Changes of somatomotor and parietal regions produced by different amounts of electrical stimulation

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ABSTRACT

Our study aims to investigate changes in electrocortical activity by observing the variations in absolute theta power in the primary somatomotor and parietal regions of the brain under three different electrical stimulation conditions: control group (without stimulation), group 24 (24 trials of stimulation) and group 36 (36 trials of stimulation). Thus, our hypothesis is that the application of different patterns of electrical stimulation will promote different states of habituation in these regions. The sample was composed of 24 healthy (absence of mental and physical impairments) students (14 male and 10 female), with ages varying from 25 to 40 years old (32.5 ± 7.5), who are right-handed (Edinburgh Inventory). The subjects were randomly distributed into three groups: control ($n = 8$), G24 ($n = 8$) and G36 ($n = 8$). We use the Functional electrical stimulation (FES) equipment (*NeuroCompact*-2462) to stimulate the right index finger extensor muscle, while the electroencephalographic signal was simultaneously recorded. We found an interaction between condition and block factors for the C3 and P3 electrode, a condition and block main effects for the C4 electrode, and a condition main effect for the P4 electrode. Our results support the hypothesis that electrical stimulation promotes neurophysiological changes. It appears that stimulus adaptation (accommodation) of specific circuits can strengthen the brain's ability to distinguish between and respond to such stimuli over time.

Functional electrical stimulation (FES) is a method that involves neuromuscular electrical stimulation that produces changes in functional activity [1,33]. The activity triggered in brain function dynamics due to electrical stimulation is not well covered in the current literature. Thus, understanding the learning process and the changes caused in the brain after electrical stimulation is essential for developing strategies to comprehend sensorimotor integration [6,29]. Through quantitative electroencephalography (qEEG), it is possible to detect changes in the brain caused by sensory, cognitive or motor stimuli [33,10,17]. Previous studies have investigated the relationship between electrical stimulation and electroencephalographic activation [33,25]. These authors observed that FES induces

transient changes, which can be detected by qEEG. The relationship between FES and qEEG has been observed in beta and alpha bands, after the application of electrical stimulation to the primary motor cortex [25,20,24].

In the current literature on studies attempting to understand the relationship between qEEG and electrical stimulation, theta band has not been observed and investigated. Theta band (4–7 Hz) is related to functions such as encoding and the retrieval of information [5] and information processing [6,16]. Although theta has been associated with information processing, no study has observed the function of theta band in purely sensorial task. In this context, our study aims to investigate changes in electrocortical activity by observing the variations in absolute theta power in the primary somatomotor and parietal regions of the brain under three different electrical stimulation conditions. Our hypothesis is that the application of different amounts of electrical stimulation quantity will provoke different absolute theta power values in the primary somatomotor and parietal regions.

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The sample was composed of 24 students (14 male and 10 female), with ages varying from 25 to 40 years old (32.5 ± 7.5), who are right-handed [22]. Subjects were screened for mental or physical impairments using previous anamnesis and clinical examinations. Only subjects who did not demonstrate these traits were chosen. They were also screened for psychoactive or psychotropic substances. All subjects signed a consent form and were aware of the experimental protocol. The individuals were not paid for participating in the study. The experiment was approved by the Ethics Committee of the Federal University of Rio de Janeiro (IPUB/UFRJ).

The subjects were randomly distributed into three groups: control ($n = 8$), G24 ($n = 8$) and G36 ($n = 8$). Subjects were seated in a comfortably sound and light-attenuated room during the task. They sat in a chair, and a table was used for arm support in order to reduce muscle artifacts. The participants were blindfolded to reduce potential visual stimuli and blinking. We used an eight channel microcomputer-controlled stimulator (Ibramed, Neuro Compact-2642), with a biphasic (fixed pulse width of 320 μs for each phase), which provided constant-current pulses to the muscle of the participants' hands. The frequency of the stimulation pulses was set in 48.8 Hz to achieve a sufficiently smooth and strong contraction of the muscles without extensive fatigue. The current amplitude was set at 2×10^{-3} A. The device provided a constant current and was used to stimulate the extension of the right index finger. The hand was secured to the table and a velcro strip was used to immobilize all other fingers, leaving only the index finger free for stimulation. The resistance of the skin was measured by a multimeter (ohmmeter) and ranged from 800 Ω (ohms) to 1500 Ω. The skin was shaved and cleaned with alcohol. The electrodes were set up at 5 cm from the lateral epicondylus on the lateral forearm side, and the other electrode was placed 12 cm from the first one, occupying the posterior forearm side, following the index finger extensor tendon's trajectory.

The experiment consisted of trials and blocks. Each trial was composed of a stimulation moment (i.e., time on) of 4.86 s of current, plus a resting moment (time off) consisting of 8.39 s without a current. Each block was composed of six trials. The control group simulated four blocks (i.e., 24 trials) with 1-min periods between each block without electrostimulation been applied. The current intensity for this group was zero. The G24 group was exposed to four blocks (i.e., 24 trials) of electrostimulation with 1-min intervals between each block, under the conditions described previously. Only the G36 group was exposed to six blocks (i.e., 36 trials) of electrostimulation with 1-min intervals between each block under the same conditions of G24. 5.693 pulses lasting 116.64 s were applied to G24. 8.539 pulses lasting 174.96 s were delivered to G36. The control group only simulated the electrostimulation procedures as described above. Simultaneously with the electrostimulation of the finger extensor muscle, electroencephalographic signals were recorded.

The International 10/20 System for electrodes [15] was used with the 20-channel EEG system Braintech-3000 (EMSAMedical Instruments, Brazil). The 20 electrodes were arranged in a nylon cap (ElectroCap Inc., Fairfax, VA, USA) yielding monopolar derivations using the earlobes reference. In addition, two 9 mm diameter electrodes were attached above and on the external corner of the right eye, in a bipolar electrode montage, to monitor eye-movement (EOG) artifacts. Impedance of EEG and EOG electrodes was kept between 5 and 10 kΩ. The data recorded had a total amplitude of less than 100 μV. The EEG signal was amplified with a gain of 22,000, analogically filtered between 0.01 Hz (high-pass) and 100 Hz (low-pass), and sampled at 240 Hz. The software *Data Acquisition* (Delphi 5.0) at the Brain Mapping and Sensory Motor Integration Lab, was employed with the following digital filters: notch (60 Hz).

To quantify reference-free data, a visual inspection and independent component analysis (ICA) were applied to remove possible sources of artifacts produced by the task. A classic estimator was applied for the power spectral density (PSD), or directly from the square modulus of the FT (Fourier Transform), which was performed by MATLAB 5.3 (Matworks, Inc.). The number of samples was 800 (4 s × 200 Hz) with rectangular windowing. Quantitative EEG parameters were extracted from 8-s periods time-locked with movement-offset or stimulation (the selected epoch started 4 s before and ended 4 s after the trigger, i.e., moment 1 and moment 2, respectively). Thereafter, all raw EEG trials were visually controlled and trials contaminated with ocular or muscle artifacts were discarded. The Fourier Transform resolution was 1/4 s to 0.25 Hz (FFT). To examine a stationary process, the "Run-test" and "Reverse-Arrangement test" were applied. Specially, the stationary process was accepted for each 4 s (epoch's duration in this period). In this manner, based on artifact-free EEG epochs, the threshold was defined by mean plus three standard deviations and epochs with a total power higher than this threshold were not integrated into the analysis.

To evaluate changes in theta power values we analyzed different electrodes: C3, C4, P3 and P4. An ANOVA two-way (repeated measures) and a Bonferroni's post hoc test were used to analyze the factors group (i.e., CG, G24 and G36) and block (i.e., first and last block) for each electrode. We use a t-test and an ANOVA one-way (repeated measures) to verify differences within factors, when we observed interaction.

In the first analysis we analyzed the C3 electrode. An interaction between condition and block factors ($p = 0.014$, $F = 4.302$) was demonstrated. To investigate the interaction we performed an ANOVA one-way among conditions (i.e., CG, G24 and G36) for each block (i.e., first and last block). There was verified a condition main effect for the first block ($p < 0.001$, $F = 88.243$) and a condition main effect for the last block ($p < 0.001$, $F = 58.474$). We found a significant difference between GC and G24, and between GC and G36 for the first ($p < 0.001$) and last ($p < 0.001$) blocks, as observed by Bon-
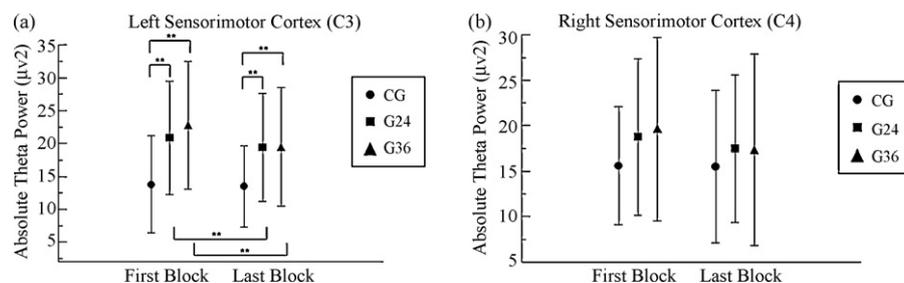
Fig. 1. Mean and standard absolute theta power between the first and last block of electrical stimulation in three experimental conditions (i.e., GC, G24 and G36) in the primary sensorimotor cortex. (a) The findings showed an interaction between condition and block factors for C3 ($p = 0.014$). (b) In the C4 electrode demonstrated a condition ($p < 0.001$) and block ($p = 0.007$) main effect.
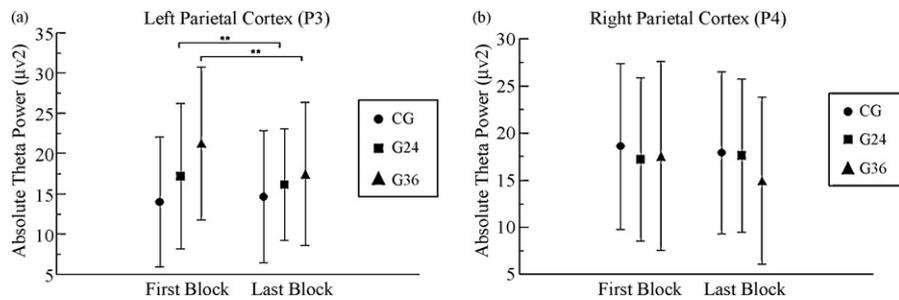
Fig. 2. Mean and standard absolute theta power between the first and last block of electrical stimulation in three experimental conditions (i.e., GC, G24 and G36) in the parietal cortex. (a) The results demonstrated an interaction between condition and block factors for P3 ($p < 0.001$). (b) In the P4 electrode demonstrated a condition main effect ($p = 0.005$).

ferroni's test. In relation to block analysis, the t-test demonstrated a significant difference between the blocks of G24 and G36 conditions ($p = 0.001$) (Fig. 1a). In the second ANOVA two-way, we observed the C4 electrode and a condition ($p < 0.001$, $F = 20.384$) and block ($p = 0.007$, $F = 7.280$) main effect was presented. A significant difference between CG and G36 ($p < 0.001$), and between CG and G24 ($p < 0.001$) was verified, as observed by Bonferroni's test (Fig. 1b). In the electrode P3 we verified an interaction between condition and block factors ($p < 0.001$, $F = 7.817$). An ANOVA one-way among conditions, for each block, was applied to examine the interaction. We found a significant difference among the three conditions (i.e., CG, G24 and G36) for the first block ($p < 0.001$, $F = 43.248$). In the last block we perceived a significant difference between CG and G36 ($p = 0.001$, $F = 6.839$). In relation to block analysis, the t-test demonstrated a significant difference between blocks for G24 ($p = 0.001$) and G36 ($p = 0.005$) (Fig. 2a). In the last analysis the P4 electrode was analyzed and it was demonstrated a condition main effect ($p = 0.005$, $F = 5.242$). The Bonferroni's test observed a significant difference between CG and G36 as observed by ($p = 0.009$) (Fig. 2b).

Our study investigates changes in electrocortical activity through absolute theta power in the primary somatomotor and parietal brain regions using three different levels of electrical stimulation conditions. Our hypothesis is that the application of different durations of electrical stimulation will lead to different absolute theta power values in these brain areas. The findings demonstrated an interaction between the condition and block factors for the C3 electrode and P3 electrode. Moreover, we found a condition and block main effect for the C4 site and a condition main effect for the P4 electrode.

In this experiment, the subjects were submitted to index finger stimulation. The index finger is primarily represented in the primary somatomotor cortex [2,28]. Therefore, we analyzed the C3 and C4 electrodes in order to detect sensorimotor modifications. Observing the C3 electrode (i.e., left hemisphere, contralateral to the stimulation site), an interaction between the condition and block factors was perceived. The interaction demonstrated that power variation is related to the combination of both independent variables (condition versus block). Examination of the interaction showed a significant difference among GC and the other experimental conditions in the first and last blocks. Otherwise, no significant difference was observed between G24 and G36. We basically observed a power increase in G36 and G24 when compared with GC. We interpreted that this activity increase represents the application of electrical stimulation. However, we did not observe an influence of the quantity of blocks of electrical stimulation in the absolute theta power. Additionally, we inspected the first and last blocks within condition G24 and G36 separately. We saw a difference between the first and the last blocks for both stimulation conditions (i.e., G24 and G36). In particular, we verified a decrease in absolute theta power in the last block when compared with the first block in G24 and G36. The activity reduction between

blocks is interpreted as a result of stimulus repetition, i.e., cortical habituation [28,29].

This region is associated with the control and learning of new motor skills [6,29] and stands out due to its important role in neuroplasticity [29]. Our findings showed an absolute theta power decrease in the last block for both conditions of electrical stimulation (i.e., G24 and G36) when compared to CG. The condition G36, which received a greater quantity of stimulation, demonstrated the highest decrease of absolute theta power. This decrease of power suggests an activity reduction, representing a habituation process due to the amount of stimulus received. In other words, the repetition of stimulation produces an attenuation of the cortical area involved [34]. Velasques et al. [33] demonstrated that the electrical stimulation in the somatomotor cortex promoted cortical adaptations that are similar to those observed when someone learns a procedural task. A minor activity in this area represents an adjustment of neural networks and a consolidation of the information learned [3,31]. Erbil and Ungan [8], observed a suppression of cortical activity, a secondary adaptation to a repetitive motor task, when subjects performed a continuous and repetitive finger flexion–extension. The researcher hypothesizing that habituation is a feature of sensory phenomena and the same modification occurs after the formation of proprioceptive afferences. We conclude that the left primary somatomotor cortex has an important role in the representation of sensory stimuli and we suggest that oscillations in absolute theta power are related to accommodation of this cortical area.

Regarding the right primary somatomotor cortex (C4 electrode), our results showed a main effect for condition and block factors. We observed a decrease of the absolute theta power in the G36 when compared to CG and G24 in the condition main effect. Furthermore, the block main effect demonstrated a power decrease between the first and last blocks. These findings may be seen as an accommodation of the right somatomotor cortex due to the longer stimulus subjects received in the last block. In other words, it can be understood as a reorganization of cortical networks based on the regular stimulation of this region. Right hemisphere information processing is driven by the external environment, and includes functions such as spatial-limb perception and limb position [28,8,12]. Previous studies verified a decrease in alpha activity in the somatomotor area during repetitive voluntary finger movements and a decrease in beta activity in the somatomotor cortex after somatosensory finger stimulation [24,27,30]. Our results support the hypothesis that electrical stimulation promotes cortical changes and we verify that during a task involving passive stimulation the results are similar to studies that observe voluntary movements. Such changes are analogous to what occurs during task execution, and the repetition of voluntary movements, which are situations correlated with the learning process [3,31]. Thus, the power decrease in the C4 electrode represents a decline of sensorimotor activation due to the learning process.

The third analysis demonstrated an interaction between condition and block in the P3 electrode. The interaction showed that the condition and moment, individually, cannot explain the absolute theta power changes in the left parietal cortex. We verified a power increase among the groups and a power decrease between the blocks. A closer inspection of the interaction indicated that conditions (i.e., CG, G24 and G36) are different in the first block. However, in the last block we only observed a difference between GC and G36. In a detailed analysis, we investigated the two blocks within each condition. A difference between the first and last blocks was found in the G24 and G36. These results demonstrated the highest power in the G36 condition followed by G24 and GC in the first block. In addition in the last block we saw that the amount of electrical stimulation did not change the absolute theta power. We interpreted that the amount of blocks of electrical stimulation did not interfere in the sensorial representation. In other words, the first time that the subjects were exposed to the stimulus generated different cortical representation in the parietal cortex. However, after a period of stimulation we perceived that the quantity of blocks stimulation did not influenced the cortical representation of the stimulus, since G24 and G36 were not different in the last block. According to this, in the G36 we perceived that a previous stimulation influences the response of a later stimulation.

The left parietal cortex is responsible for retaining the representation of sensorial information and it is an area related to the priming memory [32,7]. Priming is a type of implicit memory related to the first representation of stimulus in the brain [32,18]. More explicitly, the presentation of a stimulus activates parts particular representation or associations in memory just before carrying out an action or task [18]. Findings from studies of memory function showed that at the beginning of task execution theta power increases in the parietal regions [5,21]. Similar results are also observed in the theta band during information retention and synaptic plasticity processes [13,23]. These findings suggest that an increase in the amount of electrical stimulation may change cortical region activity, facilitating the consolidation of mental representation, contributing to the formation of an implicit memory.

Our results found an absolute theta power decrease between blocks in G24 and G36 and we observed a power increase in the G36 condition. Gevins et al. [10], suggest that an increase in the amount of blocks leads to an improvement of perceptual representation. Furthermore, an experiment found that oscillations between 5 Hz and 6 Hz reflect central retention of information related to sensory and motor stimuli [14]. Previous studies have shown an increase of theta activity in parietal areas during tasks involving working memory, after 500 ms of stimulus presentation [5,19,17]. According to these previous studies, we interpreted our findings as a reinforcement of the representation of sensorial information in this specific brain site provided by finger stimulation. Thus, our results provided evidence for a re-structure in the left parietal cortex network, produced by different amounts of electrical stimulation, which improves the integration and the representation of sensorial stimulus.

The final analysis demonstrated a main condition effect in the P4 electrode. We verified a power decrease when comparing CG with G36 as well as another significant decrease when comparing G24 with G36. The relationship between the decrease of activity in this area and the electrical stimulation can be explained by an inhibition of this region. EEG and magnetoencephalographic studies demonstrate that a somatosensory stimulus induces responses in the ipsilateral parietal cortex, as observed in our results [9,11]. In addition, recent experiments have suggested that sensorimotor events alter the neural activity of the parietal area facilitating the integration of exteroceptive and proprioceptive information [12,26,4]. Other studies identified that cortical activity is also reduced when sensory information is sent from the external environment to the cortex, including information involving repetitive hand and foot movements [4,35]. According to this, in our experiment the subjects undertook a task involving the right index finger. For this reason, the contralateral hemisphere demands that more energy be involved. This explanation may clarify the inhibition of the right parietal region. Likewise, our results demonstrated that the subjects' exposure to different amounts of stimuli lead to a cortical adaptation (habituation). We suppose that the inhibition of the right parietal cortex represents a demand of a higher activation in the contralateral area. We suggest that this paradigm must be implemented in different populations other than healthy young adults, such as healthy elderly and patients suffering from neuromuscular diseases, to elucidate the role of these areas in sensorimotor integration and to understand the changes in cortical activity produced by sensorial stimuli.

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