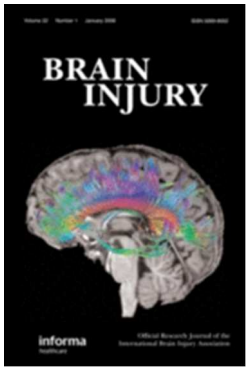

Controlled clinical trial of repeated prefrontal tDCS in chronic patients in minimally conscious state

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Controlled clinical trial of repeated prefrontal tDCS in chronic patients in minimally conscious state

Thibaut A. PhD, Wannez S. MSc, Donneau AF. PhD, Chatelle C. PhD, Gosseries O. PhD, Bruno MA. PhD, Laureys S. MD, PhD

ABSTRACT

Objectives: To assess the effects of repeated transcranial direct current stimulation (tDCS) sessions on the level of consciousness in chronic patients in minimally conscious state (MCS).

Methods: In this randomized double blind sham controlled crossover study, we enrolled 16 patients in chronic MCS. For 5 consecutive days, each patient received active or sham tDCS over the left prefrontal cortex (2mA during 20 minutes). Consciousness was assessed with the Coma Recovery Scale-Revised (CRS-R) before the first stimulation (baseline), after each stimulation (day 1 – day 5), and one week after the end of each session (day 12).

Results: A treatment effect ($p=0.013$; effect size=0.43) was observed at the end of the active tDCS session (day 5) as well as one week after the end of the active tDCS session (day 12; $p=0.002$; effect size=0.57). A longitudinal increase of the CRS-R total scores was identified for the active tDCS session ($p<0.001$) while no change was found for the sham session ($p=0.64$). Nine patients were identified as responders (56%).

Conclusion: Our results suggest that repeated (5 days) left prefrontal tDCS improve the recovery of consciousness in some chronic patients in MCS, up to one week after the end of the stimulations.

Clinical trial: NCT02019615

Keywords:

Minimally conscious state, tDCS, brain injury, disorders of consciousness, brain stimulation

INTRODUCTION

Transcranial direct current stimulation (tDCS) delivers a weak (usually 1–2 mA) electrical current through the brain using two electrodes, an anode and a cathode placed on the scalp [1]. It is presumed that anodal tDCS strengthens synaptic connections through a mechanism similar to long-term potentiation, while cathodal tDCS seems to have an opposite effect [2,3]. Better performances were observed on working memory tasks during and after active tDCS over the left dorsolateral prefrontal (DLPFC) in healthy volunteers and in patients with stroke, Parkinson's disease, and moderate traumatic brain injury [4–7]. Similarly, tDCS over the left DLPF cortex seems to have positive effects on attention in patients with stroke [8] and in patients with mild [9] or severe [10] traumatic brain injury suffering from attentional deficits.

We recently reported an improvement of the level of consciousness in patients with disorders of consciousness (DOC), especially patients in a minimally conscious state (MCS; showing reproducible but inconsistent signs of consciousness [11]), following a single session of DFPLC tDCS [12]. This finding is noteworthy as there are current very few evidence-based guidelines regarding the treatment of patients with DOC [13,14]. Until now, only amantadine has been shown to increase the pace of recovery of patients with severe traumatic brain injury in a subacute population (4-16 weeks post-injury [15]). However, if amantadine enhances the pace of recovery in subacute stage, it may not be as efficient at improving the level of consciousness in patients in a chronic stage [14]. Additionally, amantadine is associated with side effects such as epileptic seizure [16] and may, therefore, not always be supported by the patient [17]. tDCS has been widely studied and there is no severe side-effect of tDCS when applied within the safety

criteria [18]. In this context, tDCS has the advantage to have little to no side effects, even when the stimulations are repeated daily [19,20].

As the effects of a single tDCS stimulation on patients with DOC seem to last a few hours, we here aim to determine whether these short-term effects can be amplified and made more durable by the use of repeated stimulations. Indeed, previous studies on stroke patients or patients with Parkinson disease showed that repeating the number of stimulations (e.g., 5 or 10 days) could increase the duration of the effect from one week to one month [21–23]. Similarly, Angelakis et al. investigated in a prospective case series trial, the effect of repeated tDCS over the left DLPFC or the primary motor cortex in 7 chronic patients in unresponsive wakefulness syndrome (UWS – i.e., eyes opening, but no awareness of self or environment [24,25]) and 3 patients in MCS [26]. No behavioral changes were observed in patients in UWS, while the 3 patients in MCS demonstrated clinical improvement. However, among the patients in MCS, only one received tDCS over the prefrontal cortex. Therefore, in a double blind randomized sham controlled crossover study, we assessed the effects of daily sessions of tDCS over the left DLPFC on the level of consciousness in chronic patients in MCS. We hypothesized that repeated tDCS over the left DLPFC (i.e., 5 consecutive days of stimulation), as compared to sham stimulations, will improve the level of consciousness (as measured by changes in CRS-R total scores) in a sample of chronic patients in MCS. We focused on chronic patients (> 3 months post-insult) to avoid the spontaneous recovery period, which could be a confounding factor. Our second hypothesis is that the effects will last at least one week after the end of the active tDCS session, and that these effects will linearly increase over the 5 days of stimulation. Finally, we hypothesized that the number of responders to repeated tDCS sessions will increase as compared to our first study (i.e., single stimulation).

METHODS

Patients

We prospectively enrolled medically stable patients in chronic MCS (i.e., > 3 months post-insult) between January 2011 and August 2014. The sample size was based on the duration of the ethic committee approval (i.e., four years).

Inclusion criteria were patients of traumatic and non-traumatic etiology in MCS according to published diagnostic criteria [11]. We excluded patients with unclear diagnosis during prescreening assessments and patients with a metallic cerebral implant or pacemaker (in line with the safety criteria for tDCS in human subjects - 17). Patients were studied free of sedative drugs and Na+ or Ca++ channel blockers (e.g., carbamazepine) or NMDA receptor blockers (e.g., dextromethorphan) to avoid interaction with the presumed neuromodulatory effects of tDCS [27]. We did not include patients with a cranioplasty. Medications (two patients were on amantadine), physiotherapy and rehabilitation were kept unchanged throughout the experiment. If the medication and/or rehabilitation were modified during the protocol, the patients had to be excluded of the study, as well as if two consecutive assessments were missing due to clinical purposes (nursing or physical therapy cares needed). Clinically, we defined as responders the patients who showed at least one new sign of consciousness during the 5 days of tDCS, and who kept displaying this behavior one week later, as compared to baseline and sham stimulation.

Material

Each patient received both active and sham DLFPC tDCS sessions in a randomized order. A computer-generated randomization sequence was used to assign in a 1:1 ratio the first session as active tDCS or sham stimulation. For the sham session, the employed tDCS device (Neuroconn

DC Stimulator Plus, NeuroConn GmbH, Ilmenau, Germany) offers a built-in placebo mode, which is activated by an anonymous code number and includes ramp periods of 5 seconds at the beginning and the end of sham stimulation to mimic the somatosensory artifact of active tDCS. Two investigators were involved in data collection. The same investigator performed both tDCS and CRS-R assessments on the same patient. For each patient, the experimenter received two blinded codes from a third person, one for the active stimulation and one for the sham stimulation. Thus, active and sham tDCS could not be identified by the blinded experimenters who administered tDCS and CRS-R, nor by the patients.

Direct current was applied by a battery-driven constant current stimulator using saline-soaked surface sponge electrodes (7x5cm) with the anode positioned over the left DLPF cortex (F3 according to the 10–20 international system for EEG placement - 23) and the cathode placed over the right supraorbital region, as previously described [29]. During tDCS, the current was ramping up to 2 mA (in 5 seconds) from the onset of stimulation and applied for 20 minutes. For the sham condition, the same electrode placement was used as in the active condition, but the current was applied for 5 seconds only, and was then ramped down to 0 mA. Impedances were kept <10 kΩ and voltage <26 V.

tDCS was performed daily, at the same time of the day, for 5 consecutive days. tDCS and sham stimulations were tested in a random order in two different block sessions separated by one week of washout (as published elsewhere - 25 - see figure 1).

INSERT FIGURE 1

tDCS treatment effect was assessed by means of standardized CRS-R assessments performed by two trained and experienced blinded experimenters [31]. The CRS-R consists of 23 hierarchically arranged items (from reflexes – e.g., visual or auditory startle; to more complex voluntary behaviors – e.g., command following, visual pursuit, object manipulation, recognition or localization) comprising 6 subscales assessing auditory, visual, motor, verbal, communication, and arousal functions. Diagnosis is based on the presence or absence of specific behavioral responses to sensory stimuli administered in a standardized manner as described in the guidelines [31]. The lowest item on each subscale represents reflexive activity, whereas the highest items represent cognitively mediated behaviors. Before inclusion in this study, each patient was assessed at least 4 times during a one-week period in order to establish a clear diagnosis. For the protocol, CRS-R assessments were performed directly before the first baseline session and after each active tDCS and sham sessions as well as one week later.

Side effects were evaluated by the experimenters after each stimulation and included (i) the presence of redness of the skin under the electrodes; (ii) signs of discomfort, as assessed by observation of the patient's facial expression (e.g., grimace, tears); and (iii) arousal CRS-R subscale (to assess any possible sedative effect).

Standard Protocol Approvals, Registrations, and Patient Consents

Written informed consents were obtained by the legal representative of each patient. The study was approved by the ethics committee of the University and University Hospital of Liège, Belgium (ClinicalTrials.gov NCT02019615).

Statistics

Statistical analysis was performed using SAS (version 9.3 for Windows) statistical package. The effect of the treatment was analyzed based on the modification of the CRS-R total score. The differences considered in the present study were: [day 5 – baseline] and [day 12 – baseline]. In each situation, the individual data recorded during the crossover study were analyzed according to the method described elsewhere [32] and summarized hereafter. At the group level, we first looked for a period, interaction and treatment effect. The period effect refers to the calculation of active tDCS-sham stimulation response differences, which were then compared according to the order of administration using a Mann-Whitney U test. The interaction effect referred to the calculation of the mean response after active tDCS and sham session, which was then compared according to the period using a Mann-Whitney U test. If no period and interaction effect was found, then treatment effect was assessed using a Wilcoxon match-paired signed-rank test. Results were considered significant at $p<0.05$. Multiple comparisons using Bonferroni correction (6 comparisons) had to be performed for the secondary end-point assessment (i.e., assessment of CRS-R subscale change according to tDCS/sham) and results were considered significant at $p<0.0083$ (i.e., 0.05/6). To evaluate the longitudinal evolution of the CRS-R score between treatment groups, a mixed model with an undefined covariance structure was fitted to the data. The covariates included in the model were the time and the interaction with the treatment indicator. This statistical method allows the comparison of response curves between treatments while accounting for dependency of the data within each patient. The effect size was calculated using the following expression $r=z/\sqrt{2n}$ where z is the statistics obtained from the Wilcoxon signed rank test [33]. Results were considered significant at the 5% critical level ($p<0.05$). Differences between responders and non-responders were assessed using a t-test (i.e., age and time since insult) and a Chi square test (i.e., etiology).

RESULTS

We assigned 21 eligible patients to receive both active and sham tDCS in a crossover study design. Two patients were excluded from the study after the first washout period because of medical complications that required a modification of medication (one patient had a pulmonary infection and the other had epileptic seizures). Three other patients were also excluded from the study due to missing CRS-R assessments for two consecutive days (i.e., incomplete, missing or delayed – more than 1 hour – CRS-R assessments due to nursing cares or physical therapy cares for pulmonary congestion, needed after the stimulation session – see figure 2). The 5 drop-outs did not differ from the others in terms of age (p=0.443), time since injury (p=0.515) or baseline CRS-R (p=0.669).

INSERT FIGURE 2 HERE

Sixteen patients completed the study (mean age of 47 [17-74] years; 9 men; interval since insult: 85 [5-365] months; 11 post-traumatic, 5 non-traumatic - i.e., anoxic and stroke). Demographic data are reported in table 1. Nine patients first received active tDCS and seven patients first received sham stimulation. There was no significant clinical or demographic difference between the groups. No period or interaction effects were observed for the comparison between the CRS-R score at baseline and after 5 days of active tDCS, neither for the comparison between the CRS-R score at baseline and one week after the last stimulation. At the group level, a difference was observed between the two treatment conditions at day 5 (p=0.013) as well as one week after the last stimulation (p=0.002) (figure 3).

INSERT TABLE 1 AND FIGURE 3 HERE

We did not observe any significant effect of active tDCS on any of the six CRS-R subscales.

When we looked at the longitudinal change of the CRS-R scores, an improvement of the CRS-R total scores was found for the active tDCS session across time ($p<0.001$), while no change was observed under the sham session ($p=0.64$, figure 4).

At the individual level, 9 out of the 16 patients in MCS (56%) behaviorally improved only following the active tDCS (i.e., tDCS responder; new sign of consciousness never observed at baseline or during sham session). The recovery of signs of consciousness included response to command, recognition and localization of objects, automatic motor response and visual fixation/pursuit. Functional communication, which is a criterion of the emergence of MCS, was also observed in two patients after active tDCS. Behavioral improvements for tDCS responders are detailed in table 2. Four patients responded after the first stimulation session (i.e., 25%, which is similar to the 23% observed in our first study [12]). The other 5 responders improved behaviorally after 2 (n=2), 3 (n=1) or 4 days (n=2) of tDCS. When comparing responders' and non-responders' demographic data, we did not identify any difference in term of age ($p=0.788$), time since insult ($p=0.683$) or etiology ($p=0.930$).

INSERT FIGURE 4

No side effect was observed after any of the stimulation. Four patients, however, showed redness of the skin following both active and sham tDCS stimulation but it disappeared within 30 minutes. We did not find any difference for the arousal subscales before and after the stimulation sessions. No patient showed signs of discomfort. No seizures occurred during the stimulation sessions, even in patients treated for epilepsy.

DISCUSSION

We identified a positive effect of repeated tDCS over the left DPFC on level of consciousness in chronic patients in MCS. In addition, these effects lasted at least one week after the last stimulation. Our results are in line with previous studies reporting a positive short lasting effect (one or two hours) of tDCS on cognition [5,6,34,35] as well as the longer lasting effect (from one week up to one month) associated with repeated stimulations, in pain [36], stroke [22,37] and Parkinson disease [21].

As mentioned in the introduction, a previous case series study reported clinical improvement in 10 chronic (>6 months) patients with DOC after repeated tDCS over the left DLPFC (n=5) or the primary motor (n=5) cortices [26]. The only patient in MCS who received tDCS over the left DLPF cortex showed a behavioral improvement characterized by the recovery of pain localization. The reappearance of this particular behavior was not observed in our cohort of 9 responders who presented various new signs of consciousness. This finding highlights a high heterogeneity of tDCS effects on patients in MCS.

When we looked at the longitudinal changes on the CRS-R scores, we observed a significant increase over time (from day 1 to 12). In addition, we observed an increase in the effect

size when comparing the first (single) stimulation (i.e., 0.38) [12] to the fifth one (i.e., 0.43). The effect size also increased at day 12 (i.e., one week after the end of the stimulation – 0.57), suggesting that 5 days of stimulation increase the duration and the strength of tDCS clinical effects.

Some authors hypothesized that repeating tDCS every day could improve cortico-cortical excitability and therefore, strengthen the effect of the stimulation [36]. One study showed that tDCS induced greater motor evoked potential amplitude in healthy subjects when delivered every day rather than every other day [38]. This could reflect superior cumulative effects between stimulation rather than a greater response to each individual tDCS [38]. These studies are in line with our observation, since we identified an increased number of responders with the number of stimulations, together with an increased duration of the effect, lasting up to one week after the last stimulation.

Another study investigating the effect of tDCS (over the primary motor cortex) on human consciousness in REM sleep, demonstrated that tDCS could influence motor imagery during this stage of sleep [39]. The authors also suggested that, since REM sleep is involved in motor development and the preparation of movements, tDCS could be used to stimulate motor function, and especially in patients who suffered from immobilization, such as severely brain-injured patients with DOC, highlighting another potential benefit of tDCS for the rehabilitation of patients with DOC.

In our previous study using a single stimulation, we observed that 43% of MCS (in both acute and chronic stage) were responsive to tDCS [12]. However, when looking at the chronic MCS, only 23% responded to tDCS. In the present study, we noticed that 25% (n=4) of our

sample responded after the first stimulation, which is in line with our previous results. Interestingly, we observed that five other patients showed improvement after 2, 3 or even 4 days of stimulation, resulting in 56% of responders after 5 days of stimulation. These results suggest that repeating tDCS daily could increase the number of responders, and that the first session is not predictive of a future positive effect of the stimulation on the level of consciousness.

The increased effects of tDCS over the sessions could be due to an increase in NMDA receptor excitability, which could improve and strengthen cortical excitability within the stimulated area [1,2]. More distant areas also seem to be involved in tDCS responsiveness. For patients in MCS, we recently identified that responders to a single session of tDCS showed more grey matter preservation and residual metabolic activity, as compared to non-responders, in the stimulated area (i.e., left DLPFC), in the precuneus, and in the thalamus; areas known to be involved in conscious processes [40]. These results suggest that not only the stimulated area but also areas implicated in consciousness are involved in the mechanisms of action and efficacy of tDCS in patients with severe brain injured and DOC [41]. However, the stimulated area and the consciousness network need to remain at least partially preserved metabolically. Recently, another study also showed that tDCS could be used as a diagnostic tool to disentangle patients in MCS from UWS [42]. The authors identified that active tDCS induced an increase in cortical connectivity and excitability (measured by transcranial magnetic stimulations) in patients in MCS, while improvement was only observed in patients clinically diagnose in UWS who showed recovery to MCS at follow-up. This study showed that, beside the treatment effect of tDCS, this technique could be useful to detect residual connectivity markers in clinically UWS patients who may recover behavioral signs of consciousness later on.

As supported by our findings and previous studies, it is well-known that tDCS is a low risk technique [18,43]. In the 16 patients who completed the study, no seizure or sign of potential pain (e.g., grimace, tears) was observed. No complication related to the protocol occurred during the active tDCS or the sham sessions. Four patients had moderate redness of the skin that disappeared within 30 minutes. tDCS did not have any effect on the level of arousal on any of the patients. Those observations are reassuring as they suggest that tDCS may be applied safely in daily clinical practice. Nevertheless, further studies need to be performed to assess the long-term effect (e.g., 1 month, 3 months, 6months) of repeated tDCS in patients with severe brain injury.

Our study has several limitations. Firstly, we only performed one follow-up assessment after one week. It would thus be useful in future tDCS studies to conduct follow-up testing at longer intervals to determine whether treatment effects can last more than one week after treatment. In addition, even if the interaction effect was not statistically significant, it seems to be a trend toward a carry-over effect, and this suggests that future studies would need to include a longer washout period. Another limitation is the small sample of patients included. We were not able to recruit more than 21 patients for this protocol during a four-year period. Therefore, multi-centric and international studies will be necessary to replicate and confirm the results in a larger population of patients.

In conclusion, our results suggest that repeated (5 days) tDCS applied over the left prefrontal cortex seem to be safe and can enhance the level of consciousness in some chronic patients in MCS. Moreover, the effects appeared to last at least one week after the end of the stimulations. In addition, the first session was not predictive of a future positive effect of tDCS on the level of consciousness as the number of responders doubled after 5 days of repeated active tDCS as compared with the first day of stimulation. Even though our findings are based on a small

sample size, these preliminary results strongly support the need to further investigate the use of tDCS as a therapeutic intervention in patients with DOC.

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Figures and tables

Figure 1: Protocol of the study. CRS-R=Coma Recovery Scale-Revised; tDCS=transcranial direct current stimulation, BL=baseline, d=day. Active and sham tDCS sessions were randomized.

Figure 2: Study flowchart.

Figure 3: Boxplot of active tDCS (in grey) and sham tDCS (in white) at day 5 and day 12 (i.e., one week after the end of the last stimulation). Black lines represent the medians of the delta of the Coma Recovery Scale-Revised (CRS-R) total score between baseline and after tDCS (active or sham); boxes represent the interquartile range; dashed lines represent minimum and maximum. * $p<0.05$.

Figure 4: Longitudinal evolution of CRS-R total scores (median – IQR) from BL (baseline) to d12 (day 12 – one week after the end of the stimulations) for the tDCS session (black) and the sham session (grey). A significant positive linear evolution was identified for the active tDCS session but not for the sham session.

Table 1: Demographic data and Coma Recovery Scale-Revised scores.

Table 2: List of CRS-R responses recovered following tDCS in the 9 responders.

rtDCS in minimally conscious state

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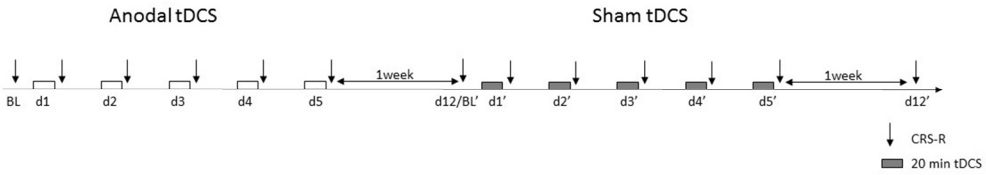

Figure 1: Protocol of the study. CRS-R=Coma Recovery Scale-Revised; tDCS=transcranial direct current stimulation, BL=baseline, d=day. Active and sham tDCS sessions were randomized.
296x53mm (96 x 96 DPI)

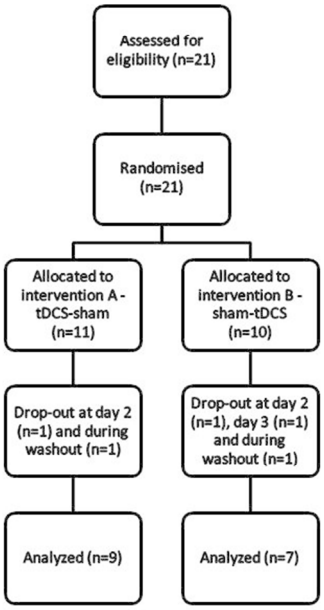

Figure 2: Study flowchart
254x190mm (96 x 96 DPI)

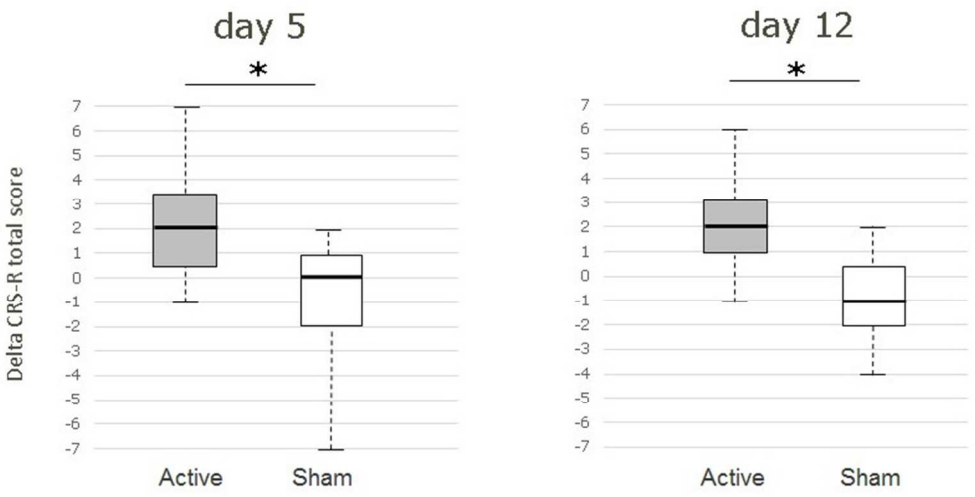

Figure 3: Boxplot of active tDCS (in grey) and sham tDCS (in white) at day 5 and day 12 (i.e., one week after the end of the last stimulation). Black lines represent the medians of the delta of the Coma Recovery Scale-Revised (CRS-R) total score between baseline and after tDCS (active or sham); boxes represent the interquartile range; dashed lines represent minimum and maximum. * $p<0.05$.
213x107mm (96 x 96 DPI)

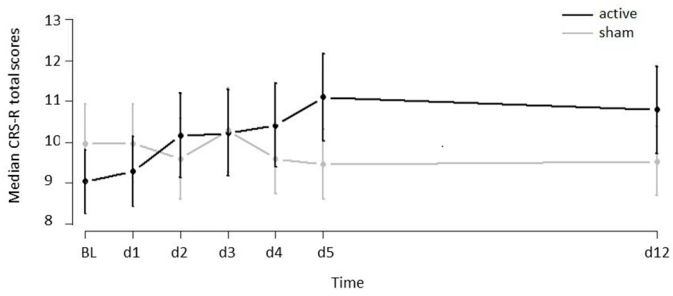

Figure 4: Longitudinal evolution of CRS-R total scores (median – IQR) from BL (baseline) to d12 (day 12 – one week after the end of the stimulations) for the tDCS session (black) and the sham session (grey). A significant positive linear evolution was identified for the active tDCS session but not for the sham session.
338x190mm (96 x 96 DPI)

Demographic data and Coma Recovery Scale-Revised scores.

ID	Age (sex)	Etiology	Structural brain lesions (MRI or CT)	Time since insult (months)	Session	Baseline CRS-R	Post d1 CRS-R	Post d2 CRS-R	Post d3 CRS-R	Post d4 CRS-R	Post d5 CRS-R	Post d12 CRS-R	Delta baseline – d5	Delta baseline – day 12
1*	17 (M)	TBI	Frontal and temporal, diffuse cortical atrophy	21	Sham	7	7	7	9	7	9	6	2	-1
					Anodal	6	9	6	11	5	9	9	3	3
2*	32 (F)	TBI	Bilateral frontal and moderate diffuse cortical atrophy. Right fronto-parietal craniotomy.	75	Anodal	11	12	14	14	14	14	14	3	3
					Sham	14	13	14	14	10	7	11	-7	-3
3*	74 (F)	Anoxic stroke	Bilateral moderate diffuse cortical atrophy	11	Sham	7	7	6	6	7	7	6	0	-1
					Anodal	6	8	6	8	8	8	8	2	2
4	35 (M)	TBI	Bilateral moderate diffuse cortical atrophy	54	Anodal	6	8	6	5	5	6	5	0	-1
					Sham	5	4	6	6	4	6	4	1	-1
5*	50 (M)	TBI	Moderate right fronto-temporal cortical atrophy.	88	Sham	13	13	10	9	14	13	13	0	0
					Anodal	13	13	14	14	14	17	17	4	4
6	40 (M)	TBI	Right posterior lesion.	243	Sham	11	12	14	15	13	12	13	1	2
					Anodal	13	12	14	12	15	15	14	2	1
7	31 (F)	TBI	Bilateral temporo-parietal diffuse cortical atrophy.	20	Anodal	8	6	6	8	9	7	9	-1	1
					Sham	9	7	7	9	8	10	7	1	-2
8	56 (F)	TBI	Frontal and temporal, diffuse cortical atrophy (L>R). Left parietal craniotomy	20	Anodal	14	14	15	14	16	15	14	1	0
					Sham	14	14	12	12	14	14	14	0	0
9*	65 (F)	CA	Bilateral severe diffuse cortical atrophy	5	Sham	3^{+}	4	4	4	3	4	5	1	2
					Anodal	5	5	5	5	7	7	7	2	2
10	49 (F)	CA	Bilateral moderate diffuse cortical atrophy (L>R)	15	Anodal	5	6	8	5	7	9	8	4	3
					Sham	8	6	6	6	7	6	6	-2	-2
11*	54 (F)	TBI	Right fronto-	21	Sham	13	13	14	13	13	13	13	0	0

			temporo-parietal cortical atrophy		Anodal	13	15	15	14	15	15	15	2	2
12*	35 (M)	CA	Bilateral severe diffuse cortical atrophy	146	Anodal	8	8	11	9	10	13	13	5	5
					Sham	13	10	10	9	9	9	9	-4	-4
13*	29 (M)	TBI	Left fronto-temporo-occipital atrophy	134	Anodal	13	15	18	19	18	20	19	7	6
					Sham	19	18	17	19	18	16	15	-3	-4
14	25 (M)	TBI	Bilateral moderate cortical atrophy	17	Sham	8	11	12	11	8	8	9	0	1
					Anodal	9	8	10	8	7	8	8	-1	-1
15	59 (M)	Anoxic stroke	Right occipital cortical atrophy	17	Anodal	8	8	14	8	8	8	11	0	3
					Sham	11	12	9	14	13	12	12	1	1
16*	42 (M)	TBI	Moderate left temporal-parietal cortical atrophy	365	Anodal	7	7	8	10	10	10	9	3	2
					Sham	9	10	9	7	9	7	7	-2	-2

Demagraphic data and Coma Recovery Scale-Revised (CRS-R) total scores of patients during anodal tDCS and sham tDCS sessions. TBI=traumatic brain injury, CA=cardiac arrest, * =responder, $^{+}$=patient previously diagnosed as MCS but UWS for the first CRS-R.

Table2: List of CRS-R responses recovered following tDCS in the 9 responders.

Items	Detailed responses during the CRS-R assessment	Number of patients
Systematic command following	4 out of 4 responses at “move your right arm”	1
Reproducible command following	3 out of 4 responses at “move your right hand/fingers”	1
Sound localization*	Turn head when presenting the own name behind the head of the patient at least twice	1
Object recognition	Recognize a comb and/or a cup, visually on 3 or more occasions	2
Automatic motor reaction	Spontaneous motor reaction (i.e., grab bed sheet)	1
Visual pursuit	Follow mirror on at least 2 occasions in the same direction	2
Visual fixation	Fixate a ball on at least 2 occasions	1
Object localization	Reaching ball with the hand on demand at 3 occasions	1
Functional communication+	Accurately respond to 6 autobiographical yes/no questions (e.g., is your name Patrick, do you have 32 years old, is your father’s name Christopher)	2

CRS-R: Coma Recovery Scale-Revised
* does not denotes MCS
+ denotes EMCS if observed on two consecutive assessments

CONSORT 2010 checklist of information to include when reporting a randomised trial*

Section/Topic	Item No	Checklist item	Reported on page No
Title and abstract			
	1a	Identification as a randomised trial in the title	P1
	1b	Structured summary of trial design, methods, results, and conclusions (for specific guidance see CONSORT for abstracts)	P1
Introduction			
Background and objectives	2a	Scientific background and explanation of rationale	P3-4
	2b	Specific objectives or hypotheses	P4
Methods			
Trial design	3a	Description of trial design (such as parallel, factorial) including allocation ratio	P6-7
	3b	Important changes to methods after trial commencement (such as eligibility criteria), with reasons	P 9
Participants	4a	Eligibility criteria for participants	P5
	4b	Settings and locations where the data were collected	P5
Interventions	5	The interventions for each group with sufficient details to allow replication, including how and when they were actually administered	P6
Outcomes	6a	Completely defined pre-specified primary and secondary outcome measures, including how and when they were assessed	P6-7
	6b	Any changes to trial outcomes after the trial commenced, with reasons	/
Sample size	7a	How sample size was determined	P5
	7b	When applicable, explanation of any interim analyses and stopping guidelines	/
Randomisation:			
Sequence generation	8a	Method used to generate the random allocation sequence	P5
	8b	Type of randomisation; details of any restriction (such as blocking and block size)	P5
Allocation concealment mechanism	9	Mechanism used to implement the random allocation sequence (such as sequentially numbered containers), describing any steps taken to conceal the sequence until interventions were assigned	P5-6
Implementation	10	Who generated the random allocation sequence, who enrolled participants, and who assigned participants to interventions	P6
Blinding	11a	If done, who was blinded after assignment to interventions (for example, participants, care providers, those	P6

		assessing outcomes) and how	
	11b	If relevant, description of the similarity of interventions	P6
Statistical methods	12a	Statistical methods used to compare groups for primary and secondary outcomes	P8
	12b	Methods for additional analyses, such as subgroup analyses and adjusted analyses	P8
Results			
Participant flow (a diagram is strongly recommended)	13a	For each group, the numbers of participants who were randomly assigned, received intended treatment, and were analysed for the primary outcome	P9
	13b	For each group, losses and exclusions after randomisation, together with reasons	P9
Recruitment	14a	Dates defining the periods of recruitment and follow-up	P5
	14b	Why the trial ended or was stopped	/
Baseline data	15	A table showing baseline demographic and clinical characteristics for each group	Table1
Numbers analysed	16	For each group, number of participants (denominator) included in each analysis and whether the analysis was by original assigned groups	Figure 2
Outcomes and estimation	17a	For each primary and secondary outcome, results for each group, and the estimated effect size and its precision (such as 95% confidence interval)	P9-10
	17b	For binary outcomes, presentation of both absolute and relative effect sizes is recommended	/
Ancillary analyses	18	Results of any other analyses performed, including subgroup analyses and adjusted analyses, distinguishing pre-specified from exploratory	P10-11
Harms	19	All important harms or unintended effects in each group (for specific guidance see CONSORT for harms)	P11
Discussion			
Limitations	20	Trial limitations, addressing sources of potential bias, imprecision, and, if relevant, multiplicity of analyses	P14
Generalisability	21	Generalisability (external validity, applicability) of the trial findings	P14-15
Interpretation	22	Interpretation consistent with results, balancing benefits and harms, and considering other relevant evidence	P14-15
Other information			
Registration	23	Registration number and name of trial registry	P7
Protocol	24	Where the full trial protocol can be accessed, if available	P7
Funding	25	Sources of funding and other support (such as supply of drugs), role of funders	/

*We strongly recommend reading this statement in conjunction with the CONSORT 2010 Explanation and Elaboration for important clarifications on all the items. If relevant, we also recommend reading CONSORT extensions for cluster randomised trials, non-inferiority and equivalence trials, non-pharmacological treatments, herbal interventions, and pragmatic trials. Additional extensions are forthcoming: for those and for up to date references relevant to this checklist, see www.consort-statement.org.