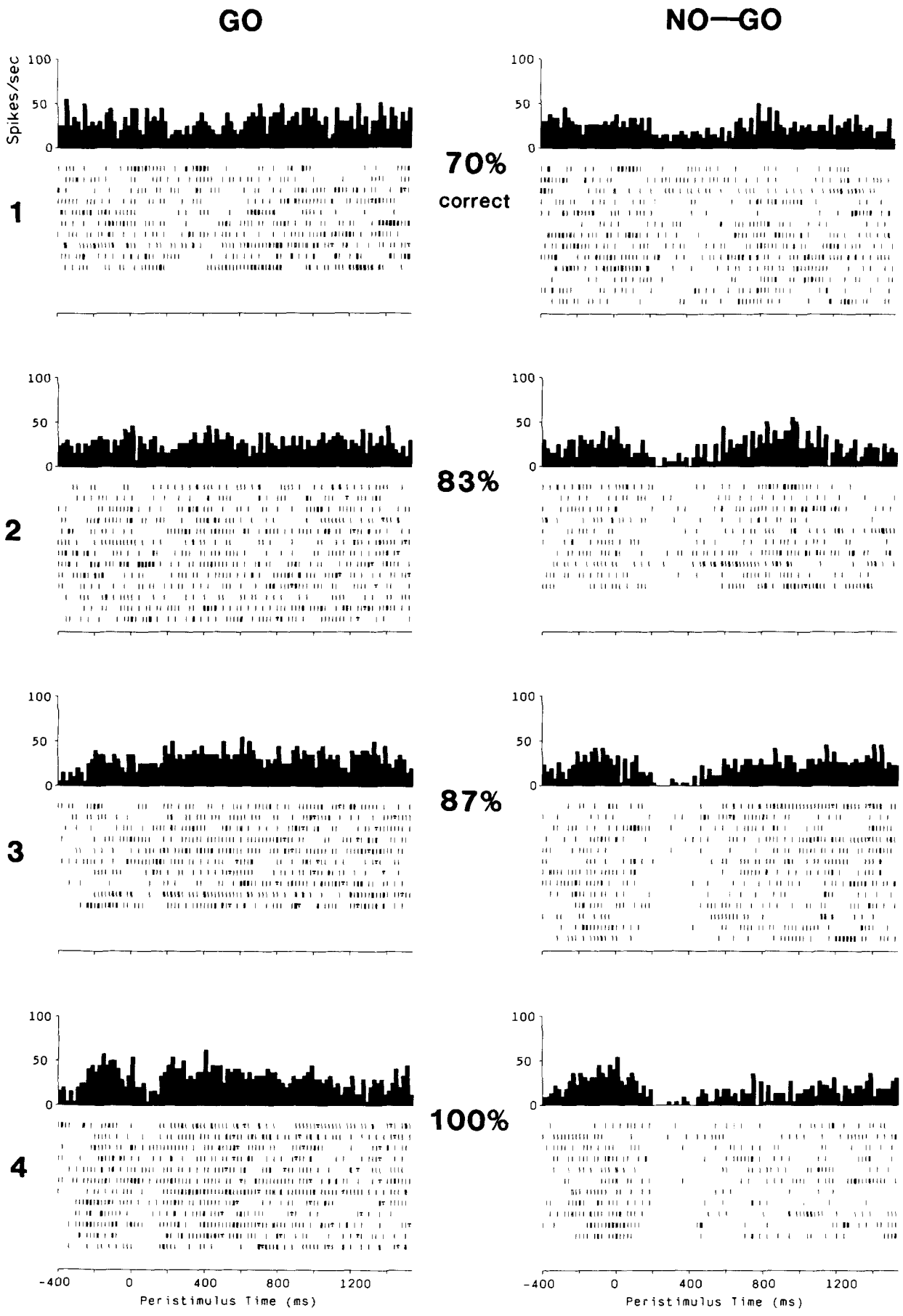
The responses of these neurons after learning consisted in 71% of experiments of a significant increase above spontaneous firing, in 19% of a decrease, and in 10% of a significant increase to one stimulus and a decrease to the other. The mean spontaneous activity of these neurons was 18.2 spikes/s, with the mean response to one stimulus equal to 37.2 spikes/s and the mean response to the other stimulus equal to 21.1 spikes/s. This value of the spontaneous activity was estimated in the period that immediately preceded each trial when the warning tone sounded, and for this reason may have given a higher value of the firing rate than when the monkey sat with no stimuli.

Transient changes in responsiveness

The responses of a neuron that showed a transient alteration in its responsiveness during learning the conditional spatial response (FR-DRO) task are illustrated in Figure 4. From the start of the experiment until approximately trial 35, the neuron responded similarly to the two stimuli used in the task, which the monkey had not seen before. However, at about this time the neuron started to show consistent and statistically significant differential responses to the two stimuli, but the differential responses remained for only approximately 29 trials, and after this, until the end of the experiment, the neuron responded equally to the two stimuli (see Fig. 4). The monkey started to perform the task correctly between trials 35 and 48, after which the monkey performed without error.

The development of similar differential responses for only a number of trials (i.e., transiently) was found in 39 of the 87 experiments (45%). The changes were shown in each case to be significant at $P < .01$ using a randomization test. The onset of these transient differential responses after the new pair of stimuli were first seen is shown in Figure 5a, and the onset relative to the monkey learning the new association is plotted in Figure 5b. The differential responses of these neurons to the stimuli did not occur randomly, but typically became evident at or about the time of learning, with the neuronal response in a number of experiments clearly occurring just before the monkey learned, as shown in Figure 5b. Because modifications of neuronal activity were found in learning both the conditional response task (24/39 experiments) and the visual discrimination task (15/39 experiments), the neurons are shown combined in Figure 5. The transient differential responses of these neurons occurred for a mean of 18 trials (SD, 17.6; range, 4–100). The mean spontaneous activity of these neurons was 23.2 spikes/s, with the mean response to one stimulus equal to 40.3 spikes/s and the mean response to the other stimulus equal to 23.9 spikes/s. Eighty-five percent of the responses consisted of a significant increase in firing over the spontaneous rate, and the remaining 15% were significant decreases.

The latency at which each of the neurons showed differential activity to the two stimuli after learning is shown in Figure 6. The latencies shown are for the new pair of stimuli, except


GO
NO—GO
Spikes/sec
100
50
0
1
70%
correct
2
83%
3
87%
4
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1200
Peristimulus Time (ms)

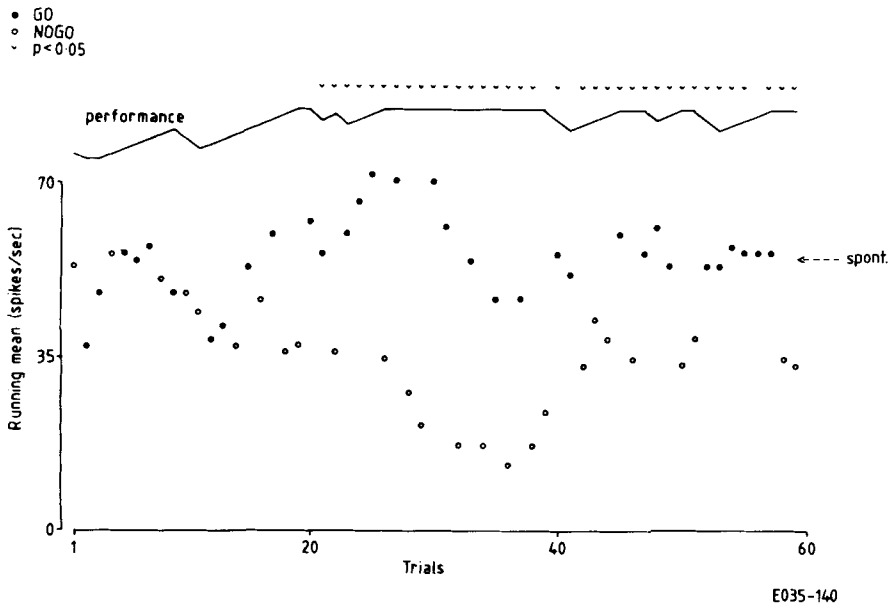


Fig. 2. The time course of the modification of neuronal activity during the learning of a visual Go/NoGo task for a neuron in which a sustained modification occurred. Open circles, the neuronal response on each trial to the NoGo stimulus; filled circles, the neuronal response on each trial to the Go stimulus. These firing rates are shown as running means calculated over 3 trials. The monkey's performance is indicated by the trace that resets when an error is made. Significant differential neuronal responses using a running randomization test are indicated by the v symbols.

where because of the considerable experience of the monkey with the familiar pair of stimuli, this latency was shorter (see the open part of the histogram in Fig. 6). The majority of the neurons had differential latencies in the range 100–200 ms (see Fig. 6). The values were similar for the neurons with sustained and with transient modifications in their responsiveness during learning. These latencies compare to typical latencies for the behavioral (touch or lick) responses in these tasks of 400–600 ms early in the learning with a new pair of stimuli, and 300–400 ms after much practice or with the familiar pair of stimuli, so that the responses of these neurons preceded and predicted the monkey's behavioral responses in this task. Further, a detailed trial-by-trial analysis of each of 26 neurons during learning showed that in 11 experiments differential firing occurred within 200 ms of the onset of the stimuli and always preceded the behavioral response latencies by at least 200 ms. This provides evidence consistent with the hypothesis that the responses of these neurons are related to the learning that occurred (see Discussion).

For nine neurons in the sample the monkey learned the conditional response task to an additional pair of new stimuli. In five of these the differential response observed to the second pair was different to that observed for the first pair, and in four there was a similar response. This indicates that any particular differential response observed with a given new pair of stimuli does not necessarily reoccur when the monkey is required to learn another new pair of associations; in addition it provides further evidence that the responses of these neurons are not related to the motor aspects of the task (see Miyashita et al., 1989).

In the remaining 30 experiments, modifications of the activity of the neuron recorded were not found.

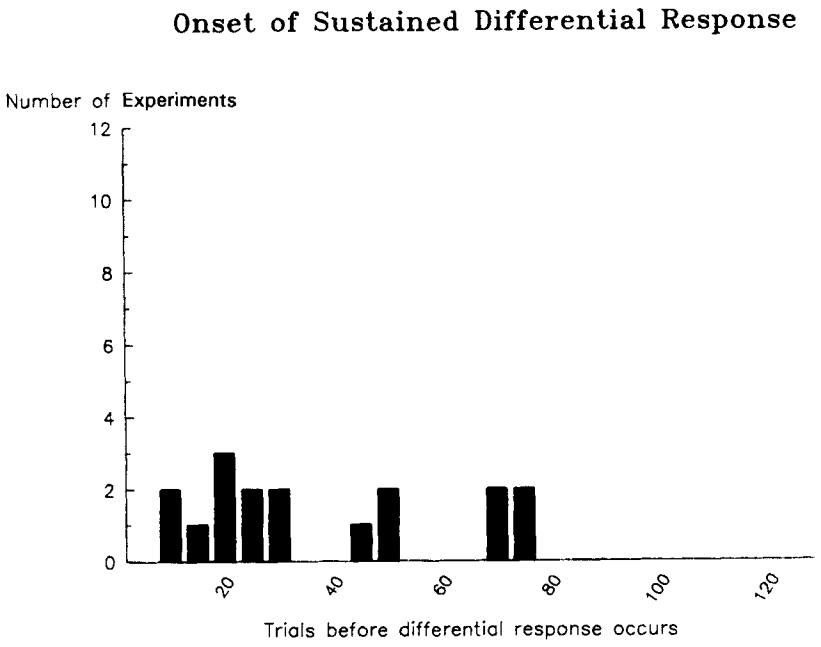
The sites at which the neurons showing response modifications during learning were recorded in the three monkeys are shown in Figure 7. Of the neurons in which response modifications occurred during learning, 14/16 tested were in the dentate gyrus, 20/26 in the CA pyramidal cell regions, 5/8 in the subiculum, and 4/4 in the parahippocampal gyrus areas TF–TH.

DISCUSSION

In this investigation, modifications of the responsiveness of neurons in the hippocampus and parahippocampal gyrus were found during the learning of conditional spatial response (FR-

Fig. 1. Rastergrams and peristimulus time histograms showing the activity of a neuron during learning of a new pair of stimuli. Each horizontal row of vertical lines represents the spikes on a single trial. The blocks 1–4 each represent 23 trials, run originally in random sequence, but sorted by trial type (Go vs NoGo) for the figure. The monkey's average performance (in percent correct) during each block of trials is indicated. The visual stimuli appeared at time 0, and were preceded by a 500 ms cue tone that ended at time 0. The monkey's touch responses are indicated by T, and errors of both omission and commission can be seen in blocks 1–3. For the first few trials of the first block of trials shown (1), the monkey's performance was at chance, and the neuron did not respond differently to the two stimuli. By the last block of trials shown, the neuron had started to respond less on the NoGo than on the Go trials, and the monkey was performing at 100% correct, that is, it had learned to touch the screen when one visual stimulus was shown (Go trials), and to not touch the screen when the other visual stimulus was shown (NoGo trials).

a



b

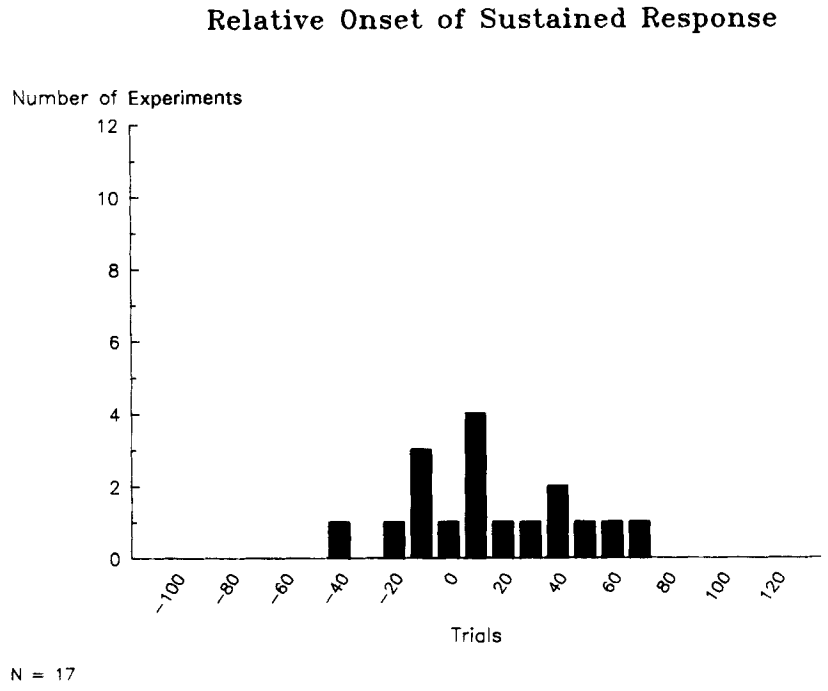


Fig. 3. (a) The onset of the differential neuronal responses after the start of the experiment in the experiments in which sustained modifications of neuronal activity were found. (b) For the same data, the onset of the sustained differential neuronal responses is shown relative to the trial (indicated as 0) at which the learning became behaviorally evident.

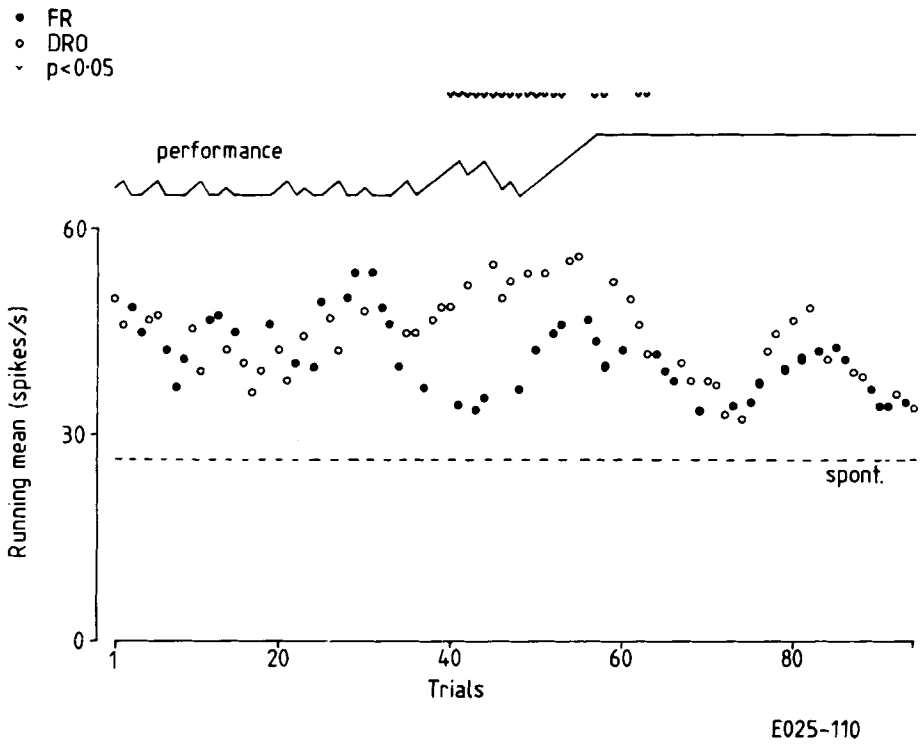


Fig. 4. The time course of the modification of neuronal activity during the learning of a conditional response task for a neuron in which a transient modification occurred. Conventions as in Figure 2. Open circles, the neuronal response to the DRO stimulus; filled circles, the neuronal response to the FR stimulus. The monkey's performance is indicated by the trace that resets when an error is made. Significant differential neuronal responses using a running randomization test are indicated by the v symbols.

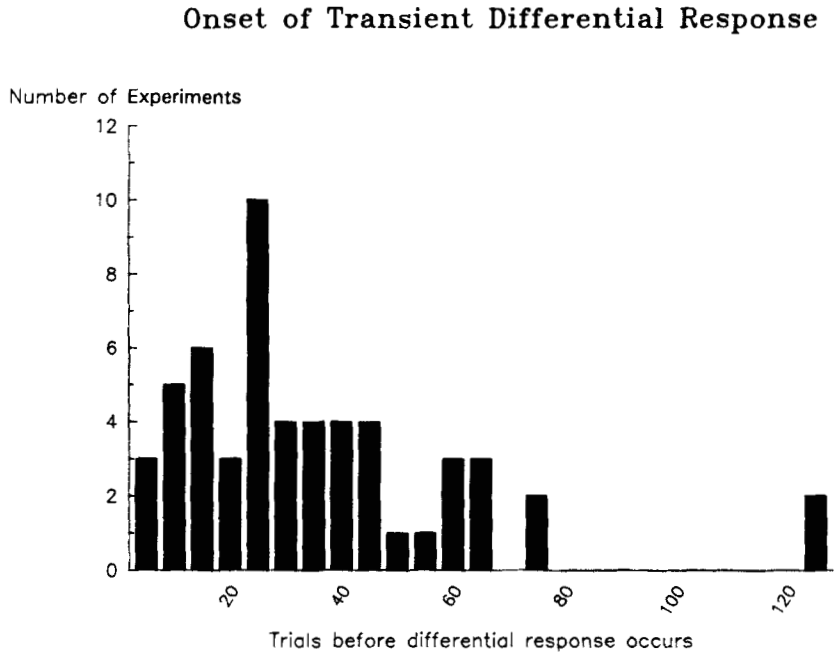
DRO) and related tasks. It was part of the experimental design that neuronal activity was measured in conditional spatial response tasks, for the learning of such conditional spatial response tasks has been shown to be impaired by damage to the hippocampal system in monkeys (Rupniak and Gaffan, 1987) and man (Petrides, 1985). The neuronal response modifications found in this study are thus on these grounds relevant to the behavioral learning that occurred during the experiments.

It is necessary to note that none of the neurons in which the effects of learning were analyzed had movement-related activity, as shown by their lack of activity either in the control tests such as the visual discrimination task with an arm movement response or the conditional spatial response task when it was being performed with a different pair of stimuli (see Miyashita et al., 1989). Further evidence is that the neurons with transient differential responses responded differentially only at about the time of learning, yet the movements once learned continued until the end of the experiment. Thus the neuronal response changes described here were not due simply to movement-related activity of the neurons, and the possibility is left open that the modifications in neuronal activity are related to the learning that occurred.

The modifications in neuronal activity that occurred were found at about the time when the monkey learned the task. As shown in Figures 3b and 5b, in a considerable number of experiments the modifications in the neuronal responses just preceded the time at which the learning became evident in the behavior of the monkey. Thus, on these grounds the modifications observed were in many experiments sufficiently early during the learning to be part of the mechanism of the learning. A statistical analysis confirmed that the differential neuronal activity was more likely to start close to the trial when the behavioral learning became evident than at a random trial in the recording period (chi-square = 27.7, $df = 11, P < .01$). The results also show that neuronal response changes do not occur only just before or at the time when the learning becomes evident in behavior, but that some neurons show alterations after the first behavioral evidence of learning. This suggests the reasonable possibility that some hippocampal neurons reflect the improvement in the learning that takes place over a number of trials, and that hippocampal neurons do not show response alterations only in the initial stages of learning.

The latencies at which these neurons responded differentially to the stimuli were for most neurons between 100 and 200 ms (see Fig. 6), compared to behavioral response latencies of 400–600 ms during learning, and 300–400 ms after much practice with a particular pair of stimuli. (The EMGs corresponding to these behavioral responses would occur 80–100 ms before the response was detected behaviorally.) Further, a detailed trial-by-trial analysis of each of 26 neurons during learning showed that in 11 experiments differential firing occurred within 200 ms of the onset of the stimuli and always preceded the behavioral response latencies by at least 200 ms. The responses of the majority of these neurons thus preceded and predicted the learned responses of the monkeys in this

a



b

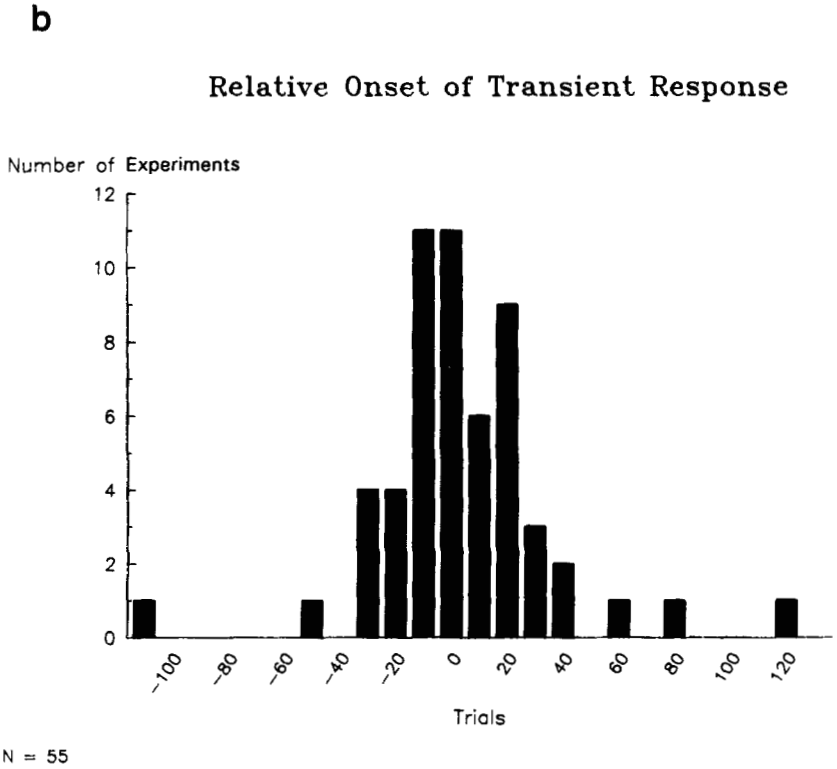


Fig. 5. (a) The onset of the differential neuronal responses after the start of the experiment in the experiments in which transient modifications of neuronal activity were found. (b) The onset of the transient differential neuronal responses shown relative to the trial at which the learning became behaviorally evident.

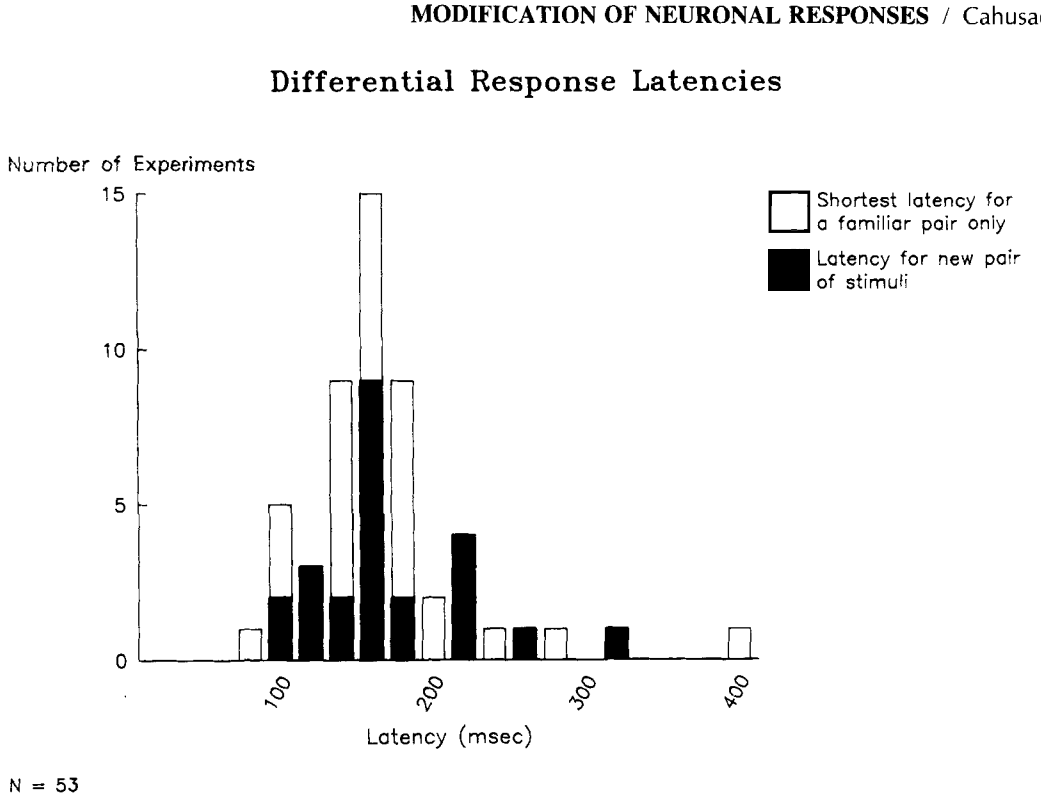


Fig. 6. The latency at which the response became differential to the two stimuli for each of the neurons that showed modification of its responsiveness during learning. The latencies shown are for the new pair of stimuli, except where because of the considerable experience of the monkey with the familiar pair of stimuli, the latency with the familiar pair was shorter (see the open part of the histogram).

task, and thus occurred sufficiently fast to be involved in the learning of the monkeys in these experiments.

It was found that while in 22% of experiments neurons formed relatively long-term differential responses to the stimuli used in the task, with differential responses that lasted for at least 50 trials and as long as the recording continued (often more than 1 hour), in more experiments (45%) neurons started to show differential responses at about the time of learning, but showed this effect only transiently for a number of trials, after which differential responses did not continue. These neurons with transient differential responses are of great interest, for their responses are consistent with the suggestion that there are competitive interactions between hippocampal neurons that ensure that only a limited number, and not all, neurons in a population become specialized during learning for any one learned association or event (Rolls, 1989a–c; 1990a–c; 1987). It is clearly essential to have a mechanism in a neuronal population involved in learning that ensures that not all neurons become devoted to only one learned item. The way in which it has been suggested that this operates in the hippocampus in a process involving competition is as follows (for further details, including neuronal network simulations, see Rolls, 1989a–c; 1990a–c; 1991; 1987).

Hippocampal neurons (e.g., dentate granule or CA1 pyramidal cells) that receive relatively large proportions of conjunctively afferent inputs are activated by these inputs. The most strongly activated neurons then through the inhibitory interneurons tend to inhibit the other neurons in the population to produce a state by competition in which only the neurons receiving the strongest inputs would remain strongly active. This competitive mechanism thus increases the differences in the firing rates of the activated population of neurons, and is a process that may utilize a nonlinear (e.g., sigmoid) output activation function of the neurons (e.g., Grossberg, 1987). The synapses from active axons to activated dendrites would then show long-term potentiation (consistent with evidence on the hippocampus, McNaughton, 1984; Levy, 1985; Brown et al., 1989; 1990). This competitive learning process could be helped by a decrease in synaptic strength from inactive axons to activated dendrites (heterosynaptic long-term depression), or by a decrease in synaptic strength from active axons to inactive dendrites (homosynaptic long-term depression), as described by Rolls (1989c). This competitive learning would also be helped by the nonlinearity inherent in the operation of the NMDA receptors (Rolls, 1989c). The changes described in this paper are consistent with a competitive learning process, in that for the majority of the neurons (23/36) that showed transient changes during learning, an increase of activity was seen initially to one of the stimuli, and then over the next 10–20 trials the activity to this stimulus dropped back down to the level of the other stimulus. This is consistent with inhibition of such neurons that may have started to learn by other (sustained) hippocampal neurons that performed better in the competition. This could force the neurons that show transient learning into a state in which reverse learning (long-term depression) can occur. The operation of such competitive learning provides one way in which it can be ensured in a neuronal network that

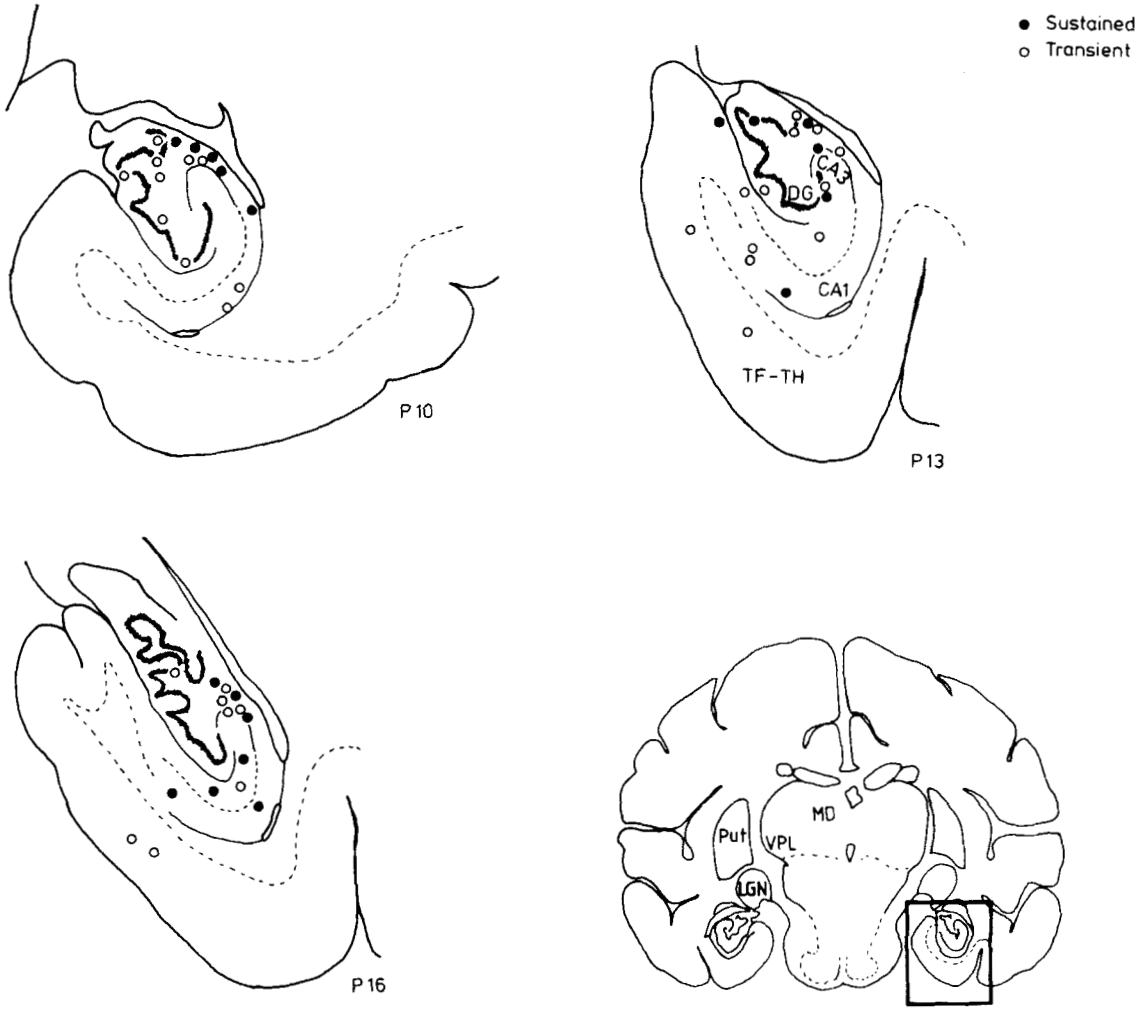


Fig. 7. Recording sites shown on coronal sections of the brain taken at three different planes, the number of mm indicated posterior (P) to the posterior wing of the sphenoid reference (see Aggleton and Passingham, 1981). CA1 and CA3, subfields of hippocampal cells; DG, dentate gyrus; LGN, lateral geniculate nucleus; MD, mediodorsal nucleus of the thalamus; Put, putamen; TF-TH, parahippocampal gyrus; VPL, ventral posterolateral nucleus of the thalamus. Filled circles, recording sites of neurons that showed sustained modifications of activity during learning. Open circles, recording sites of neurons that showed transient modifications of activity during learning. The neurons were recorded in both hemispheres of three monkeys.

different associations are learned by different neurons, and that every neuron does not become allocated to the same learned association or event (see Rolls, 1989a–c; 1990a–c; 1987).

At the systems level, we understand the results described here in the following way (for details and references see Rolls, 1989a–c; 1990a–c; 1987). Inputs reach hippocampal neurons from the cerebral cortex via the parahippocampal gyrus, entorhinal cortex, and perforant path. These inputs include specifications of the stimuli being seen, from the temporal lobe visual cortical areas, and of the positions of the limbs and body in space and the movements being made, from the parietal cortex. When a particular visual stimulus repeatedly occurs conjunctively with the specification of a movement (when, for example, the monkey reaches before learning has occurred to touch a particular visual stimulus and does this repeatedly because reward is obtained), then the conjunction of these inputs is detected by hippocampal neurons so that they come to respond to this combination of inputs. The ways in which only a proportion of hippocampal neurons may learn this association is described above and elsewhere (Treves and Rolls, 1992). Once this association has been formed by the hippocampus, it then sends this output not only back to the cerebral cortex, where it may help to build representations of the new visual stimuli being seen (see Rolls, 1989a,b), but also via the fornix/mammillary body/anterior thalamic nucleus route to the cingulate cortex, from which there are connections to the supplementary motor area. By this route, access to the motor system may be provided so that the appropriate motor responses can be produced after the learning has occurred. Part of the reason why the hippocampus is involved in this type of conditional spatial response learning may be

that an arbitrary association of a stimulus with a spatial response must be learned and applied in a particular context, i.e., that an episodic memory must be formed (see further Rolls, 1990a; 1991).

Although in this investigation alterations of neuronal responses were found in the hippocampus in a conditional spatial response task for the learning of which the hippocampus is essential (Rupniak and Gaffan, 1987), neuronal response modifications were also found during the learning of a visual discrimination task. In the case of visual discrimination tasks, the association required is between a (visual) stimulus and reinforcement (e.g., the taste of food), and this type of association depends on other brain structures such as the amygdala and orbitofrontal cortex (see Rolls, 1985; 1990d; 1987). It is therefore suggested that insofar as the hippocampal activity reflects associations between visual stimuli and reinforcers, it does so because this information may be useful, or potentially useful, as part of an episodic memory (e.g., that food was associated with a visual stimulus in a particular context). On the other hand, when there is a general rule, that a visual stimulus is normally associated with a reinforcement, then the amygdala and orbitofrontal cortex are sufficient (Rolls, 1990d).

It should be noted that although in this investigation modifications were found during learning of neuronal responses recorded in the hippocampus, this does not prove that the synaptic changes relevant to the learned modification actually took place in the hippocampus. It is possible for example that the learning took place at neurons earlier in information processing than in those from which recordings were made. However, synaptic modifications within the hippocampus accounting for the neuronal response modifications observed are quite likely for a number of reasons. First, the hippocampo-fornical system is necessary for the conditional spatial response learning the monkeys performed (Rupniak and Gaffan, 1987). Second, the hippocampus provides one of the regions in the brain where inputs from cortical areas as diverse as the temporal lobe visual cortical areas and the parietal lobe can be brought together (see Rolls, 1985; 1989a,b; 1990a). Consistent with this hypothesis that inputs from different cortical areas are brought together in the hippocampus, hippocampal neurons respond in conditional spatial response tasks to combinations of particular stimuli with particular responses, as described here and elsewhere (Miyashita et al., 1989). Third, synaptic modification is a property of hippocampal neurons (see McNaughton, 1984; Levy, 1985; Kelso et al., 1986; Cahusac et al., 1991). Fourth, the transient modifications described in the greater proportion of the neurons described here have a simple explanation if the hippocampus is the site of a competitive learning mechanism. However, it will be of great interest in future studies to record in the brain regions from which the hippocampus receives its inputs during the type of learning described here, to determine whether the learned modifications in neuronal responses described are a result of synaptic modifications within the hippocampus, or in structures afferent to the hippocampus and parahippocampal gyrus. In either case, the modifications of the neuronal responses described here do appear to be part of the neuronal basis for certain types of learning in which the hippocampus is involved.

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References

Aggleton JP, Passingham RE (1981) Stereotaxic surgery under X-ray guidance in the Rhesus monkey, with special reference to the amygdala. Exp Brain Res 44:271–276.

Brown TH, Ganong AH, Kairiss EW, Keenan CL, Kelso SR (1989) Long-term potentiation in two synaptic systems of the hippocampal brain slice. In Neural models of plasticity: experimental and theoretical approaches (Byrne JH, Berry WO, eds), pp 266–306. San Diego: Academic Press.

Brown TH, Kairiss EW, Keenan CL (1990) Hebbian synapses: biophysical mechanisms and algorithms. Annu Rev Neurosci 13: 475–511.

Bruning JL, Kintz BL (1977) Computational handbook of statistics. 2nd ed. Glenview, IL: Scott, Foresman.

Cahusac PMB, Miyashita Y, Rolls ET (1989) Responses of hippocampal formation neurons in the monkey related to delayed spatial response and object-place memory tasks. Behav Brain Res 33: 229–240.

Cahusac PMB, Rolls ET, Miyashita Y (1987) A comparative study of responsive neurons in different subareas of the primate hippocampal formation. Neurosci Lett 29(Suppl):S103.

Feigenbaum JD, Rolls ET (1991) Allocentric and egocentric spatial information processing in the hippocampal formation of the behaving primate. Psychobiology 19:21–40.

Gaffan D (1985) Hippocampus: memory, habit and voluntary movement. Philos Trans R Soc Lond B 308:87–99.

Gaffan D (1987) Amnesia, personal memory and the hippocampus: experimental neuropsychological studies in monkeys. In Cognitive neurochemistry (Stahl SM, Iversen SD, Goodman EC, eds), pp 46–56. Oxford: Oxford University Press.

Gaffan D, Harrison S (1984) Reversal learning by fornix-transected monkeys. Q J Exp Psychol 36B:223–234.

Gaffan D, Saunders RC (1985) Running recognition of configural stimuli by fornix-transected monkeys. Q J Exp Psychol 37B:61–71.

Gaffan D, Harrison S (1989) Place memory and scene memory: effects of fornix transection in the monkey. Exp Brain Res 74:202–212.

Grossberg S (1987) Competitive learning: from interactive activation to adaptive resonance. Cognitive Science 11:23–63.

Levy WB (1985) Associative changes in the synapse: LTP in the hippocampus. In Synaptic modification, neuron selectivity, and nervous system organization (Levy WB, Anderson JA, Lehmkuhle S, eds), pp 5–33. Hillsdale, NJ: Erlbaum.

McNaughton BL (1984) Activity dependent modulation of hippocampal synaptic efficacy: some implications for memory processes. Ch. 13. In Neurobiology of the hippocampus (Seifert W, ed), pp 233–252. London: Academic Press.

Merrill EG, Ainsworth A (1972) Glass-coated platinum-plated tungsten microelectrodes. Med Biol Eng 10:662–672.

Milner B (1972) Disorders of learning and memory after temporal lobe lesions in man. Clin Neurosurg 19:421–446.

Mishkin M (1978) Memory in monkeys severely impaired by combined but not separate removal of amygdala and hippocampus. Nature 273:297–298.

Mishkin M (1982) A memory system in the monkey. Philos Trans R Soc B 298:85–95.

Mishkin M, Petri HL (1984) Memories and habits: some implications

for the analysis of learning and retention. In Neuropsychology of memory (Squire LR, Butters N, eds), pp 287–296. New York: Guilford.

Miyashita Y, Rolls ET, Cahusac PMB, Niki H, Feigenbaum JD (1989) Activity of hippocampal neurons in the monkey related to a conditional spatial response task. J Neurophysiol 61:669–678.

Parkinson JK, Murray EA, Mishkin M (1988) A selective mnemonic role for the hippocampus in monkeys: memory for the location of objects. J Neurosci 8:4059–4167.

Petrides M (1985) Deficits on conditional associative-learning tasks after frontal- and temporal-lobe lesions in man. Neuropsychologia 23:601–614.

Rolls ET (1985) Connections, functions and dysfunctions of limbic structures, the prefrontal cortex, and hypothalamus. In The scientific basis of clinical neurology (Swash M, Kennard C, eds), pp 201–213. London: Churchill Livingstone.

Rolls ET (1987) Information representation, processing and storage in the brain: analysis at the single neuron level. In The neural and molecular bases of learning (Changeux J-P, Konishi M, eds), pp 503–540. Chichester: Wiley.

Rolls ET (1989a) Functions of neuronal networks in the hippocampus and neocortex in memory. In Neural models of plasticity: experimental and theoretical approaches (Byrne JH, Berry WO, eds), pp 240–265. San Diego: Academic Press.

Rolls ET (1989b) The representation and storage of information in neuronal networks in the primate cerebral cortex and hippocampus. In The computing neuron (Durbin R, Miall C, Mitchison G, eds), pp 125–159. Wokingham, England: Addison-Wesley.

Rolls ET (1989c) Functions of neuronal networks in the hippocampus and cerebral cortex in memory. In Models of brain function (Cotterill RMJ, ed), pp 15–33. Cambridge: Cambridge University Press.

Rolls ET (1990a) Functions of the primate hippocampus in spatial processing and memory. In Neurobiology of comparative cognition (Olton DS, Kesner RP, eds), pp 339–362. Hillsdale, NJ: Erlbaum.

Rolls ET (1990b) Functions of neuronal networks in the hippocampus and of backprojections in the cerebral cortex in memory. In Brain organization and memory: cells, systems and circuits (McGaugh JL, Weinberger NM, Lynch G, eds), pp 184–210. New York: Oxford University Press.

Rolls ET (1990c) Theoretical and neurophysiological analysis of the functions of the primate hippocampus in memory. Cold Spring Harbor Symposia in Quantitative Biology 55:995–1006.

Rolls ET (1990d) A theory of emotion, and its application to understanding the neural basis of emotion. Cogn Emotion 4:161–190.

Rolls ET (1991) Functions of the primate hippocampus in spatial and nonspatial memory. Hippocampus 1:258–261.

Rolls ET, Burton MJ, Mora F (1976) Hypothalamic neuronal responses associated with the sight of food. Brain Res 111:53–66.

Rolls ET, Miyashita Y, Cahusac PMB, Kesner RP, Niki H, Feigenbaum J, Bach L (1989) Hippocampal neurons in the monkey with activity related to the place in which a stimulus is shown. J Neurosci 9:1835–1845.

Rolls ET, Sanghera MK, Roper-Hall A (1979) The latency of activation of neurones in the lateral hypothalamus and substantia innominate during feeding in the monkey. Brain Res 164:121–135.

Rolls ET, O'Mara S (1992) Neurophysiological and theoretical analysis of how the hippocampus functions in memory. In Brain mechanisms of perception: from neuron to behavior (Ono T, Squire LR, Raichle M, Perrett D, Fukuda M, eds). New York: Oxford University Press. in press.

Rupniak NMJ, Gaffan D (1987) Monkey hippocampus and learning about spatially directed movements. J Neurosci 7:2331–2337.

Scoville WB, Milner B (1957) Loss of recent memory after bilateral hippocampal lesions. J Neurol Neurosurg Psychiatr 20:11–21.

Siegel S, Castellan NJ (1989) Non-parametric statistics for the behavioural sciences. 2nd ed. New York: McGraw-Hill.

Smith ML, Milner B (1981) The role of the right hippocampus in the recall of spatial location. Neuropsychologia 19:781–793.

Squire L (1986) Mechanisms of memory. Science 232:1612–1619.

Squire L, Zola-Morgan S (1983) The neurology of memory: the case for correspondence between the findings for human and nonhuman primates. In The physiological basis of memory. 2nd ed. (Deutsch JA, ed), pp 199–268. New York: Academic Press.

Squire LR, Shimamura AP, Amaral DG (1989) Memory and the hippocampus. In Neural models of plasticity: theoretical and empirical approaches (Byrne J, Berry WO, eds), pp 208–239. New York: Academic Press.

Treves A, Rolls ET (1992) Computational constraints suggest the need for two distinct input systems to the hippocampal CA3 network. Hippocampus 2:189–199.

Woodward RH, Goldsmith PL (1964) Cumulative sum techniques: mathematical and statistical techniques for industry. ICI Monograph No. 3. Edinburgh: Oliver & Boyd.

Zola-Morgan S, Squire LR, Amaral DG (1986) Human amnesia and the medial temporal region: enduring memory impairment following a bilateral lesion limited to field CA1 of the hippocampus. J Neurosci 6:2950.