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Cathodal transcutaneous spinal direct current stimulation (tsDCS) improves motor unit recruitment in healthy subjects

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HIGHLIGHTS

- We studied the effects of spinal direct current stimulation on motor unit recruitment.
- Cathodal tsDCS improves motor unit recruitment and shortens peripheral silent period.
- Anodal and sham tsDCS have no significant effect.
- tsDCS could provide a novel therapeutic tool for several diseases characterized by abnormal motor unit recruitment.
- tsDCS could also modulate supraspinal activities in a polarity-specific manner.

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ABSTRACT

Transcutaneous spinal direct current stimulation (tsDCS) is a new promising technique for modulating spinal cord function in humans. However, its effects on corticospinal pathways and lower motorneuron excitability are poorly understood. We studied the effects of tsDCS on motor unit recruitment by evaluating changes in motor unit number (MUNE) and peripheral silent period (PSP) after sham (s-tsDCS), anodal (a-tsDCS) and cathodal (c-tsDCS) tsDCS applied either over the cervical or the lower thoracic spinal cord in healthy subjects. For the calculation of MUNE we used the multipoint incremental technique recording from either the ulnar nerve innervated abductor digiti minimi (ADM) or the median nerve innervated abductor pollicis brevis (APB) muscle. c-tsDCS dramatically increases MUNE values following cervical polarization, while sham and anodal polarization have no significant effect (APB: $F_{(4,99)} = 26.4$, $p < 0.001$, two-way repeated measures ANOVA with "time" and "stimulation" as factors; ADM: $F_{(4,99)} = 22.1$, $p < 0.0001$). At the same time, c-tsDCS dampened PSP respect to sham and anodal conditions ($p < 0.0001$). Interestingly, also thoracic c-tsDCS significantly improved motor unit recruitment compared with both s-tsDCS and a-tsDCS (APB: $F_{(4,99)} = 20.1$, $p < 0.0001$; ADM: $F_{(4,99)} = 16.6$, $p < 0.0001$). Our data in healthy subjects suggest that tsDCS, possibly also through supraspinal effects, could provide a novel therapeutic tool in managing several pathological conditions characterized by reduced motor unit recruitment, such as stroke and spinal cord injuries.

1. Introduction

Transcutaneous spinal direct current stimulation (tsDCS) [11,13] is a noninvasive technique for modulating spinal cord function in humans [22] and animals [3,4,6]. DC stimulation intensity ranges from 1.5 to 2.5 mA, with effects lasting for minutes [12,23,26,39] and is well tolerated by subjects. After the first reports [12,13], this technique has come into increasingly

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widespread use [21,22]. Previous studies showed that thoracic anodal tsDCS depresses the cervico-medullary component (P30) of somatosensory potentials evoked by stimulation of posterior tibial nerve (PTN-SEP) and modulates post-activation H-reflex dynamics [23,39]. tsDCS also modulates the flexion reflex in the human lower limb [13] and is able to increase pain tolerance in healthy subjects [35]. Different from transcranial direct current stimulation (tDCS), anodal tsDCS has probably an overall inhibitory effect on spinal cord activity [12,13,26,35]; by analogy with the effects of direct currents on peripheral nerves, it has been hypostasized that anodal tsDCS leads to a hyperpolarizing "anodal block" [10]. Conversely, there is an extensive debate whether cathodal tsDCS has or not a significant and polarity-specific effect on segmental activity [26].

Here, we assessed possible changes in motor unit recruitment following application of tsDCS on spinal cord. That could be of particular interest in rehabilitation strategies of several neurological disorders characterized by abnormal motor unit recruitment or spinal cord dysfunction [21]. To this end, we evaluated changes in motor unit number score (MUNE) and peripheral silent period (PSP) before and at different time points following direct polarization of either cervical or thoracic spinal cord in a sample of healthy subjects.

2. Methods

2.1. Transcutaneous spinal DC stimulation (tsDCS)

With participants lying supine on a comfortable couch, tsDCS (2.5 mA, 20 min) was delivered by a constant current programmable electrical stimulator (HDCStim^{TM}, Newronika®, Italy) connected to a pair of rectangular electrodes, one on the cervical spinal cord, between the 6th cervical and the first thoracic vertebra with the major axis parallel to spinal cord, and the other above the right shoulder (cervical tsDCS); in a second experiment, to address the hypothesis of supraspinal effects of current polarization, we placed the active electrode over the spinous process of the tenth thoracic vertebra, from 9th to 11th vertebra. tsDCS electrodes were thick (6 mm) rectangular pieces of saline-soaked synthetic sponge (7×5 cm, 35 cm^2). We applied current at a density of 0.071 mA/cm^2 and delivered a total charge density of 85.7 mC/cm^2 below the threshold values for tissue damage [25,27].

2.2. Motor unit number estimation (MUNE)

Changes in motor unit function and recruitment were assessed and quantified by using Motor Unit Number Estimation (MUNE; [14,15]). Particularly, we adopted the multipoint incremental method, first proposed by Wang [37,38] and recently modified by Shefner and colleagues [34]; this technique is reproducible, easily to perform and highly sensitive in predicting motor unit changes over time. Besides, multipoint stimulation is the most commonly used method of incremental stimulation [15] and its results well correlate with those obtained using other MUNE techniques, such as the *F*-wave method [36,37].

As described by Shefner [34], recording electrodes were placed on either the ulnar nerve innervated abductor digiti minimi (ADM) or the median nerve innervated abductor pollicis brevis (APB) muscle, using the standard belly-tendon montage. There were three stimulus locations used for each muscle; for the median nerve, stimulus points were 2 cm proximal to the wrist crease, 4 cm proximal to the first stimulation site and in the antecubital fossa. For the ulnar nerve, locations were 2 cm proximal to the wrist crease, 4 cm proximal to the first site and 1 cm proximal to the ulnar groove at the elbow. For the distal location we recorded the maximal compound muscle action potential ($CMAP_{max}$). For all the stimulation points we recorded three different traces, the first one with an amplitude of at least 25 μV, then increasing until a clearly defined incremental response (of more than 25 μV incremental amplitude) was obtained at each step. For the calculation of MUNE, the amplitude of the third response at each site was summed, then divided by 9 to yield the average single motor unit action potential (SMUP) amplitude. This amplitude was divided into the $CMAP_{max}$ amplitude to yield the MUNE. Recording only three clearly identifiable surface-recorded motor unit action potentials at each point is shown to avoid the alternation phenomenon [37], a critical limitation of a number of others MUNE techniques.

2.3. Peripheral silent period (PSP)

In a sub-group of six volunteers, spinal motorneuron excitability was assessed by the duration of the so-called peripheral silent period (PSP) using bipolar supramaximal electrical stimulation (constant current square wave pulses of 0.2 ms duration) of both the ulnar and median nerve at the wrist. For each subject, 10 trials were recorded at approximately 20% of maximum voluntary contraction and at twice the intensity to evoke the maximal M response [30].

2.4. Subjects and experimental procedure

Twelve healthy volunteers (six women and six men, mean age ± SD: 25.8 ± 5.9 years) were enrolled in the study, which was previously approved by the institutional review board and conducted in accordance with the declaration of Helsinki.

Subjects were studied before and after anodal, cathodal and sham tsDCS (a-tsDCS, c-tsDCS, s-tsDCS). They all underwent to both cervical and thoracic current polarization. Different polarities were tested in random order and at least 1 week elapsed among different sessions. The subjects were blinded about tsDCS polarity.

MUNE and PSP values were recorded before tsDCS (T_0), immediately after tsDCS offset (T_1), and at 60 min after tsDCS offset (T_2). For all the electrophysiological recordings we chose the left side to avoid interference from the reference placed over the contralateral shoulder.

2.5. Data analysis

Each MUNE and PSP value is expressed throughout the text as percentage changes from baseline. tsDCS-induced changes in each variable were tested with a two-way repeated measure analysis of variance (ANOVA) (STATISTICA 5.5, StatSoft Inc.) with main factors "stimulation", three levels (anodal, cathodal and sham), and "time", three levels (T_0, T_1 and T_2), followed by Holm–Sidak post-hoc method. A one-way independent measures ANOVA with factor "stimulation" (three levels: anodal, cathodal and sham) was run to compare data for anodal, cathodal and sham tsDCS at each time point.

The p values ≤ 0.05 were considered statistically significant.

3. Results

Fig. 1A shows traces recorded from ADM in the same subject at T_1 after sham, anodal and cathodal tsDCS. Baseline MUNE values are in line with those reported by Shefner, ranging from 221 ± 74 to 214 ± 54 for median and ulnar nerves, respectively, and did not differed among experimental sessions ($p > 0.1$ for all the comparisons). *M*-wave amplitude did not changed from baseline ($p > 0.1$, one-way ANOVA on simple data). PSP duration at baseline