Effect of anodal high-definition transcranial direct current stimulation on the pain sensitivity in a healthy population

a double-blind, sham-controlled study

Kold, Sebastian; Graven-Nielsen, Thomas

Published in:
Pain

DOI (link to publication from Publisher):
10.1097/j.pain.0000000000002187

Publication date:
2021

Document Version
Accepted author manuscript, peer reviewed version

PAIN Publish Ahead of Print
DOI: 10.1097/j.pain.0000000000002187

The effect of anodal high definition transcranial direct current stimulation on the pain sensitivity in a healthy population: a double-blind, sham-controlled study

Sebastian Kold, Thomas Graven-Nielsen[*,1]

[1]Center for Neuroplasticity and Pain (CNAP), Aalborg University, Denmark

[*]Corresponding Author: Prof. Thomas Graven-Nielsen, PhD, DMSc. Center for Neuroplasticity and Pain (CNAP), Department of Health Science and Technology, Faculty of Medicine, Aalborg University, Fredrik Bajers Vej 7 D3, DK-9220 Aalborg, Denmark, Phone: +45 9940 9832, Fax: +45 9815 4008, E-mail: tgn@hst.aau.dk

ABSTRACT

High-definition transcranial direct current stimulation (HD-tDCS) of brain areas related to pain processing may provide analgesic effects evident in the sensory detection and pain thresholds. The somatosensory sensitivity was assessed following HD-tDCS targeting the primary motor cortex (M1) and/or the dorsolateral prefrontal cortex (DLPFC). Eighty-one (40 females) subjects were randomly assigned to one of four anodal HD-tDCS protocols (20 min) applied on three consecutive days: Sham-tDCS, DLPFC-tDCS, M1-tDCS, and DLPFC&M1-tDCS (simultaneous tDCS of DLPFC and M1). Subjects and experimenter were blinded to the tDCS protocols. The somatosensory sensitivity were assessed each day, before and after each tDCS by detection and pain thresholds to thermal and mechanical skin stimulation, vibration detection thresholds, and pressure

pain thresholds. Subjects were effectively blinded to the protocol, with no significant difference in rates of whether they received real or placebo tDCS between the four groups. Compared with the Sham-tDCS, none of the active HD-tDCS protocols caused significant changes in detection or pain thresholds. Independent of tDCS protocols, pain and detection thresholds except vibration detection were increased immediately after the first tDCS protocol compared with baseline ($P < 0.05$). Overall, the active stimulation protocols were not able to induce significant modulation of the somatosensory thresholds in this healthy population compared to sham-tDCS. Unrelated to the HD-tDCS protocol a decreased sensitivity was found after the first intervention, indicating a placebo effect or possibly habituation to the QST assessments. These findings add to the increasing literature of null-findings in the modulatory effects of HD-tDCS on the healthy somatosensory system.

Keywords: quantitative sensory testing; High definition transcranial direct current stimulation; Non-invasive brain stimulation

INTRODUCTION

Transcranial direct current stimulation (tDCS) is a non-invasive brain stimulation method where recent advances have increased the spatial resolution by using smaller electrodes in e.g. a ring configuration, instead of the larger pad-electrodes[58]. This high definition (HD) tDCS have shown advantages over the traditional methodology in terms of accuracy of the current delivery and effect duration[27, 32, 58].

The physiological action of tDCS is a subthreshold shift of the resting membrane potential, towards hyperpolarisation or depolarisation. Anodal stimulation increases the excitability, and cathodal stimulation inhibits the excitability of the underlying cortical target[30, 34]. The

excitability change induced by tDCS is reflected in the responsiveness to synaptic inputs and spontaneous neuronal firing rate. This has been demonstrated by modifications in the somatosensory and corticomotor excitability[32, 56], outlasting the tDCS time when using adequate stimulation protocols[33]. The excitability changes have been demonstrated to entail behavioural effects, including changes in working memory and semantic processing[8, 27]. Moreover, HD-tDCS has shown clinical potential providing analgesia in e.g. fibromyalgia, migraine and musculoskeletal pain conditions, although it is still unclear how excitability changes in select cortical areas, can induce an analgesic effect[2, 13, 52]. In other conditions such as stroke and depression, tDCS compared with sham tDCS have shown functional improvements with facilitation of recovery of function after stroke and improved depression and mood questionnaire scores[17, 43].

The use of tDCS in pain conditions includes anodal/cathodal stimulation of the somatosensory cortex, although showing modest efficacy in modulating the pain perception in healthy adults[21, 57]. Anodal stimulation of the dorsolateral prefrontal cortex (DLPFC), have been investigated in pain patients with moderate effectiveness[11, 19, 46]. The most commonly used stimulation protocol is 20-min anodal stimulation of the primary motor cortex (M1), with the cathode located contralaterally over the supraorbital prefrontal cortex[33]. This configuration received the highest recommendations to relieve pain in the evidence-based guidelines[34]. A single tDCS period may be insufficient to produce changes in cortical excitability[33] and tDCS possibly need to be administered several consecutive days to provoke neuroplastic changes[34]. Moreover, multiple tDCS sessions could have additive effects, on both the duration and the efficacy[22].

These uncertainties pertaining to the tDCS methodology have requested more studies investigating the different tDCS configurations with larger sample size and a rigorous methodology

[30, 35, 39, 45, 61]. This is expected to provide better understanding of the mechanisms involved in the neuromodulation, and strengthen the clinical potential of the technology [30].

Investigating the efficacy of interventions targeting the somatosensory system is commonly done by quantitative sensory testing (QST) to compare the pain and somatosensory sensitivity before and after the intervention [24, 48, 50]. The changes in QST measurements are useful to understand pain processes in healthy subjects, as the respective assessments reflect the function of specific nerve fibers [48].

QST has been used to assess the effect of various pharmacological interventions on the somatosensory pain and detection thresholds where e.g. opioids is able to increase the cold pain tolerance with a large effect size [16]. Similarly, non-opioid interventions, such as the tricyclic antidepressant modulates the pressure pain tolerance and heat pain tolerance in healthy subjects with medium effect sizes whereas the detections thresholds of these modalities were not significantly affected [41].

This study aimed to investigate the pain and somatosensory sensitivity before and after different spatial HD-tDCS protocols in healthy subjects. In a sham-controlled design, three different configurations of active HD-tDCS were assessed with stimulation to the 1) DLPFC, 2) M1, and 3) simultaneously to M1 and DLPFC. Each HD-tDCS protocol was delivered for 20-min on three consecutive days. The temporal effects of the HD-tDCS protocols were assessed by pain and sensory thresholds before and after the daily HD-tDCS-session. It was hypothesised that the analgesic effects of HD-tDCS reflected as increased sensory and pain thresholds was 1) demonstrated in the active versus sham HD-tDCS protocols, 2) with the M1+DLPFC protocol being superior to the others, and 3) with increasing efficacy over the three days.

MATERIALS AND METHODS

Participants

Eighty-one healthy participants (40 female) in the age 18-55 years were included in this study conducted at Center for Neuroplasticity of Pain (CNAP) at Aalborg University, Denmark between 18/12/2018 and 02/18/2020. The sample size was calculated based on detecting a large effect size (partial $\eta^2 = 0.14$)[20], with 80% power and an alpha level of 0.05. The effect size used in the sample size calculation were based on the latest meta-analysis review investigating the modulatory effect of tDCS on pain thresholds, which showed a medium positive effect of tDCS (effect size: 0.47, 95% CI:0.13 to 0.82), corresponding to a partial η2 = 0.06 (95% CI: 0.01-0.14) [20, 24]. As it was hypothesized that HD-tDCS would provide stronger modulatory effects than the conventional tDCS configuration assessed in the meta-analysis review, it was decided to aim for the high end of the effect size spectrum. [27]. The relationship between effect size (Cohen's d and partial η2) and the sample size needed to detect differences between the groups with 80% power and an alpha level of 0.05 was estimated for large (Cohen's d = 0.8, partial η2 = 0.14, n = 15), medium (0.5, 0.06, 30), and small (0.2, 0.01, 161) effect sizes. The calculations are made using the software G*Power 3.1.9.2, using the MANOVA: Repeated measures, within-between interaction with 4 groups and 6 number of measurements

Exclusion criteria included pregnancy, drug addiction, use of opioids, antipsychotics, benzodiazepines or any previous or current neurological, musculoskeletal, rheumatic, malignant, inflammatory or mental illnesses. As the experimental outcome was psychometrics of sensory perceptions, individuals with current or prior pain conditions, recent alcohol intake or sleep deprivation were excluded. All participants received written and verbal information about the study and all signed a consent form before the first experimental session. The study was performed

according to the Helsinki Declaration, approved by the North Denmark Region Committee on Health Research Ethics (VN-20180085), and was registered at ClinicalTrials.gov (NCT04165876).

Experimental design

A randomized double-blind sham-controlled longitudinal study including four groups ('Sham-tDCS': 20 min sham tDCS; 'DLPFC-tDCS': 20 min anodal tDCS of DLPFC; 'M1-tDCS': 20 min anodal tDCS of M1; and 'DLPFC&M1-tDCS': 20 min anodal multichannel tDCS of DLPFC and M1 simultaneously). The experimental design consisted of three consecutive days of tDCS sessions with quantitative sensory assessments (vibration, tactile, pressure and thermal sensitivity) before and after each tDCS-stimulation protocol (Fig. 1). Thus, a total of six QST sessions were included (Day1pre, Day1post, Day2pre, Day2post, Day3pre, and Day3post). Stimulation sessions were separated by 24 h. Participants sat in a reclined position during the entire experimental protocol.

Figure 1.

Participants were randomly allocated to the four groups with at least 20 per group. Subjects were blinded to the stimulation protocol throughout the study and received uniform instructions prior to each session. The four protocols were programmed in the tDCS software by a third person, and named secretly, so the experimenter could run the preconfigured stimulation programs without knowing the parameters. Further it was not possible to identify the type of action of the protocols during the administration hereof, resulting in the experimenter being blinded to the stimulation type the participant received. The group numbers were linked to the respective stimulation protocols after the primary statistical analysis were conducted.

Blinding procedure

Participants were explained that they would be assigned to one of four groups, which would determine the specific configuration of HD-tDCS they would receive. Additionally it was explained that one of these configurations would be a sham-stimulation, designed to have no effects, but would simulate the potential physiological sensation of the real stimulations. The participants were asked whether they believed they had received 'real' stimulation or sham-stimulation after the tDCS-procedure on each day. The response was scored as 0 if the participant believed that they had received an active stimulation protocol and 1 if they believed that they had received the sham stimulation. The average value of their responses was calculated across the three days, and defined as the Protocol-index.

High definition transcranial direct current stimulation

The HD-tDCS was administered using the non-invasive 32-channel neuro-stimulator (Starstim 32, Neuroelectrics, Spain) with 3.14 cm^2 Ag/AgCl gelled electrodes in a neoprene cap (NE056 Headcap R, Neuroelectrics, Spain) following the international 10-10 EEG system. The protocol lasted approximately 35 minutes; 15 minutes preparing the setup with appliance of the electrode-cap, conductive gel and electrodes, and 20 minutes of HD-tDCS. The cortical targets for stimulation are chosen based on suggestions from similar studies[27, 34]. The electrical current was distributed between the electrodes in the respective ways: DLPFC-tDCS: Anode F3= 2000 µA and cathodes AF3= -667 µA, FC5= -667 µA, FC1= -667 µA. M1-tDCS:Anode C3= 2000 µA and cathodes CP1= -500 µA, FC1= -500 µA, FC5= -500 µA, CP5= -500µA. DLPFC&M1-tDCS: Anodes C3= 2000 µA, F3= 2000 µA and cathodes AF3= -800 µA, CP1= -800 µA, FC1= -800 µA, FC5= -800 µA, CP5= -800 µA. The electrode montages and their corresponding electric field distribution is shown in figure 2 (estimated by the Neuroelectrics stimulation software).

For active-tDCS, direct current was ramped up to the target amplitude over 30 s, and delivered continuously for 19 minutes before ramping down over 30 s. In the Sham-tDCS the stimulation was ramped up over 30 s to a total intensity of anode C3= 2000 µA and cathodes CP1=-500 µA, FC1=-500 µA, FC5=-500 µA, CP5 = -500 µA. Subsequently the tDCS was turned off and after 19 min of no stimulation the stimulation was turned on again and ramped down over 30 s[18]. The electrical current in the active stimulation was so weak that participants were not supposed to distinguish active stimulation from sham[7, 23].

Figure 2.

Thermal sensitivity

A 3×3 cm (9 cm^2) contact thermode (Medoc Advanced Medical Systems, Israel) was used to assess the thermal sensitivity on the skin above flexor carpi radialis muscle of the right arm. Each stimulus series began at 32°C and detection and pain thresholds were assessed, respectively. The temperature ascended and descended in ramps, and the participant's cold detection threshold (CDT) and heat detection threshold (HDT) was recorded, by them pressing a button the first time they felt a change in temperature. Cold pain thresholds (CPT) and heat pain thresholds (HPT) were assessed by the participants indicating the moment, the thermal sensation first became painful. CDT and WDT were expressed as the difference from 32°C. The measurements were repeated three times, within the temperature range of 0-50 °C, and the average was used for analysis.

Tactile sensitivity

The Tactile Detection Threshold (TDT) was determined on the skin above the right flexor carpi radialis muscle using a set of Von Frey filaments (Touch Test® Sensory Evaluators, North Coast Medical Inc, USA). The filaments are made by nylon fibre of various diameters providing a range

of forces of from 0.02 g to 300 g. Five threshold determinations were performed, each with a series of ascending and descending stimulus intensities. The final mechanical detection threshold (MDT) was defined as the geometric mean of these five series of supra- and sub-threshold stimuli intensities[47].

Mechanic pain sensitivity

The mechanical pain threshold (MPT) was determined at the skin above the right flexor carpi radialis muscle using a set of seven weighted pinprick stimulators (PinPrick, MRC Systems GmbH, Germany) with a contact area of 0.25 mm tip diameter that exert forces between 8 mN and 512 mN. Five threshold determinations were made, each with a series of ascending and descending stimulus intensities. The final threshold was defined as the geometric mean of the five supra- and subthreshold readings[47].

Sensitivity to vibration

Vibration Detection Threshold (VDT) was determined with a Rydel-Seiffer tuning fork (64 Hz, 8/8 scale, Uniplex, England). Vibrating at 64 Hz at maximum vibration amplitude, decreasing over time, the tuning fork was placed over the prominence of the distal part of the ulna in the right arm. Subjects were asked to indicate the exact moment the vibratory sensation disappeared, at which time the VDT were registered as the score from 1-8, where vibration could no longer be detected; 8 being the least amplitude causing a sensation of vibration[60]. The assessment was repeated three times and the mean value across the three trials were extracted for further analysis.

Pressure pain sensitivity

A hand-held pressure algometer (Somedic, Hörby, Sweden) with a 1-cm^2 probe was used to record the pressure pain thresholds (PPTs). The pressure was increased gradually at a rate of 30 kPa/s. The measurement was repeated three times at the flexor carpi radialis muscle of the right arm and at the medial part of the tibialis anterior muscle on the left leg. The participants indicated the exact time the pressure sensation changed from strong pressure to a painful sensation with a button press, and the force exerted was registered. The mean value across three trials was defined as the PPT used for further analysis.

Statistics

Data are presented as mean ± standard deviation (SD) in text and tables, and mean and standard error of the mean (SEM) in figures. Significance was accepted at $P < 0.05$. The Protocol-index was analysed in a one-way analysis of variance (ANOVA) with *Groups* (Sham-tDCS, DLPFC-tDCS, M1-tDCS and DLPFC&M1-tDCS) as between subject factor to identify differences. As psychophysical data were not normally distributed evaluated by the Shapiro-Wilk's test of normality for the modalities WDT, MPT, PPT and VDT, a log-transformation was conducted and used for further analysis. A one-way ANOVA was performed on the baseline values of each of assessment modalities with *Groups* (Sham-tDCS, DLPFC-tDCS, M1-tDCS and DLPFC&M1-tDCS) as between group factor to analyse baseline differences.

Three different two-way mixed-model ANOVA were performed for each sensory modality. The first analysis included the factors *Time* (Day1pre, Day1post, Day2post and Day3post) as within subject factors and *Groups* (Sham-tDCS, DLPFC-tDCS, M1-tDCS and DLPFC&M1-tDCS) as between group factor to analyse differences between the baseline and all post-tDCS assessments. A Bayesian two-way mixed-model ANOVA with the same factors were also performed to add the

meaningfulness of the frequentist statistics presenting with tendencies for significance ($P < 0.1$). The methods, results and discussion of these analyses are included in the supplementary material (available at http://links.lww.com/PAIN/B254).

The second ANOVA included factors *Time* (Day1pre, Day2pre and Day3pre) as within subject factors and *Groups* (Sham-tDCS, DLPFC-tDCS, M1-tDCS and DLPFC&M1-tDCS) as between group factor to analyse differences between the pain and sensory modalities of the three days pre-tDCS. The second analyses avoids the immediate effects of tDCS intervention acting as confounders.

To identify whether the immediate effect of the tDCS altered over the course of the three days, the delta values between pre-tDCS assessments and post-tDCS assessments (post-tDCS minus pre-tDCS) were calculated for each of the days. These delta-values were included in the third ANOVA with the factor *Time* (Day1, Day2 and Day3) as within subject factors, and the factor *Groups* (Sham-tDCS, DLPFC-tDCS, M1-tDCS and DLPFC&M1-tDCS) were used as between subject factors. If significant main effects or interactions, post hoc analysis was done using a Bonferroni test to correct for multiple comparison. As the two-way mixed model ANOVA assumes sphericity, that the data did not fulfil, a Greenhouse-Geisser correction is utilized.

RESULTS

Of the 81 participants, one participant had missing data at the Day3post session, and two subjects were excluded from the pressure pain threshold assessment, as they reported to have misunderstood the instructions. No adverse effects of the interventions were reported. Demographics of participants in the four groups, including gender, handedness, age, weight and height are presented in Table 1.

Table 1.

Blinding efficacy

The Protocol-index indicating the average rate that the participants believed to have received the sham-tDCS across the three sessions was reported as 0.33±0.07 (Sham-tDCS), 0.30±0.06 (DLPFC-tDCS), 0.22±0.07 (M1-tDCS), and 0.27±0.06 (DLPFC&M1-tDCS). There was no significant differences between groups when analysed by a one-way ANOVA ($F(3, 76) = 0.44$, $P= 0.73$).

Somatosensory and pain sensitivity across groups before tDCS

Baseline somatosensory and pain sensitivity were not significantly different between groups except for the vibration detection threshold (Day1pre in Fig. 3 and Fig. 4), analysed by a one-way ANOVA: CDT ($F(3, 77)= 0.43$, $P=0.74$, partial $\eta^2=0.02$), WDT ($F(3, 77)= 0.51$, $P=0.67$, partial $\eta^2=0.02$), CPT ($F(3, 77)= 0.89$, $P=0.45$, partial $\eta^2=0.03$), HPT ($F(3, 77)= 1.26$, $P=0.30$, partial $\eta^2=0.05$), TDT ($F(3, 77)= 2.35$, $P=0.08$, partial $\eta^2=0.08$), MPT ($F(3, 77)= 0.33$, $P=0.80$, partial $\eta^2=0.01$), PPT ($F(3, 75)= 1.74$, $P=0.17$, partial $\eta^2=0.07$), and VDT ($F(3, 77)= 4.85$, $P=0.004$, partial $\eta^2=0.16$). Post-hoc comparison revealed that the VDT of the DLPFC&M1-tDCS group (7.37±0.48) was lower than the M1-tDCS (7.85±0.30, $P<0.01$) and DLPFC-tDCS (7.79±0.44, $P=0.02$) groups.

Figure 3.

Thermal detection and pain thresholds after tDCS on each day

The ANOVA with factors *Group* (Sham-tDCS, DLPFC-tDCS, M1-tDCS and DLPFC&M1-tDCS) and *Time* (Day1pre, Day1post, Day2post and Day3post) revealed that there was no significant interaction between the two factors on any of the thermal modalities (Fig. 3): CDT ($F(6.83, 173.05)=1.0$, $P=0.43$, partial $\eta^2=0.04$), HDT ($F(8.42, 213.42)=1.22$, $P=0.29$, partial $\eta^2=0.05$), CPT

(F(5.56, 140.95)=0.86, P=0.52, partial η^2=0.03), and HPT (F(7.72, 181.76)=1.50, P=0.17, partial η^2=0.06).

Unrelated to the groups, the thresholds did change over the time for all modalities: CDT (F(2.28, 173.05)=6.36, P <0.01, partial η^2=0.08), HDT (F(2.81, 213.42)=23.95, P <0.01, partial η^2=0.24), CPT (F(1.86, 140.95)=3.07, P=0.05, partial η^2=0.039), and HPT (F(2.39, 181.76)=8.52, P <0.01, partial η^2=0.10). Post-hoc analysis revealed that CDT was higher at Day1pre (P <0.01, -1.8±0.2 °C) than Day1post (-2.4±0.2 °C), Day2post (-2.3±0.2 °C), and Day3post (-2.4±0.2 °C). The same was the case for WDT for which Day1pre (P<0.01, 2.16±0.1 °C) was lower than Day1post (2.66±0.12 °C), Day2post (2.73±0.13 °C), and Day3post (2.81±0.14 °C). For CPT, Day1post (15.0±1.1°C) was higher than Day3post (P=0.02; 12.8±1.1 °C). For HPT, both Day1pre (42.2±0.4 °C) and Day1post (42.3±0.4 °C) was lower than Day3post (P <0.01, 43.3±0.3 °C).

Thermal detection and pain thresholds before tDCS on each day

The ANOVA included the two factors *Group* (Sham-tDCS, DLPFC-tDCS, M1-tDCS, and DLPFC&M1-tDCS) and *Time* (Day1pre, Day2pre, and Day3pre). There was no significant interaction between Group and Sessions: CDT (F(5.01, 128.63)=1.83, P=0.11, partial η^2=0.07), HDT (F(5.69, 146.07)=0.069, P=0.65, partial η^2=0.03), CPT (F(4.27, 109.60)=0.48, P=0.76, partial η^2=0.02), and HPT (F(5.35, 137.26)=0.37, P=0.88, partial η^2=0.01).

Independent of the Group factor, the modalities CDT and HPT, did differ across time: CDT (F(1.67, 128.63)=13.02, P <0.01, partial η^2=0.15) and HPT (F(1.78, 137.26)=5.34, P=0.01, partial η^2=0.07). Post-hoc analysis showed that CDT was higher on Day1pre (P<0.01, -1.8±1.2 °C) than Day2pre (-2.2±0.2 °C) and Day3pre (-2.5±0.2 °C). For HPT Day1pre (42.2±0.4 °C) was lower than Day3pre (43.0±0.3 °C, P=0.01).

Changes in thermal detection and pain thresholds on each day due to tDCS

An ANOVA between *Groups* and *Time* (Day1, Day2 and Day3) on the delta values (post minus pre tDCS) of the thermal modalities, revealed that there was no significant interaction between the two factors: delta-CDT ($F(5.46, 138.31)=0.71$, $P=0.63$, partial $\eta^2=0.03$), delta-HDT ($F(5.66, 143.28)=0.45$, $P=0.84$, partial $\eta^2=0.02$), delta-CPT ($F(5.17, 131.09)=1.14$, $P=0.34$, partial $\eta^2=0.04$), and delta-HPT ($F(5.83, 147.60)=0.69$, $P=0.65$, partial $\eta^2=0.03$).

Unrelated to the factor Group, the delta-CDT was significant across time (CDT: $F(1.82, 138.31)=10.41$, $P<0.01$, partial $\eta^2=0.12$). Post-hoc analysis showed that the delta-CDT, were different on Day1 ($P<0.01$, -0.6±0.2 °C) compared to Day2 (-0.1±0.1 °C) and Day3 (0.1±0.1 °C).

Figure 4.

Mechanical detection and pain thresholds after tDCS on each day

An ANOVA with factors *Groups* (Sham-tDCS, DLPFC-tDCS, M1-tDCS and DLPFC+M1-tDCS) and *Time* (Day1pre, Day1post, Day2post and Day3post) revealed that there was significant interaction between the two factors in MPT ($F(7.78, 197.14)=2.07$, $P=0.04$, partial $\eta^2=0.08$) (Fig. 4).

The other mechanical modalities showed no significant interaction between the two factors: TDT ($F(7.62, 190.46)=1.91$, $P=0.06$, partial $\eta^2=0.07$), PPT ($F(7.58, 187.02)=1.32$, $P=0.24$, partial $\eta^2=0.05$) and VDT ($F(8.15, 206.47)=1.00$, $P=0.44$, partial $\eta^2=0.04$) (Fig. 4). Post hoc analysis of the MPT showed that that the groups did not differ significantly in the interaction between the two factors.

The factor Time unrelated to Group were significant for all mechanical modalities except VDT: TDT ($F(2.54, 190.46)=5.91$, $P<0.01$, partial $\eta^2=0.07$), MPT ($F(2.59, 197.14)=2.87$, $P=0.05$, partial $\eta^2=0.04$) and PPT ($F(2.53, 187.02)=2.49$, $P=0.07$, partial $\eta^2=0.03$). Post hoc analysis showed

that the TDT were lower on Day1pre (P<0.05, 0.43±0.05 g) compared to Day1post (0.53±0.05 g) and Day2post (0.61±0.09 g). For MPT and PPT the sessions did not differ significantly in the post hoc analysis.

Mechanical sensitivity before tDCS on each day

The ANOVA with factors *Group* (Sham-tDCS, DLPFC-tDCS, M1-tDCS and DLPFC+M1-tDCS) and *Time* (Day1pre, Day2pre and Day3pre) showed that there was no significant interaction between the two factors for any of the mechanical modalities: TDT ($F(5.29, 135.73)=1.20$, $P=0.31$, partial $\eta^2=0.05$), MPT ($F(5.30, 136.11)=1.54$, $P=0.18$, partial $\eta^2=0.06$), PPT ($F(5.65, 141.35)=1.53$, $P=0.18$, partial $\eta^2=0.06$) and VDT ($F(5.45, 139.75)=1.31$, $P=0.26$, partial $\eta^2=0.05$).

Unrelated to the Group factor, MPT and PPT did differ across time: MPT ($F(1.77, 136.11)=6.30$, $P<0.01$, partial $\eta^2=0.08$) and PPT ($F(1.89, 141.35)=4.95$, $P=0.01$, partial $\eta^2=0.06$).

Post hoc analysis showed that the MPT was higher on Day3pre (215.93±18.85 mN) than Day2pre (P<0.05, 178.25±17.22 mN). The PPT was higher on Day3pre (408.55±17.52 kPa) than Day1pre (P<0.05, 378.47±14.88 kPa).

Changes in mechanical sensitivity on each day due to tDCS

An ANOVA including the factors *Group* and *Time* (Day1, Day2 and Day3) using the delta values of the mechanical modalities, revealed that there was no significant interaction between the two factors: delta-TDT ($F(5.58, 141.38)=0.37$, $P=0.89$, partial $\eta^2=0.01$), delta-MPT ($F(5.55, 140.51)=1.14$, $P=0.34$, partial $\eta^2=0.07$), delta-PPT ($F(5.66, 139.50)=1.71$, $P=0.13$, partial $\eta^2=0.04$), and delta-VDT ($F(5.52, 139.73)=0.94$, $P=0.47$, partial $\eta^2=0.04$).

Similarly there was no significant difference in the factor Time: delta-TDT ($F(1.86, 141.38)=0.95$, $P=0.38$, partial $\eta^2=0.01$), delta-MPT ($F(1.85, 140.51)=2.07$, $P=0.13$, partial $\eta^2=0.01$),

delta-PPT ($F(1.89, 139.50)=0.80$, $P=0.45$, partial $\eta^2=0.03$), and delta-VDT ($F(1.84, 139.73)=1.95$, $P=0.15$, partial $\eta^2=0.03$).

DISCUSSION

This is the first comprehensive study to assess the effects of three different HD-tDCS protocols delivered across three days on the pain and somatosensory sensitivity in a healthy population with a successful blinded study design and sham protocol. Overall, no significant effect on the pain and somatosensory sensitivity could be detected between the active and sham stimulation protocols. Unrelated to the group factor a desensitisation was observed in the post-tDCS assessments compared to baseline for all modalities except the vibration modality.

Effects of tDCS on the somatosensory and pain sensitivity

Earlier studies have reported the opportunity to modulate the somatosensory system using tDCS, primarily studied in pain conditions[30, 52, 59]. It was hypothesised that the analgesic effect of anodal tDCS seen in chronic pain patients, would be reflected as underlying changes in the somatosensory detection and pain thresholds in healthy subjects. Potentially as a result of activation of endogenous opioids[13, 14], a decrease of excitability in the neural network related to pain processing[15, 30, 34], or other means of inhibition of the afferent corticospinal signalling related to the stimuli[56]. It was further presumed, that the results of the stimulation protocols on the somatosensory thresholds, could provide insight in the mechanisms at play when chronic pain patients experience an analgesic effect of the intervention. Assessing a more homogenous group of subjects with healthy central nervous systems were thought to strengthen the reliability of this endeavor. However, it is important to note that while the findings of healthy subjects may provide

better insights in the involved mechanisms, these mechanisms are not necessarily transferable to patients with chronic pain conditions, as they may have a sensitized central nervous system.

In the present study the three active HD-tDCS protocols did not modulate any of the somatosensory modalities significantly different from sham-stimulation. These findings are in line with Jürgens et al.[29] who in their single-blinded cross-over study, did not see any significant changes in the thermal detection and pain thresholds between the M1-tDCS and the Sham-tDCS groups. These results do however contradict a number of earlier studies, which reported that tDCS modulate selected somatosensory parameters[5–7, 44]. Reidler et al.[44] and Boggio et al.[6] demonstrated increased mechanical detection and pain thresholds. These studies used a double-blinded cross-over design, with only a single session of conventional anodal tDCS administered for 20 min[44] and 5 min[6], respectively. Interestingly, the increase in pressure pain and tactile detection thresholds reported in those studies are similar to the increases seen in the present study (PPT increase ~17.6%, TDT increase ~24%). However, the subjects receiving the sham-condition in both Reidler et al. and Boggio et al. showed a negative effect or no changes. Likewise for thermal modalities in studies from Bachmann et al.[31] and Borckardt et al.[7], where cold detection thresholds increased in the active M1-tDCS group and little to no increase was found in the sham-tDCS group, thus ending with positive effects of tDCS.

In the present study the subjects, including those administered sham-tDCS, showed an overall increase in sensory detection and pain thresholds in all modalities after tDCS except for the vibration modality. These increases in thresholds could indicate a strong placebo effect of the sham-condition. This finding is in line with other studies, that have also demonstrated strong placebo effects of sham protocols of non-invasive brain stimulation[9, 14, 42, 53, 54], but also contrasts previous positive effects of tDCS. A noteworthy difference to consider, is that both Reidler et al.[44], Boggio et al.[6], and Bachmann et al.[5] utilized a cross-over design, in which the subjects

experience both the real and the sham-tDCS. Borckardt et al.[7], did not use a cross-over design but reported, that the participants who received active stimulation rated the scalp pain associated with the stimulation significantly higher than the participants receiving sham-tDCS, suggesting an unsuccessful blinding. In the present study, the group that received sham-stimulation did not have a prior tDCS experience to compare the sensation with, which may have been pivotal for the successful blinding results that were achieved in the present study. The absence of prior experience is lost in a cross-over study, which may weaken the blinding[23]. The effective blinding in the present study may offer insight to why a stronger placebo effect were seen here than in earlier studies[5, 25, 44], and subsequently why no main effect were present between the tDCS the active and sham protocols.

An alternative explanation to the decreased sensitivity seen across the groups could be habituation to the sensory testing. As the participants were exposed to the same assessments several times over the course of the three days of experimentation, the novelty and salience decreased, which may have resulted in lowered attention and consequently less intense sensory experiences[31].

Stimulation paradigm

One of the main differences between the present study and the findings from earlier studies is the tDCS-technology. The HD-tDCS affords higher specificity[1, 12], which should enhance the effects of the stimulation[27, 32]. This is due to the concentric ring configuration stimulating a smaller area of the scalp with higher intensity than the conventional tDCS protocol. Despite this, the HD-tDCS M1 stimulation in the present study were not able to modulate the somatosensory pain and detection thresholds, better than the sham-tDCS. This may be explained by a number of unexplored caveats

of HD-tDCS that can be counterproductive for the modulating the somatosensory system. First of all, the increased specificity of the HD-tDCS configuration is not necessarily positive. Very little is known about the neurophysiological mechanism underlying the analgesic effect of tDCS, including why M1-stimulation appears to be the most effective configuration[34]. It is conceivable that the electrical field of conventional tDCS modulate nearby cortical areas surrounding M1, and possibly deeper structures, that is necessary to produce the modulatory effects. Contrary to the conventional tDCS, the HD-tDCS protocols does not lead electrical current through the brain to an anode located contralaterally, but instead have the cathodes mounted in a concentric ring around the anode ipsilaterally, which may result in a more superficial electrical current distribution.

The two tDCS protocols targeting the DLPFC demonstrated no significant effect compared to the sham stimulation. DLPFC tDCS have previously shown, to induce changes of the affective state of patients suffering from depression [17]. As the affective component is an important aspect of the chronic pain experience, DLPFC stimulation has become a developing area of interest [10, 49]. However, it is possible that the low and brief levels of pain induced with the QST approach is less useful to assess the tDCS effects, as these induce little to no affective reaction. This is in line with the findings from a study of tricyclic antidepressants, where the pain thresholds were not significantly affected whereas the pain tolerance was increased, potentially due to the lack of an affective component in pain thresholds [41].

The increased stimulation intensity reaching the cortex using HD-tDCS may also be counterproductive. Some studies report, that electrical stimulation for too long or with too high amplitude result in a decreased or even reversed effects of the stimulation at the motor-cortical excitability [28, 37]. This reverse effect is attributed endogenous homeostatic mechanisms, which are well preserved in a healthy population[51].

No effects of HD-tDCS in healthy subjects

Majority of prior tDCS studies aiming to modulate the pain sensitivity has been done in pain patients[34, 38]. It is well accepted that chronic pain conditions alter the central nervous system in various ways, including changes of the corticospinal signalling and excitability, cortical reorganisation, and impairment of homeostatic control[3, 4, 36, 40]. This leads to the question, whether a sensitised central nervous system is more easily modulated by brain stimulation methods than the healthy system. It is possible that the functional homeostatic control of the healthy participants counteracted the modulatory effects of the tDCS intervention[26]. This theory would be in line with the findings of the present study, where no significant modulatory effects were identified after the active tDCS protocols. This may provide insight into why tDCS seemingly has lower effect on a healthy population than a chronic pain patient population[38]. It is important to note, that despite tDCS is not inducing any apparent changes to the somatosensory thresholds in the present study, it does not disprove that tDCS can be utilized to induce an analgesic effect in a clinical chronic pain population.

Pain through sensory testing

The nature of the tests that are used here to evaluate the somatosensory and pain sensitivity may have been a limitation. Only detection and pain thresholds were assessed, which implies low levels of pain induced for brief periods. By including assessments of pain tolerance, entailing more intense pain, a more nuanced insight of the HD-tDCS effects on the somatosensory system would have been established. Further an assessment entailing a more intense pain experience, may have added an affective component that is also minimal in these experimental pain stimuli.

Sample size

An important methodological limitations to consider, is the sample size. It is possible that the sample size of N=20 per group is insufficient, considering the variability of the psychophysical measurements in the present study exceeded the power estimation. The effect size of the difference between the four groups in thermal detection and pain thresholds ranged from 0.03 partial η^2 (CPT) to 0.06 partial η^2 (HPT) when assessing the effect between baseline and post-tDCS on each day. Similarly for the mechanical detection and pain thresholds the effect size ranged from 0.04 partial η^2 (VDT) to 0.08 partial η^2 (MPT). These effect sizes are considered to be low to medium[20], which are lower than what was assumed needed to detect significant differences from the power calculation. However, the effect sizes reported here are similar to studies with comparable methodologies[44, 55]. Despite the lack of effect size, this study includes a much larger total population size of 81 participants, than the comparable studies, which included between 8 and 41 subjects[55]. This fact strengthens the results of study, and the findings should not be dismissed despite conflicting other studies.

CONCLUSION

The most promising electrode configurations for HD-tDCS in order to induce neuroplastic changes in specific areas of the brain were assessed in healthy people. The active HD-tDCS configurations were not significantly more effective than sham-stimulation in modulating the somatosensory and pain sensitivity. Such a lack of findings may be due to a strong placebo effect, or potentially due to the healthy central nervous system being less susceptible to neuromodulation than an already sensitized central nervous system, such as in chronic pain patients or otherwise experimentally perturbed.

ACKNOWLEDGEMENTS

The authors declare that there are no conflicts of interest regarding the publication of this paper.

REFERENCES

1. Alam M, Truong DQ, Khadka N, Bikson M (2016) Spatial and polarity precision of concentric high-definition transcranial direct current stimulation (HD-tDCS). Phys Med Biol 61:4506–4521. doi: 10.1088/0031-9155/61/12/4506

2. Antal A, Terney D, Kühnl S, Paulus W (2010) Anodal Transcranial Direct Current Stimulation of the Motor Cortex Ameliorates Chronic Pain and Reduces Short Intracortical Inhibition. Journal of Pain and Symptom Management 39:890–903. doi: 10.1016/j.jpainsymman.2009.09.023

3. Arendt-Nielsen L, Fernández-de-las-Peñas C, Graven-Nielsen T (2011) Basic aspects of musculoskeletal pain: from acute to chronic pain. Journal of Manual & Manipulative Therapy 19:186–193. doi: 10.1179/106698111X13129729551903

4. Arendt-Nielsen L, Morlion B, Perrot S, Dahan A, Dickenson A, Kress HG, Wells C, Bouhassira D, Mohr Drewes A (2018) Assessment and manifestation of central sensitisation across different chronic pain conditions. European Journal of Pain 22:216–241. doi: 10.1002/ejp.1140

5. Bachmann CG, Muschinsky S, Nitsche MA, Rolke R, Magerl W, Treede R-D, Paulus W, Happe S (2010) Transcranial direct current stimulation of the motor cortex induces distinct

changes in thermal and mechanical sensory percepts. Clinical Neurophysiology 121:2083–2089. doi: 10.1016/j.clinph.2010.05.005

6. Boggio PS, Zaghi S, Lopes M, Fregni F (2008) Modulatory effects of anodal transcranial direct current stimulation on perception and pain thresholds in healthy volunteers: Modulation of pain threshold with transcranial direct current stimulation. European Journal of Neurology 15:1124–1130. doi: 10.1111/j.1468-1331.2008.02270.x

7. Borckardt JJ, Bikson M, Frohman H, Reeves ST, Datta A, Bansal V, Madan A, Barth K, George MS (2012) A Pilot Study of the Tolerability and Effects of High-Definition Transcranial Direct Current Stimulation (HD-tDCS) on Pain Perception. The Journal of Pain 13:112–120. doi: 10.1016/j.jpain.2011.07.001

8. Brückner S, Kammer T (2017) Both anodal and cathodal transcranial direct current stimulation improves semantic processing. Neuroscience 343:269–275. doi: 10.1016/j.neuroscience.2016.12.015

9. Burke MJ, Kaptchuk TJ, Pascual-Leone A (2019) Challenges of Differential Placebo Effects in Contemporary Medicine: The Example of Brain Stimulation. Ann Neurol 85:12–20. doi: 10.1002/ana.25387

10. Chalah MA, Riachi N, Ahdab R, Mhalla A, Abdellaoui M, Créange A, Lefaucheur J-P, Ayache SS (2017) Effects of left DLPFC versus right PPC tDCS on multiple sclerosis fatigue. Journal of the Neurological Sciences 372:131–137. doi: 10.1016/j.jns.2016.11.015

11. Choi Y-H, Jung S-J, Lee CH, Lee S-U (2014) Additional Effects of Transcranial Direct-Current Stimulation and Trigger-Point Injection for Treatment of Myofascial Pain Syndrome:

A Pilot Study with Randomized, Single-Blinded Trial. The Journal of Alternative and Complementary Medicine 20:698–704. doi: 10.1089/acm.2013.0243

12. DaSilva AF, Truong DQ, DosSantos MF, Toback RL, Datta A, Bikson M (2015) State-of-art neuroanatomical target analysis of high-definition and conventional tDCS montages used for migraine and pain control. Front Neuroanat 9:1–12. doi: 10.3389/fnana.2015.00089

13. DosSantos MF, Love TM, Martikainen IK, Nascimento TD, Fregni F, Cummiford C, Deboer MD, Zubieta J-K, DaSilva AFM (2012) Immediate Effects of tDCS on the μ-Opioid System of a Chronic Pain Patient. Front Psychiatry 3:1–6. doi: 10.3389/fpsyt.2012.00093

14. DosSantos MF, Martikainen IK, Nascimento TD, Love TM, DeBoer MD, Schambra HM, Bikson M, Zubieta J-K, DaSilva AF (2014) Building up Analgesia in Humans via the Endogenous μ-Opioid System by Combining Placebo and Active tDCS: A Preliminary Report. PLoS ONE 9:1–9. doi: 10.1371/journal.pone.0102350

15. DosSantos MF, Oliveira AT, Ferreira NR, Carvalho ACP, Rosado de Castro PH (2018) The Contribution of Endogenous Modulatory Systems to TMS- and tDCS-Induced Analgesia: Evidence from PET Studies. Pain Research and Management 2018:1–14. doi: 10.1155/2018/2368386

16. Eisenberg E, Midbari A, Haddad M, Pud D (2010) Predicting the analgesic effect to oxycodone by ‘static’ and ‘dynamic’ quantitative sensory testing in healthy subjects. PAIN 151:104–109. doi: 10.1016/j.pain.2010.06.025

17. Ferrucci R, Bortolomasi M, Vergari M, Tadini L, Salvoro B, Giacopuzzi M, Barbieri S, Priori A (2009) Transcranial direct current stimulation in severe, drug-resistant major depression. Journal of Affective Disorders 118:215–219. doi: 10.1016/j.jad.2009.02.015

18. Flood A, Waddington G, Cathcart S (2016) High-Definition Transcranial Direct Current Stimulation Enhances Conditioned Pain Modulation in Healthy Volunteers: A Randomized Trial. The Journal of Pain 17:600–605. doi: 10.1016/j.jpain.2016.01.472

19. Fregni F, Boggio PS, Lima MC, Ferreira MJL, Wagner T, Rigonatti SP, Castro AW, Souza DR, Riberto M, Freedman SD, Nitsche MA, Pascual-Leone A (2006) A sham-controlled, phase II trial of transcranial direct current stimulation for the treatment of central pain in traumatic spinal cord injury: Pain 122:197–209. doi: 10.1016/j.pain.2006.02.023

20. Fritz CO, Morris PE, Richler JJ (2012) Effect size estimates: Current use, calculations, and interpretation. Journal of Experimental Psychology: General 141:2–18. doi: 10.1037/a0024338

21. Gallo S, Maskaljunas T, Lammes B, Henriques C, Suttrup J, Keysers C, Gazzola V (2017) Stimulation of the somatosensory cortex using HD-tDCS modulates others' pain perception; Combined evidence from behaviour, EEG, fMRI data. Brain Stimulation 10:466. doi: 10.1016/j.brs.2017.01.366

22. Gálvez V, Alonzo A, Martin D, Loo CK (2013) Transcranial direct current stimulation treatment protocols: should stimulus intensity be constant or incremental over multiple sessions? The International Journal of Neuropsychopharmacology 16:13–21. doi: 10.1017/S1461145712000041

23. Garnett EO, den Ouden D-B (2015) Validating a Sham Condition for Use in High Definition Transcranial Direct Current Stimulation. Brain Stimulation 8:551–554. doi: 10.1016/j.brs.2015.01.399

24. Giannoni-Luza S, Pacheco-Barrios K, Cardenas-Rojas A, Mejia-Pando PF, Luna-Cuadros MA, Barouh JL, Gnoatto-Medeiros M, Candido-Santos L, Barra A, Caumo W, Fregni F

(2020) Non-invasive motor cortex stimulation effects on quantitative sensory testing (QST) in healthy and chronic pain subjects: a systematic review and meta-analysis. Pain Publish Ahead of Print. doi: 10.1097/j.pain.0000000000001893

25. Grundmann L, Rolke R, Nitsche MA, Pavlakovic G, Happe S, Treede R-D, Paulus W, Bachmann CG (2011) Effects of transcranial direct current stimulation of the primary sensory cortex on somatosensory perception. Brain Stimulation 4:253–260. doi: 10.1016/j.brs.2010.12.002

26. Hassanzahraee M, Zoghi M, Jaberzadeh S (2018) How different priming stimulations affect the corticospinal excitability induced by noninvasive brain stimulation techniques: a systematic review and meta-analysis. Reviews in the Neurosciences 29:883–899. doi: 10.1515/revneuro-2017-0111

27. Hill AT, Rogasch NC, Fitzgerald PB, Hoy KE (2018) Effects of single versus dual-site High-Definition transcranial direct current stimulation (HD-tDCS) on cortical reactivity and working memory performance in healthy subjects. Brain Stimulation 11:1033–1043. doi: 10.1016/j.brs.2018.06.005

28. Jamil A, Batsikadze G, Kuo H-I, Labruna L, Hasan A, Paulus W, Nitsche MA (2017) Systematic evaluation of the impact of stimulation intensity on neuroplastic after-effects induced by transcranial direct current stimulation. The Journal of Physiology 595:1273–1288. doi: 10.1113/JP272738

29. Jürgens TP, Schulte A, Klein T, May A (2012) Transcranial direct current stimulation does neither modulate results of a quantitative sensory testing protocol nor ratings of suprathreshold

heat stimuli in healthy volunteers: tDCS in experimental pain. European Journal of Pain 16:1251–1263. doi: 10.1002/j.1532-2149.2012.00135.x

30. Knotkova H, Nitsche MA, Cruciani RA (2013) Putative physiological mechanisms underlying tDCS analgesic effects. Front Hum Neurosci 7:628. doi: 10.3389/fnhum.2013.00628

31. Kucyi A, Salomons TV, Davis KD (2013) Mind wandering away from pain dynamically engages antinociceptive and default mode brain networks. PNAS 110:18692–18697. doi: 10.1073/pnas.1312902110

32. Kuo H-I, Bikson M, Datta A, Minhas P, Paulus W, Kuo M-F, Nitsche MA (2013) Comparing Cortical Plasticity Induced by Conventional and High-Definition 4 × 1 Ring tDCS: A Neurophysiological Study. Brain Stimulation 6:644–648. doi: 10.1016/j.brs.2012.09.010

33. Lefaucheur J-P (2016) A comprehensive database of published tDCS clinical trials (2005–2016). Neurophysiologie Clinique/Clinical Neurophysiology 46:319–398. doi: 10.1016/j.neucli.2016.10.002

34. Lefaucheur J-P, Antal A, Ayache SS, Benninger DH, Brunelin J, Cogiamanian F, Cotelli M, De Ridder D, Ferrucci R, Langguth B, Marangolo P, Mylius V, Nitsche MA, Padberg F, Palm U, Poulet E, Priori A, Rossi S, Schecklmann M, Vanneste S, Ziemann U, Garcia-Larrea L, Paulus W (2017) Evidence-based guidelines on the therapeutic use of transcranial direct current stimulation (tDCS). Clinical Neurophysiology 128:56–92. doi: 10.1016/j.clinph.2016.10.087

35. Luedtke K, Rushton A, Wright C, Geiss B, Juergens TP, May A (2012) Transcranial Direct Current Stimulation for the Reduction of Clinical and Experimentally Induced Pain: A

Systematic Review and Meta-analysis. The Clinical Journal oF Pain 28:452–461. doi: 10.1097/AJP.0b013e31823853e3

36. Martino ED, Petrini L, Schabrun S, Graven-Nielsen T (2018) Cortical Somatosensory Excitability Is Modulated in Response to Several Days of Muscle Soreness. The Journal of Pain 19:1296–1307. doi: 10.1016/j.jpain.2018.05.004

37. Monte-Silva K, Kuo M-F, Hessenthaler S, Fresnoza S, Liebetanz D, Paulus W, Nitsche MA (2013) Induction of Late LTP-Like Plasticity in the Human Motor Cortex by Repeated Non-Invasive Brain Stimulation. Brain Stimulation 6:424–432. doi: 10.1016/j.brs.2012.04.011

38. Mylius V, Borckardt JJ, Lefaucheur J-P (2012) Noninvasive cortical modulation of experimental pain: Pain 153:1350–1363. doi: 10.1016/j.pain.2012.04.009

39. O'Connell NE, Marston L, Spencer S, DeSouza LH, Wand BM (2018) Non-invasive brain stimulation techniques for chronic pain. Cochrane Database Syst Rev 4:CD008208. doi: 10.1002/14651858.CD008208.pub5

40. Parker RS, Lewis GN, Rice DA, McNair PJ (2016) Is Motor Cortical Excitability Altered in People with Chronic Pain? A Systematic Review and Meta-Analysis. Brain Stimulation 9:488–500. doi: 10.1016/j.brs.2016.03.020

41. Poulsen L, Arendt-Nielsen L, Brøsen K, Nielsen KK, Gram LF, Sindrup SH (1995) The hypoalgesic effect of imipramine in different human experimental pain models. Pain 60:287–293. doi: 10.1016/0304-3959(94)00142-2

42. Rabipour S, Wu AD, Davidson PSR, Iacoboni M (2018) Expectations may influence the effects of transcranial direct current stimulation. Neuropsychologia 119:524–534. doi: 10.1016/j.neuropsychologia.2018.09.005

43. Raffin E, Siebner HR (2014) Transcranial brain stimulation to promote functional recovery after stroke. Curr Opin Neurol 27:54–60. doi: 10.1097/WCO.0000000000000059

44. Reidler JS, Mendonca ME, Santana MB, Wang X, Lenkinski R, Motta AF, Marchand S, Latif L, Fregni F (2012) Effects of Motor Cortex Modulation and Descending Inhibitory Systems on Pain Thresholds in Healthy Subjects. The Journal of Pain 13:450–458. doi: 10.1016/j.jpain.2012.01.005

45. Roche N, Geiger M, Bussel B (2015) Mechanisms underlying transcranial direct current stimulation in rehabilitation. Annals of Physical and Rehabilitation Medicine 58:214–219. doi: 10.1016/j.rehab.2015.04.009

46. Roizenblatt S, Fregni F, Gimenez R, Wetzel T, Rigonatti SP, Tufik S, Boggio PS, Valle AC (2007) Site-specific Effects of Transcranial Direct Current Stimulation on Sleep and Pain in Fibromyalgia: A Randomized, Sham-controlled Study. Pain Practice 7:297–306. doi: 10.1111/j.1533-2500.2007.00152.x

47. Rolke R, Andrews K, Magerl W (2010) A standardized battery of Quantitative Sensory Testing according to the protocol of the German Research Network on Neuropathic Pain (DFNS). 27

48. Rolke R, Magerl W, Campbell KA, Schalber C, Caspari S, Birklein F, Treede R-D (2006) Quantitative sensory testing: a comprehensive protocol for clinical trials. European Journal of Pain 10:77–77. doi: 10.1016/j.ejpain.2005.02.003

49. Seminowicz DA, de Martino E, Schabrun SM, Graven-Nielsen T (2018) Left DLPFC rTMS Reduces the Development of Long-Term Muscle Pain: PAIN 1. doi: 10.1097/j.pain.0000000000001350

50. Smith SM, Dworkin RH, Turk DC, Baron R, Polydefkis M, Tracey I, Borsook D, Edwards RR, Harris RE, Wager TD, Arendt-Nielsen L, Burke LB, Carr DB, Chappell A, Farrar JT, Freeman R, Gilron I, Goli V, Haeussler J, Jensen T, Katz NP, Kent J, Kopecky EA, Lee DA, Maixner W, Markman JD, McArthur JC, McDermott MP, Parvathenani L, Raja SN, Rappaport BA, Rice ASC, Rowbotham MC, Tobias JK, Wasan AD, Witter J (2017) The Potential Role of Sensory Testing, Skin Biopsy, and Functional Brain Imaging as Biomarkers in Chronic Pain Clinical Trials: IMMPACT Considerations. The Journal of Pain 18:757–777. doi: 10.1016/j.jpain.2017.02.429

51. Thapa T, Schabrun SM (2018) Test-Retest Reliability of Homeostatic Plasticity in the Human Primary Motor Cortex. Neural Plasticity 2018:1–9. doi: 10.1155/2018/6207508

52. Treister R, Lang M, Klein M, Oaklander AL (2015) Transcranial direct current stimulation (tDCS) for treating chronic pain – Preliminary results of open-label, self-administered, at-home treatment. Journal of the Neurological Sciences 357:e244. doi: 10.1016/j.jns.2015.08.851

53. Turi Z, Bjørkedal E, Gunkel L, Antal A, Paulus W, Mittner M (2018) Evidence for Cognitive Placebo and Nocebo Effects in Healthy Individuals. Scientific Reports 8:1–14. doi: 10.1038/s41598-018-35124-w

54. Turi Z, Mittner M, Paulus W, Antal A (2017) Placebo Intervention Enhances Reward Learning in Healthy Individuals. Scientific Reports 7:1–15. doi: 10.1038/srep41028

55. Vaseghi B, Zoghi M, Jaberzadeh S (2014) Does anodal transcranial direct current stimulation modulate sensory perception and pain? A meta-analysis study. Clinical Neurophysiology 125:1847–1858. doi: 10.1016/j.clinph.2014.01.020

56. Vaseghi B, Zoghi M, Jaberzadeh S (2015) The effects of anodal-tDCS on corticospinal excitability enhancement and its after-effects: conventional vs. unihemispheric concurrent dual-site stimulation. Frontiers in Human Neuroscience 9. doi: 10.3389/fnhum.2015.00533

57. Vaseghi B, Zoghi M, Jaberzadeh S (2015) Differential effects of cathodal transcranial direct current stimulation of prefrontal, motor and somatosensory cortices on cortical excitability and pain perception – a double-blind randomised sham-controlled study. European Journal of Neuroscience 42:2426–2437. doi: 10.1111/ejn.13043

58. Villamar MF, Volz MS, Bikson M, Datta A, DaSilva AF, Fregni F (2013) Technique and Considerations in the Use of 4x1 Ring High-definition Transcranial Direct Current Stimulation (HD-tDCS). Journal of Visualized Experiments. doi: 10.3791/50309

59. Villamar MF, Wivatvongvana P, Patumanond J, Bikson M, Truong DQ, Datta A, Fregni F (2013) Focal Modulation of the Primary Motor Cortex in Fibromyalgia Using 4×1-Ring High-Definition Transcranial Direct Current Stimulation (HD-tDCS): Immediate and Delayed Analgesic Effects of Cathodal and Anodal Stimulation. The Journal of Pain 14:371–383. doi: 10.1016/j.jpain.2012.12.007

60. Whitton TL, Johnson RW, Lovell AT (2005) Use of the Rydel—Seiffer graduated tuning fork in the assessment of vibration threshold in postherpetic neuralgia patients and healthy controls. European Journal of Pain 9:167–171. doi: 10.1016/j.ejpain.2004.05.001

61. Zhu C, Yu B, Zhang W, Chen W, Qi Q, Miao Y (2017) Effiectiveness and safety of transcranial direct current stimulation in fibromyalgia: A systematic review and meta-analysis. Journal of Rehabilitation Medicine 49:2–9. doi: 10.2340/16501977-2179

FIGURE LEGENDS

Figure 1. Experimental timeline. Quantitative sensory testing (QST) of pain and sensory profiles was done before and after each transcranial direct current stimulation (tDCS) session on three days.

Figure 2. *The electrode montages and the electrical field distribution of the three active stimulation paradigms (A: DLPFC, B: M1, C: M1+DLPFC). The models are made using the modelling program in Neuroelectrics stimulation software.*

Figure 3. Mean (±SEM) cold detection threshold (CDT), warm detection threshold (WDT), cold pain threshold (CPT), and heat pain threshold (HPT) over 3 days before (non-shaded) and after (shaded) tDCS. CDT and WDT represents the temperature change required for the participants to notice an increase or decrease in temperature from the baseline (32 C°). CPT and HPT represents the temperature at which the participants identified the temperature as painful.

Figure 4. Mean (±SEM) tactile detection threshold (TDT), pressure pain threshold (PPT), mechanical pain threshold (MPT), and vibration detection threshold (VDT) over 3 days before (non-shaded) and after (shaded) tDCS. TDT represents the pressure needed for the participants to detect the touch of the Von Frey filament. PPT and MPT represents the threshold at which the participants identified the pressure and mechanical stimulation as painful. VDT represents the

amplitude of vibration of a tuning fork at which the participants could no longer detect the vibration.

Table 1. Mean (±SD*) characteristics and distribution of participants.*

Group	Gender (N)		Handedness (N)		Age (years)	Height (cm)	Weight (kg)
	Male	Female	Right	Left			
Sham-tDCS	12	8	16	4	26.5±7.0	176.8±9.1	76. 8±12.1
DLPFC-tDCS	9	12	19	2	23.3±3.3	172.6±11.9	69.2±14.9
M1-tDCS	10	10	18	2	24.5±4.1	174.2±9.4	69.9±9.5
DLPFC&M1-tDCS	10	10	20	0	26.1±6.9	173.2±9.8	74.2±17.5
Total	41	40	73	8	25.1±5.6	174.2±10.1	72.5±14.0