EFFECT OF STIMULUS PARAMETERS IN THE TREATMENT OF SEIZURES BY ELECTRICAL STIMULATION IN THE KAINATE ANIMAL MODEL

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Preliminary results from animal and clinical studies demonstrate that electrical stimulation of brain structures can reduce seizure frequency in patients with refractory epilepsy. Since most researchers derive stimulation parameters by trial and error, it is unclear what stimulation frequency, amplitude and duration constitutes a set of optimal stimulation parameters for aborting seizure activity in a given patient. In this investigation, we begin to quantify the independent effects of stimulation parameters on electrographic seizures, such that they could be used to develop an efficient closed-loop prosthesis that intervenes before the clinical onset of a seizure and seizure generalization. Biphasic stimulation is manually delivered to the hippocampus in response to a visually detected electrographic seizure. Such focal, responsive stimulation allows for anti-seizure treatment delivery with improved temporal and spatial specificity over conventional open-loop stimulation paradigms, with the possibility of avoiding tissue damage stemming from excessive exposure to electrical stimulation. We retrospectively examine the effects of stimulation frequency (low, medium and high), pulse-width (low and high) and amplitude (low and high) in seizures recorded from 23 kainic acid treated rats. We also consider the effects of total charge delivered and the rate of charge delivery, and identify stimulation parameter sets that induce after-discharges or more seizures. Among the stimulation parameters evaluated, we note 2 major findings. First, stimulation frequency is a key parameter for inhibiting seizure activity; the anti-seizure effect cannot be attributed to only the charge delivered per phase. Second, an after-discharge curve shows that as the frequency and pulse-width of stimulation increases, smaller pulse amplitudes are capable of eliciting an after-discharge. It is expected that stimulation parameter optimization will lead to devices with enhanced treatment efficacies and reduced side-effect profiles, especially when used in conjunction with seizure prediction or detection algorithms in a closed-loop control application.

Keywords: Epilepsy; responsive focal stimulation; bi-phasic stimulation; kainate animal model.

1. Introduction
Neuromodulation and deep brain electrical stimulation are emerging as promising treatment options for anti-epileptic drug (AED) resistant epileptic patients. Epilepsy, a chronic neurological disorder characterized by recurrent, spontaneous seizures, affects nearly 50 million people worldwide. Although current AEDs are effective in many patients, an estimated 25% of epileptic patients do not experience an anti-seizure benefit outweighing the risks and side effects associated with the drug-based therapies. Approximately 50% of those receiving anticonvulsants may suffer major side effects during the course of therapy. In highly refractory and selective cases of epilepsy, neurosurgeons may also resect brain tissue containing the seizure foci or disconnect the corpus callosum to prevent generalization of ictal activity during seizures (callosotomy). Alternative, less invasive treatments are being tried along the lines of neuromodulation. Evidence from a number of animal and clinical studies has been gathered about electrical stimulation...
holding great promise as a neuromodulation therapeutic mode for epilepsy.3–13

The use of electrical stimulation for treating epilepsy is hindered by a lack of insight into the mechanisms of therapeutic neural stimulation coupled with an incomplete knowledge of ictogenesis.14,15 With varying degrees of success, several studies have examined the effects of continuous and periodic stimulation for controlling seizures.3–13 Based on the findings from these studies, preliminary human studies were instantiated to evaluate the efficacy and safety of stimulation for treating epilepsy. Velasco et al. (2001)10 reported promising results with hippocampal stimulation in uncontrolled human studies. Results from well-controlled studies of the NeuroPace Responsive Neurostimulator System (RNS) and the Cyberonics Vagus Nerve Stimulation (VNS) system also demonstrate that electrical stimulation of the epileptogenic zone and the vagus nerve, respectively, can terminate epileptiform after-discharges in select patient populations. However, only about 37% of the patients implanted with a VNS device experience greater than 50% reduction in seizure frequency. Currently the VNS involves intermittent stimulation of the left cervical vagus nerve, usually for 30 seconds followed by 3–5 minutes of off-time, with a stimulus of 20–30 Hz, 250–500 µsec and an intensity comfortably tolerated by patients (typically between 0.5 and 3 mA).16,17 The intensity is set during the “stimulation adjustment period,” during which the amplitude is progressively increased to the maximum comfortably tolerated level. Additional increases in intensity (i.e., current amplitude) are also made if the patient stops responding to the existing parameters.17

The limited successes of existing stimulation paradigms have provided us with an impetus for seeking optimal stimulation parameters for more effective suppression of seizures. Since most researchers derive stimulation parameters by trial and error,18 it is unclear which stimulation frequencies, amplitudes and durations constitute an optimal stimulation parameter set for aborting seizure activity. In addition, very few studies have analyzed the effects of responsive stimulation on seizure activity, especially in contrast to the effects of continuous or intermittent stimulation using a defined set of parameters.

In this investigation, we seek to delineate the independent effect of various stimulation parameters in aborting seizures in a kainate animal model of temporal lobe epilepsy. We systematically examine the effects of stimulation frequency (low, medium and high), pulse-width (low and high) and pulse amplitude (low and high). The stimulation parameters are empirically chosen based on literature, while respecting safety limits for charge density.25,26 There are two major considerations that must be made when evaluating the effect of stimulation parameters: (1) the subject’s tolerance to the stimulation and (2) the objective electrographic measures of anti-seizure efficacy derived from the hippocampal local-field potential (LFP) recordings. An effective stimulus must not lead to more seizures. Stimulation is delivered manually when a seizure is observed. Such responsive stimulation provides for temporal specificity, and may avoid the possibility of tissue damage caused due to excessive exposure of tissue to stimulation.

Materials and Methods

2. Materials and Methods

2.1. Animal model

All animal care and handling procedures were approved by the Institutional Animal Care and Use Committee (IACUC) and followed principles set forth in the Guide for the Care and Use of Laboratory Animals, a National Institutes of Health publication.21

Recording and stimulating electrodes were bilaterally implanted in the hippocampi of 250–350 g female Long-Evans rats. Electrographic and clinical seizures were then elicited by systemic injection of kainic acid.22 This model has also been previously used to test the effects of continuous stimulation on the subthalamic nucleus (STN), amygdala and cortex.23,24

2.2. Surgery and treatment

Thirty female Long-Evans rats (250–350 g) were anesthetized (Induction: 5% isoflurane in 2 L/min O2; Maintenance: 0.5–3% isoflurane in 2 L/min O2) and bilaterally implanted in the dentate gyrus with
Fig. 1. Stainless steel Plastics One electrode triodes are bilaterally implanted in the hippocampi of anesthetized rats.

stainless steel Plastics One electrode triodes, as shown in Fig. 1 (coordinates: 4.0 mm posterior to bregma, 2.5 mm lateral to midline, and 3.3 mm ventral to dura). Each electrode triode comprises one 125 µm diameter recording and two 125 µm diameter stimulating electrodes. Electrodes were secured to the skull using dental acrylate and threaded to a percutaneous Plastics One connector. One animal was neurologically impaired postoperatively, and six animals died during surgery or lost their head-cap prior to kainic acid treatment. As a result, the data from 23 out of 30 animals are reported in this study.

After a 1–2 week post-surgical recovery period, a kainic acid treatment protocol was used to induce status epilepticus. A kainate solution (2.5 mg/ml kainic acid in 0.9% NaCl) was administered intraperitoneally in repeated, low doses (0.2 ml per 100 g) every hour until each rat experienced convulsive status epilepticus for >3 hours. Seizure activity was carefully monitored and marked according to the Racine scale: R = 0, no motor seizure activity; R = 1, oral facial movements only; R = 2, head noding; R = 3, forelimb clonus; R = 4, rearing; R = 5, rearing and falling.

2.3. Electrochemical measurements

To determine the charge carrying capacity of the stimulating electrodes, cyclic voltammetry (CV) was performed (using an Autolab Potentiostat/Galvanostat PGSTAT12 and a 3 electrode configuration). Measurements were made using a three electrode cell configuration (the stimulating electrode served as a working electrode, a saturated calomel electrode served as a reference electrode and a large surface-area platinum wire served as a counter electrode) with 1x phosphate-buffered saline (PBS) solution serving as the electrolyte (pH 7.4). Autolab was configured to sweep voltages of the electrode at a constant rate within potential limits of hydrolysis. The current flow between the electrode and the auxiliary or the counter electrode was monitored. The potential was swept from −0.6 V to 0.8 V at a sweep rate of 50 mV/sec. The charge storage capacity of the electrode is calculated from the area under the I-E curve and was found to be 200 µC/cm² for the Plastics One electrodes (Fig. 2).

2.4. Recording and stimulating system

Hippocampal local field potentials (LFPs) from kainate-treated rats were amplified, digitized at 2 kHz and band-pass filtered from 1 to 655 Hz using a Pinnacle Technologies setup. For all subjects, LFP-synchronized video recordings were obtained for the treatment duration (Fig. 3).

A custom-built constant-current programmable biphasic stimulator was constructed and used to deliver the stimulation when required. The current amplitude was chosen as the maximum current that could be safely applied without inducing neural damage. Various studies have evaluated the safety criteria for implantable stimulating electrodes. Stuart Cogan showed that macroelectrodes (with surface areas of 100,000 µm² or greater) exhibit high-charge phase thresholds and low-charge density thresholds, while microelectrodes (with surface areas of 10,000 µm² or less) exhibit the opposite behavior.
Robert Shannon\textsuperscript{28} presented a model, based on a large body of data collected by McCreery\textsuperscript{29,30} and others, regarding the safety of electrical stimulation. According to Shannon, the maximum current that can be applied without inducing neural damage is estimated using Eq. (1):

\[ Q = \sqrt{(A \ast 10^6)}, \]

where \( Q \) is the charge per phase in \( \mu \)C, \( A \) is the electrode surface area in \( \text{cm}^2 \), and \( k \) is a constant of 1.5.\textsuperscript{28} Using Eq. (1), the maximum deliverable charge per phase for the Plastics One electrodes is approximately 0.062 \( \mu \)C/ph (\( A \approx 12,000 \mu \text{m}^2 \)). Amplitudes chosen for this study were within these limits for the electrodes.

When a seizure is visually detected, both implanted sites are simultaneously stimulated with a 5-second train of rectangular constant-current bipolar pulses. The stimulation frequency, pulse-width, and amplitude of the stimulus were selected in accordance with the protocol being tested: (a) stimulation frequency—low, medium or high (5, 60 or 130Hz), (b) pulse-width — short or long (60 or 240\( \mu \)sec) and (c) amplitude-low or high (150 or 300\( \mu \)A). These stimulation parameters are empirically chosen based on literature, while respecting safety limits for charge density.\textsuperscript{19,20,28}

This study was conducted in four phases (three phases shown in Table 1). In Phase 1, rats were stimulated using a stimulation frequency of 5 Hz (25 pulses in 5 sec), 60 Hz (300 pulses in 5 sec) or 130 Hz (650 pulses in 5 sec). Each applied pulse had a 60\( \mu \)sec width and a 150\( \mu \)A biphasic current amplitude. The pulse train was delivered for 5 sec upon visual and electrographic determination of a seizure. In Phase 2, the rats were stimulated using the same parameters as Phase 1, but with a 240\( \mu \)sec pulse-width instead of a 60\( \mu \)sec pulse-width. The Phase 2 stimulation protocol delivers 4x more charge than Phase 1 within the
5 sec window. In Phase 3, rats were stimulated using the same parameters as Phase 1, but with a 300 µA instead of a 150 µA biphasic current amplitude. The Phase 3 stimulation protocol delivers 2x more charge than Phase 1 within a 5 sec window. The cumulative charge delivered denotes the sum of the absolute values of charge delivered in the anodic and cathodic phases. Each rat was treated only with a particular set of stimulus parameters. In phase 4, we characterized a strength-duration curve for elicitation of ADs. One week after the KA treatment, five rats were used to determine the maximum stimulus amplitudes that induced an AD. For different stimulation frequencies (5, 60 and 130 Hz), the pulse-widths and amplitudes of the constant-current waveforms were varied (pulse-width: 60, 120, 240 and 480 µsec; amplitude: 30 to 2000 µA) until an AD was observed.

In order to quantify the independent effects of stimulation parameters on seizure activity, electrographic responses, including the time for a seizure to stop after stimulation (sec), the change in mean and absolute amplitudes (%) of LFP data and the change in inter-spike interval (ISI in %) were measured (Fig. 4). While the change in mean amplitude denotes a change in the dc potential of the surroundings, a change in the absolute amplitude value denotes the change in the seizure characteristics.

**Table 1. Study design.**

<table>
<thead>
<tr>
<th>Study phase</th>
<th>Protocol</th>
<th>Freq (Hz)</th>
<th>Amp (µA)</th>
<th>Pulse width (µsec)</th>
<th># Animals</th>
<th>Charge per phase (µC)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Standard (std)</td>
<td>5</td>
<td>150</td>
<td>60</td>
<td>3</td>
<td>0.009</td>
</tr>
<tr>
<td></td>
<td>Med freq-60</td>
<td>60</td>
<td>150</td>
<td>60</td>
<td>3</td>
<td>0.009</td>
</tr>
<tr>
<td></td>
<td>High freq-130</td>
<td>130</td>
<td>150</td>
<td>60</td>
<td>3</td>
<td>0.009</td>
</tr>
<tr>
<td>2</td>
<td>Std, High pw</td>
<td>5</td>
<td>150</td>
<td>240</td>
<td>3</td>
<td>0.036</td>
</tr>
<tr>
<td></td>
<td>High pw-60</td>
<td>60</td>
<td>150</td>
<td>240</td>
<td>3</td>
<td>0.036</td>
</tr>
<tr>
<td></td>
<td>High pw-130</td>
<td>130</td>
<td>150</td>
<td>240</td>
<td>3</td>
<td>0.036</td>
</tr>
<tr>
<td>3</td>
<td>Std, High amp</td>
<td>5</td>
<td>300</td>
<td>60</td>
<td>3</td>
<td>0.018</td>
</tr>
<tr>
<td></td>
<td>High amp-60</td>
<td>60</td>
<td>300</td>
<td>60</td>
<td>1</td>
<td>0.018</td>
</tr>
<tr>
<td></td>
<td>High amp-130</td>
<td>130</td>
<td>300</td>
<td>60</td>
<td>1</td>
<td>0.018</td>
</tr>
</tbody>
</table>

Fig. 4. Electrographic measures (change in amplitude, inter-spike interval and time to stop a seizure) as measures of stimulation efficacy.
2.5. **Statistical analysis**

The amplitudes were expressed in μA, and the ISI and time to stop a seizure in seconds. Since a Gaussian data distribution cannot be assumed, a non-parametric Kruskal-Wallis one-way analysis of variance on ranks test was used to compare the efficacies of the different stimulation protocols. Regression analysis was used to determine significant differences (if any) between stimulation response curves for treated groups in order to quantify the effects of stimulation parameters on therapeutic efficacy.

3. **Results**

In kainate-treated animals, stereotypicprogressions of seizure activity were observed. The earliest evidence of KA-induced convulsant activity was the occurrence of brief 1-sec sharp wave bursts. These
repetitive paroxysms typically began a few minutes after the first administered KA dose. No clinical manifestations of seizures were seen during this time (R = 0). After about 30 minutes, the LFP data gradually included high amplitude spikes or multiple spike-and-wave complexes lasting < 1 sec. On occasions, these bursts were also associated with staring or head jerking (R = 1, 2). After 2 to 3 KA doses, rapid electrographic progression to high amplitude synchronous activity, along with tonic and clonic seizures (R = 3, 4 and 5), was observed.

In each rat, stimulation was not delivered to randomly selected control seizures and delivered to the others upon visual detection of seizure onset (Fig. 4). A linear trend was observed between the time to stimulation and seizure duration, however it was not significant (p = 0.5460).

### 3.1. Experiment 1: Effect of stimulation frequency

Nine KA-treated rats (120 seizures) were stimulated at a low, medium or high stimulation frequency (5, 60 or 130 Hz) for 5 seconds with a 60 µsec pulse-width and a 150µA biphasic current amplitude. All three parameter sets delivered 9 nC of charge per phase, but different cumulative charges at different rates (Table 1). Various response types were observed using the three parameter sets, including depression of activity, no effect on seizure activity and after-discharge elicitation (as shown in Fig. 5).

Further analyses were performed to quantify the change in mean electrographic seizure amplitude and the time taken for seizure activity to stop after applying the differing sets of stimulation parameters. When no stimulus was delivered, the mean seizure duration was 88.71 ± 7.5 sec. Although none of the stimulation protocols completely aborted seizure activity, there was no effect on seizure activity and after-discharge elicitation (as shown in Fig. 5).

<table>
<thead>
<tr>
<th>Stimulation frequency</th>
<th>Charge delivered per phase (µC)</th>
<th>Cumulative charge delivered (µC)</th>
<th>Mean time for seizure to stop (sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5 Hz</td>
<td>0.009</td>
<td>0.22</td>
<td>18.86 ± 16.41</td>
</tr>
<tr>
<td>60 Hz</td>
<td>0.009</td>
<td>2.70</td>
<td>67.46 ± 56.56</td>
</tr>
<tr>
<td>130 Hz</td>
<td>0.009</td>
<td>5.85</td>
<td>40.38 ± 32.82</td>
</tr>
</tbody>
</table>

*Pulse-width = 60 µsec; Pulse amplitude = 150 µA.

Seizures stimulated with a 60 Hz frequency also showed a reduction in spike amplitude (2.65%). The change in spike amplitude and absolute value of amplitude was significantly larger in the 130 Hz frequency group after stimulation (Table 3). The change in spike intervals was not significant between the 3 groups.

In a few R = 4 (i.e., Class IV) seizures stimulated with a 60 Hz frequency, stimulation intensified and extended the duration of the seizures, resulting in R = 5 seizure classifications. The large variability in the time to stop a seizure in the 60 Hz group (Table 2) is indicative of these extended seizures periods.

### 3.2. Experiment 2: Effect of pulse-width

In this experiment, rats were treated with 5 seconds of bilateral hippocampal stimulation using a 5, 60 or 130 Hz stimulation frequencies, a 240 µsec
Table 3. Median responses to different stimulation frequencies.

<table>
<thead>
<tr>
<th>Stimulation frequency</th>
<th>Change in mean amplitude (%)</th>
<th>Change in abs in spike amplitude (%)</th>
<th>Change in inter-spike interval (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5 Hz</td>
<td>−4.94</td>
<td>50.66</td>
<td>60.95</td>
</tr>
<tr>
<td>60 Hz</td>
<td>−2.71</td>
<td>58.54</td>
<td>−2.65</td>
</tr>
<tr>
<td>130 Hz</td>
<td>−5.40</td>
<td>95.29</td>
<td>28.46</td>
</tr>
</tbody>
</table>

*Pulse-width = 60 µsec; Pulse amplitude = 150 µA.

Table 4. Charge delivered per phase, cumulative charge delivered and mean time taken for a seizure to stop after stimulation with differing stimulation frequencies (mean ± std).

<table>
<thead>
<tr>
<th>Stimulation frequency</th>
<th>Charge per phase (µC)</th>
<th>Cumulative charge delivered (µC)</th>
<th>Mean time to stop (sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5 Hz</td>
<td>0.036</td>
<td>0.88</td>
<td>21.92 ± 20.78</td>
</tr>
<tr>
<td>60 Hz</td>
<td>0.036</td>
<td>10.80</td>
<td>48.52 ± 42.40</td>
</tr>
<tr>
<td>130 Hz</td>
<td>0.036</td>
<td>23.41</td>
<td>59.79 ± 36.44</td>
</tr>
</tbody>
</table>

*Pulse-width = 240 µsec; Pulse amplitude = 150 µA.

3.3. Experiment 3: Effect of pulse amplitude

When the stimulation intensity (i.e., pulse amplitude) was doubled to 300 µA, rats stimulated at 60 and 130 Hz frequencies displayed highly abnormal motor movements that were atypical of KA-induced seizure activity. Because of the discomfort caused by these current intensities, they were not further evaluated. At 300 µA intensity and 5 Hz frequency also, many stimulations suppressed the seizure and then an after-discharge (AD) was observed after a short silent period. Comparing the three stimulations delivered at 5 Hz — one with 60 µsec pulse-width, 150 µA; second with 240 µsec pulse-width, 150 µA, and third with 60 µsec pulse-width, 300 µA current, we observe that all treatments were equally effective in reducing the time taken for the seizure to stop (Table 5).

3.4. Experiment 4: Strength-duration curves

From Experiment 3, we found that increasing the stimulation amplitude can invoke abnormal motor manifestations and elicit ADs (kindling effect). As a result, Experiment 4 was designed to examine these thresholds, i.e. the minimum amplitude, at a given stimulation frequency and pulse-width, that would induce an AD. For different stimulation frequencies (5, 60 and 130 Hz), the pulse-widths and amplitudes of the constant-current waveforms were varied...
Table 5. Charge delivered per phase, cumulative charge delivered and mean time taken for a seizure to stop after stimulation with differing stimulation frequencies (mean ± std).

<table>
<thead>
<tr>
<th>Stimulation frequency</th>
<th>Charge per phase (µC)</th>
<th>Cumulative charge delivered (µC)</th>
<th>Mean time for seizure to stop (sec)</th>
</tr>
</thead>
<tbody>
<tr>
<td>No stimulation</td>
<td>—</td>
<td>—</td>
<td>88.71 ± 7.5*</td>
</tr>
<tr>
<td>5 Hz, 60 µsec, 150 µA</td>
<td>0.009</td>
<td>0.23</td>
<td>18.86 ± 16.41</td>
</tr>
<tr>
<td>5 Hz, 240 µsec, 150 µA</td>
<td>0.036</td>
<td>0.9</td>
<td>21.92 ± 20.78</td>
</tr>
<tr>
<td>5 Hz, 60 µsec, 300 µA</td>
<td>0.018</td>
<td>0.45</td>
<td>20.45 ± 17.71</td>
</tr>
</tbody>
</table>

Standard deviations are across rats and seizures with control seizure duration.

*pulse-width: 60, 120, 240 and 480 µsec; amplitude: 30 to 2000 µA) until an AD was observed.

We observed that as the pulse-width is increased, the amplitude of current required to induce an AD is decreased (Fig. 8). Also, with the 5 Hz stimulation frequency, much higher amplitude currents could be used to stimulate without causing an AD, when compared to the 60 or 130 Hz stimulation.

Strength-duration curves were produced to summarize data from Experiment 4. They are characterized by a rheobase current (the lowest current intensity with an indefinite pulse duration which just stimulated muscles or nerves, and in our application an intensity capable of invoking an AD if applied with an infinite pulse-width) and chronaxie time (the pulse-width at which the current intensity is twice that of the rheobase current). Using the rheobase current (I_r) and chronaxie time (t_c), Eq. (2) was used to construct strength-duration curves for eliciting ADs:

\[ I = I_r (1 + t_c/t), \]  \tag{2}

where \( I \) is the threshold current at pulse-width, \( I_r \) is the rheobase current and \( t_c \) is the chronaxie time. An inverse power relation is observed between the maximum stimulation amplitude and the stimulation frequency, and between the maximum stimulation amplitude and the pulse-width duration. When constructed in this manner, the AD strength-duration curve gives a maximum current amplitude that can be applied at a given frequency and pulse-width so that it does not elicit an after-discharge, thereby providing investigators with a set of guidelines for choosing safe stimulation parameters.

4. Discussion

The purpose of this research is to evaluate the therapeutic and rehabilitative effects of responsive neurostimulation at the beginning of seizures. The results presented within this report serves as a first step for developing effective neurostimulation protocols for use in a closed-loop epilepsy prosthesis. In this paper, we begin to quantify the independent effects of biphasic, constant-current stimulation parameters on seizures in KA-treated rats (stimulation frequency, pulse amplitude and pulse-width) to determine which individual or combination of parameters are most effective at controlling seizures.
The most important findings are as follows: (a) stimulation frequency is a key parameter for inhibiting seizure activity, while charge delivered per phase has less of a pronounced effect, and (b) an after-discharge curve shows that as the pulse-width increases, small current amplitudes can elicit an AD. This is an important by-product of the methodology for establishing a maximum stimulation threshold.

Our data indicate that although none of the applied stimulation parameter sets completely abort seizure activity, the time taken for a seizure to stop was significantly lower in the 5 Hz stimulation groups compared to 60 or 130 Hz stimulation groups. Similar observations have been made by Chkhenkeli et al., who found that short-duration, high frequency stimulation of the caudate nucleus produces an enhancement of the epileptiform state, while low frequency stimulation reduces the frequency of sharp transient interictal spikes and truncates epileptic discharges. This observation is in agreement with the after-discharge curve produced in Experiment 4. The AD curve shows that as the stimulation frequency increases, the current magnitude required for eliciting an AD decreases (that is, smaller amplitudes can elicit ADs and produce an opposite effect of seizure control).

The exact nature of the anti-convulsant action of electrical stimulation is not fully understood. Generally, accepted theories center around the following hypotheses: (1) preferential release of inhibitory neurotransmitters due to stimulation and/or (2) depolarization block, in which the stimulated neurons are inactivated because of stimulus-induced membrane hyperpolarization. It has also been suggested that stimulation with a high frequency has a lesion-like effect (depolarization block), while low-frequency stimulation induces long-term depression (LTD) of stimulated neurons in the peri-electrode space. Some limitations of our study include results associated with only a particular model of epilepsy, the manual stimulation at the time of visual detection of a seizure onset, and lack of histology to access electrode position and damage to tissue with various stimulation parameters. Future work involves testing the stimulation model in a closed-loop setting with non-lesional refractory motor epilepsy, the stimulation model in a closed-loop setting with non-lesional refractory motor epilepsy, and responsive stimulation that would address the limitations of the current investigation. Such responsive stimulation would suppress seizures by delivering charge automatically in response to electrographic activity. The possibility of damage due to excessive exposure of tissue to stimulation might then also be reduced. Besides, in some patients, it is also observed that continuous stimulation could lead to an increased neuronal threshold for effective treatment by stimulation, that is, it increases the necessary amount of delivered charge to achieve the same response, which in turn may also increase the possibility of neuronal damage.

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