# Effects of Low and High Frequency Electrical Stimulation on Rat Hippocampal Place Cells

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**Abstract**

This paper describes experimental results on interaction between place cell in rat hippocampal CA1 area and low (1 Hz) or high (100 Hz) frequency electrical stimulation of the Schaffer collaterals. High frequency unilateral stimulation of Schaffer collaterals impaired both random wandering and spatial navigation tasks, motivated by intracranial self-stimulation as a reward. However, low frequency stimulation did not disrupt its behaviors. The complex-spike activity of CA1 neuron was suppressed by high frequency stimulation and, as a result, the place field disappeared just after stimulation. Within an hour, both neuronal activity and task performance were recovered to the control level. The place field recovered was different from the control ones. Low frequency stimulation on the same electrode also modified neuronal activity and place field. These suggest that place dependency in the CA1 neurons may be implemented through the Schaffer collaterals of CA3, and that neuronal information correlated to the long-term potentiation or long-term depression might be related to the place processing.

## 1. Introduction

The hippocampus is one of the central sites in learning and memory. Both long-term potentiation (LTP) and long-term depression (LTD) at excitatory synapses contribute to memory process [13]. The LTP can be induced by high frequency electrical stimulation [3], and the LTD by low frequency electrical stimulation [12]. The physiological evidences of the LTP and LTD suggest different mechanisms of induction in synaptic responses, depending on a pattern of cell discharge[8].

Behaviorally, high frequency stimulation of the rat hippocampal perforant path impaired spatial learning using circular platform [1], although some negative results were reported in different spatial task [4, 14]. This suggests that synaptic transmission in neural

circuit involving dentate gyrus may be modulated by artificial electrical stimulation.

On the other hand, it is well-known that the hippocampal CA1 neuron encodes spatial information in the form of place cells [15, 16]. We reported that the place cells have characteristic response of plasticity influenced by new spatial navigation learning and displacement of rewarding area [9, 10]. This change of place dependency may be implemented by a physiological mechanism to LTP. It is reported that the electrical potential evoked by the perforant path stimulation was enhanced by introduction of a new environment or new spatial task [14, 18]. However, there are few reports to investigate direct correlation between place cells in behaving animals and electrical tetanic stimulation. It was suggested that LTP and LTD are reversible modifications of the same Schaffer collateral synapses [8]. In this paper we study effects of electrical stimulation on the hippocampal CA1 neurons of the rats that learn a spatial navigation task motivated by intracranial self-stimulation as a reward.

## 2. Materials and Methods

General methods for the spatial tasks are described elsewhere [7, 11]. In brief, five male albino Wistar rats were anesthetized with sodium pentobarbital (40 mg/kg, i.p.) and then implanted bilaterally stimulating electrodes (enamel-coated stainless-steel wire of 200 $\mu$m in diameter) aimed at the lateral hypothalamus for intracranial self-stimulation (ICSS). After one week recovery, the rats were trained to self-stimulate by lever pressing in an operant chamber. Current intensity for ICSS was set to produce a rate of 40 to 60 lever presses per min (0.5 sec train of 0.3 msec negative square pulses at 100 Hz, 100–250 $\mu$A). The ICSS-trained animals were then again anesthetized and implanted with a bipolar stimulating electrode aimed at Schaffer collateral. The stimulating electrode was positioned at a point that produced evoked potential at CA1 pyramidal layer (AP: 2.8 mm, L: 2.0 mm from bregma). Also the animal was implanted with a movable recording electrode assembly consisting of a bundle of 4 or 8 microwires (Formvar insulated, 25 $\mu$m diameter nicrome) aimed at the dorsal CA1 hippocampal area (AP: 4.0 mm, L: 2.0 mm from bregma). After recovery, the animals were used for the spatial behavior tests in a circular open-field (150 cm diameter with a 45 cm high wall). The open-field was enclosed by a black curtain (180 cm in diameter and 200 cm in height). An electric bulb at 3 o'clock and a speaker emitting white noise at 9 o'clock were set on the ceiling of the enclosure as environmental cues. To detect the locomotion of the rat, a small light placed on the animal's headstage was tracked at the rate of 10 frames/sec with a video camera

(CT-10, Toyo Sangyo, Toyama). Whenever the rat entered a circular reward area delimited by a program, then ICSS was delivered as reward. Two kinds of tasks were employed in the open-field: 1) A random wandering (RW) task, in which a reward area (70 cm in diameter) was successively located at randomly selected sites. 2) A spatial navigation (SN) task in which two reward areas (40 cm in diameter) were fixed and a reward was delivered when a rat entered them alternately. A session was terminated after the animal received 50 ICSS, or after 300 sec elapsed, whichever occurred first. The rat was exposed to the RW task in the open-field for 1–2 weeks until they explored the field entirely. Unit activity was recorded by a conventional system consisting of a preamplifier and a window discriminator , and was processed by a microcomputer. Spikes were counted by counter I/O board at every 10 msec intervals.

## 3. Results

### 3.1 Effects of high frequency electrical stimulation

Figure 1A shows an example of locomotion and firing rate of the hippocampal CA1 complex-spike activity for 5 min during the RW task, where the firing rate is expressed by the cumulated number of spike recorded divided by the cumulated time spent in each pixel. The animal tended slightly to move along the wall despite the demand for random wandering (ICSS =17, traveled distance=5991 cm). The neuronal activity shows a large place field (firing rate=1.03±1.47 spikes/sec). The spatial ratio is 0.27, where the value is defined to be pixel that was active divided by pixel that was stayed. Then we applied one-time electrical stimulation to the Schaffer collaterals at 100 Hz and 100 pulses of 200 $\mu$sec duration. The stimulus intensity was 6V, which was optimum value adjusted at the implantation of the stimulating electrode. Immediately after the application, the RW task was tested (Fig. 1Ba). The animal would move to acquire the ICSS during the task less than the control (ICSS=12). However, ability of movement did not change (traveled distance=5539 cm). The trajectory shows the fixed circular forms along the wall. The neuronal activity was suppressed by stimulation (firing rate=0.33±2.06 spikes/sec; spatial ratio=0.08). At following times of 10, 20, and 30 min, the movement and motivation did not change (ICSS=21, 24 and 28; traveled distance=5830, 6587 and 7797 cm, respectively). Accordingly, the spontaneous activity and place fields were recovered (firing rate=0.23±0.53, 0.31±0.66 and 0.57±0.98 spikes/sec; spatial ratio=0.1, 0.13 and 0.2, respectively) (Figs. 1Bb, c and d). Figure 1Be shows the trajectory and place field at 60 min after tetanic stimulation. The trajectory looked similar to the control (ICSS=32,

traveled distance=8394 cm). The place field was still relative large (firing rate=0.45± 0.81 spikes/sec; spatial ratio=0.17), but seemed to be different from the control.

After high frequency stimulation, a SN task was conducted, in which animals were trained in 10 trials per day for two days. Again, high frequency electrical stimulation to the Schaffer collaterals was applied under the same conditions as in the previous tasks. Figure 2 shows an example of the trajectory of movement and firing rate map as a function of time measured. At control immediately before the application, the place field was large and at the direction of 6 o'clock in the SN task (spatial ratio=0.22; ICSS=50 for 207 sec, traveled distance=7764 cm) (Fig. 2A). The mean frequency was 1.03±1.46 spikes/sec. After stimulation, stereotypical patterns of trajectory were completely disturbed (spatial ratio=0.04, ICSS=41 for 5 min, traveled distance=9383 cm) (Fig. 2Ba).

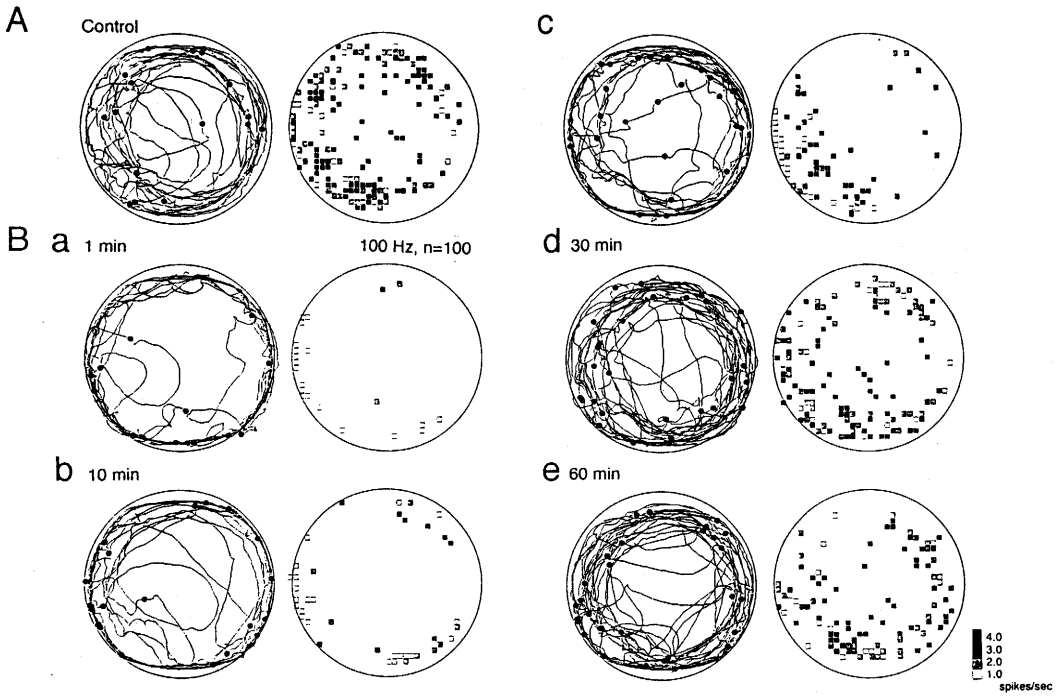


**Fig. 1.** Changes of exploration patterns and hippocampal complex-spike firing due to high frequency electrical stimulation (100 Hz, 100 pulses) of Schaffer collaterals. Left: exploration tracing. Small dots indicate locations of reward delivery. Right: firing rate map. A, control in RW task. The neuron had wide place field (acquired rewards: 17 for 5 min). B, effects of high frequency stimulation (100 Hz, 100 pulses of 200 $\mu$sec duration). a-e, 1,10,20,30, and 60 min after stimulation. The movement and neuronal activity were suppressed by stimulation. After an hour, both appeared to be recovered.

The spontaneous firing rate was suppressed (0.34±2.8 spikes). At following times of 10, 20, and 45 min, movement and motivation did not change (spent time=198, 204 and 201 sec for 50 ICSS; traveled distance=7373, 7515 and 7108 cm, respectively). Spontaneous activity and place field were recovered (firing rate=0.05±0.22, 0.22±0.69, 0.75±1.29 spikes/sec; spatial ratio=0.02, 0.06 and 0.13, respectively) (Figs. 2Bb, c and d). Figure 2Be shows the trajectory and place field at 60 min after tetanic stimulation. Movement, spontaneous firing and the ICSS number were recovered (spent time=200 sec for 50 ICSS; traveled distance=7461 cm; firing rate=0.79±1.47 spikes/sec; spatial ratio= 0.17). However, the place field was different from the control. Similar results were obtained in 3 of five animals.

### 3.2 Effects of low frequency stimulation

Figure 3A shows effects of low frequency electrical stimulation on place fields during

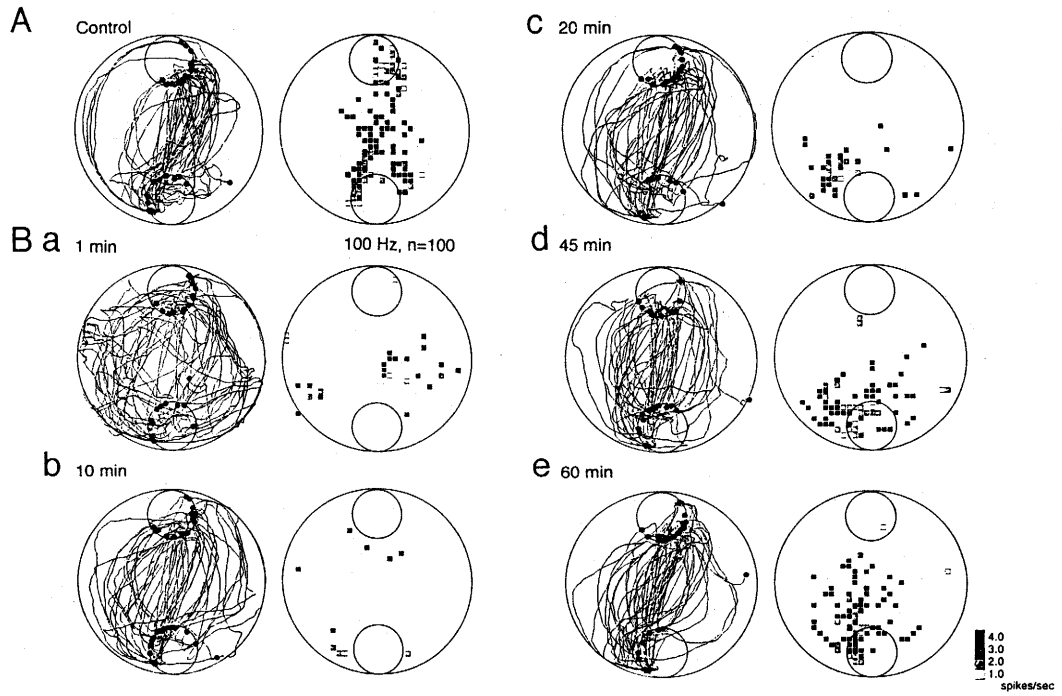


**Fig. 2.** Changes during SN task by high electrical stimulation to Schaffer collateral. Left: exploration tracing. Right: firing rate map. A, control in SN record. B, effects of high frequency stimulation, a-e: 1,10,20,45, and 60 min after stimulation. The movement and neuronal activity were disrupted by stimulation. Others descriptions as Fig. 1.

the RW task. Before application of stimulation, the animal trail covered over entire open field during the RW task (ICSS =31, traveled distance=8630 cm). The neuronal activity shows a large place field (firing rate=0.98±1.34 spikes/sec). The spatial ratio is 0.31. We applied one-time electrical stimulation to the Schaffer collaterals at 1 Hz and 100 pulses. Ability of movement did not change so much through the time (ICSS=21, traveled distance=5034 cm). The neuronal activity were suppressed by stimulation (firing rate= 0.52±0.89 spikes/sec; spatial ratio=0.11). At following times of 10, 20, and 30 min, movement and motivation did not change (ICSS=25, 30 and 42; traveled distance=6009, 6946 and 8273 cm, respectively). Accordingly, spontaneous activity and place fields were recovered (firing rate=0.75±1.11, 0.87±1.22 and 0.67±1.06 spikes/sec; spatial ratio= 0.22, 0.28 and 0.24, respectively) (Figs. 3Bb, c and d). Figure 3Be shows the trajectory and place field at 60 min after low frequency stimulation. The trajectory looked similar to the control (ICSS=33, traveled distance=7801 cm), and the spontaneous firing

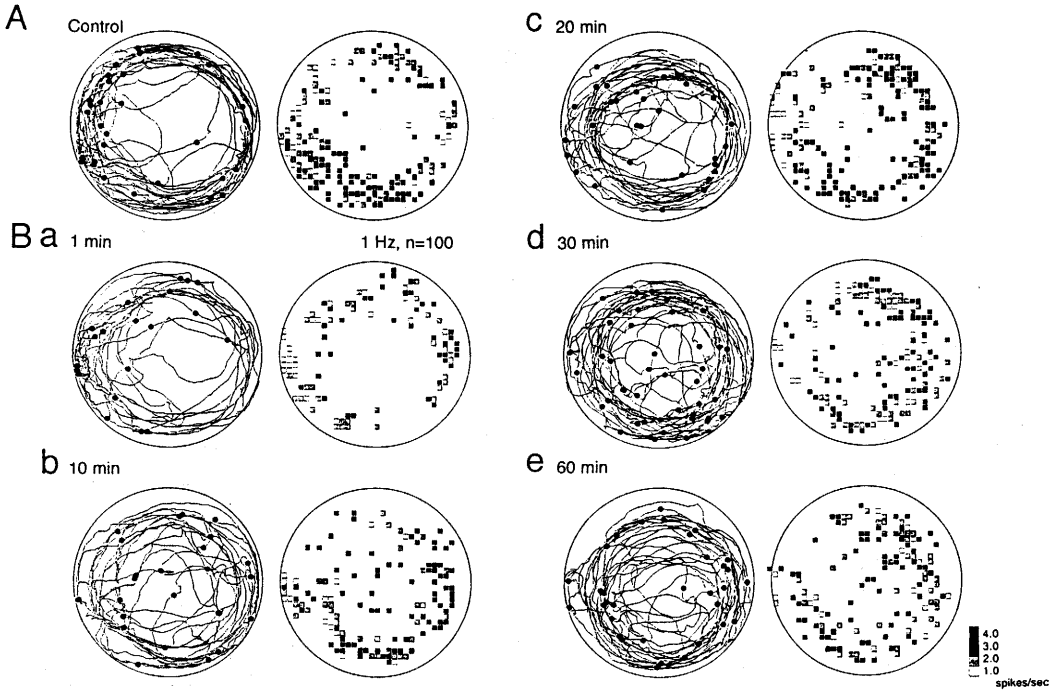


**Fig. 3.** Changes of exploration patterns and hippocampal complex-spike firing due to low frequency electrical stimulation (1 Hz, 100 pulses) of Schaffer collaterals. A, control in RW task. B, effects of low frequency stimulation (1 Hz, 100 pulses of 200 μsec duration). a-e, 1,10,20,30, and 60 min after stimulation. The movement and neuron activity were suppressed by stimulation. After an hour, both appeared recover. Others descriptions as Fig. 1.

frequency was recovered almost to the control level (0.60±0.97 spikes/sec). The place field was still relatively large and similar as the control (spatial ratio=0.22).

Low frequency electrical stimulation to the Schaffer collaterals was applied during the SN task. Figure 4 shows an example of the trajectory of movement and firing rate map. At the control immediately before the application, the place field was restrict and at the direction of 12 o'clock (spatial ratio=0.09; ICSS=22, traveled distance=7270 cm) (Fig. 4A). The mean frequency was 0.45±1.39 spikes/sec. After the stimulation, the behavior did not change (Fig. 4Ca-c). (spent time=144, 132 and 145 sec for 50 ICSS; traveled distance=6543, 6149 and 6591 cm, respectively). Spontaneous activity and place fields were recovered (firing rate=0.34±0.88, 0.26±0.78, 0.68±2.09 spikes/sec; spatial ratio=0.09, 0.07 and 0.14, respectively) (Figs. 4Cb, c and d). Figure 4Cc shows the trajectory and place field at 60 min after tetanic stimulation. Movement, spontaneous firing and the ICSS number were recovered (spent time, 146 sec for 50 ICSS; traveled distance=6591 cm; firing rate=0.68±2.09 spikes/sec; spatial ratio=0.14). Similar results

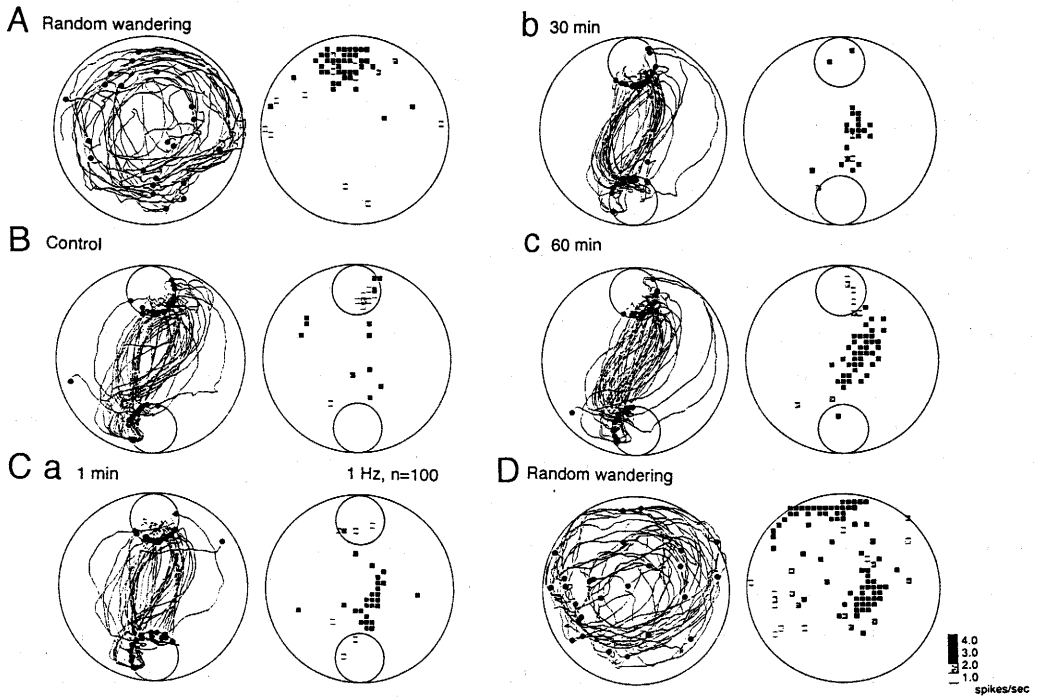


**Fig. 4.** Changes during SN task by low frequency stimulation to Schaffer collaterals. A, place field in RN task. B, place field in SN task. C, effects of low frequency stimulation, a-e: SN task, 1,30,45, and 60 min after stimulation. Others descriptions as Fig. 1.

were obtained in 4 of five animals.

**4. Discussions**

Our results showed that high frequency stimulation disrupted the spatially learned behavior, and suppressed the spontaneous unit activity and, as a result, the place field. The behavioral impairment was consistent with previous reports in which the electrical stimulation of the angular bundle disrupted spatial learning in a circular maze either prior to or immediately after learning [14]. Concerning the neuronal activity, it was confirmed that behavioral impairment corresponded to disappearance of the place field. It is reported that the stimulation saturated synaptic transmissions around the stimulating electrode [1, 2]. Such saturation might block synaptic transmission from CA3 to CA1 through the Schaffer collaterals. These suggest that LTP itself is essential for learning because saturation impaired acquisition of the spatial learning task.

After recovery from the stimulation, the place field appeared to be re-organized, showing a different shape from the control. The artificial electrical high frequency stimulation may induce a global and uniform disturbance in synaptic transmission to targeting neurons. If the stimulation induces temporally loss of memory stored in the neural circuit, the spatial information could be re-encoded according to the recovery of behavior, since the animal performed relatively complete spatial navigation task after recovery. This process will be carried out by the persisting information in dentate gyrus, CA3 and others. The new configuration in neural information might be formed according to performance of the corresponding spatial task.

The LTP usually lasts a long time from weeks to months [2]. In our experiment, however, the deficit effects lasted only one hour. This suggest an possibility of induction of short-term potentiation which decays in about one hour. Sharp et al. reported short-term exploratory modulation in changes of population EPSPs, which lasts only 20–40 min [17]. It is unclear that our phenomena is directly correlated with the LTP, since the evoked potentials were not measured over two hours. Dudek and Bear found that several hundred stimuli delivered to the Schaffer collaterals at low frequencies produced a sustained depression of modest but significant magnitude [5, 6]. The LTD is frequency dependent and the same number of pulses at higher frequencies can produce LTP [5]. There were few reports on behavioral effects of the LTD [19]. Our data showed fails of behavioral deficits, but modulation of spatial information processing. The fails of behavior might be small number of pulses in stimulation.

The LTP and LTD reversibly affect synaptic effectiveness by acting at a common

site [6]. Both LTP and LTD modulated spatial information processing in hippocampal CA1. Further study to record evoked potential and analyze precise behavior will be needed.

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