Bortoletto, M.C. Pellicciari, C. Rodella, C. Miniussi (2015). The interaction with task-induced activity is more important than polarization: a tDCS study. Brain Stimulation, 8;2: 269-76. doi: 10.1016/j.brs.2014.11.006

The Interaction With Task-induced Activity is More Important Than Polarization: A tDCS Study

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Background: Anodal transcranial direct current stimulation (A-tDCS) is a non-invasive technique in which cortical polarization can be used to increase excitability and facilitate learning through the modulation of neuroplasticity. Although the facilitatory effects of A-tDCS are well documented, there is evidence that they are not always present and may even be reversed during task execution.
Objective: In this study, we explored the interaction between A-tDCS and task execution. We aimed to test how the excitability induced by the task interacts with the excitability induced by A-tDCS and determines the behavioral outcome.
Methods: We performed an experiment in which A-tDCS or a control stimulation (Ctrl) were combined with one of two motor practices (MP), one inducing learning and increasing cortical excitability (F-MP) and the other neither inducing learning nor changing cortical excitability (S-MP). Six blocks of MP were performed while the primary motor cortex was stimulated. Moreover, one block of F-MP was performed before the stimulation (baseline) and one after. In an additional experiment, motor evoked potentials (MEPs) were recorded before the baseline block (TMS-pre) and after the MP (TMS-post).
Results: We observed that A-tDCS reduced learning when participants performed the F-MP and facilitated learning for the S-MP. MEPs data paralleled behavioral results, confirming that the effects generated by A-tDCS depend on the excitability changes induced by the task.
Conclusions: Our results demonstrate that tDCS-induced plasticity is task-dependent, and the concurrent combination of A-tDCS with another excitability-increasing event, e.g., motor practice, may trigger non-additive mechanisms, hindering neuroplasticity.

Introduction

The possibility of influencing brain activity and steadily enhancing behavioral performance through external intervention has long fascinated neuroscientists. In recent years, we have witnessed the rapid development of non-invasive brain stimulation techniques that allow interaction with brain function. Nevertheless, despite an ever-increasing enthusiasm for the use of these techniques, we are also realizing that the basic knowledge of their mechanisms of action is not sufficient to predict their outcome; rather, their interaction with brain activity should be considered.

One of these techniques, transcranial direct current stimulation (tDCS), has received great interest because it has great potential use in basic research and clinical applications. The tDCS technique [1,2] involves the application to the brain of an electric field that modulates neuronal activity [3]. Generally, its effects are polarity dependent: anodal tDCS (A-tDCS) increases neuronal excitability and promotes long-term potentiation (LTP), whereas cathodal tDCS induces the opposite effects [4e6].

The application of A-tDCS over an area subtending a task usually facilitates the learning of that task[7,8]. It has been suggested that this effect may be due to a gating mechanism, i.e., the possibility of inducing additive neuroplastic changes by modulating the excitability of the target neurons[9,10]. In the motor system, evidence that A-tDCS over the primary motor cortex (M1) facilitates learning has been found with many different tasks, e.g., the isometric pinch force task [11,12], implicit sequence learning [13], explicit sequence learning [14,15], visuo-motor learning [16] and force-field tasks [17,18].

Nevertheless, there is evidence that in some cases A-tDCS effects may not be present [19,20] or even reverse. During resting state, AtDCS has been shown to induce non-linear physiological responses at increased duration and intensity of stimulation [21,22], or by simultaneous application of A-tDCS with another excitability enhancing stimulation [23]. Moreover, a few studies have reported a decrease in cortical excitability and in LTP-like effects when AtDCS was applied during a task, like muscular contraction [24] or at the end of a simple excitability-increasing motor task [25]. Although these latter data have been explained differently, they may instead be the result of a mechanism that so far has not been considered in this context, and non-linear non-additive phenomena may occur when tDCS is associated with a task.

A recent proposal attempts to explain the effects of non-invasive brain stimulation during a behavioral task in the framework of neural noise [26], suggesting that the outcome of applying tDCS depends on the interaction between task-induced and tDCS-induced activity, rather than solely on the polarity of the stimulation. To verify this hypothesis, we utilized two simple motor practice tasks, in which use-dependent learning may or may not be induced, through multiple repetitions of the same movement. We tested both the behavioral (Experiment 1) and neurophysiological (Experiment 2) effects of applying A-tDCS during the motor tasks to evaluate the interaction of A-tDCS-induced activity with the task induced activity. Our prediction was that if A-tDCS interacts with the state of the stimulated cortex, we should find different outcomes depending on the task; alternatively, if the effects of A-tDCS depend only on the polarization of the cortical activity, we should find facilitatory effects, independently of the task. In line with the predictions of the neural noise model, we found that the activity of the motor system at the time of stimulation determined the effect of A-tDCS on motor learning. This supports the suggestion that the effect of tDCS may be predicted more precisely by taking into account the level of neural activity induced by the task when applying tDCS.

Methods

Participants

Eighty-eight young, healthy, right-handed non-musicians with no history of psychiatric, neurological or other relevant medical diseases participated in the study. Forty-eight participants participated in Experiment 1 (25 females, aged 19e31 years, mean age ¼ 22.31 years). Forty participants took part in Experiment 2 (20 females, aged 18e33 years, mean age ¼ 24.20 years). Each participant had one testing session in one experiment and did not take part in more than one experiment. Handedness was assessed using the Edinburgh Handedness Inventory test [27]. We applied a single-blind study, in which participants were not aware of the type of stimulation they received. The protocol was performed in accordance with safety procedures for non-invasive brain stimulation [28,29] and was approved by the Ethics Committee of IRCCS Centro San Giovanni di Dio Fatebenefratelli,
Brescia, Italy.

Procedure

Both experiments consisted of four sessions with parallel procedures. Four conditions were created by changing the type of motor practice (Fast MP and Slow MP) and the type of stimulation

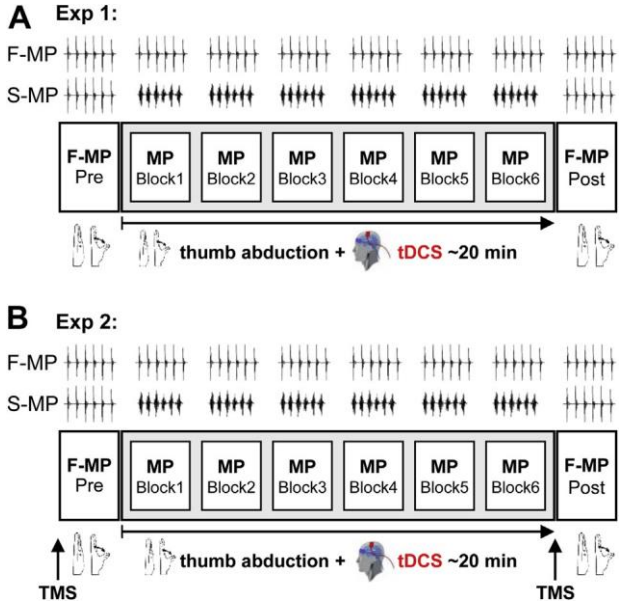

Figure 1. A: Paradigm of Experiment 1. The MP task consisted of fast thumb abduction movements (F-MP) of the left hand, which induce learning or of slow thumb abduction movements (S-MP), which do not induce learning. In both cases, six blocks of MP (corresponding to 20 min) were performed during tDCS, which was either anodal tDCS or Control stimulation. Moreover, two blocks with fast thumb abduction were performed, one before and one after tDCS. B: The paradigm of Experiment 2 was identical to Experiment 1 with the exception that TMS-elicited MEPs were recorded in two blocks, one before the baseline block and one after the MP.

(A-tDCS and Control stimulation-Ctrl), as it will be described in the motor practice and stimulation sessions respectively.

Participants were comfortably seated on an armchair, looking at a computer screen placed in front of them. Their left arm was adducted at the shoulder and flexed 90 at the elbow, and the semi pronated forearm rested on a flat armrest. The forearm, wrist and fingers IIeV were fixed in a cast, leaving the thumb free for movements in the horizontal direction.

The location of the right M1 representation of the abductor pollicis brevis muscle (APB) was identified using transcranial magnetic stimulation (TMS) (Magstim SuperRapid, Whitland, UK). The TMS coil was held tangential to the scalp and moved around the motor hand area in 0.5 cm steps until the position to elicit motor evoked potentials (MEPs) in the left APB was found.

The participants were then given instructions for the motor task and briefly practiced the movement 10 times. The motor task consisted of repeatedly performing thumb abduction movements with the left hand as fast as possible. The movements were paced by a brief 1000 Hz tone at a rate of 0.25 Hz. The signal of the accelerometer mounted on the participant's left thumb was displayed as visual feedback of the movement acceleration. Before each movement, the thumb had to be relaxed into a resting position, leaning against a soft support that corresponded to the starting position. The use of the support eliminated shifting of the thumb's starting position during practice as a possible cause for performance change.

Experiment 1 consisted of one block of 40 fast thumb abductions (Baseline), 20 min of motor practice (MP) concurrent to the application of tDCS, and one block of 40 fast thumb abductions (Post) (Fig. 1A). Experiment 2 (Fig. 1B) followed the same procedure as in Experiment 1, with 2 additional blocks of TMS-elicited MEPs recording, one before the baseline block (TMSpre) and one after the MP (TMS-post). The participants received visual feedback on the movement acceleration and verbal encouragement from the experimenter for the entire duration of the experiment.

Movement acceleration was measured during the entire experiment using a monoaxial accelerometer (model: 352C23, voltage sensitivity ¼ 5 mV/g; PCB piezotronic) mounted on the thumb's interphalangeal joint to detect acceleration along the abduction e adduction axis. The raw signal was amplified with a gain factor of 100 (model: 480E09 signal conditioner; PCB piezotronic), digitized (A/D rate: 500 Hz) and fed into the laboratory computer for online visual display and offline analysis.

At the end of the experimental session, all participants were asked to complete a questionnaire on the sensations they experienced in association with the tDCS [30]. The participants gave a score from 0 ¼ none to 4 ¼ strong to rate different skin sensations during the stimulation (irritation, pain, burning, heat, itch, iron taste and fatigue) and the influence of stimulation on their performance.

Motor practice

Thumb abduction movements with the left hand at a rate of 0.25 Hz were performed during the MP. The MP was varied only for the speed of movement. Fast MP (F-MP) consisted of fast thumb abductions known to induce learning and plasticity in M1 [31,32]. In contrast, the Slow MP (S-MP) consisted of slow ramp-like thumb abduction movements, which do not induce learning [32,33]. In this case, the participants were instructed to perform the movements at a natural speed, and the experimenter verified that the peak acceleration was less than 1/3 of the peak acceleration in the F-MP.

In all experiments, the MP was performed in six blocks of 40 movements alternating with 40-s breaks for a total duration of 20 min (duration of each block: 2.40 min). A relatively low movement rate was set to prevent muscle fatigue, as suggested in a previous work [34].

tDCS

A battery-driven DC stimulator (BrainStim) delivered a constant current flow of 1.5 mA via two conducting electrodes covered with a saline-soaked sponge. In the A-tDCS condition the anodal electrode was positioned over the right M1 as individuated by TMS (Electrode dimension: 25 cm^2, Current density: 0.06 mA/cm^2). The reference (i.e., cathodal) electrode was positioned over the left eyebrow. A bigger electrode was chosen as a reference (electrode dimension: 35 cm^2, current density: 0.042 mA/cm^2) so that the stimulation of the prefrontal cortex was minimized [35] and current shunting was minimized with a distance between electrodes of more than 8 cm. The current ramped up and down over the first and last 8 s of stimulation and was applied for 20 min in A-tDCS. In the Ctrl, a sham stimulation was applied in which A-tDCS stimulation was on only in the first and last 20 s (Experiment 1) or no stimulation was applied (Experiment 2).

TMS

In Experiment 2, TMS-elicited MEPs were recorded in two blocks, one before the baseline block (TMS-pre) and one after the MP (TMS-post). TMS was carried out by a 70 mm figure-of-eight coil. The coil was placed tangentially to the scalp with the handle pointing backwards and laterally at a 45 angle away from the midline, and positioned for inducing maximal MEPs from the left APB. MEPs were recorded via two pairs of Ag/AgCl-coated surface electrodes in belly-tendon montage from the left APB and from the first dorsal interosseous (FDI), as a control; signals were sampled at 5 kHz and band-pass filtered at 10e1000 Hz. In each block, 20 single TMS pulses were delivered at random intervals (0.25e0.125 Hz) at the stimulation intensity eliciting an MEP of about 1 mV (68% of the maximum stimulator output on average). Complete muscle relaxation was monitored throughout the experimental session. A TMS neuronavigation system (SofTaxic, EMS, Bologna, Italy) was used to ensure a high degree of reproducibility across blocks.

Behavioral data

Movement acceleration was measured at the first peak of the accelerometer signal and was baseline-corrected to the 100 ms preceding the beginning of the movement. The mean peak acceleration was calculated for each block of fast thumb abductions and slow thumb abductions. The values of acceleration in the block of MP and in the Post block were normalized to the mean peak acceleration in the baseline block.

We collapsed the behavioral data from the two experiments and analyzed them as a whole, after including the factor Experiment (Exp.1 and Exp.2) in each statistics and checking that no substantive effects or interactions involving this factor were revealed.

To test if performance was modulated by the protocol, we compared the normalized acceleration in the Post to 1 by means of single sample t-tests. Then, our hypotheses were verified through a factorial-design ANOVA including Stimulation (A-tDCS, Ctrl) and Motor Practice (Fast and Slow) as factors. Finally, to calculate the online effect of A-tDCS on the motor practice, we run mixed-design ANOVAs including Stimulation (A-tDCS, Ctrl) as a between-subject factor and Block (from 1 to 6) as a within-subject factor, separately for F-MP and S-MP.

For all analyses, the Kolmogorove Smirnov Test was applied to test for normal distribution of variables, the Greenhousee Geisser correction was used when appropriate and post-hoc comparisons were performed using the Fisher's least significant difference method.

Neurophysiological data

Changes in M1 excitability were evaluated using MEP amplitude, measured peak-to-peak after removing MEPs preceded by muscle activity. The mean MEP value was calculated separately for the two blocks of TMS, i.e., TMS-pre and TMS-post, both for APB and for FDI. For each muscle, data were compared in a single mixed-design ANOVA including Stimulation (A-tDCS, Ctrl) and Motor Practice (Fast and Slow) as a between-subject factors and Time (TMS-Pre and TMS-Post) as a within-subject factor. The Kolmogorove Smirnov Test was applied to test for normal distribution of variables and post-hoc comparisons were performed using the Fisher's least significant difference method.

Correlation between behavioral and neurophysiological data

We investigated whether the MEPs were a good measure of the changes induced by the protocol by correlating them with the behavioral changes. We carried out a Pearson's correlation of the normalized peak acceleration in the post block (Post/Pre) with the normalized MEPs post block (TMS-Post/TMS-Pre), separately for APB and FDI. We considered the data from Experiment 2, for which we had both behavioral and physiological measures.

Questionnaires

For each type of sensation, the sensations scores extracted from the questionnaires were summed to obtain a global sensation score for each task and group condition (max score ¼ 28). For this index

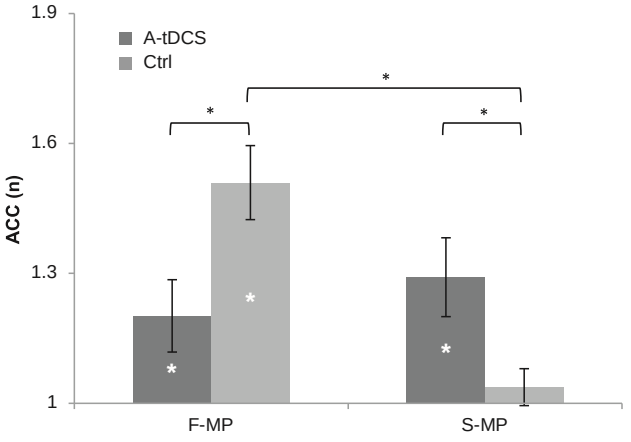

Figure 2. Behavioral results. For each condition, the normalized movement acceleration (ACC (n)) of the Post block is displayed. The black columns represent the A-tDCS condition; the gray columns represent the Ctrl condition. Error bars indicate the mean SEM. Black asterisks indicate a significant difference compared with the other conditions ($P < 0.05$, ANOVA followed by Fisher's LSD post-hoc tests); White asterisks indicate a significant difference between the Post and the baseline ($P < 0.05$, separate t tests for each condition). Although a small increment is evident when participants performed the F-MP ($P < 0.05$), A-tDCS reduced learning compared to the Ctrl stimulation. Differently, when participants performed the S-MP, A-tDCS increased learning compared with the Ctrl condition.

and for the scores on the influence of the stimulation on the motor performance, we applied a generalized linear model with Poisson distribution, which is suitable for discrete dependent variables, including Stimulation (A-tDCS, Ctrl) and Motor Practice (Fast and Slow) as factors.

Results

Behavioral

Overall, we observed that MP leads to motor learning and performance improvement (single sample t-tests). Performance improved after the F-MP both for the Ctrl condition [t_{22} ¼ 5.97, $P < 0.001$] and for the A-tDCS condition [t_{22} ¼ 2.41, $P < 0.05$]. After the S-MP, peak acceleration increased in the A-tDCS condition [t_{22} ¼ 3.20, $P < 0.005$] and not in the Ctrl condition [t_{22} ¼ 0.87, P ¼ 0.39].

These results were confirmed in the (factorial) ANOVA main effect of Motor Practice (Fast vs. Slow), showing that stronger performance improvement occurred after F-MP than after S-MP [$F_{1,84}$ ¼ 6.02, $P < 0.05$]. Crucially, the effect of Motor Practice was dependent on the concurrent Stimulation (A-tDCS vs. Ctrl) [Motor Practice Stimulation; $F_{1,84}$ ¼ 12.94, $P < 0.001$], as shown in Fig. 2. The post-hoc analyses showed that when participants performed the F-MP, A-tDCS reduced movement acceleration compared to the Ctrl condition ($P < 0.01$). Vice versa, A-tDCS increased the peak acceleration compared to the Ctrl condition for the S-MP ($P < 0.05$). Therefore, applying A-tDCS, during the execution of the fast motor learning task resulted in a negative effect and stalled motor learning, whereas applying A-tDCS during the slow motor learning task facilitated motor learning. Moreover, movement acceleration was higher after the F-MP than after the S-MP in the Ctrl condition ($P < 0.001$), whereas no difference was found when A-tDCS was applied (P ¼ 0.42). These results confirmed that the MP per se had different effects on performance improvement, so that fast movements but not slow movements induced learning and shows that the application of A-tDCS changes this pattern.

Looking at the online effects of the stimulation on the motor performance (mixed-design ANOVA on F-MP), we found that the FMP was associated with motor learning [main effect Block (1e6): $F_{5,210}$ ¼ 12.57, ε ¼ 0.56, $P < 0.001$], with the peak acceleration generally increasing from the first block to the last

block of practice. Importantly, the detrimental effect of A-tDCS in the F-MP condition occurred from the beginning of the stimulation (Fig. 3). This was supported by the main effect Stimulation when analyzing motor performance between A-tDCS and Ctrl over the six F-MP blocks in which the stimulation was delivered [$F_{1,42}$ ¼ 4.93, P < 0.05]. Interaction was not significant in this analysis.

Regarding the blocks of S-MP (mixed-design ANOVA on S-MP), we found no significant effects related to the motor practice and the stimulation, nor in the main effects nor in the interactions (all P > 0.06). The lack of a significant learning effect in these blocks suggests that participants complied with the task instructions and performed movements at a constant low speed. Importantly, this pattern was not different between A-tDCS and Ctrl conditions, supporting that the effects observed after the MP could hardly be related to differences in the training across groups.

Neurophysiological data

The results on MEPs confirmed that the A-tDCS interacted with the cortical excitability changes induced by the task, as shown by the results on the APB MEPs. First, the interaction

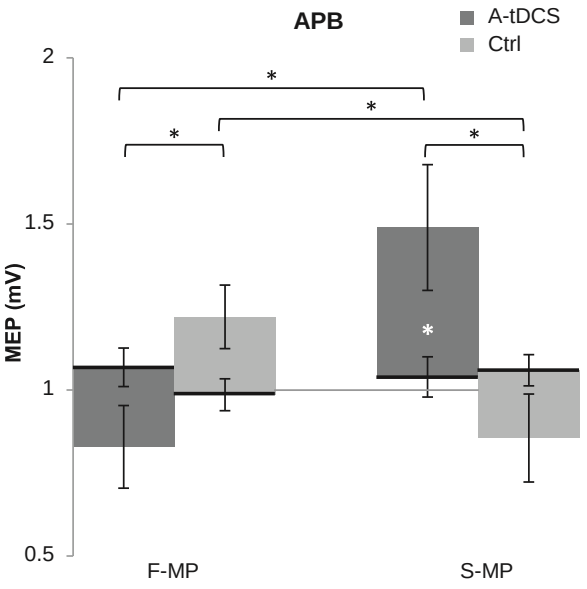

Figure 4. Physiological results. MEPs from the APB muscle are displayed before and after the F-MP and the S-MP, for both A-tDCS and Ctrl. The black horizontal lines represent the MEP value in the TMS-Pre blocks and the columns represent the change in the MEP value in the TMS-Post blocks. The dark gray columns represent the A-tDCS condition; the light gray columns represent the Ctrl condition. Error bars indicate the compared with the other conditions and white asterisks indicate a significant mean SEM. Black asterisks indicate a significant difference in the TMS-Post block significant difference between the TMS-Post and the TMS-Pre (P < 0.05, ANOVA followed by Fisher's LSD post-hoc tests). After the F-MP, MEPs increased in the Ctrl condition compared to A-tDCS. Vice versa, after the S-MP MEP increased if A-tDCS was applied compared to the Ctrl condition.

Stimulation (A-tDCS vs. Ctrl) Motor practice (Fast vs. Slow) was significant [$F_{1,36}$ ¼ 7.35, P < 0.05]. Post-hoc showed that the application of A-tDCS in the S-MP increased MEPs compared with the Ctrl condition (P < 0.05) and compared to the A-tDCS F-MP. Crucially, the interaction Stimulation Motor practice Time (TMS-Pre vs. TMS-Post) was also significant [$F_{1,36}$ ¼ 20.12, P

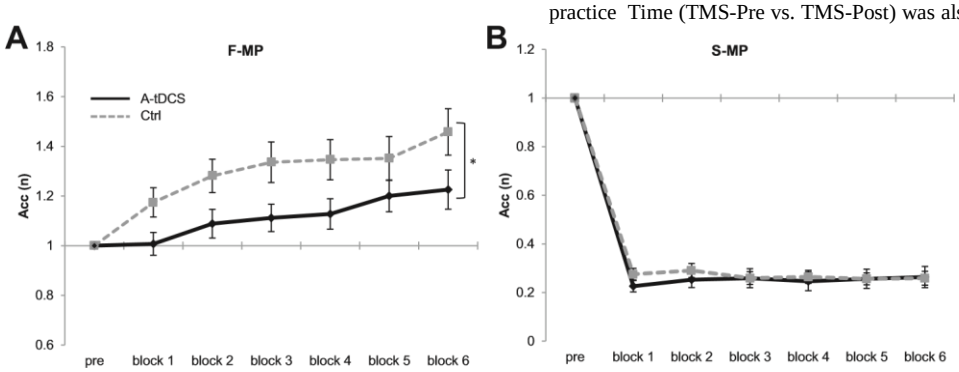

Figure 3. Online effects on behavior. The black diamonds and the continuous line represent the A-tDCS condition; the gray squares and the dashed line represent the Ctrl condition. Error bars indicate the mean SEM. The black asterisks indicate significant main effects of Blocks and Stimulation (P < 0.05, ANOVA) A: The movement acceleration for the fast thumb abduction blocks during F-MP is displayed. The movement acceleration steadily increased from the first block to the last block. If A-tDCS was applied, movement acceleration was overall reduced compared to the Ctrl condition. B: The movement acceleration for the slow thumb abduction blocks during S-MP is displayed. No difference in acceleration was found between the Ctrl and A-tDCS conditions nor any other changes.

< 0.001], as shown in Fig. 4. The interaction was driven by effects in the TMS-Post blocks across conditions, whereas no differences were found across the baseline blocks. After the F-MP, MEPs were higher in the Ctrl condition compared to A-tDCS (P < 0.001), whereas after the S-MP they were higher for the AtDCS compared to the Ctrl condition (P < 0.001). Moreover, MEPs were increased after the F-MP compared to after the S-MP in the Ctrl condition (P < 0.05), confirming that the two MPs employed in the experiments substantially differed for their effects on cortical excitability. When the A-tDCS was applied we observed the opposite pattern, i.e., MEPs were increased for the S-MP compared to the F-MP (P < 0.001). Last, when comparing the TMS-pre with the TMS-post block for each stimulation and task condition we

found that MEPs increased in the TMS-Post when AtDCS had been applied during the S-MP.

These effects were specific for the effector involved in the motor practice, as the MEPs recorded from the FDI only showed two main effects: an increase in the A-tDCS condition compared to Ctrl (Main effect Stimulation [$F_{1,36}$ ¼ 4.41, P < 0.05]) and a decrease after the MP (Main effect Time [$F_{1,36}$ ¼ 8.56, P < 0.01]). No significant interactions were found. These results suggest that the two MPs employed in the experiments substantially differ for their effects on cortical excitability of the muscle involved in the task, i.e., APB, and that the interaction of the stimulation with these excitability changes may determine the final behavioral outcome.

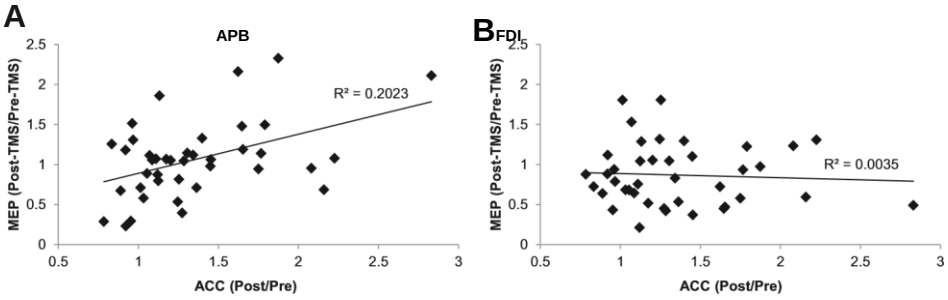

Figure 5. Correlation between MEP amplitude and behavioral data. The correlation between MEP changes (MEP amplitude in TMS-Post/TMS-Pre) and performance improvement (peak acceleration in Post/Pre block) is displayed for APB (A) and for FDI(B). The correlation is significant for APB (P < 0.05), while it is not for FDI.

Correlation between behavioral and neurophysiological data

We found that behavioral changes held significant positive correlation with changes in MEPs for the APB (r ¼ 0.45, P < 0.01) and not for the FDI (r ¼0.06, P ¼ 0.72), as shown in Fig. 5. This suggests that the higher the increment of cortical excitability of the muscle employed in the task, the stronger the improvement in the performance.

Questionnaires

The global sensation perception was significantly higher in the A-tDCS (mean ¼ 4.61) than in the Ctrl (mean ¼ 3.00) [W_1 ¼ 9.64, P < 0.01]. Nevertheless, the interference of the stimulation with the ability of participants to perform the task, according to their reports, was not different across conditions (A-tDCS: S-MP ¼ 0.36 and F-MP ¼ 0.41; Ctrl: S-MP ¼ 0.17 and F-MP ¼ 0.33; all W_1 < 0.99, P > 0.32). Therefore, it is unlikely that the effects were related to differences in the skin sensations induced by the tDCS.

Discussion

Our results highlight the task-dependency of tDCS effects by showing that A-tDCS may either facilitate or inhibit motor learning based on the state of cortical activation at the time of stimulation. In our experiments, A-tDCS was combined with a fast MP task that increases cortical excitability in M1 and improves performance [31,36] as also confirmed by changes in MEPs amplitude. In this case, we found that A-tDCS hindered learning, as indicated by the worsened motor performance during and after A-tDCS application compared to the Ctrl condition. Conversely, A-tDCS significantly enhanced performance when combined with a slow MP task, that does not induce changes in cortical excitability and motor learning per se [32] as showed by MEPs results. Interestingly, the effects on learning resembled the cortical excitability changes revealed by MEPs, highlighting similar effects of tDCS at neurophysiological and behavioral level. These data suggest that the concurrent combination of A-tDCS with another excitability-increasing event can reverse the commonly expected polarity-dependent facilitatory effect of A-tDCS.

To date, the majority of studies have reported that A-tDCS over M1 improves motor learning [13,16,37], and this effect has been related to a gating mechanism, i.e., a decrease in the activity of intracortical inhibitory circuits, eventually leading to long-term changes in synaptic strength [9,10]. To the best of our knowledge, this experiment is the first to show a detrimental effect of A-tDCS on motor learning when the stimulation is simultaneous with motor training. The interaction between the task-induced increase in excitability and the stimulation-induced increase in excitability may explain these findings.

It is worth noting that the motor task employed in this study is different from the previous ones in relation to the cortical activation recruited by the task [38]. Many of the previously employed tasks [14,15,39] involved complex movement sequences and visuo-motor coordination that activate broad brain networks, including motor and premotor areas and subcortical structures [40]. The evidence suggests that learning these tasks produces slow changes in M1 that follow rapid changes in other structures [40]. Conversely, the thumb abduction task in this study is primarily based on activity within M1. Indeed, M1 activity is crucial for the codification of single digit movements and basic movement parameters such as effector selection and the speed of movement [41]. Crucially, repeated finger movements leading to motor learning are associated with increased excitability of M1, even after a few minutes of training [31]. This increase is specific to the motor representation of
the moved finger and not of unmoved fingers [32]. Therefore, in contrast to previous studies, the thumb abduction task relies mainly on M1 activation and may lead to a rapid increase in cortical excitability in M1.

To the best of our knowledge, only one other study has assessed the application of tDCS during simple repetitive finger movements and reported facilitatory effects of A-tDCS [42]. Although it may seem at odds with our results, the differences in movement parameters should be considered. In Galea and Celnik's study, the thumb movements were slower than the thumb abductions employed in our learning-inducing F-MP and much more similar to our S-MP, for which we also found A-tDCS facilitation. Given that the firing rate of neurons in M1 is directly proportional to the movement speed [41,43,44], the F-MP most likely induced much stronger activation in the neural population coding for the thumb abduction than in the S-MP in the present experiment and in Galea and Celnik's (2009) motor task.

Thesedatasuggestthatthestateofthe M1neuralpopulationatthe time of stimulation may be a critical factor for the final effects of tDCS. Accordingly, there is evidence that the combination of A-tDCS with motor activity may have non-additive effects [25] and reduce cortical excitability [24]. Moreover, Schabrun and colleagues have shown that the concurrent combination of two excitability enhancing protocols, e.g., A-tDCS and neuromuscular electrical stimulation, does not induce any cortical excitability enhancement [45].

What type of mechanisms can explain the non-additive or even detrimental effects of A-tDCS during excitability-increasing motor tasks? Understanding the basis of the interaction between tDCS and the concurrent task-associated neural activity could result in optimized tDCS protocols and new strategies to enhance learning.

Although fatigue may be the most direct explanation, we can discard it because the low movement rate used in the F-MP should not induce muscle

fatigue [34]. Additionally, the reduction of learning during the first blocks of the task further weakens this explanation.

A second possible explanation at the local level could be based on metaplasticity phenomena, i.e., activity-dependent mechanisms in which the neural activity at one point in time can modulate the induction of plasticity in a following point in time [46]. These mechanisms, such as homeostatic plasticity, have been previously shown to alter the induction of plasticity by non-invasive brain stimulation techniques, even after a few minutes of stimulation [10,47]. Nevertheless, a crucial aspect of homeostatic plasticity refers to compensatory mechanisms that develop with intense and prolonged plasticity-inducing protocols rather than being instantaneous [9]. When plasticity is inducted in a high proportion of neurons, as in our study, homeostatic plasticity may require from a few to tens of minutes to occur, as shown in in-vitro [48] and in non-invasive brain stimulation studies in healthy participants [23]. In contrast, in this experiment, we found that A-tDCS hindered motor learning even at the earliest stages of training, i.e., as soon as A-tDCS and MP were combined. In this situation, a structure active during "high" excitability levels can be hypothesized: a mechanism that would use a recurrent inhibitory circuit acting during A-tDCS. However, such a circuit would maintain the system within a normal functional range, thus still enabling learning, as in the sham condition. However, this level of learning was not present.

An alternative framework can be used to explain both the facilitatory and the inhibitory effects of A-tDCS on learning. According to the framework of "neural noise", the effects of tDCS depend on the strength of the signal and on the signal-to-noise ratio [26,49e51] where the signal is the neural activity operational to the task and the noise is random neural activity. Because tDCS involves the relatively indiscriminate modulation of a large number of neurons, its impact can be easily understood as a change in the relationship between noise and signal in the neural activity. As tDCS adds neuronal activity to the ongoing neural activity, its effects are state-dependent; neurons that are already activated at a high level by the task (signal) may receive less benefit from an excitatory stimulation, because they are already close to the saturation level. Neurons that are less activated and close to the threshold (signal and/or noise) receive the most benefit from an excitatory stimulation, because their activity can cross the threshold and affect learning processes. Importantly, such changes are magnified by the presence of a system threshold, representing the minimum intensity of a signal for inducing learning. The activity that is below threshold is less relevant for the system and therefore it is somehow further filtered out at neuronal level (e.g., see lateral inhibition mechanisms) while all

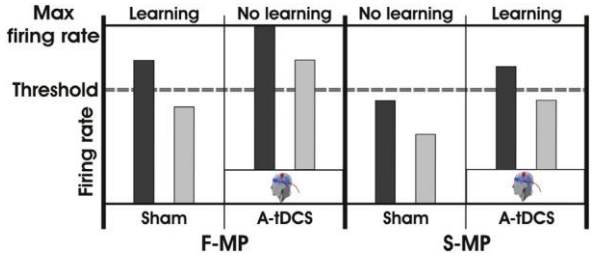

Figure 6. The relationship between the signal (black) related to the task goal activity, i.e., thumb abductions in the slow (S) and fast (F) motor practice (-MP), and other activities that are not associated with the thumb abductions, which can be defined as neuronal noise (gray). The plots on the left depict fast thumb abductions (F-MP), and the plots on the right depict slow thumb abductions (S-MP). In the right plots of each task condition, the final behavioral outcome results from the interaction between the two neuronal patterns: the task-induced activity and A-tDCS induced activity. The threshold represents the minimum intensity of a signal for inducing learning. We can say that the difference between the signal and noise over the threshold will determine the level of accuracy speed in performing the task and therefore the final learning effect.

the suprathreshold activity will influence the final performance. As a result, both facilitatory and inhibitory effects of A-tDCS may be expected. In other words, tDCS may be considered as a tool that injects activity and interacts with the resources involved in performing the task, thus facilitating or hindering task execution depending on the activity pattern of the system at the time of stimulation. Clearly the noise hypothesis is not mutually exclusive with all neurophysiological essential mechanisms that are at the basis of tDCS-induced effects, and its account is mainly valid for the so-called online procedures, i.e., tDCS concurrent with the task.

In relation to our study, the neural activity in M1 during MP may be modeled as a mixture of signal and noise. The level of the signal and the signal-to-noise ratio depend on the specific characteristics of the motor task. Specifically, in the F-MP, the signal is strong because the neurons encoding for thumb abduction have high frequency discharge; in the S-MP, the signal is weak. In the former case, the signal does not benefit from A-tDCS as much as the less activated noise. Therefore, an increase in the noise reaching the threshold will decrease the signal-to-noise ratio and impair performance, as shown in the left panel of Fig. 6. In the latter case, however, the signal is weak and benefits from A-tDCS, and therefore, the performance is improved (Fig. 6 right panel). In conclusion, depending on the neural activation induced by the task in the target area, A-tDCS may facilitate or inhibit learning.

Overall, our results suggest that the effects of tDCS are task dependent and may be non-additive when the task itself induces a strong neural activation in the target area, whereas they are beneficial for mild task-related neural activation. Therefore we can hypothesize that such interaction between task-activation and tDCS-activation may be explained through a neural noise framework.

Acknowledgments

We thank Piercarlo Mauri and Chiara Fanciullacci for their assistance in conducting the experiments.

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