NeuroRehabilitation 39 (2016) 401–411
DOI:10.3233/NRE-161371
IOS Press

Transcranial direct current stimulation (tDCS) of the primary motor cortex and robot-assisted arm training in chronic incomplete cervical spinal cord injury: A proof of concept sham-randomized clinical study

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Abstract.
BACKGROUND: After cervical spinal cord injury, current options for treatment of upper extremity motor functions have been limited to traditional approaches. However, there is a substantial need to explore more rigorous alternative treatments to facilitate motor recovery.
OBJECTIVE: To demonstrate whether anodal-primary motor cortex (M1) excitability enhancement (with cathodal-supra orbital area) (atDCS) combined with robot-assisted arm training (R-AAT) will provide greater improvement in contralateral arm and hand motor functions compared to sham stimulation (stDCS) and R-AAT in patients with chronic, incomplete cervical spinal cord injury (iCSCI).
METHODS: In this parallel-group, double-blinded, randomized and sham-controlled trial, nine participants with chronic iCSCI (AIS C and D level) were randomized to receive 10 sessions of atDCS or stDSC combined with R-AAT. Feasibility and tolerability was assessed with attrition rate and occurrence of adverse events, Changes in arm and hand function were assessed with Jebson Taylor Hand Function Test (JTHFT), Amount of Use Scale of Motor Activity Log (AOU-MAL), American Spinal Injury Association Upper Extremity Motor Score and Modified Ashworth Scale (MAS) at baseline, after treatment, and at two-month follow-up.

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RESULTS: None of the participants missed a treatment session or dropped-out due to adverse events related to the treatment protocol. Participants tended to perform better in JTHFT and AOU-MAL after treatment. Active group at post-treatment and two-month follow-up demonstrated better arm and hand performance compared to sham group.
CONCLUSION: These preliminary findings support that modulating excitatory input of the corticospinal tracts on spinal circuits may be a promising strategy in improving arm and hand functions in persons with incomplete tetraplegia. Further study is needed to explore the underlying mechanisms of recovery.

Keywords: Spinal cord injury, arm, motor recovery, non-invasive brain stimulation, rehabilitation-robotics

1. Introduction

Incomplete tetraplegia is the most frequent neurologic category after spinal cord injury (SCI). Each year nearly 12,000 people suffer from SCI in the United States, and about 50 percent of these are reported to be injuries to cervical spine (National Spinal Cord Injury Statistical Center, 2011). Survivors after injury are usually left with severe motor impairments which not only affect walking but also cause paralysis of arm and hand functions resulting in impairments with self-care (i.e., feeding, bathing), transfers, work and decreased participation in social activities. Therefore, regaining arm and hand functions are highly linked to improved quality of life among adults with tetraplegia (Anderson, 2004; Snoek, Ijzerman, Hermens, Maxwell, & Biering-Sorensen, 2004).

While the immediate damage to the cervical spinal cord is to the ascending and descending spinal tracts and the cell bodies within the cord, following injury both animal and human studies have demonstrated structural and functional reorganization at remote cortical and subcortical structures (Kriz, Kozak, & Zedka, 2012; Laubis-Herrmann, Dichgans, Bilow, & Topka, 2000; Lotze, Laubis-Herrmann, Topka, Erb, & Grodd, 1999; Topka, Cohen, Cole, & Hallett, 1991). The spinal cord and cortex become atrophic and axonal integrity is reduced (Freund et al., 2012; Henderson, Gustin, Macey, Wrigley, & Siddall, 2011) and these changes are considered to be an obstacle to achieve maximum functional recovery. Although the roles of residual corticospinal tract fibers, primary motor cortex and spinal neurons are critical in regaining sensorimotor functions in the arm and hands, treatment options aiming to restore functions in the upper limb are usually focused on the extremity, using functional electrical stimulation (Freund et al., 2012), exercise (Hicks et al., 2003), massed practice(Sadowsky & McDonald, 2009) or compensation techniques to accomplish daily tasks (Garber & Gregorio, 1990). Unfortunately none of these rehabilitation strategies has proven to be a gold standard in recovery of impaired arm and hand functions. One alternative rehabilitation approach would be to combine treatment modalities that will facilitate afferent input to sensorimotor cortex and increase the executive activity of the primary motor cortex (M1) and corticospinal tracts. In this context, transcranial direct current stimulation has gained popularity in clinical studies as an add-on neuromodulation technique that uses weak, direct electric current to induce changes in cortical excitability. These changes occur in a polarity-specific manner; while anodal tDCS applied over M1 induces facilitatory effects, cathodal tDCS leads to inhibitory effects. Studies have shown, in a single session of tDCS with current intensities of at least 0.6 mA up to 2 mA applied over 5–20 minutes, modulation of motor cortex excitability is possible and lasts for up to 60 minutes after stimulation (Bastani & Jaberzadeh, 2012; Nitsche et al., 2003; Nitsche, Liebetanz, Tergau, & Paulus, 2002; Nitsche & Paulus, 2001). When applied daily, repeated anodal tDCS sessions over five days can induce motor skill acquisition that persist for several weeks (Reis et al., 2009). In addition, when applied before motor training, anodal tDCS has been shown to enhance CST excitability significantly compared to during or after motor training (Cabral et al., 2015).

Training intensity has a profound effect on motor recovery, and rehabilitation robotic devices have the potential to deliver high-dosage, high-intensity, repetitive therapy procedures in a way that is semi-automated and less labor-intensive (Pehlivan et al., 2014; Zariffa et al., 2011).

In the current 'proof of concept' study we aimed to respond the following questions: (i) is the combination treatment feasible and tolerable as indexed by participation, adverse effects and attrition rate? (ii) Is there an increased treatment effect in the active group compared to the control group immediately after the end of the treatment? (iii) finally, if there is a treatment effect (question ii), are the effects lasting when analyzing motor function over

time? We hypothesized that adults with chronic tetraplegia caused by incomplete cervical spinal cord injury (iCSCI) can improve their upper-limb voluntary movement by participating in a therapeutic program that combines non-invasive brain stimulation with robotic-assisted training. The overall aim of this proof-of-concept preliminary trial is to ultimately provide initial data, including effect sizes and variances, to be used to design a well powered trial. We therefore collected extensive data in each subject as to maximize the amount of information that will be provided for designing future trials. We also submit as preliminary data detailed tables with information on all outcomes which may be useful for future trials and meta-analyses.

2. Material and methods

2.1. Subjects

Nine adults with chronic iCSCI (eight male, one female; age between 36–63 years) participated and eight participants have completed this research study (see Table 1 for demographic and clinical data). Main inclusion criteria were age between 18–65 years; diagnosis of chronic incomplete cervical spinal cord injury as defined by the American Spinal Injury Association Impairment scale classification (AIS C and D) and at least 6 months post-injury; minimal finger motor function (i.e., being able to perform isolated thumb and index finger movement such as pinch grip). Exclusion criteria included neuropsychiatric comorbidities; traumatic brain injury (TBI); involvement in any specific exercise program (e.g., Neuromuscular electrical stimulation, functional electrical stimulation) within the previous 3 months; planned alteration in upper-extremity therapy or medication for muscle tone during the course of the study; contraindications to tDCS such as metal in the head or implanted brain medical devices. Subjects were also excluded if they had prior history of seizure; use of medications containing sodium channel blocker such as carbamezapine; any joint contracture or severe spasticity in the affected upper extremity, as determined by a Modified Ashworth Score great then and equal to 3 out of 4; and history of substance abuse. All subjects were provided written informed consent to participate in the study using a form that was approved by the Institutional Review Board of the University of Texas Health Science Center at Houston.

2.2. Study design

This study used a parallel-group, double-blinded, randomized, sham-controlled trial model. Subjects were randomly assigned to one of two groups: active tDCS with robotic-assisted training or sham tDCS with robotic-assisted training. A computer generated randomization order was used to assign subjects in a 1 : 1 fashion into active or sham tDCS group. For each subject, the researcher received an anonymous code from an independent research assistant. Thus, the researcher who administered the tDCS and the patients were both blinded through all stimulation sessions.

After randomization, the target arm for training was selected based on residual function in the arm and hand. For example, if the subject had asymmetric strength in the upper extremities, the arm with lesser impairment was selected; or if one side had nearly normal function, then the opposite side with more impairment was chosen for treatment. Each patient underwent clinical and functional assessment at baseline, after the intervention, and at two-month follow up. All assessments were administered by the same senior occupational therapist that was blinded to the type of intervention the subjects had received.

2.3. Intervention

2.3.1. Transcranial direct current simulation (tDCS)

tDCS is a form of non-invasive cortical stimulation and has the potential to alter cortico-spinal excitability. Direct current was transferred by two saline-soaked surface sponge electrodes ($7 \times 5\text{cm} = 35\text{cm}^2$ active area) and delivered by a battery-driven stimulator device (medical tDCS for clinical trials device, Soterix Medical ®, NY). To stimulate the primary motor cortex (M1) the anode electrode (increasing cortical excitability) was placed over C3/C4 (according to the 10–20 international electroencephalogram electrode system) contralateral to the targeted arm. The cathode (i.e., reference) electrode was placed over contralateral supraorbital area as shown in Fig. 1.

During tDCS, subjects in the active group received 20 minutes of 2 mA anodal direct current with resulting current density of $0.0571\ \text{mA/cm}^2$, whereas for sham stimulation in the control group, first 30 seconds the current was ramped up to 2 mA and during last 30 seconds ramped down. These parameters for sham stimulation were shown in previous reports as mimicking same somatosensory artifact, i.e., tingling

Table 1
Subjects demographics and clinical assessment scores

Intervention	Gender	Age, y	Neurological lesion level	AIS	Time since injury, mo	ASIA UEMS
Active	M	48	C3-C5	D	38	10
Active	M	36	C4-C6	D	8	21
Active	M	53	C4-C6	D	7	17
Active	M	62	C4-C6	C	48	17
Sham	F	52	C4-C6	C	205	11
Sham	M	63	C3-C6	D	47	13
Sham	M	50	C6-C7	C	244	14
Sham	M	58	C3-C4	D	72	14

of active stimulation without producing measurable effect on cortical excitability (Gandiga, Hummel, & Cohen, 2006). During stimulation subjects were seated in their own wheelchair or a regular chair with comfortable back support.

2.3.2. Robotic-assisted training

Immediately after cortical stimulation, repetitive movement training was provided by the MAHI Exo-II exoskeleton (Fitle et al., 2015) operated in constraint mode. In this mode, the robotic device opposes the subject's movement by adding a force that requires the subject to provide greater work in order to initiate and maintain movement. Single degree of freedom movement for elbow, forearm and wrist was repeated at high intensity. Treatment was progressed gradually by increasing the number of repetitions and amount of resistance applied to each movement. On a computer screen graphic feedback about performance was given after each attempt in order to maintain motivation. Standardized rest breaks were given in order to avoid fatigue. During the study period, subjects did not participate in any other occupational therapy program involving arm and hand training. Details of the robotic training protocol are published in a previous study from our lab (Yozbatiran et al., 2012).

2.4. Clinical, functional and safety assessments:

After subjects were randomized into active or control groups, a blinded evaluator performed baseline clinical and functional assessment and repeated them within a week after the last treatment session and at the two-month follow up. To minimize the effect of trunk and limb position on test scores, standardized positions were used during all testing.

2.4.1. Measurement of safety, tolerability and feasibility

For tDCS safety, during each tDCS administration, subjects were closely monitored and asked to report and rate symptoms such as headache, neck pain, scalp pain, tingling, itching, burning sensation, skin redness, sleepiness, trouble concentrating, acute mood change and others on a ordinal scale from 1–4. The questionnaire has been standardized by Brunoni et al., 2011 for reporting of commonly seen tDCS adverse events (Brunoni et al., 2011). For tolerability subjects were asked to score their level of perceived fatigue at the beginning and end of each session on a scale ranging from 0 = no fatigue to 100 = extreme fatigue.

2.4.2. Testing for arm and hand function

The Jebsen-Taylor Hand Function Test was used as the primary outcome measure of arm and hand functions. The test (Jebsen, Taylor, Trieschmann, Trotter, & Howard, 1969) has shown to have good to excellent interrater and intrarater reliability (Beebe & Lang, 2009) and capacity for detecting performance change in activities that resemble daily life activities. Time to perform 7 everyday activities, (e.g. writing, feeding) is tested. We excluded the writing task due to heavy dependence on side with less impairment. Administration of the JTHFT subtests discontinued after 120 sec if the subject could not complete the task by that time. Scores were recorded in number of items completed/total time (in seconds). This method of recording had superiority compared to traditional recording of total time only. Thus change in number of items completed within 120 seconds could be reflected as an increase or decrease of performance.

2.4.3. Self-report of arm function with Motor Activity Log (MAL)

Subjects were asked to report 'how much' and 'how well' they have used their arm during 30 daily activities such as brushing hair, drinking from a glass, picking up phone. Two scores are typically generated from this self-report; for the purpose of this study, only the amount of use was used as we were interested in increased activity of the arm in daily life (Taub et al., 1993). A score of 0.50 was used to report as

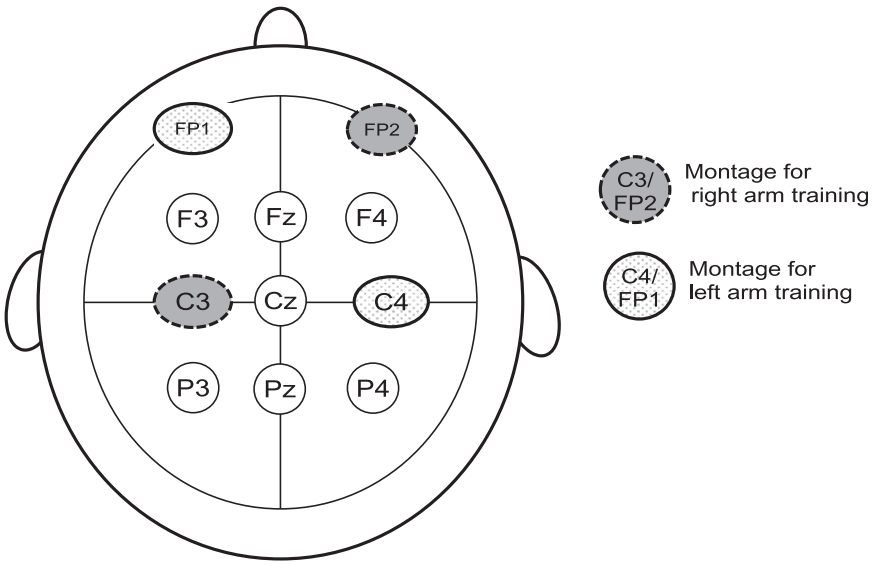

Fig. 1. 1 × 1 tDCS electrode montage. In subjects who were trained for their left arm, anodal (active) electrode was placed on the right primary motor cortex (C4) with cathode (reference) electrode placed over left supraorbicular cortex (FP1).

minimal clinically important difference (MCID) (van der Lee et al., 1999).

2.4.4. *Testing for arm strength*

Strength of selected muscles (C5–T1: biceps, triceps,wrist extensor, finger flexor, finger abductor) in both upper extremities was scored using the Medical Research Council grade (0 = absent, 5 = normal) in accordance with International Standards for Neurologic Classification of SCI (ISNCSCI) exam. Despite standardized supine position, the manual muscle testing was performed with the subject in sitting position either in their own chair or in a standard chair.

2.4.5. *Testing of muscle tone*

Modified Ashworth Scale (MAS) was used to measure the resistance during passive elbow flexion and extension, pronation and supination, wrist flexion and extension and finger extension. Sum of each score was reported (Bohannon & Smith, 1987).

2.5. *Data analysis*

The current project was preliminary in nature and aimed to provide initial data for future trials. We did tested a few hypotheses according to the following method:

(i) is the combination treatment feasible and tolerable as indexed by participation and adverse effects? For this question we provide qualitative and descriptive data. Feasibility measurements regarding whether it was possible to combine both interventions and whether the combination affected how the intervention was being administered were observed. Tolerability was measured by adherence to the trial protocol (attrition rate), level of perceived fatigue and adverse events.

(ii) is there an increased treatment effect in the active group compared to the control group when comparing the two groups favoring the active group immediately after the end of the

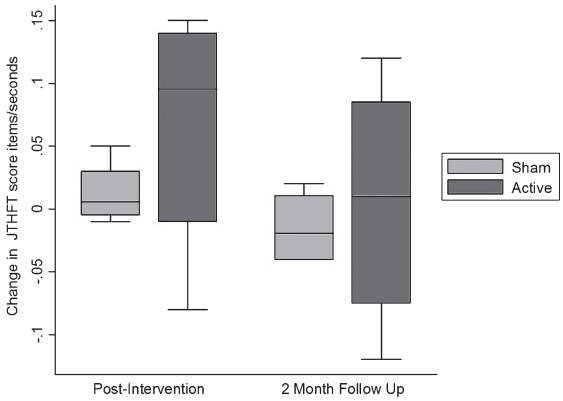

Fig. 2. Change in JTHFT scores from baseline to post-treatment and from baseline to 2-month follow-up.

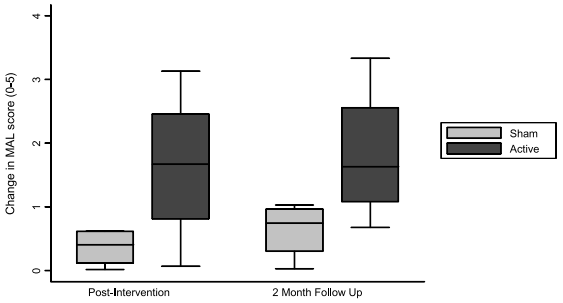

Fig. 3. Mean ± SE of MAS-AOU from baseline to post-treatment and 2-month follow-up. After ten sessions 4/4 participants (100%) achieved a clinically meaningful outcome of 0.52 points at immediate posttreatment and after 12 months, respectively. In the control group 2/4 participants (50%) at 2 weeks and 3/4 participants (75%) at 2-months achieved a clinically meaningful outcome.

treatment? For this question we tested differences between pre-test and post-test scores between the two groups using a between-group analysis with the Mann-Whitney U test.

(iii) if there is a treatment effect (question ii), are the effects lasting – when analyzing follow-up motor function? For this question we built an ANOVA model to test whether there is a significant difference between time (baseline, immediately after and follow-up) and group (active vs. sham tDCS). For the group by time interaction we used repeated measures ANOVA.

For both questions (ii) and (iii), we choose JTHFT, MAL-AOU, ASIA UEMS and MAS as dependent variables. Given the exploratory nature of these comparisons, we did not correct for multiple comparisons. Therefore α level of significance was set at 0.05. Analyses were carried out using Stata (StataCorp, College Station, TX, USA) Proportional change of motor impairment and functional scores from baseline to post intervention assessments at post-treatment assessment (post 1) and follow-up (post 2) was calculated by ([post-pre]/pre*100).

3. Results

The groups did not differ with respect to age, and gender distribution. Table 1 summarizes the demographics and clinical characteristics of the subjects.

The mean age of the patients in the active group was 49.7 ± 5.4 years and 55.7 ± 2.9 years in the sham group. At baseline the mean time since injury was significantly higher in the sham group, 141.2 ± 48.2 months compared to active group 25.2 ± 10.4 months (Mann-Whitney U test, $p = 0.04$). ASIA UEMS, JTHFT, MAL-AOU and MAS scores were not significantly different in both groups (Mann-Whitney U test, $p > 0.05$), Table 2.

In accordance with our three questions the results are presented in the following order:

3.1. Is the combination treatment feasible and tolerable as indexed by participation, adverse effects and attrition rate?

Nine subjects were randomized into active tDCS and sham tDCS group and only one subject withdrew from the study. After 5 training session the subject in the active group dropped out from the study due to transportation problems.

In the remaining group all sessions were well tolerated and none of the subjects requested the stimulation to be terminated or needed medical intervention during or after stimulation. Tolerance level was measured with VAS in response to induced fatigue at each session. On a scale from 0–100 subjects in both groups had very similar perceived fatigue level; active tDCS 11.8 ± 1.4 versus sham tDCS 12.0 ± 1.5 ($p = 0.9$). Observed (skin redness) and self-reported (tingling, sleepiness, trouble concentrating, headache, neck pain, scalp pain, scalp burning sensation and acute mood changes) side effects were usually mild and reported in both groups. Most common symptoms were tingling, skin redness and sleepiness. Frequency of tDCS related adverse events are shown in Table 3.

3.2. Is there a treatment effect when comparing the two groups favoring the active group immediately after the end of the treatment?

3.2.1. Arm and hand function

Immediately after treatment, arm and hand function as measured with JTHFT demonstrated larger improvement in the active tDCS group 40.7 ± 29.1% as compared to 6.7 ± 4.5% in the sham group (Fig. 2). The biggest improvement in the active group occurred for the 'feeding' subtest. In contrast to all other subtests, the 'feeding' test depends more on proximal arm movement rather than finger dexterity. However, the change in total JTHFT score did not show a statistically significant difference between groups ($Z = 0.577$, $p = 0.564$; Cohen's $d = 1.31$).

Table 2
Baseline clinical and functional assessments in active and sham tDCS treated subjects

	Active group ($n=4$)	Sham group ($n=4$)	
	Median (IQR range)	Median (IQR range)	p-value
ASIA UEMS (0–25)	17.0 (13.5,21.0)	14.0 (11.0,14.5)	0.343
JTHFT	0.27 (0.22,0.4)	0.23 (0.03,0.43)	0.386
MAL-AOU (0–5)	2.50 (1.47,3.6)	1.84 (0.18,3.49)	0.248
MAS (0–4)	0.75 (0.38,1.19)	0.22 (0.00,0.88)	0.124

Table 3
Frequency of adverse effects in active and sham groups

	Active group		Sham group	
Sensation	Number of subjects/number of sessions (% of total sessions)		Number of subjects/number of sessions (% of total sessions)	
Tingling	3/28	(70%)	3/21	(52.5%)
Skin Redness	4/7	(17.5%)	4/9	(22.5%)
Sleepiness	1/7	(17.5)	1/1	(2.5%)
Trouble concentrating	1/3	(7.5%)	–	
Headache	1/2	(5%)	–	
Neck pain	1/2	(5%)	–	
Scalp pain	–		1/1	(2.5%)
Scalp Burns	–		–	–
Acute Mood changes	–		–	–

3.2.2. Motor activity log – amount of use

Treatment groups had very similar MAL-AOU scores at baseline (Z = –0.577, $p = 0.564$). Immediately after treatment subjects in the active group demonstrated higher amount of use score after 10 sessions and maintained the scores at follow up. Absolute mean MAL-AOU scores changed from 1.5 ± 0.6 to 3.2 ± 0.7 (post 1) and 3.3 ± 0.7 (post 2) in active stimulation group versus sham group (from1.6 ± 0.9 to 1.9 ± 1 (post 1) and 2.2 ± 0.9 (post 2). However the MAL-AOU score immediately after treatment did not show a significant difference between groups (Z = 0.871, $p = 0.384$, Cohen's $d = 1.38$), Fig. 3.

3.2.3. ASIA upper extremity motor scores

Two subjects in the active group and three subjects in the sham group received training on their pre-injury dominant side. At baseline the groups had similar muscle strength scores (Z = –1.169, $p = 0.242$). Immediately after treatment the change in ASIA UEMS score did not significantly differ between groups (Z = –0.744, $p = 0.457$, Cohen's $d = 0.88$).

3.2.4. Muscle tone

Average mean MAS scores for active group demonstrated bigger decrease in spasticity as measured with MAS (from 1.00 ± 0.3 to 0.7 ± 0.3 (post 1) and 0.7 ± 0.2 (post 2) but neither of the groups showed a significant difference in muscle tone from baseline to post-treatment (Z = 1.648, $p = 0.099$, Cohen's $d = 1.32$).

3.3. If there is a treatment effect (question ii), are the effects lasting when analyzing follow-up motor function?

Analysis for the outcome variables JTHFT, MAL-AOU, ASIA-UEMS and MAS revealed no significant interaction effect, showing that the improvement seen immediately after did not last for the follow-up. Repeated ANOVA revealed no significant group-by-time interaction for total JTHFT scores F(2,5) = 0.429, $p = 0.673$, MAL-AOU scores F(2,5) = 1.675, $p = 0.277$; ASIA UEMS scores F(2,5) = 0.732, $p = 0.526$; muscle tone F (2,5) = 2.355, $p = 0.190$.

4. Discussion

4.1. Tolerability and safety of combination therapy

The current 'proof of concept' study has shown that the combination of non-invasive brain stimu-

lation to repetitive robotic training is feasible and tolerable. Attrition rate was low and withdrawal of one subject was not caused by treatment related adverse event that resulted with refusal to continue the treatment. Moreover in those that were able to have reliable transportation, the compliance rate was 100% and no loss to follow-up. None of the subjects missed a session due to additional adverse events such as excessive fatigue, discomfort or muscle soreness resulting from intensive training.

In our observation participants in active and sham groups demonstrated overall similar subjective findings. Adverse events related to tDCS were very limited and similar to those reported previously (for review see (Brunoni et al., 2011). In both groups the most frequent symptoms were tingling, mild skin irritation on the supraorbital area beneath reference electrode followed by less common symptoms such as headache, neck pain, sleepiness, trouble in concentration. Scalp burn and acute mood changes were not observed.

4.2. Efficacy of combination therapy

Our study demonstrated that combining non-invasive brain stimulation with repetitive robotic peripheral training has a potential to augment arm and hand functions compared to a control group receiving only repetitive arm training. Addition of ten sessions of anodal tDCS of M1 and cathodal tDCS of supraorbital area to high intensity repetitive training could be associated with better outcomes immediately after treatment with a lasting effect of at least 2-months. At baseline, the groups had similar hand function as measured with the Jebsen-Taylor Hand Function test and Motor Activity Log (MAL). All of the subjects had minimal movements in thumb and index fingers, which translated into pinch grip, manipulation and moving small objects such as turning pages, picking up a penny and paper clips, or stacking checkers. After ten sessions of robotic-assisted arm training, all patients demonstrated improvements in arm and hand functions with greater gain in the active tDCS group. Half of the of the subjects in the active group have shown a 50% proportional improvement in the JTHFT performance while none of the patients in the sham group reached this level. Similarly, the increase in amount of use of the trained hand in daily activities (MAL items) has also been greater in the active stimulation group. Immediately after ten sessions four of four participants (100%) achieved a clinically meaningful outcome of 0.50 points, whereas this ratio was two out of four in the control group. Improvement in arm use continued for control group and three out of four persons exceeded MCID at two-months. In addition to the measured functions, patients in the active group reported overall improved sensation, such as "my whole arm feels more alive. I started to feel my arm more" in the trained arm (3 out of 4), improved fine motor skills, such as "I am able to swipe the phone (touch screen) with my thumb for the first time since the injury" (1 out of 4), "I can use this hand to put my hat on without difficulty" (1 out of 4). One subject even started to move the ipsilateral toe, which he was not able to before. Interestingly, almost all of these changes occurred on the second week of their treatment with a predominant change in their fine motor skills. In the sham group, only one subject reported considerable change in arm and hand movements; extending his arm out from the car window with ease to insert the parking ticket into the machine. Before training, he needed to get out of his car and use his other hand. Given the relatively short treatment duration, these observations evoke very interesting questions about the underlying mechanisms of facilitation of neuroplastic changes at cortical and spinal levels when subjects are exposed to high intensity repetitive training alone or combined with the facilitatory effect of motor cortex anodal and supraorbital cathodal tDCS. Combination therapies with tDCS have been successfully applied in a single session or over repeated sessions to modulate cortical excitability and enhance motor behavior in stroke patients (Hummel et al., 2005). Because persons with incomplete tetraplegia may have similar mechanisms of recovery as persons with hemiplegia since both may have altered and inappropriate sensory input and motor output, incorporating intense activity, repeated practice, attention and somatosensory augmentation concurrent with movement practice may facilitate neural plasticity and functional recovery (Backus, 2010).

Priming of the motor cortex to augment functional benefits with peripheral training is relatively new, and therapeutic effects of motor cortex stimulation as an intervention alone or combined with peripheral training is reported by only a few researchers. Belci et al., 2004, applied 10 Hz of paired rTMS over 5 sessions in chronic incomplete SCI and found improvement in ASIA motor and sensory scores and hand function sustained at 3 weeks follow-up (Belci, Catley, Husain, Frankel, & Davey, 2004). In another study, Gomes-Osman & Field-Fote, 2015,

researchers have shown that 3 sessions of combined 10-Hz repetitive transcranial magnetic stimulation (rTMS, over motor cortex) and repetitive task practice (RTP) resulted with higher improvement in hand function compared to sham-rTMS and RTP(Gomes-Osman & Field-Fote, 2015). Similarly, Kuppuswamy et al. (2011) have demonstrated improvement in arm and hand function immediately after 5 sessions of active 5-Hz rTMS sensorimotor cortex stimulation. In addition to upper limb motor recovery, other researchers have also looked into benefits of combination therapies to enhance gait functions in spinal cord injury. In a study by Kumru et al., 15 daily sessions of 20-Hz rTMS over leg motor area combined with gait rehabilitation resulted with improvement in walking speed, lower limb spasticity and lower limb muscle strength (Kumru et al., 2013).

As reported in the aforementioned studies, combining the effects of cortical stimulation with peripheral training can produce favorable functional outcomes. Although we observed similar results in our study, our design differed in type and dose of motor cortex stimulation and active exercises. First, instead of rTMS we used tDCS. tDCS has been shown to be a safe, portable noninvasive brain stimulation technique capable to modulate excitability of targeted brain regions by altering neuronal transmembrane potentials. In patients with chronic incomplete SCI, the magnitude of change in the CST excitability has been shown to be significant with 20 minutes of 2 mA anodal tDCS compared to 1 mA anodal tDCS. In addition, tDCS allows for very effective sham stimulation. None of our patients were able to distinguish whether they were receiving active or sham stimulation. Second, we used repetitive robotic-assisted training instead of repetitive task practice. Robotic-assisted training with MAHI Exo-II exoskeleton device has produced positive outcomes in other studies from our lab with spinal cord injury patients (Kadivar et al., 2012; Yozbatiran et al., 2012) High intensity repetitive training of elbow flexion/ extension, forearm pronation/ supination, wrist flexion/extension and ulnar/radial deviation was successfully delivered. Within subjects' number of repetition reached up to 1000 on average in one-hour. All subjects were trained in the constraint mode, and the amount of challenge during each session was experimentally modulated by the researcher. Notably, some of the changes in hand functions occurred despite an absence of specific training of finger movements.

4.3. Study limitations

Whilst this study used only a small group of patients and there is no substantive evidence for the use of tDCS in motor recovery in iSCI, it is reasonable to speculate that augmentation of corticospinal activity in spared fibers via primary motor cortex stimulation with anodal tDCS had positive effects on hand fine motor skills that result in improved performance in daily activities such as picking and moving small objects, feeding, grasping and releasing objects. However, effects of improved motor cortex and corticospinal tract activity on relearning motor skills, generalization of motor outcomes and overcoming learned non-use after spinal cord injury may all have contributed to this improvement. Therefore the differential effect of each of these contributing factors should be explored in future studies. Furthermore, we also suggest using outcome measures that are specifically designed to reflect changes in the quality of movement, and exclude compensatory movements which do not necessarily reflect neuroplasticity.

5. Conclusions

In conclusion, in this pilot study we have demonstrated that combination therapy protocol using 20 minutes of 2 mA anodal tDCS over M1 followed by 60 minutes of high intensity repetitive training with a robotic exoskeleton is safe, tolerable and feasible in treatment of impaired arm and hand functions in chronic incomplete spinal cord injury. Our findings show promise in improving arm and hand function with this combination therapy. However, there is an increased need to explore the effects of the suggested protocol in larger sample size with homogenous groups, especially in patients with similar time lapse since injury, impairment level (AIS level), and baseline hand motor function.

Acknowledgments

This study was supported by a pilot research grant from Mission Connect a project of TIRR Foundation and by The Institute for Rehabilitation and Research at Memorial Hermann. Also, we wish to thank Jay Karri for his kind assistance.

Conflict of interest

The authors have no conflict of interest to report.

References

Anderson, K. D. (2004). Targeting recovery: Priorities of the spinal cord-injured population. *Journal of neurotrauma*, *21*(10), 1371-1383.

Backus, D. (2010). Exploring the potential for neural recovery after incomplete tetraplegia through nonsurgical interventions. *PM R*, *2*(12 Suppl 2), S279-S285. doi:10.1016/j.pmrj.2010.10.004

Bastani, A., & Jaberzadeh, S. (2012). Does anodal transcranial direct current stimulation enhance excitability of the motor cortex and motor function in healthy individuals and subjects with stroke: A systematic review and meta-analysis. *Clinical neurophysiology : Official Journal of the International Federation of Clinical Neurophysiology*, *123*(4), 644-657. doi:10.1016/j.clinph.2011.08.029

Beebe, J. A., & Lang, C. E. (2009). Relationships and responsiveness of six upper extremity function tests during the first six months of recovery after stroke. *Journal of Neurologic Physical Therapy : JNPT*, *33*(2), 96-103. doi:10.1097/NPT.0b013e3181a33638

Belci, M., Catley, M., Husain, M., Frankel, H. L., & Davey, N. J. (2004). Magnetic brain stimulation can improve clinical outcome in incomplete spinal cord injured patients. *Spinal Cord*, *42*(7), 417-419. doi:10.1038/sj.sc.3101613

Bohannon, R. W., & Smith, M. B. (1987). Interrater reliability of a modified Ashworth scale of muscle spasticity. *Physical Therapy*, *67*(2), 206-207. Retrieved from http://ptjournal.apta.org/content/67/2/206.full.pdf

Brunoni, A. R., Amadera, J., Berbel, B., Volz, M. S., Rizzerio, B. G., & Fregni, F. (2011). A systematic review on reporting and assessment of adverse effects associated with transcranial direct current stimulation. *The International Journal of Neuropsychopharmacology / Official Scientific Journal of the Collegium Internationale Neuropsychopharmacologicum (CINP)*, *14*(8), 1133-1145. doi:10.1017/S1461145710001690

Cabral, M. E., Baltar, A., Borba, R., Galvao, S., Santos, L., Fregni, F., & Monte-Silva, K. (2015). Transcranial direct current stimulation: Before, during, or after motor training? *Neuroreport*, *26*(11), 618-622. doi:10.1097/WNR.0000000000000397

Freund, P., Schneider, T., Nagy, Z., Hutton, C., Weiskopf, N., Friston, K.,. . . Thompson, A. J. (2012). Degeneration of the injured cervical cord is associated with remote changes in corticospinal tract integrity and upper limb impairment. *PloS one*, *7*(12), e51729. doi:10.1371/journal.pone.0051729

Fitle, K. D., Pehlivan, A. U., & O'Malley, M. K. (2015). A robotic exoskeleton for rehabilitation and assessment of the upper limb following incomplete spinal cord injury. *Robotics and Automation (ICRA), 2015 IEEE International Conference on.* 2015:4960-4966. doi: 10.1109/ICRA.2015.7139888.

Gandiga, P. C., Hummel, F. C., & Cohen, L. G. (2006). Transcranial DC stimulation (tDCS): A tool for double-blind sham-controlled clinical studies in brain stimulation. *Clin Neurophysiol*, *117*(4), 845-850. doi:10.1016/j.clinph.2005.12.003

Garber, S. L., & Gregorio, T. L. (1990). Upper extremity assistive devices: Assessment of use by spinal cord-injured patients with quadriplegia. *The American Journal of Occupational Therapy : Official Publication of the American Occupational Therapy Association*, *44*(2), 126-131. Retrieved from http://ajot.aota.org/article.aspx?articleid=1879368

Gomes-Osman, J., & Field-Fote, E. C. (2015). Improvements in hand function in adults with chronic tetraplegia following a multiday 10-Hz repetitive transcranial magnetic stimulation intervention combined with repetitive task practice. *J Neurol Phys Ther*, *39*(1), 23-30. doi:10.1097/NPT.0000000000000062

Henderson, L. A., Gustin, S. M., Macey, P. M., Wrigley, P. J., & Siddall, P. J. (2011). Functional reorganization of the brain in humans following spinal cord injury: Evidence for underlying changes in cortical anatomy. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *31*(7), 2630-2637. doi:10.1523/JNEUROSCI.2717-10.2011

Hicks, A. L., Martin, K. A., Ditor, D. S., Latimer, A. E., Craven, C., Bugaresti, J., & McCartney, N. (2003). Long-term exercise training in persons with spinal cord injury: Effects on strength, arm ergometry performance and psychological well-being. *Spinal Cord*, *41*(1), 34-43. doi:10.1038/sj.sc.3101389

Hummel, F., Celnik, P., Giraux, P., Floel, A., Wu, W. H., Gerloff, C., & Cohen, L. G. (2005). Effects of non-invasive cortical stimulation on skilled motor function in chronic stroke. *Brain*, *128*(Pt 3), 490-499. doi:10.1093/brain/awh369

Jebsen, R. H., Taylor, N., Trieschmann, R. B., Trotter, M. J., & Howard, L. A. (1969). An objective and standardized test of hand function. *Archives of Physical Medicine and Rehabilitation*, *50*(6), 311-319.

Kadivar, Z., L. Sullivan, J., P. Eng, D., U. Pehlivan, A., K. O. Malley, M., Yozbatiran, N.,. . . & Francisco, G. E. (2012). RiceWrist robotic device for upper limb training: Feasibility study and case report of two tetraplegic persons with spinal cord injury. *International Journal of Biological Engineering*, *2*(4), 27-38. doi:10.5923/j.ijbe.20120204.01

Kriz, J., Kozak, J., & Zedka, M. (2012). Primary motor cortex inhibition in spinal cord injuries. *Neuro Endocrinology Letters*, *33*(4), 431-441. doi:NEL330412A10

Kumru, H., Benito, J., Murillo, N., Valls-Sole, J., Valles, M., Lopez-Blazquez, R.,. . . Vidal, J. (2013). Effects of high-frequency repetitive transcranial magnetic stimulation on motor and gait improvement in incomplete spinal cord injury patients. *Neurorehabil Neural Repair*, *27*(5), 421-429. doi:10.1177/1545968312471901

Kuppuswamy, A., Balasubramaniam, A. V., Maksimovic, R., Mathias, C. J., Gall, A., Craggs, M. D., & Ellaway, P. H. (2011). Action of 5Hz repetitive transcranial magnetic stimulation on sensory, motor and autonomic function in human spinal cord injury. *Clin Neurophysiol*, *122*(12), 2452-2461. doi:10.1016/j.clinph.2011.04.022

Laubis-Herrmann, U., Dichgans, J., Bilow, H., & Topka, H. (2000). Motor reorganization after spinal cord injury: Evidence of adaptive changes in remote muscles. *Restorative Neurology and Neuroscience*, *17*(4), 175-181. Retrieved from http://content.iospress.com/articles/restorative-neurology-and-neuroscience/rnn00155

Lotze, M., Laubis-Herrmann, U., Topka, H., Erb, M., & Grodd, W. (1999). Reorganization in the primary motor cortex after spinal cord injury–A functional Magnetic Resonance (fMRI) study. *Restorative Neurology and Neuroscience*, *14*(2-3), 183-187. Retrieved from http://

content.iospress.com/articles/restorative-neurology-and-neuroscience/rnn00078

Nitsche, M. A., Liebetanz, D., Antal, A., Lang, N., Tergau, F., & Paulus, W. (2003). Modulation of cortical excitability by weak direct current stimulation–technical, safety and functional aspects. *Supplements to Clinical Neurophysiology*, *56*, 255-276.

Nitsche, M. A., Liebetanz, D., Tergau, F., & Paulus, W. (2002). Modulation of cortical excitability by transcranial direct current stimulation. *Der Nervenarzt*, *73*(4), 332-335.

Nitsche, M. A., & Paulus, W. (2001). Sustained excitability elevations induced by transcranial DC motor cortex stimulation in humans. *Neurology*, *57*(10), 1899-1901.

Pehlivan, A. U., Sergi, F., Erwin, A., Yozbatiran, N., Francisco, G. E., & O'Malley, M. K. (2014). Design and validation of the RiceWrist-S exoskeleton for robotic rehabilitation after incomplete spinal cord injury. *Robotica*. Retrieved from http://www.scopus.com/inward/record.url?eid=2-s2.0-84902716924&partnerID=40&md5=c1cd2f0879ba62b1fc6667976ac689b0

Reis, J., Schambra, H. M., Cohen, L. G., Buch, E. R., Fritsch, B., Zarahn, E.,. . . & Krakauer, J. W. (2009). Noninvasive cortical stimulation enhances motor skill acquisition over multiple days through an effect on consolidation. *Proceedings of the National Academy of Sciences of the United States of America*, *106*(5), 1590-1595. doi:10.1073/pnas.0805413106

Sadowsky, C. L., & McDonald, J. W. (2009). Activity-based restorative therapies: Concepts and applications in spinal cord injury-related neurorehabilitation. *Developmental Disabilities Research Reviews*, *15*(2), 112-116. doi:10.1002/ddrr.61

Snoek, G. J., Ijzerman, M. J., Hermens, H. J., Maxwell, D., & Biering-Sorensen, F. (2004). Survey of the needs of patients with spinal cord injury: Impact and priority for improvement in hand function in tetraplegics. *Spinal Cord*, *42*(9), 526-532. doi:10.1038/sj.sc.3101638

Taub, E., Miller, N. E., Novack, T. A., Cook, E. W. 3rd, Fleming, W. C., Nepomuceno, C. S.,. . . Crago, J. E. (1993). Technique to improve chronic motor deficit after stroke. *Archives of Physical Medicine and Rehabilitation*, *74*(4), 347-354.

Topka, H., Cohen, L. G., Cole, R. A., & Hallett, M. (1991). Reorganization of corticospinal pathways following spinal cord injury. *Neurology*, *41*(8), 1276-1283.

van der Lee, J. H., Wagenaar, R. C., Lankhorst, G. J., Vogelaar, T. W., Deville, W. L., & Bouter, L. M. (1999). Forced use of the upper extremity in chronic stroke patients: Results from a single-blind randomized clinical trial. *Stroke*, *30*(11), 2369-2375. Retrieved from http://www.ncbi.nlm.nih.gov/pubmed/10548673

Yozbatiran, N., Berliner, J., O'Malley, M. K., Pehlivan, A. U., Kadivar, Z., Boake, C., & Francisco, G. E. (2012). Robotic training and clinical assessment of upper extremity movements after spinal cord injury: A single case report. *Journal of Rehabilitation Medicine*, *44*(2), 186-188. Retrieved from http://www.scopus.com/inward/record.url?eid=2-s2.0-84856257654&partnerID=40&md5=d6e36ea005022ea337d209fcb904bb7e

Zariffa, J., Kapadia, N., Kramer, J. L., Taylor, P., Alizadeh-Meghrazi, M., Zivanovic, V.,. . . Steeves, J. D. (2011). Effect of a robotic rehabilitation device on upper limb function in a sub-acute cervical spinal cord injury population. *IEEE...International Conference on Rehabilitation Robotics : [proceedings]*, *2011*, 5975400. doi:10.1109/ICORR.2011.5975400