



Cerebellar tDCS does not affect performance in the N-back task

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ABSTRACT

The *N-back task* is widely used in cognitive research. Furthermore, the cerebellum's role in cognitive processes is becoming more widely recognized. Studies using transcranial direct current stimulation (tDCS) have demonstrated effects of cerebellar stimulation on several cognitive tasks. Therefore, the aim of this study was to investigate the effects of cerebellar tDCS on cognitive performance by using the N-back task. The cerebellum of 12 participants was stimulated during the task. Moreover, the cognitive load was manipulated in $N = 2$, $N = 3$, and $N = 4$. Every participant received three tDCS conditions (anodal, cathodal, and sham) divided over three separated days. It was expected that anodal stimulation would improve performance on the task. Each participant performed 6 repetitions of every load in which correct responses, false alarms, and reaction times were recorded. We found significant differences between the three levels of load in the rate of correct responses and false alarms, indicating that subjects followed the expected pattern of performance for the N-back task. However, no significant differences between the three tDCS conditions were found. Therefore, it was concluded that in this study cognitive performance on the N-back task was not readily influenced by cerebellar tDCS, and any true effects are likely to be small. We discuss several limitations in task design and suggest future experiments to address such issues.

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The N-back task is a widely used cognitive task that measures working memory capacity (Gevins & Cutillo, 1993; Jaeggi, Buschkuhl, Perrig, & Meier, 2010; Jonides et al., 1997; Veltman, Rombouts, & Dolan, 2003). In its basic form, stimuli are sequentially presented, and the participant has to decide whether the currently presented stimulus is the same as the one presented one, two, or more trials before. By increasing the number of trials between the current trial and the relevant trial before, referred to as N , the task becomes more difficult, which is known as increasing the cognitive load. Imaging studies have shown involvement of the left prefrontal cortex (PFC) in the N-back task (D'Esposito et al., 1998; Owen, McMillan, Laird, & Bullmore, 2005). With increasing N , activity in this area increases as well (Veltman et al., 2003). Moreover, stimulation of the PFC using transcranial magnetic stimulation (TMS) has shown to

modulate performance on the N-back task (Mottaghy, 2006).

Transcranial direct current stimulation (tDCS) is an emerging technique to investigate the relationship between specific brain areas and behavior (Nitsche & Paulus, 2011). Several studies on various cognitive tasks have observed modulatory effects of tDCS on task performance. Both anodal and cathodal stimulation on various brain areas have been found to have modulatory effects on various cognitive tasks (Jacobson, Koslowsky, & Lavidor, 2012).

With respect to the N-back task, a few studies have observed improvements of performance after anodal stimulation of the left PFC in the N-back task on accuracy (Fregni et al., 2005; Martin et al., 2013) and reaction time (Teo, Hoy, Daskalakis, & Fitzgerald, 2011). tDCS changes cortical excitability by delivering a weak current (between 1 and

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2 mA) through the scalp, which can have prolonged effects on task performance (Dayan, Censor, Buch, Sandrini, & Cohen, 2013). For example, anodal stimulation over the primary motor cortex enhances performance on a motor task as reaction times decreased over time (Nitsche et al., 2003). tDCS on the PFC has been shown to affect performance in several cognitive tasks, including the N-back task (Gladwin, den Uyl, Fregni, & Wiers, 2012; Martin et al., 2013), but also see the recent review by Horvath, Forte, and Carter (2015). Enhancement of performance in the N-back task using tDCS on the left PFC was observed with higher cognitive loads only, which may indicate the contribution of the PFC in complex cognitive and working memory tasks.

Over the past few decades, interest in the role of the cerebellum in cognition, in addition to its known importance in motor control, has increased (Hayter, Langdon, & Ramnani, 2007; Schmahmann & Sherman, 1998). Anatomically, the cerebellum is reciprocally connected to various areas of the cortex, including the motor cortex and the prefrontal cortex via independent loops (Kelly & Strick, 2003), which suggests that the cerebellum supports the motor and cognitive processes carried out by these cortical areas (Ramnani, 2006). A cerebellar hemisphere is connected to the contralateral hemisphere of the cortex. Lesion studies confirm the idea of cerebellar involvement in cognition, by showing that right posterior damage to the cerebellum leads to cognitive deficits, in particular executive function, verbal working memory, and attentional processes (Timmann & Daum, 2007). Patients with cerebellar lesions have lower scores in attention and working memory tasks than healthy subjects (Gottwald, Wilde, Mihajlovic, & Mehdorn, 2004). In addition, children with cerebellar tumors show impairment of development of cognitive functions (Scott et al., 2001).

More evidence of cerebellar involvement in cognition comes from neuroimaging studies. Positron emission tomography (PET) and functional magnetic resonance imaging (fMRI) studies show cerebellar activity in many tasks involving various cognitive processes like selective attention, visual and phonological working memory, and semantic memory retrieval (Cabeza & Nyberg, 1997; Stoodley, 2011). In a memory task, increases in cognitive load are related to more cerebellar activation (Kirschen, Chen, Schraedley-Desmond, &

Desmond, 2004). In that study, participants first had to memorize a set of stimuli of increasing load (two or six letters) and later had to decide which of two shown stimuli was present in the set they saw earlier.

Cerebellar activity during an auditory version and a visual version of the N-back task has also been reported in fMRI studies (Hautzel, Mottaghy, Specht, Muller, & Krause, 2009; Salmi et al., 2010). In the visual version, participants performed a two-back task with both letters and abstract figures. In both tasks, left and right cerebellar activity was observed. In the auditory task, participants performed an N-back task with different pitched chords. Changing the task from a one-back task to a two-back task increased cognitive load. Significant load-dependent activations were observed in both the left and the right cerebellum. More cerebellar activation, particularly on the right side, was observed with higher cognitive loads (Jonides et al., 1997; Kirschen et al., 2004; Salmi et al., 2010). Finally, as with the PFC, TMS on the right superior cerebellum increases the reaction times (but not accuracy) of a working memory task (Desmond, Chen, & Shieh, 2005).

The goal of the present study is to examine the effects of cerebellar tDCS on the N-back task. Similar to the previously observed effects of anodal left PFC stimulation (Fregni et al., 2005; Teo et al., 2011), we hypothesized that anodal right cerebellar stimulation would improve performance as indicated by more hits, fewer false alarms, and/or faster reaction times than for sham stimulation or cathodal stimulation. Cathodal stimulation might even be detrimental to performance, increasing, for instance, reaction times. We also expect a bigger effect of tDCS with a higher cognitive load. A within-subjects design is used to avoid confounds of individual differences.

Method

Participants

Twelve healthy people (6 females) gave informed consent prior to their participation in this study, which consisted of three experimental sessions. Ages ranged between 18 and 45 years ($M = 29.9$ years, $SD = 11.0$ years). All subjects came from the general population, (had) attended at least a high school, and were without any known neurological or psychiatric disturbances.

Participants did not receive a reward for their participation. All procedures performed were in accordance with the ethical standards of the institutional research committee and with the 1964 Helsinki declaration and its later amendments. The study took place at the Department of Neuroscience at the Erasmus Medical Centre in Rotterdam.

Task and stimuli

The N-back task was implemented in MatLab (R2010a, Version 7.10.0.499) based on the version used by Hoy et al. (2013) and by Thurling et al. (2012) and presented on a laptop (model Sony Vaio VPCEA3S1E, 14").

The experiment consisted of three sessions run on separate days. In a single session, 18 blocks of 48 trials each were presented. The participant started a block by pressing a key, allowing him or her to take a break between blocks. In a single trial, a single letter was presented for 500 ms in the center of the screen, followed by a blank screen for 1000 ms. The letter was an A, B, C, D, or E. The participants were instructed to press a key on the keyboard of the laptop as fast as possible when they thought the letter was the same as that N trials earlier. The participants had to respond within 1 s after onset of the trial. The value of N determined the load of the N-back task. Within each block, 25 trials required a key press to be denoted a correct trial—that is, in 25 trials the letter was the same as that N trials before.

The load of the block (N) was given before each block of 48 trials. The load could be two, three, or four. Each of the three loads was presented six times in each session (referred to as repetitions). The order of the different loads was pseudorandomized across blocks so that no load was presented twice in a row. The order of loads was the same for all three sessions.

For each key press, it was determined whether it was a correct response (hit), or an incorrect response (false alarm). The reaction time, relative to the onset of a trial, was also determined.

Transcranial direct current stimulation (tDCS)

In an experimental session, subjects received anodal, cathodal, or sham cerebellar tDCS. tDCS was delivered by a DC stimulator (Neuroconn GmbH, Ilmenau, Germany) connected to a pair of 12-mm

sintered silver/silver chloride (Ag/AgCl) ring electrodes. The stimulation electrode was placed over the right cerebellar hemisphere (3 cm lateral to the inion), and the reference electrode was placed on the left buccinator muscle (similar to Verhage, Avila, van der Geest, Frens, & Donchin, 2014). Anodal or cathodal direct current at 2 mA intensity was started 3 min before the first block and lasted the whole session. When stimulation started, all participants felt the current under both electrodes as a mild itching sensation.

This sensation disappeared after a few seconds. In the sham condition, current was only applied for 30 s to give participants the same sensation without affecting brain processes (Gandiga, Hummel, & Cohen, 2006; Nitsche & Paulus, 2000).

In all three groups, a gradual ramp up and ramp down of the current in 30 seconds reduced unpleasant side effects. Participants could not distinguish sham and real tDCS conditions. tDCS started three minutes prior to the task in order for stimulation to be applied throughout performance of the task.

Design

Before performing the actual experiment, participants performed 30 practice trials for each load. During these practice trials, feedback was provided. When a false alarm was detected, a red "X" was displayed in the center of the screen. When they missed a target, the word "miss" was displayed. When a correct response was made, nothing was displayed. After the practice session, the actual experimental session started, and the tDCS stimulator was turned on. The stimulation was administered during the experiment for 20 min.

Each participant ran three experimental sessions. Across these sessions, they received three tDCS conditions (anodal, cathodal, and sham), separated by at least five days between the sessions, to avoid carryover effects of the stimulation. The order of tDCS stimulation was randomized according to a Latin square design and was counterbalanced across subjects.

Data analysis

For each block of 48 trials, the number of hits and false alarms was calculated, as well as the average reaction time of the correct responses. The reaction times represent the reaction times on the hits,

not on the false alarms. Data were analyzed in SPSS 19 using repeated measures analyses of variance (ANOVAs) with three within-subjects factors: load (3 levels: $N = 2$, $N = 3$, or $N = 4$), repetition (6 levels), and tDCS condition (3 levels: anodal, cathodal, or sham). In case of sphericity violations, we report corrected estimations of the degrees of freedom. Post hoc tests were done using Bonferroni correction. The three outcomes measures (hits, false alarms, and reaction times) were analyzed separately. All values are reported as means \pm standard deviations. The threshold of significance was set at 5% ($\alpha = .05$).

Results

Figure 1 shows the task performance over the six blocks per load (N) for the three conditions of tDCS stimulation. There were main effects of load on task performance (Table 1). On average, increasing the load reduced the number of hits, $F(1.13, 12.41) = 51.95$, $p < .001$, $\eta_p^2 = .83$, and increased the number

of false alarms, $F(2, 22) = 18.07$, $p < .001$, $\eta_p^2 = .62$. There was no main effect of load on reaction times, $F(2, 22) = 0.106$, $p = .80$, $\eta_p^2 = .01$.

The main effects of tDCS stimulation on performance were not significant [hits: $F(2, 22) = 0.17$, $p = .80$, $\eta_p^2 = .02$; false alarms, $F(2, 22) = 1.12$, $p = .34$, $\eta_p^2 = .09$; reaction times, $F(2, 22) = 1.13$, $p = .30$, $\eta_p^2 = .09$]. Furthermore, none of the interactions involving tDCS stimulation were significant (all $p > .30$ with effect sizes < 0.1). In addition, we found no effect of either stimulation condition when compared directly to sham using paired t tests (Table 2).

The main effects of repetition on performance were also not significant [hits: $F(5, 55) = 1.56$, $p = .18$, $\eta_p^2 = .12$; false alarms: $F(5, 55) = 1.82$, $p = .124$, $\eta_p^2 = .14$; reaction times, $F(5, 55) = 1.46$, $p = .20$, $\eta_p^2 = .12$], indicating that performance did not improve over blocks. The interaction between load and repetition was significant for hits, $F(10, 110) = 22.77$, $p < .001$, $\eta_p^2 = .23$, and false alarms, $F(2, 22) = 18.07$, $p < .01$, $\eta_p^2 = .62$, but not for reaction

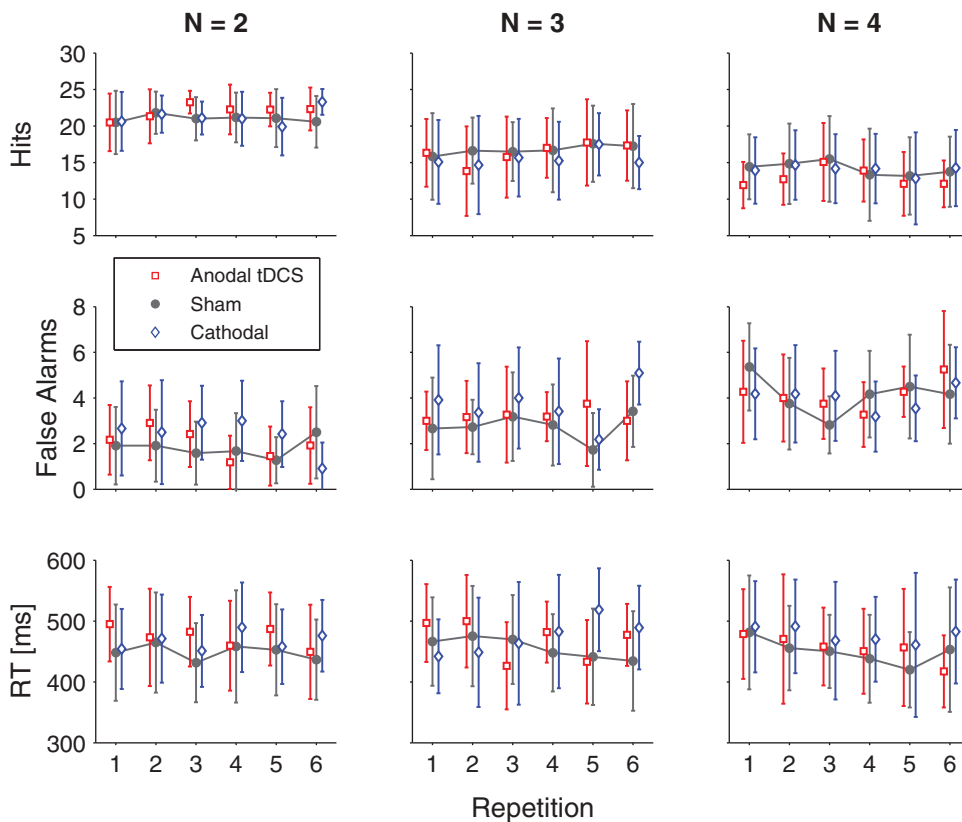


Figure 1. N-back task performance over the six repetitions of a block with a specific load ($N = 2$, $N = 3$, or $N = 4$), separated for the three cerebellar transcranial direct current stimulation (tDCS) stimulation conditions (anodal, sham, cathodal). Hits and false alarms are scored per block of 48 trials. Each point shows the average of the 12 participants, and error bars denote standard error of the means. To view a color version of this figure, please see the online issue of the Journal.

Table 1. Overall average performance per block for each of the three loads.

Performance	Load (N)		
	N = 2 Mean ± SD	N = 3 Mean ± SD	N = 4 Mean ± SD
Hits**	21.43 ± 0.54	16.20 ± 0.86	13.71 ± 0.75
False alarms**	2.07 ± 0.30	3.22 ± 0.43	4.08 ± 0.50
Reaction times (ms)	463 ± 7	466 ± 7	461 ± 14

Note. A total of 25 of the 48 trials in a block required a key press to be denoted as a hit.

**Effect of load $p < .001$.

times, $F(10, 110) = 0.68$, $p = .60$, $\eta_p^2 = .06$. The interactions were assessed comparing the effect of repetition for each load separately, yielding no effects of repetition on the hits for either load. For the lowest load (N = 2), more false alarms were found in the first block than in the five subsequent blocks of 48 trials, which was not observed in the other loads.

Discussion

The aim of this study was to examine the effects of tDCS over the cerebellum on performance in the N-back task. Previous research showed involvement of the cerebellum in this task (Hautzel et al., 2009; Jonides et al., 1997; Kirschen et al., 2004; Owen et al., 2005), especially with higher loads (Jonides et al., 1997; Kirschen et al., 2004). Based on the observations, improved performance after anodal left prefrontal tDCS, especially with higher cognitive loads (Fregni et al., 2005; Teo et al., 2011), we hypothesized that right anodal cerebellar tDCS would have similar effects.

As expected, increasing the load decreased performance: Participants in our study had fewer hits and made more false alarms. However, task performance was not significantly modulated by anodal or cathodal cerebellar tDCS. The statistical effect sizes of the direct comparison between anodal or cathodal stimulation and sham were small (between 0.02 and 0.24, see Table 2) and were also smaller than the effect sizes of other studies that did report an effect of tDCS stimulation (Jacobson et al., 2012). Therefore, we conclude that in our

study, tDCS over the right cerebellum does not critically influence performance in the N-back task.

Several previous studies using different cognitive tasks have observed performance changes with cerebellar tDCS. For instance, cerebellar tDCS has been shown to improve scores and reaction times on a Sternberg task (Ferrucci et al., 2008) and on the Paced Auditory Serial Subtraction Test (PASST; Pope & Miall, 2012) or to impair performance in the Digit Span Task (Boehringer, Macher, Dukart, Villringer, & Pleger, 2013). However, a recent review meta-analysis suggests that the tDCS effects on cognitive processes may be not as prominent as proposed in the literature (Horvath et al., 2015). Therefore, an explanation for our results is that also cerebellar tDCS does not have modulating effects on cognitive processes.

Another explanation is that the cerebellum is not critically involved in learning the N-back task. It could be that this type of memory task relies much more upon processes in the prefrontal cortex as suggested by, for instance, imaging studies (D'Esposito et al., 1998; Owen et al., 2005; Veltman et al., 2003). In the internal network model proposed by Ito (2002), the cerebellum and PFC are connected, but serve different memory processes: The PFC is involved in explicit memory, and the cerebellum relates to implicit memory. One could argue that the N-back task is more explicit than implicit in nature, and therefore the cerebellum is less involved. In turn, tDCS would then have little to no effect on performance. This can be tested by stimulating the PFC or the cerebellum in a within-subjects design allowing for a direct comparison between cerebellar and PFC stimulation effects.

A within-subject design seems to be important in tDCS studies that investigate working memory. Studies using a between-subjects design often fail to observe an effect due to between-subject variability. For instance, Lally and colleagues observed that a group of subjects who received anodal tDCS stimulation on the prefrontal cortex in the N-back did not differ from a separate sham control group over time (Lally, Nord, Walsh, & Roiser, 2013).

Table 2. Performance per stimulation condition and the statistics of the direct comparison between anodal or cathodal stimulation to sham.

Performance	Stimulation condition			Anodal vs. sham			Cathodal vs. sham		
	Sham (Mean ± SD)	Anodal (Mean ± SD)	Cathodal (Mean ± SD)	<i>t</i>	<i>p</i>	Cohen's <i>d</i>	<i>t</i>	<i>p</i>	Cohen's <i>d</i>
Hits	17.18 ± 3.96	17.26 ± 3.62	16.74 ± 3.70	$t(11) = 0.11$	$p = .91$	0.02	$t(11) = 0.39$	$p = .70$	-0.11
False alarms	3.03 ± 1.04	3.30 ± 0.97	3.56 ± 1.42	$t(11) = 0.94$	$p = .38$	0.04	$t(11) = 0.93$	$p = .37$	0.07
Reaction times (ms)	457 ± 64	460 ± 61	473 ± 60	$t(11) = 0.21$	$p = .84$	0.04	$t(11) = 1.14$	$p = .28$	0.24

There are several limitations to our study. First, our sample size was small, which may well have well contributed to the absence of statistical significance. Other studies on the effects of cortical tDCS on working memory did find effects with a small sample size (12 to 15 subjects; Horvath et al., 2015). However, in our study, the effect sizes of the analyses regarding tDCS stimulation were all small according to traditional metrics. This suggests that cerebellar tDCS does not seem to improve performance in the N-back memory task.

Another limitation could be the particular tDCS methodology we applied. Our present set-up using small electrodes of 1.13 cm² was based on a previous study in our lab in which we showed effects of cerebellar tDCS on saccadic eye movement learning (Avila et al., 2015). Our protocol was also comparable with other cerebellar tDCS protocols. Other protocols do exist, however, and some of them are more commonly used than others. However, research on the effectiveness of various tDCS protocols is beyond the scope of this study (Gandiga et al., 2006).

Future research should focus on optimizing tDCS effects for motor and cognitive tasks. TDCS shows an anodal-excitation and cathodal-inhibition effect for motor studies; however, for cognitive studies the polarity effect is not so distinct (Jacobson et al., 2012). This diverse effect extends to cerebellar tDCS studies investigating cognitive tasks. Several studies have shown that anodal and cathodal tDCS can have similar, dissimilar, or even no results in cognitive tasks (Boehringer et al., 2013; Ferrucci et al., 2012; Ferrucci et al., 2008; Pope & Miall, 2012).

In conclusion, we found that cerebellar tDCS does not seem to improve performance in the N-back memory task. Since the number of subjects was rather small in our study, we cannot rule out the possibility that effects of cerebellar tDCS do exist. If these effects do exist, they are likely to be small. It could be worthwhile to compare prefrontal tDCS to cerebellar tDCS directly in a future study using more subjects.

Disclosure statement

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