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Systematic study of the effects of stimulus parameters and stimulus location on afterdischarges elicited by electrical stimulation in the rat

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Summary Electrical brain stimulation is used in a variety of clinical situations, including cortical mapping for epilepsy surgery, cortical stimulation therapy to terminate seizure activity in the cortex, and in deep brain stimulation therapy. However, the effects of stimulus parameters are not fully understood. In this study, we systematically tested the impact of various stimulation parameters on the generation of motor symptoms and afterdischarges (ADs). Focal electrical stimulation was delivered at subdural cortical, intracortical, and hippocampal sites in a rat model. The effects of stimulus parameter on the generation of motor symptoms and on the occurrence of ADs were examined. The effect of stimulus irregularity was tested using random or regular 50 Hz stimulation through subdural electrodes. Hippocampal stimulation produced ADs at lower thresholds than neocortical stimulation. Hippocampal stimulation also produced significantly longer ADs. Both in hippocampal and cortical stimulation, when the total current was kept constant with changing pulse width, the threshold for motor symptom or AD was lowest between 50 and 100 Hz and higher at both low and high frequencies. However, if the pulse width was fixed, the threshold did not increase above 100 Hz and it apparently continued to decrease through 800 Hz even if the difference did not reach statistical significance. There was no significant difference between random and regular stimulation. Overall, these results indicate that electrode location and several stimulus parameters including frequency, pulse width, and total electricity are important in electrical stimulation to produce motor symptoms and ADs.

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Introduction

Electrical deep brain stimulation is a promising alternative therapy for patients with medically refractory focal seizures who are not candidates for surgical resection. Several stimulation therapies are currently being investigated, e.g., stimulation of the anterior nucleus of the thalamus (Mirski et al., 1997; Hodaie et al., 2002; Kerrigan et al., 2004; Theodore and Fisher, 2004; Takebayashi et al., 2007; Osorio et al., 2007; Fisher et al., 2010; Gigante and Goodman, 2011), stimulation of the hippocampus (Velasco et al., 2001a,b; Tellez-Zenteno et al., 2006; Velasco et al., 2007; Boon et al., 2007; McLachlan et al., 2010), and subdural stimulation of the cortex (Kossoff et al., 2004; Chkhenkeli et al., 2004; Kinoshita et al., 2005; Osorio et al., 2005; Elisevich et al., 2006; Schrader et al., 2006; Salanova and Worth, 2007). In these studies, different stimulus parameters have been used. For example, 40—1000 μs pulse width, 0.9—500 Hz frequency, and 0.5—15 mA intensity therapeutic stimuli were delivered either through subdural or hippocampal depth electrodes in clinical trials of the responsive neurostimulation system (RNS) (Kossoff et al., 2004; Fountas et al., 2005; Skarpaas and Morrell, 2009). However, the rationale for applying a given set of parameters is not fully based on experimental evidence.

Apart from its use in brain stimulation therapies, electrical stimulation is also used for cortical mapping during brain surgery in areas close to or at eloquent cortex. Direct cortical high-frequency stimulation of sufficient strength in humans elicits epileptiform activity in the form of afterdischarges (ADs). ADs in general remain localized in the human brain, but sometimes they spread and produce clinical epileptic seizures even when stimulating non-epileptogenic cortex (Lüders and Noachtar, 2000).

To determine the safest and most effective stimulation method, systematic evaluation of different stimulation parameters must be carried out in experimental animals (Boon et al., 2007). There are a number of studies on electrical stimulation at hippocampus or amygdala to create kindling model. Usually, stimulation parameters used to induce kindling are 0.2—1.0 mA amplitude at 60—100 Hz for 1—2 s trains (Goddard et al., 1969; LaSalle, 1981; Fisher, 1989). Massive electrical stimulation is also used to induce rapid kindling (Lothman and Williamson, 1993). In the few studies on the use of focal cortical stimulation to induce AD, the parameters applied were 0.5—5 mA, 0.5—10 ms pulse duration, and 1—5 s trains of alternative square wave pulses with a frequency of 10—100 Hz (Adrian, 1936; Pinsky and Burns, 1962; Ajmone-Marsan, 1972; Fisher, 1989). However, no systematic studies have been conducted on the effects of various stimulation parameters on the induction of seizure and AD activity. Therefore, we investigated the effects of different stimulation parameters, including stimulation frequency, pulse width, intensity, stimulus regularity, and electrode location on the threshold for motor symptom and ADs induction in anesthetized rats. The results may have application in developing animal models of epilepsy using cortical or hippocampal electrical stimulation. In addition, the results may be used to find the safest and most effective way to use therapeutic electrical stimulation of the cortex or hippocampus.

Methods

Surgical procedures

Seven male Sprague-Dawley rats (300—450 g) were used (Charles River Labs, USA). They were anesthetized with pentobarbital (50 mg/kg, i.p.), and placed in a stereotactic frame. The scalp was cut along the midline to expose the skull, and subdural stainless steel screw electrodes (0.06 in. diameter, MX-080-2, Small Parts Inc., FL, USA) were implanted bilaterally in the frontal cortex (anterior: 3 mm, lateral: ±3 mm from Bregma, Paxinos), the right motor area (anterior: −1 mm, right: 2.5 mm), bilateral sensory areas (anterior: −1 mm, lateral: ±5 mm), and bilateral occipital areas (anterior: −5 mm, lateral: ±3 mm). To confirm whether the tip of the screw electrode was set in the subdural space, the dura was removed to make the surface of the cortex visible through the drilled bone hole. A reference screw electrode for electroencephalographic (EEG) recordings was placed at the frontal bone. Intracortical twisted wire electrodes were also implanted stereotaxically into the bilateral frontoparietal cortex (anterior: −1 mm, lateral: 3.5 mm, depth: less than 2 mm from the cortical surface). The twisted wire electrodes consisted of quadruple Teflon-insulated stainless steel wires (0.008 in. diameter; 316SS8T, Medwire, NY, USA). The distance between adjacent active points at the uninsulated tips of the electrodes was 1.0 mm. In addition, twisted depth electrodes were implanted bilaterally in the hippocampi (anterior: −5.3 mm, lateral: ±5 mm, depth: 5 mm). Fig. 1A shows a diagram of the implanted electrodes. The electrodes were connected to a socket, which was fixed onto the scalp using dental cement. Stimulation and recordings were performed through the same electrodes using a switching device. The research protocol and the use of animals were approved by the Cleveland Clinic Foundation Institutional Animal Care and Use Committee.

Determination of the motor symptom threshold, AD threshold, and AD duration

Two weeks after electrode implantation, electrical stimulation examination was conducted. A Grass S88 dual stimulator was used to deliver stimuli (Grass Instruments, RI, USA). EEG activity was recorded from all electrodes, including the stimulating electrodes, using a Vanguard digital EEG monitoring system. A 5-s train of biphasic square pulses was used. For cortical stimulation, stimuli were delivered bipolarly between the two tips of twisted wires of the intracortical depth electrodes in the right frontoparietal cortex (Fig. 1A, electrode D1/2), and between the subdural electrodes over the right motor and sensory area (Fig. 1A, electrode RFP1 and RFP2). Stimulus intensity was initially 0.1 mA and was increased every minute in increments of 0.1 mA up to 1 mA, in 0.2 mA increments between 1 and 2 mA, and 0.5 mA increments between 2 and 15 mA. The threshold of motor symptom onset (motor symptom threshold) was defined as the lowest stimulus intensity that induced motor symptoms. Cortical ADs were considered focal when the spread was limited to the anterior half of the right hemisphere (Fig. 1B). The AD threshold