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Research report

Neuronal codes for the inhibitory control of impulsive actions in the rat infralimbic cortex

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HIGHLIGHTS

- Inactivation of the infralimbic cortex (IL) impaired impulse control in rats.
- More than 30% of recorded IL units were determined to be impulse control-related units.
- Several types of impulse control-related units were identified.
- Units with firing that was related to impulse control and attentional function were also detected in the IL.

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ABSTRACT

Poor impulse control is a debilitating condition observed in various psychiatric disorders and could be a risk factor for drug addiction, criminal involvement, and suicide. The rat infralimbic cortex (IL), located in the ventral portion of the medial prefrontal cortex, has been implicated in impulse control. To elucidate the neurophysiological basis of impulse control, we recorded single unit activity in the IL of a rat performing a 3-choiceserial reaction time task (3-CSRTT) and 2-choice task (2-CT), which are animal models for impulsivity. The inactivation of IL neuronal activity with an injection of muscimol (0.1 μg /side) disrupted impulse control in the 3-CSRTT. More than 60% (38/56) of isolated IL units were linked to impulse control, while approximately 30% of all units were linked to attentional function in the 3-CSRTT. To avoid confounding motor-related units with the impulse control-related units, we further conducted the 2-CT in which the animals' motor activities were restricted during recording window. More than 30% (14/44) of recorded IL units were linked to impulse control in the 2-CT. Several types of impulse control-related units were identified. Only 16% of all units were compatible with the results of the muscimol experiment, which showed a transient decline in the firing rate immediately before the release of behavioral inhibition. This is the first study to elucidate the neurophysiological basis of impulse control in the IL and to propose that IL neurons control impulsive actions in a more complex manner than previously considered.

1. Introduction

Impulsive behavior is broadly defined as "actions that are poorly conceived, prematurely expressed, unduly risky, or inappropriate to the situation and that often result in undesirable outcomes" [1]. A lack of impulse control could be a risk factor for drug addiction [2], criminal involvement [3], and suicide [4,5]. Moreover, disrupted impulse control has been observed in psychiatric disorders such as attention-deficit/hyperactivity disorder [6,7], antisocial or bor-

Abbreviations: AP, anterior-posterior; DV, dorsal-ventral; GABA, gamma-aminobutyric acid; IL, infralimbic cortex; ITI, intertrial interval; L, lateral; mPFC, medial prefrontal cortex; 2-CT, 2-choice task; 3-CSRTT, 3-choice serial reaction time task.

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derline personality disorders [8,9], and bipolar disorder [10,11]. Thus, elucidating the neural basis of impulse control would be an important contribution to modern society.

The rat medial prefrontal cortex (mPFC) has been implicated in various cognitive functions [12]. A previous study demonstrated that inactivation of the prelimbic cortex, a dorsal component of the mPFC, impaired behavioral inhibition [13]. Moreover, in vivo unit recording using freely moving rats revealed that one-third of recorded units in the prelimbic cortex were linked to behavioral inhibition [14,15]. On the other hand, many pharmacological and lesion studies have supported the hypothesis that the neural activities of the infralimbic cortex (IL), a ventral portion of the mPFC, rather than those of the prelimbic cortex [16] inhibit impulsive behavior [17–22]. However, the electrophysiological properties of the IL involved in impulse control remain poorly understood. The aim of the present study was to bridge the gap between the abundance of pharmacological/lesion studies and paucity of electrophysiological studies.

In order to elucidate the neurophysiological basis of impulse control, we first performed single unit recordings in the rat IL during the 3-choice serial reaction time task (3-CSRTT), an established task to assess impulsive action and attentional function [16,20,23,24]. In this task, a light is briefly flashed in the aperture of one of three holes and animals are required to make a nose poke response into the lit hole (i.e., correct response) to get a food pellet. Nose poke responses before the presentation of the light stimulus are described as premature responses, which result in a time-out period and are regarded as an impulsive action (i.e., disruption of impulse control) [23]. We compared the pre-response activities of the same neuronal unit in correct versus premature trials. Since neurons in the rat mPFC have also been shown to be involved in attentional function [25,26], we herein compared pre-stimulus firing rates in correct versus incorrect trials for the same unit. We employed the 2-choice task (2-CT), in which animal movements were controlled during the recording window, to further confirm our assertion that IL neural activities are linked to behavioral inhibition. In this task, animals are required to keep pushing the center panel until a light stimulus is presented briefly in one of two holes, whereas animals are allowed to move freely while waiting for a light stimulus in the 3-CSRTT.

2. Materials and methods

2.1. Subjects

Male Wistar/ST rats were obtained from Nippon SLC Co., Ltd. (Hamamatsu, Japan). Animals were housed under alternating light/dark cycles (lights on from 7 p.m. to 7 a.m.) at approximately 21 °C and a relative humidity of 40–50%. When the rats were 9 weeks old (weighing 270–290 g), their food intake was restricted and their body weights were maintained at 85% of free-feeding conditions. Daily food intake (CE-2; CLEA JAPAN Inc., Tokyo, Japan) in the home cage was 10–15 g during the training period and 10–12 g during the experimental period. Water was available ad libitum. This study was approved by the Care and Use of Laboratory Animals of the Animal Research Committee of Hokkaido University.

2.2. Three-choice serial reaction time task (3-CSRTT)

The sequence of tasks employed in the 3-CSRTT (Fig. 1A) has been described previously [23,27]. These experiments were performed in aluminum operant chambers controlled by a computer program written in the MED-PC language (Med Associates Inc., VT, USA). The curved rear wall of each chamber contained three holes, with an infrared photocell beam to detect nose poke responses, and a 2.8 W bulb at the rear of each hole.

When each trial of the task started, the house light was illuminated. After a 5-s waiting period, one of the three holes was briefly illuminated for 1 s. A computer randomized the order of illuminations of the 3 holes to prevent the rats from predicting the next target hole. Nose pokes during the waiting period were recorded as premature responses. The time between the start of the waiting period and a premature response was recorded as the premature response latency. Nose pokes into the lit hole while it was illuminated or within 5 s of a limited hold were recorded as correct responses and were rewarded with the delivery of a food pellet (45 mg each, dustless precision pellets, Bio-serv, NJ, USA). Nose pokes into non-lit holes were recorded as incorrect responses. The percent accuracy was calculated as 100 × (the number of correct responses)/(the number of correct and incorrect responses). The correct response latency (the time between a hole light illumination and correct response) and reward latency (the time between a correct response and nose poke into the food magazine) were also measured. When a rat failed to nose poke within the time limit, it was recorded as an omission. After the delivery and collection of the food pellet by a rat, the house light was switched off for 2 s (intertrail interval: ITI) to allow the rat to eat the pellet before the next trial started automatically. The start of the next trial was signaled by the illumination of the house light. Additional nose pokes into any of the 3 holes prior to food collection were recorded as perseverative responses. Premature responses, incorrect responses, omissions, and perseverative responses resulted in a 5-s time-out period during which the house light and hole light were extinguished. Each session consisted of 100 trials, and all rats in the present study finished 100 trials within 40 min. We conducted training sessions once daily and 6 sessions per week.

At the beginning of the training schedule, the light stimulus lasted 30 s, which was progressively reduced to 1 s (15, 10, 5, 3, 2, 1.5, and 1 s) depending on individual performances. When each rat attained >80% accuracy and had <20 omissions in a session, the stimulus duration was reduced in the next session. Training was finished when a rat reached the target phase (stimulus duration of 1 s) and showed stable performances. We set the criteria for determining stable performances as follows: the change in premature responses stayed within ±25%, the accuracy stayed within ±5%, and <20 omissions occurred in at least 3 consecutive sessions. After training was completed, the stimulus duration was fixed at 1 s regardless of performance.

2.3. Two-choice task (2-CT)

These experiments were performed in the same apparatus used in the 3-CSRTT, but equipped with a hinged aluminum panel in front of the center hole. The task procedure employed in the 2-CT is described in Fig. 4A. When each trial of the task started, the house light was illuminated. Rats were required to push the center panel using their nose within 5 s (holding period) to start the choice period. In the choice period, a computer randomized the order of illumination of the two holes to prevent the rats from predicting the next target hole. When the rat released its nose from the center panel before the illumination of hole light, it was recorded as premature withdrawal. The time between the start of the panel push and a premature withdrawal was recorded as the premature withdrawal latency. Nose pokes into the lit hole while it was illuminated or within 5 s of limited hold were recorded as correct responses and were rewarded with the delivery of a food pellet. Nose pokes into the non-lit hole were recorded as incorrect responses. The percent accuracy was calculated as 100 × (the number of correct responses)/(the number of correct and incorrect responses). The correct response latency (the time between a hole light illumina-