Journal of Child Neurology
2014, Vol. 29(2) 232-239

DOI: 10.1177/0883073813492385
jcn.sagepub.com
SAGE

Cathodal Transcranial Direct Current Stimulation in Children With Dystonia: A Sham-Controlled Study

Scott J. Young, PhD[1], Matteo Bertucco, PhD[1], and Terence D. Sanger, MD, PhD[2]

Abstract

Increased motor cortex excitability is a common finding in dystonia, and transcranial direct current stimulation can reduce motor cortex excitability. In an earlier study, we found that cathodal direct-current stimulation decreased motor overflow for some children with dystonia. To investigate this observation further, we performed a sham-controlled, double-blind, crossover study of 14 children with dystonia. We found a significant reduction in overflow following real stimulation, when participants performed the experimental task with the hand contralateral to the cathode. While these results suggest that cathodal stimulation may help some children to reduce involuntary overflow, the size of the effect is small. Further research will need to investigate ways to increase the magnitude of the effect of cathodal transcranial direct current stimulation.

Keywords

dystonia, child, transcranial direct current stimulation, overflow, electromyogram

Received March 12, 2013. Received revised May 1, 2013. Accepted for publication May 6, 2013.

Childhood dystonia is "a movement disorder in which involuntary sustained or intermittent muscle contractions cause twisting and repetitive movements, abnormal postures, or both."[1] Current treatment options have limited effectiveness,[2] so new interventions are needed for treating dystonia in children.

Research has shown that cortical inhibition is impaired in dystonia,[3-5] and that methods to restore inhibition can help alleviate dystonic symptoms. For example, transcranial magnetic stimulation can reduce symptoms of dystonia.[6-8] Transcranial direct current stimulation is a more recent and more easily used technology that also shows promise.[9-12]

In an earlier uncontrolled study,[13] we investigated the effects of cathodal transcranial direct current stimulation of the motor cortex on dystonic symptoms in 10 children. We did not see any change in the clinical measure of dystonia, but several participants had reduced motor overflow in an electromyogram tracking task. This result suggested that there might be a subset of children for whom cathodal transcranial direct current stimulation can reduce unwanted muscle activity.

We performed the present controlled double-blinded study to confirm this result. Participants attended 2 experimental visits. They received real cathodal stimulation on 1 visit, and they received sham stimulation on the other; the actual stimulation was blinded from investigators and participants. We measured 2 outcome measures with an electromyogram tracking task: (1) voluntary control of the first dorsal interosseous muscle and (2) overflow of muscle activity between the 2 hands.

Methods

Participants

Participants were 14 children with primary or secondary dystonia affecting 1 or both hands. The characteristics of the children are outlined in Table 1. All participants were recruited from the movement disorders clinic at Children's Hospital Los Angeles. A pediatric neurologist (TDS) diagnosed participants based on history and clinical examination, according to standard definitions.[1] Children were excluded if there was clinical evidence of spasticity or corticospinal injury in the upper extremities (hyper-reflexia, a spastic catch, or pyramidal distribution weakness). For example, children with hemiplegic

[1] Department of Biomedical Engineering, University of Southern California, Los Angeles, CA, USA
[2] Departments of Biomedical Engineering, Child Neurology, and Biokinesiology & Physical Therapy, University of Southern California and Children's Hospital of Los Angeles, Los Angeles, CA, USA

Corresponding Author:
Terence D. Sanger, MD, PhD, Department of Biomedical Engineering, University of Southern California, 1042 Downey Way, DRB 140, Los Angeles, CA 90089, USA.
Email: tsanger@usc.edu

Table 1. Characteristics of Participants.

Participant	Age	Gender	Diagnosis	Left Arm BAD Scale Score	Right Arm BAD Scale Score	Cathode Location
D1	7	M	Dyskinetic cerebral palsy	3	3	Left
D2	8	M	Tetraplegic cerebral palsy	1	3	Left
D3	9	F	Dyskinetic cerebral palsy	2	2	Right
D4	9	M	Ataxic cerebral palsy	1	1	Right
D5	11	F	Right hemiplegic cerebral palsy	2	1	Right
D6	11	M	Cerebral palsy	2	3	Right
D7	12	M	Cerebral palsy	3	3	Left
D8	13	F	Idiopathic dystonia	1	3	Left
D9	14	F	Cerebral palsy due to kernicterus	3	3	Right
D10	14	M	Dyskinetic cerebral palsy	3	3	Right
D11	15	M	Idiopathic dystonia	1	1	Left
D12	17	M	Cerebral palsy	4	4	Left
D13	18	M	Right hemidystonia from traumatic brain injury at 3 years of age	0	3	Left
D14	19	M	Cerebral palsy	4	4	Right

Abbreviations: BAD Scale, Barry-Albright Dystonia Scale; F, female; M, male.

cerebral palsy were included if symptoms included only hemidystonia. The University of Southern California Institutional Review Board approved the study protocol. All parents gave informed written consent for participation and authorization for use of protected health information, and all children gave written assent. The study was registered with clinicaltrials.gov (NCT01460771).

Measurement Procedure

Each participant attended 2 experimental sessions of approximately 2 hours each. Participants received real transcranial direct current stimulation in 1 of the sessions and sham stimulation in the other session. Sessions were separated by at least 1 week, and the order of sessions (ie, real or sham stimulation) was randomized and counterbalanced. All participants were rated on the Barry-Albright Dystonia Scale[14] at the beginning and end of each session. Participants sat in a chair or their own wheelchair and placed both hands flat on the surface of a table with palms down in a comfortable position. The table was adjusted to a comfortable height for each participant. To ensure isometric activation, the index finger of the task hand was constrained to prevent abduction, using a plastic block attached to the surface of the table. For participants who could not maintain their hands flat on the table, a bandage was wrapped around the fingers to prevent abduction.

Surface electromyography electrodes (DE-2.1 electrodes with Bagnoli-8 amplifier, Delsys Incorporated, Boston, MA, USA) with a band-pass filter of 20 to 450 hertz and amplification of 1000 times were placed over the bellies of the first dorsal interosseous and abductor digiti minimi muscles on both hands. The electromyography signals were sampled at 1 kilohertz using an analog to digital interface (Power 1401, Cambridge Electronic Design Limited, Cambridge, UK) controlled by custom data acquisition software. Electromyography signals were processed on-line for display and data analysis with a high-pass Butterworth filter (fourth order, 1 hertz cutoff), then a Bayesian filter, and finally a low-pass Butterworth filter (second order, 5 hertz cutoff). The Bayesian filter produces a smooth output that estimates the drive underlying the electromyogram signal, while also allowing fast low-latency changes in the filtered signal.[15]

Prior to the start of the experiment, we measured the maximum voluntary isometric contraction for each of the 4 muscles. The signal from each electrode was displayed as visual feedback for the participant. The participant performed 3 attempts of 5 seconds of maximum contraction for each muscle with encouragement and visual feedback. Maximum voluntary contraction was quantified as the maximum mean electromyogram measured over a 200-millisecond period. After measuring the maximum voluntary contraction values, we measured the resting activity by recording the electromyograms for 55 seconds while participants attempted to maintain relaxation.

Transcranial Direct Current Stimulation

A Magstim NeuroConn Direct Current Stimulator Plus Model 0021 (The Magstim Company Limited, Whitland, UK), with 5-centimeter by 7-centimeter saline-soaked electrodes, was used for stimulation. The cathode was placed over motor cortex at the C3 or C4 scalp location according to the international 10-20 electroencephalography placement system.[16] The cathode was placed on the side of the scalp contralateral to (1) the hand most affected by dystonia or (2) if both hands were similarly affected, the child's preferred hand. The anode was placed on the forehead, contralateral to the cathode. We performed the stimulation with the most effective parameters outlined by Monte-Silva and colleagues,[17] using 9 minutes at 1 milliampere, a 20-minute pause, and an additional 9 minutes at 1 milliampere. For the sham condition, the stimulator ramped down to 0 milliamperes after 30 seconds of 1-milliampere stimulation. An experimenter who did not take part in data collection or analysis operated the stimulator. Participants and all other experimenters did not know the type of stimulation used during the visit.

We altered the stimulation pattern for 4 participants due to skin discomfort or time constraints. We performed a single 9-minute stimulation for the real stimulation visits of participant D14 and participant D7, and for both visits of participant D4. We also reduced the current to 750 microamperes during the last 4 minutes of the second 9-minute stimulation for participant D9's real stimulation visit.

Electromyogram Tracking Task

The electromyogram from either the right or left first dorsal interosseous muscle controlled the vertical position of a small circular cursor on a screen placed in front of the participant. Gain was adjusted so that

the cursor remained at the bottom of the screen when the muscle was at rest and the top of the screen at 20% of maximum voluntary contraction. Prior to testing the task, participants practiced moving the cursor on the screen with the first dorsal interosseous muscle for 1 minute on each side. During this time, the experimenter monitored the participant's performance and made adjustments or suggestions to ensure that the participant understood the relation between muscle contraction and cursor movement. No target was shown during practice.

Following practice, each participant performed 2 trials of a tracking task with each first dorsal interosseous muscle. During each trial, the computer monitor displayed a target (horizontal line) and a circular cursor, as shown in Figure 1. The target moved vertically, jumping between the bottom of the screen (0% of maximum voluntary contraction, or relaxation) and the middle of the screen (10% of maximum voluntary contraction) with an interval of 5 seconds. Participants tracked the target with the cursor by isometrically activating the first dorsal interosseous muscle. Each trial had a duration of 55 seconds (ie, 5 repetitions of 5 seconds of relaxation followed by 5 seconds of activation, with an additional 5 seconds of relaxation at the end). Participants were instructed to maintain all nontask muscles at rest during the task, but they were given feedback only from the active first dorsal interosseous muscle. For each first dorsal interosseous muscle, participants performed 2 trials of the task before stimulation, and 2 trials following stimulation. Participants performed the task with the hand contralateral to the cathode in most visits, and there was a time interval of 2 to 5 minutes between the end of stimulation and the first trial after stimulation. For participant D9, we set the target to move with an interval of 10 seconds, because this participant had difficulty moving to the target within 5 seconds.

Analysis

We measured 2 aspects of participant performance in the electromyogram tracking task: tracking error and overflow muscle activity. We defined tracking error as the absolute difference between the target position and the cursor position, expressed in units of normalized electromyogram. We defined normalized electromyogram as the ratio of the activation of each muscle to its maximum voluntary contraction. We also truncated the electromyogram for each muscle at 20% of maximum voluntary contraction to match the range of normalized electromyogram values displayed on the screen for the task muscle. Tracking error allowed us to measure how well individuals could modulate muscle activity on demand, as well as determine whether participants performed the task similarly before and after stimulation. See Figure 2 for the muscle activity from sample trials.

We defined overflow for each nontask muscle as the normalized electromyogram of that muscle, because the task goal was to keep all nontask muscles relaxed. We quantified overflow as the mean overflow across all 3 nontask muscles—the abductor digiti minimi muscle of the task hand and the first dorsal interosseous and abductor digiti minimi muscles of the nontask hand (see Figure 2 for sample output).

For analysis, we used the mean values of task error and overflow for each 5-second period of target activation or relaxation. To check the agreement of our electromyogram-based measures with the clinical manifestation of dystonia, we calculated the correlation of tracking error and overflow in all trials before stimulation with the Barry-Albright Dystonia Scale score of participants' arms.

To test for changes due to stimulation, we paired all mean values after stimulation with the matching measurement before stimulation to find the differences for each combination of participant, task hand (contralateral and ipsilateral to cathode), and stimulation type (real or sham). We measured individual and group confidence intervals for each task hand and stimulation type, as shown in Figures 3 and 4.

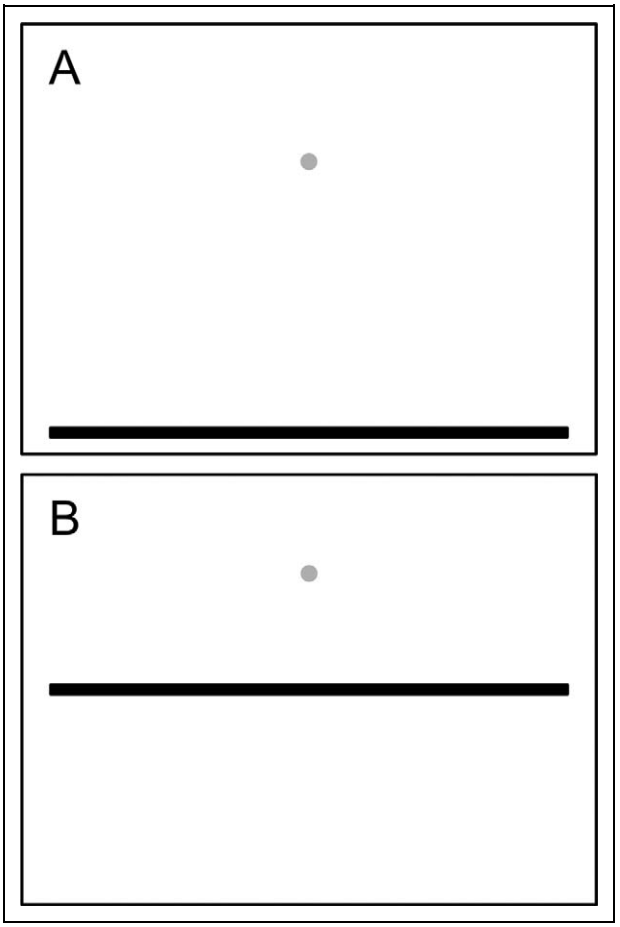

Figure 1. Electromyogram tracking task used in this experiment. The horizontal bar acted as a target, jumping between the bottom of the screen (A) and the middle of the screen (B) with an interval of 5 seconds. Participants tracked the target with the cursor (gray circle), activating their first dorsal interosseous muscle to move the cursor upward and relaxing their first dorsal interosseous to move the cursor downward. The muscle activation for the bottom of the screen was 0% of maximum voluntary contraction, and muscle activation for the middle of the screen was 10% of maximum voluntary contraction.

We used a linear mixed-effects model to test for group effects of task hand (contralateral vs ipsilateral to the cathode), stimulation type (real vs sham stimulation), and interaction between task hand and stimulation type. Participants were considered as a random factor. Analysis was performed using the lme function from the nlme package[18,19] of the R statistical computing environment.[20] The R model was lme (difference $\sim$ taskHand*stimType, random $= \sim 1|$participant). We used a criterion of $P < .05$ to signify a significant difference. For overflow, we also used a linear mixed-effects model to test for the effect of stimulation type (real vs sham) within the hand contralateral to the cathode. The R model was lme(difference $\sim$ stimType, random $= \sim 1|$participant).

Results

Participants D4, D7, and D9 had discomfort during the stimulation, but all were comfortable after we adjusted the

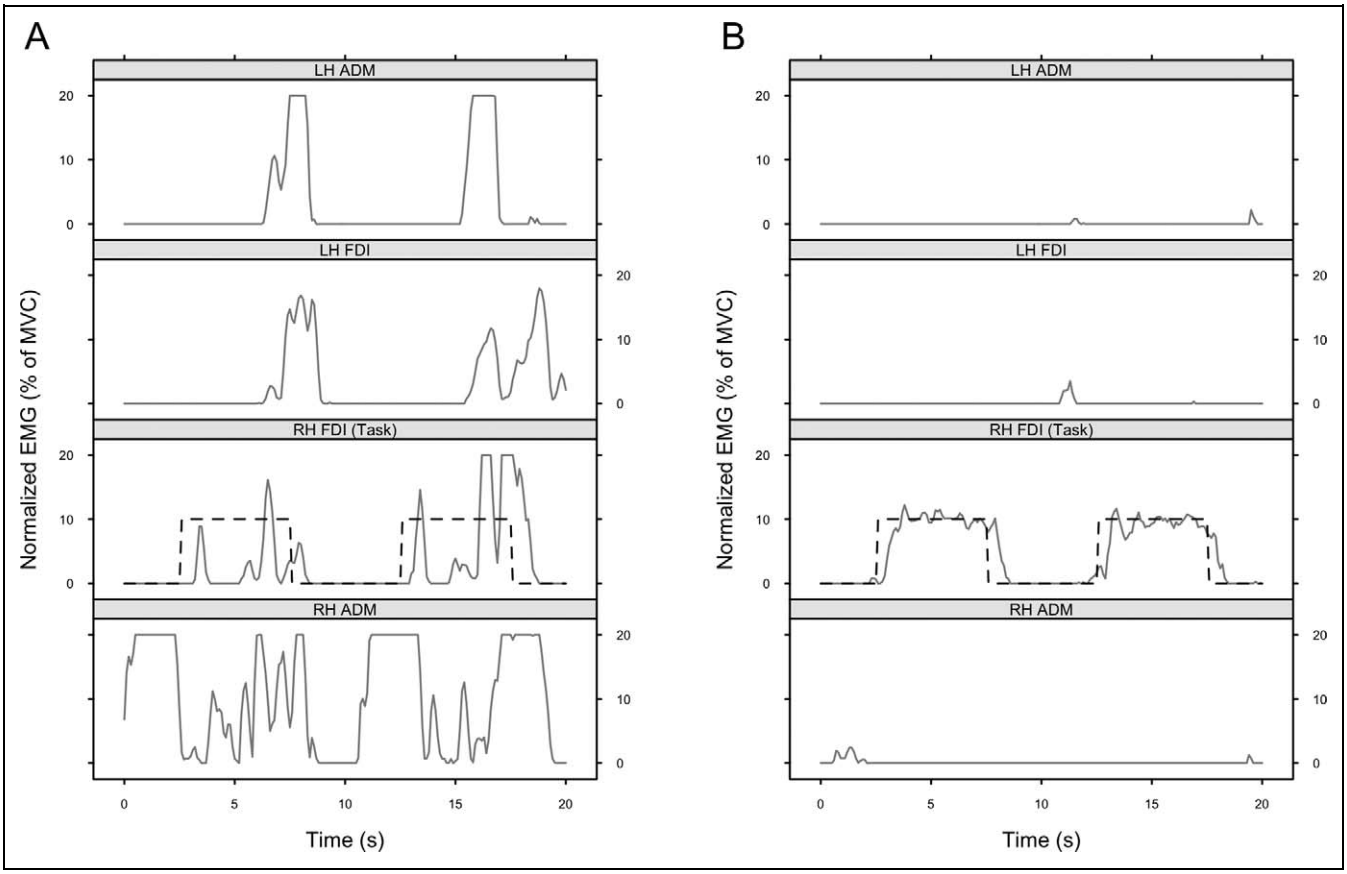

Figure 2. Representative muscle activation during the electromyogram tracking task. Each panel plots target location for the task muscle (black dashed line) and normalized muscle activation for all 4 muscles (solid gray lines) during 20 seconds of a single trial. All muscle activation traces are truncated at 20% of maximum voluntary contraction, as was done during the analysis. (A) This participant had difficulty activating the task muscle to reach the target, and there was substantial overflow in all nontask muscles. (B) This participant was able to perform the task well, and there was little overflow in nontask muscles. ADM, abductor digiti minimi; EMG, electromyogram; FDI, first dorsal interosseous; LH, left hand; MVC, maximum voluntary contraction; RH, right hand.

stimulation settings, as noted above. No other participants reported discomfort, and there were no other adverse events. There was no change in the Barry-Albright Dystonia Scale score in any participant.

The mean tracking error prior to stimulation was 4.2% of maximum voluntary contraction while the hand contralateral to the cathode performed the task, and 4.1% of maximum voluntary contraction while the hand ipsilateral to the cathode performed the task. Individual participant means prior to stimulation ranged from 1.4% to 8.6% of maximum voluntary contraction while the hand contralateral to the cathode performed the task, and from 1.4% to 10.3% of maximum voluntary contraction while the hand ipsilateral to the cathode performed the task. Tracking error correlated with the Barry-Albright Dystonia Scale score ($r = .590$, 95% confidence interval: .387, .738). The mean difference in tracking error after stimulation is shown in Figure 3 for all participants and for the group. Group means were not significantly different than 0 for any combination of stimulation type and task hand. For the group, there were no significant effects of task hand, stimulation type, nor interaction between task hand and stimulation type, for all effects, $F(1, 1059) < 0.806$ and $P > .369$.

The mean overflow prior to stimulation was 4.1% of maximum voluntary contraction while the hand contralateral to the cathode performed the task and 2.3% of maximum voluntary contraction while the hand ipsilateral to the cathode performed the task. Individual participant means prior to stimulation ranged from less than 0.1% to 16.5% of maximum voluntary contraction while the hand contralateral to the cathode performed the task, and from less than 0.1% to 12.0% of maximum voluntary contraction while the hand ipsilateral to the cathode performed the task. Overflow correlated with the Barry-Albright Dystonia Scale score ($r = .500$, 95% confidence interval: .273, .674). The mean difference in overflow after stimulation is shown in Figure 4 for all participants and for the group. The group mean for the real stimulation visits when the hand contralateral to the cathode performed the task was significantly lower than 0 (estimated difference –1.5% of maximum voluntary contraction; 95% confidence interval: –2.2%, –0.7%). There were significant effects for task hand (estimate 0.59% of maximum voluntary contraction; 95% confidence interval: 0.41%, 0.77%); $F(1, 1063) = 42.1$, $P < .001$, for stimulation type (estimate 0.18% of maximum voluntary contraction; 95% confidence interval: 0.36%, 0.002%); $F(1, 1063) =$

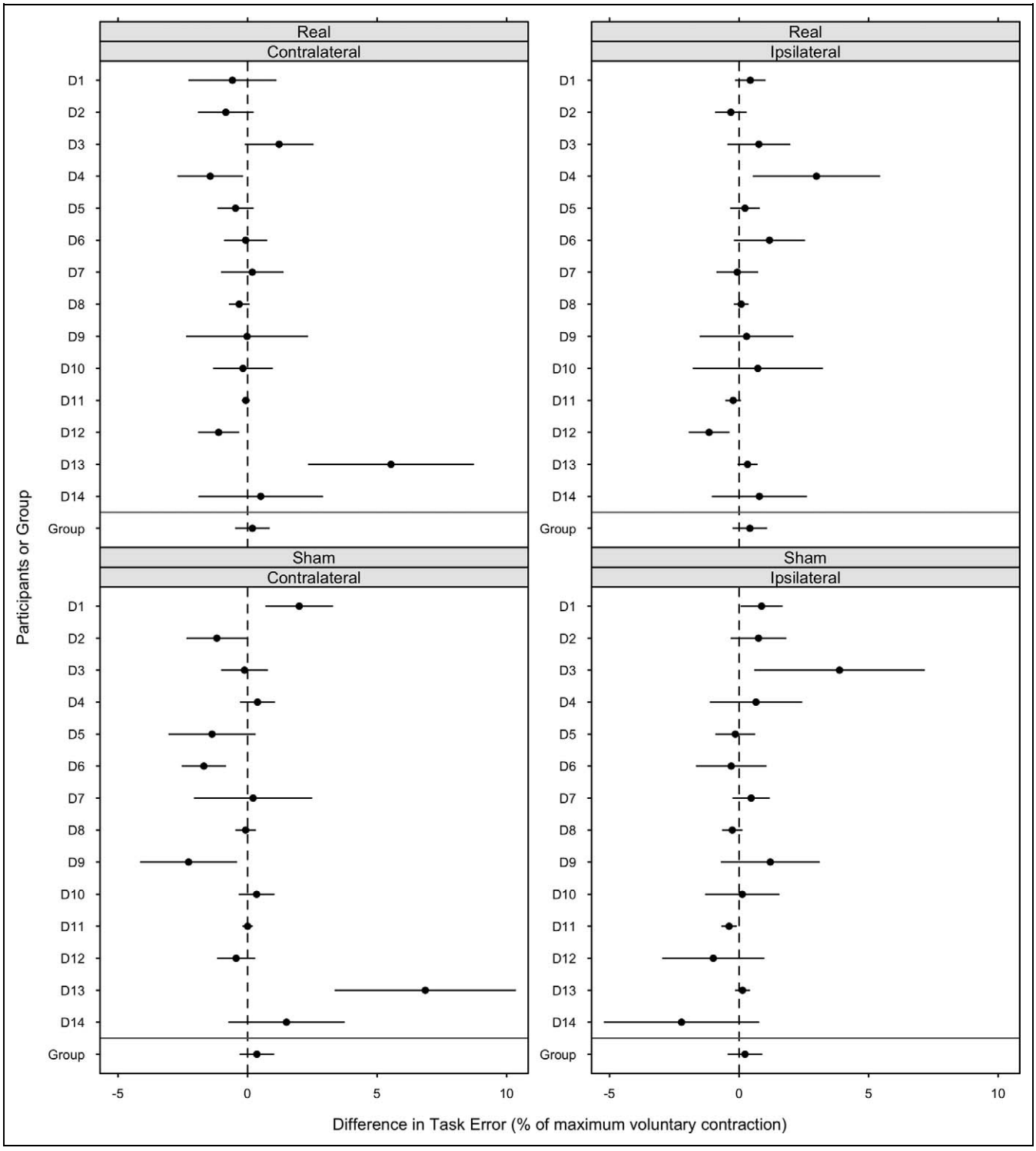

Figure 3. Mean difference in task error for individual participants and the group. Participants are arranged vertically, with the group mean from the mixed-effects model at the bottom of each panel. Panels are organized by the task hand (contralateral hand in left column, and ipsilateral hand in right column) and stimulation type (real stimulation on the top row, and sham stimulation on the bottom row). Differences are shown horizontally, with a negative value (to the left) indicating lower task error after stimulation. Circles indicate means, and horizontal lines represent 95% confidence intervals of the means. The mean group difference in task error is not significantly different from zero for any combination of task hand or stimulation type.

3.95, $P = .047$, and for the interaction between task hand and stimulation type (estimate 0.19% of maximum voluntary contraction; 95% confidence interval: 0.36%, 0.009%); $F(1, 1063) = 4.26$, $P = .039$. The interaction effect indicates that the differences between the values for ipsilateral hand and contralateral hand are greater in the real visits than they are in the

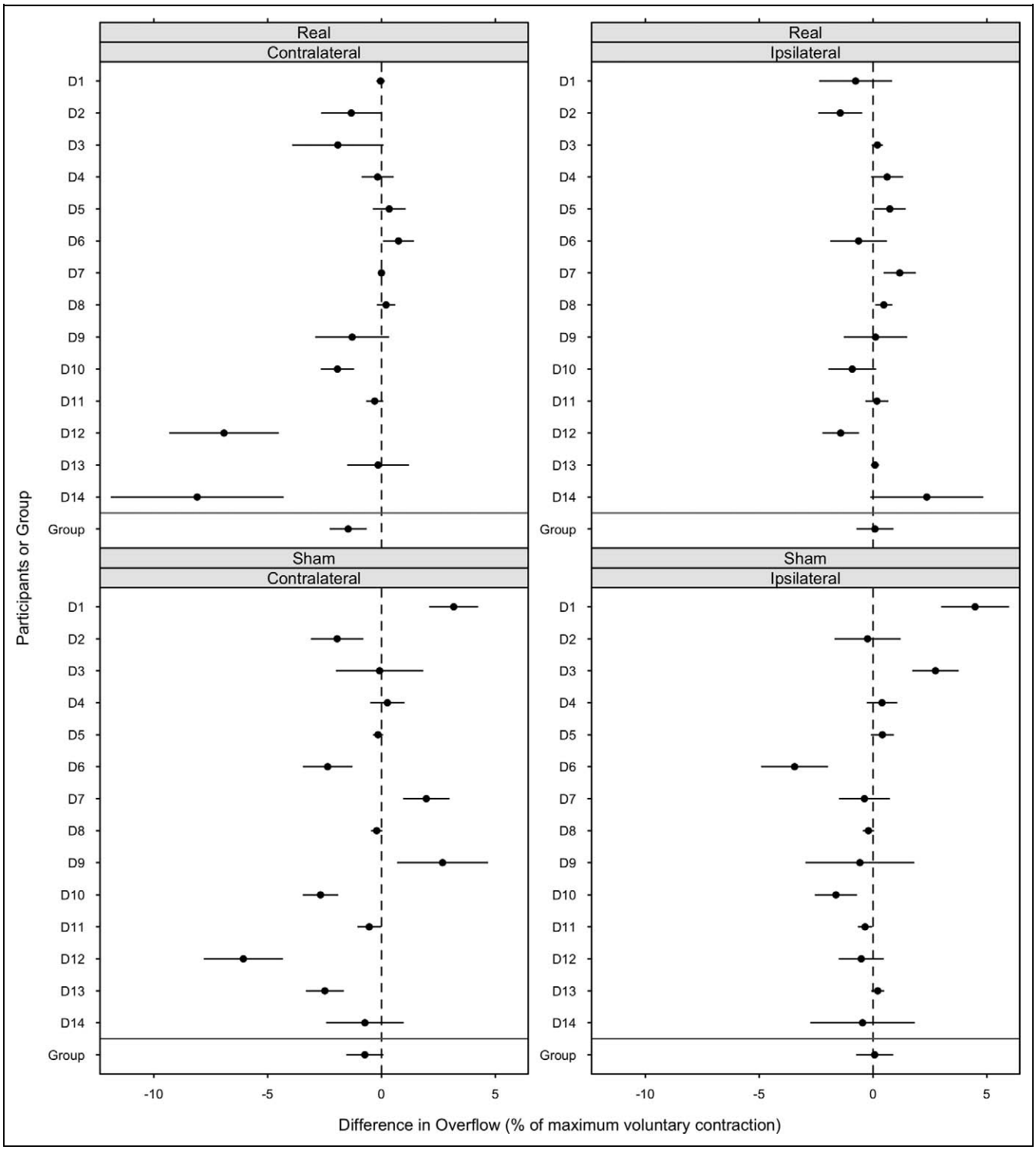

Figure 4. Mean difference in overflow for individual participants and the group. Participants are arranged vertically, with the group mean from the mixed-effects model at the bottom of each panel. Panels are organized by the task hand (contralateral hand in left column, and ipsilateral hand in right column) and stimulation type (real stimulation on the top row, and sham stimulation on the bottom row). Differences are shown horizontally, with a negative difference (to the left) indicating lower overflow after stimulation. Circles indicate means, and horizontal lines represent 95% confidence intervals of the means. The mean group difference was significantly lower than zero when the hand contralateral to the cathode performed the task during the real stimulation visits.

sham visits. To investigate the effect of stimulation further, we fit a model to the overflow that occurred only while the contralateral hand performed the task. There was a significant effect of stimulation type (estimate 0.36% of maximum voluntary contraction; 95% confidence interval: 0.62%, 0.10%); $F(1, 525) = 7.34$, $P = .007$.

Discussion

In this double-blind, sham-controlled study, cathodal transcranial direct current stimulation of the motor cortex reduced motor overflow by a small amount in children with primary or secondary dystonia. These results are similar to the results of our earlier open-label study: stimulation reduced overflow in a subset of children, but it did not adversely affect voluntary muscle control.[13] Therefore, it is possible that transcranial direct current stimulation could be useful for some children with dystonia.

Although the decrease in overflow we observed was statistically significant, it is not clear whether it was clinically meaningful. The measures we used are related to the clinical incidence of dystonia, but they are not perfectly correlated.[21] As well, there were no noticeable differences in the Barry-Albright Dystonia Scale, and most participants in this study did not have a statistically significant decrease in overflow. Future work will need to identify ways to increase the effect size.

In addition to the decrease in overflow following real stimulation, we also observed a smaller, not statistically significant, decrease in overflow following sham stimulation. As a result, it is possible that other factors, such as the placebo effect, could have contributed to the decrease in overflow. One possible contribution was the order of task completion. In most visits, participants performed the task with the hand contralateral to the cathode first. This was a practical approach, to ensure that the participant was able to complete the task with their most-affected hand, but it may have contributed to the statistical difference between ipsilateral and contralateral hands.

The present study and our previous study[13] have shown small effects of cathodal stimulation for a subset of children with dystonia. Other studies of cathodal stimulation for dystonia have also shown either no improvement[9,10] or improvement in isolated cases.[11,12] There are at least 2 potential avenues for increasing effectiveness for dystonia. One option is stronger or repeated stimulation over multiple days. Another option could be other forms of transcranial current stimulation. For example, a recent case study showed strong effects of transcranial alternating current stimulation in torticollis.[22]

Acknowledgments

We thank Rebecca Sheehan-Stross and Andrea Mogollon Carrillo for assistance with recruitment. We thank Diana Ferman, physician assistant, and Dr Quyen Luc for assistance with neurological examinations. We also thank several members of the laboratory for assistance with operating the stimulator. This research was performed at the University of Southern California. Preliminary results were presented at the Society for Neuroscience Annual Meeting in Washington, DC, in November 2011, the Neural Control of Movement Annual Meeting in Venice, Italy, in April 2012, and the Movement Disorders Society International Congress in Dublin, Ireland, in June 2012.

Author Contributions

SJY conceived the experiment, developed the apparatus and methods, performed the experiment, designed, performed, and reviewed the statistical analysis, and wrote the first draft and reviewed the manuscript. MB conceived the experiment, developed the apparatus and methods, performed the experiment, designed and reviewed the statistical analysis, and reviewed the manuscript. TDS conceived the experiment, developed the apparatus and methods, clinically evaluated the participants, designed and reviewed statistical the analysis, and reviewed the manuscript.

Declaration of Conflicting Interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The authors disclosed receipt of the following financial support for the research, authorship and/or publication of this article: This study was funded in part by Grant NS052236 from the National Institute of Neurological Disorders and Stroke. Additional funding was provided by the Crowley-Carter Foundation and the Don and Linda Carter Foundation.

Ethical Approval

The University of Southern California Institutional Review Board approved the study protocol. All parents gave informed written consent for participation and authorization for use of protected health information. All children gave written assent. The study was registered with clinicaltrials.gov (NCT01460771).

References

1. Sanger TD, Delgado MR, Gaebler-Spira D, et al. Classification and definition of disorders causing hypertonia in childhood. *Pediatrics*. 2003;111:e89-e97.
2. Sanger TD. Hypertonia in children: how and when to treat. *Curr Treat Options Neurol*. 2005;7:427-439.
3. Ridding MC, Sheean G, Rothwell JC, et al. Changes in the balance between motor cortical excitation and inhibition in focal, task specific dystonia. *J Neurol Neurosurg Psychiatry*. 1995;59:493-498.
4. Beck S, Richardson SP, Shamim EA, et al. Short intracortical and surround inhibition are selectively reduced during movement initiation in focal hand dystonia. *J Neurosci*. 2008;28:10363-10369.
5. Hallett M. Neurophysiology of dystonia: the role of inhibition. *Neurobiol Dis*. 2011;42:177-184.
6. Siebner HR, Tormos JM, Ceballos-Baumann AO, et al. Low-frequency repetitive transcranial magnetic stimulation of the motor cortex in writer's cramp. *Neurology*. 1999;52:529-537.
7. Murase N, Rothwell JC, Kaji R, et al. Subthreshold low-frequency repetitive transcranial magnetic stimulation over the premotor cortex modulates writer's cramp. *Brain*. 2005;128:104-115.
8. Lefaucheur J, Fenelon G, Menard-Lefaucheur I, et al. Low-frequency repetitive TMS of premotor cortex can reduce painful axial spasms in generalized secondary dystonia: a pilot study of three patients. *Neurophysiol Clin*. 2004;34:141-145.

9. Benninger DH, Lomarev M, Lopez G, et al. Transcranial direct current stimulation for the treatment of focal hand dystonia. *Mov Disord*. 2011;26:1698-1702.
10. Buttkus F, Baur V, Jabusch H, et al. Single-session tDCS-supported retraining does not improve fine motor control in musician's dystonia. *Restorative Neurol Neurosci*. 2011;29:85-90.
11. Buttkus F, Baur V, Jabusch H, et al. Retraining and transcranial direct current stimulation in musician's dystonia—a case report. *Mov Disord*. 2010;25:1758-1760.
12. Buttkus F, Weidenmuller M, Schneider S, et al. Failure of cathodal direct current stimulation to improve fine motor control in musician's dystonia. *Mov Disord*. 2010;25:389-394.
13. Young SJ, Bertucco M, Sheehan-Stross R, Sanger TD. Cathodal transcranial direct current stimulation in children with dystonia a pilot open-label trial [published online ahead of print October 3, 2012]. *J Child Neurol.* doi:10.1177/0883073812460092.
14. Barry MJ, VanSwearingen JM, Albright AL. Reliability and responsiveness of the Barry-Albright Dystonia Scale. *Dev Med Child Neurol*. 1999;41:404-411.
15. Sanger TD. Bayesian filtering of myoelectric signals. *J Neurophysiol*. 2007;97:1839-1845.
16. Herwig U, Satrapi P, Schonfeldt-Lecuona C. Using the international 10-20 EEG system for positioning of transcranial magnetic stimulation. *Brain Topogr*. 2004;16:95-99.
17. Monte-Silva K, Kuo M, Liebetanz D, et al. Shaping the optimal repetition interval for cathodal transcranial direct current stimulation (tDCS). *J Neurophysiol*. 2010;103:1735-1740.
18. Pinheiro J, Bates D, DebRoy S, Sarkar D, R Core team. nlme: linear and nonlinear mixed effects models. *R package version* 3. 1-96. 2009.
19. Pinheiro J, Bates D. *Mixed-Effects Models in S and S-PLUS*. New York: Springer; 2000.
20. R Core Development Team. *R: A Language and Environment for Statistical Computing*. Vienna, Austria: R Foundation for Statistical Computing; 2010. http://www.R-project.org.
21. Young SJ, Doornik JV, Sanger TD. Finger muscle control in children with dystonia. *Mov Disord*. 2011;26:1290-1296.
22. Angelakis E, Liouta E, Andreadis N, et al. Transcranial alternating current stimulation reduces symptoms in intractable idiopathic cervical dystonia: a case study [published online ahead of print November 10, 2012]. *Neurosci Lett.* doi:10.1016/j.neulet.2012.11.007.