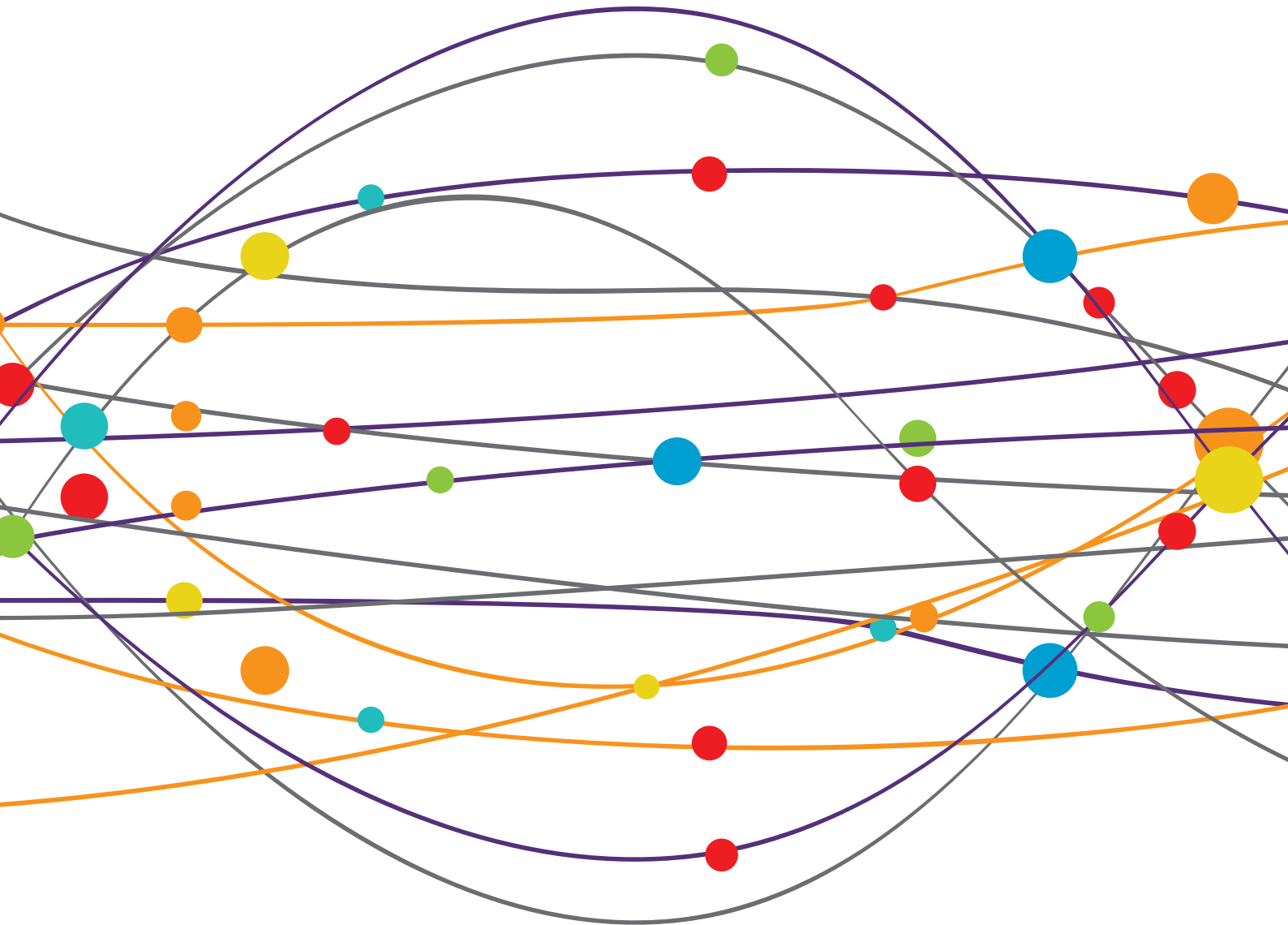


NON-INVASIVE BRAIN STIMULATION IN THE STUDY AND MODULATION OF METAPLASTICITY IN NEUROLOGICAL DISORDERS

EDITED BY: Mariagiovanna Cantone, Giuseppe Lanza, Federico Ranieri,
George M. Opie and Carmen Terranova
PUBLISHED IN: Frontiers in Neurology





frontiers

Frontiers eBook Copyright Statement

The copyright in the text of individual articles in this eBook is the property of their respective authors or their respective institutions or funders. The copyright in graphics and images within each article may be subject to copyright of other parties. In both cases this is subject to a license granted to Frontiers.

The compilation of articles constituting this eBook is the property of Frontiers.

Each article within this eBook, and the eBook itself, are published under the most recent version of the Creative Commons CC-BY licence.

The version current at the date of publication of this eBook is CC-BY 4.0. If the CC-BY licence is updated, the licence granted by Frontiers is automatically updated to the new version.

When exercising any right under the CC-BY licence, Frontiers must be attributed as the original publisher of the article or eBook, as applicable.

Authors have the responsibility of ensuring that any graphics or other materials which are the property of others may be included in the CC-BY licence, but this should be checked before relying on the CC-BY licence to reproduce those materials. Any copyright notices relating to those materials must be complied with.

Copyright and source acknowledgement notices may not be removed and must be displayed in any copy, derivative work or partial copy which includes the elements in question.

All copyright, and all rights therein, are protected by national and international copyright laws. The above represents a summary only. For further information please read Frontiers' Conditions for Website Use and Copyright Statement, and the applicable CC-BY licence.

ISSN 1664-8714

ISBN 978-2-88971-260-1

DOI 10.3389/978-2-88971-260-1

About Frontiers

Frontiers is more than just an open-access publisher of scholarly articles: it is a pioneering approach to the world of academia, radically improving the way scholarly research is managed. The grand vision of Frontiers is a world where all people have an equal opportunity to seek, share and generate knowledge. Frontiers provides immediate and permanent online open access to all its publications, but this alone is not enough to realize our grand goals.

Frontiers Journal Series

The Frontiers Journal Series is a multi-tier and interdisciplinary set of open-access, online journals, promising a paradigm shift from the current review, selection and dissemination processes in academic publishing. All Frontiers journals are driven by researchers for researchers; therefore, they constitute a service to the scholarly community. At the same time, the Frontiers Journal Series operates on a revolutionary invention, the tiered publishing system, initially addressing specific communities of scholars, and gradually climbing up to broader public understanding, thus serving the interests of the lay society, too.

Dedication to Quality

Each Frontiers article is a landmark of the highest quality, thanks to genuinely collaborative interactions between authors and review editors, who include some of the world's best academicians. Research must be certified by peers before entering a stream of knowledge that may eventually reach the public - and shape society; therefore, Frontiers only applies the most rigorous and unbiased reviews.

Frontiers revolutionizes research publishing by freely delivering the most outstanding research, evaluated with no bias from both the academic and social point of view. By applying the most advanced information technologies, Frontiers is catapulting scholarly publishing into a new generation.

What are Frontiers Research Topics?

Frontiers Research Topics are very popular trademarks of the Frontiers Journals Series: they are collections of at least ten articles, all centered on a particular subject. With their unique mix of varied contributions from Original Research to Review Articles, Frontiers Research Topics unify the most influential researchers, the latest key findings and historical advances in a hot research area! Find out more on how to host your own Frontiers Research Topic or contribute to one as an author by contacting the Frontiers Editorial Office: frontiersin.org/about/contact

NON-INVASIVE BRAIN STIMULATION IN THE STUDY AND MODULATION OF METAPLASTICITY IN NEUROLOGICAL DISORDERS

Topic Editors:

Mariagiovanna Cantone, Sant'Elia Hospital, Italy

Giuseppe Lanza, University of Catania, Italy

Federico Ranieri, University of Verona, Italy

George M. Opie, University of Adelaide, Australia

Carmen Terranova, University of Messina, Italy

Citation: Cantone, M., Lanza, G., Ranieri, F., Opie, G. M., Terranova, C., eds. (2021). Non-Invasive Brain Stimulation in the Study and Modulation of Metaplasticity in Neurological Disorders. Lausanne: Frontiers Media SA.
doi: 10.3389/978-2-88971-260-1

Table of Contents

- 04 Editorial: Non-invasive Brain Stimulation in the Study and Modulation of Metaplasticity in Neurological Disorders**
Mariagiovanna Cantone, Giuseppe Lanza, Federico Ranieri,
George M. Opie and Carmen Terranova
- 07 Neuroplasticity and Epilepsy Surgery in Brain Eloquent Areas: Case Report**
Pedro Jesus Serrano-Castro, Bienvenido Ros-López,
Victoria Eugenia Fernández-Sánchez, Natalia García-Casares,
Luis Muñoz-Becerra, Pablo Cabezudo-Garcia, Maria José Aguilar-Castillo,
Maria Vidal-Denis, Esperanza Cruz-Andreotti, Maria Jose Postigo-Pozo,
Guillermo Estivill-Torrús and Guillermo Ibañez-Botella
- 15 Novel TMS for Stroke and Depression (NoTSAD): Accelerated Repetitive Transcranial Magnetic Stimulation as a Safe and Effective Treatment for Post-stroke Depression**
Jessica Frey, Umer Najib, Christa Lilly and Amelia Adcock
- 22 Prediction of Motor Recovery in the Upper Extremity for Repetitive Transcranial Magnetic Stimulation and Occupational Therapy Goal Setting in Patients With Chronic Stroke: A Retrospective Analysis of Prospectively Collected Data**
Toyohiro Hamaguchi, Naoki Yamada, Takuya Hada and Masahiro Abo
- 31 How to Design Optimal Accelerated rTMS Protocols Capable of Promoting Therapeutically Beneficial Metaplasticity**
Alix C. Thomson and Alexander T. Sack
- 37 Transcranial Direct Current Stimulation to Enhance Cognitive Impairment in Parkinson's Disease: A Systematic Review and Meta-Analysis**
Diana M. A. Suarez-García, Johan S. Grisales-Cárdenas, Máximo Zimmerman
and Juan F. Cardona
- 45 Facilitative Effects of Transcranial Direct Current Stimulation on Semantic Memory Examined by Text-Mining Analysis in Patients With Schizophrenia**
Chika Sumiyoshi, Zui Narita, Takuma Inagawa, Yuji Yamada, Kazuki Sueyoshi,
Yumi Hasegawa, Aya Shirama, Ryota Hashimoto and Tomiki Sumiyoshi
- 54 Neurobiological After-Effects of Low Intensity Transcranial Electric Stimulation of the Human Nervous System: From Basic Mechanisms to Metaplasticity**
Sohaib Ali Korai, Federico Ranieri, Vincenzo Di Lazzaro, Michele Papa
and Giovanni Cirillo
- 68 Repetitive Transcranial Magnetic Stimulation at Different Sites for Dysphagia After Stroke: A Randomized, Observer-Blind Clinical Trial**
Lida Zhong, Jinzhu Rao, Jing Wang, Fang Li, Yang Peng, Huiyu Liu,
Yan Zhang and Pu Wang



Editorial: Non-invasive Brain Stimulation in the Study and Modulation of Metaplasticity in Neurological Disorders

Mariagiovanna Cantone^{1*}, Giuseppe Lanza^{2,3}, Federico Ranieri⁴, George M. Opie⁵ and Carmen Terranova⁶

¹ Department of Neurology, Sant'Elia Hospital, ASP Caltanissetta, Caltanissetta, Italy, ² Department of Surgery and Medical-Surgical Specialties, University of Catania, Catania, Italy, ³ Department of Neurology IC, Oasi Research Institute-IRCCS, Troina, Italy, ⁴ Unit of Neurology, Department of Neuroscience, Biomedicine and Movement Sciences, University of Verona, Verona, Italy, ⁵ Discipline of Physiology, Adelaide Medical School, The University of Adelaide, Adelaide, SA, Australia, ⁶ Department of Clinical and Experimental Medicine, University of Messina, Messina, Italy

Keywords: transcranial magnetic stimulation, direct current stimulation, neuroplasticity, stroke rehabilitation, depression treatment

Editorial on the Research Topic

Non-invasive Brain Stimulation in the Study and Modulation of Metaplasticity in Neurological Disorders

OPEN ACCESS

Edited and reviewed by:

Thomas Platz,
University of Greifswald, Germany

*Correspondence:

Mariagiovanna Cantone
m.cantone@asp.cl.it
orcid.org/0000-0002-9072-4971

Specialty section:

This article was submitted to
Neurorehabilitation,
a section of the journal
Frontiers in Neurology

Received: 07 June 2021

Accepted: 10 June 2021

Published: 30 June 2021

Citation:

Cantone M, Lanza G, Ranieri F,
Opie GM and Terranova C (2021)
Editorial: Non-invasive Brain
Stimulation in the Study and
Modulation of Metaplasticity in
Neurological Disorders.
Front. Neurol. 12:721906.
doi: 10.3389/fneur.2021.721906

This Research Topic, which consists of 8 articles by a total of >40 authors, addresses different aspects of metaplasticity in acquired neurological and psychiatric disorders. Metaplasticity refers to the activity-dependent modulation of synaptic plasticity. This pivotal determinant of learning, memory, and other functions represents a higher order of synaptic plasticity that acts on the threshold for modifying synaptic strength (1). However, our understanding of the cellular and molecular mechanisms underlying distinct forms of synaptic plasticity, including metaplasticity, remains limited. Moreover, impaired synaptic plasticity, the so-called “maladaptive plasticity,” has been associated with the pathogenesis and trajectory of several brain diseases, including contributions to the dysfunctional remodeling of underlying neural networks (2–5).

Given its role in regulating synaptic plasticity, alterations to metaplastic mechanisms are likely to represent an important element of many neurological disorders. Until relatively recently, though, investigation of these processes was limited to invasive techniques in animal models. However, the development of non-invasive brain stimulation techniques (NIBS) has meant that it is now possible to induce and modulate metaplasticity in human subjects. Excitingly, there is a rapidly growing constellation of novel interventions that have been developed using NIBS, many of which are showing promise as therapeutic tools for treating neurological and neuropsychiatric disorders, despite our still limited understanding of the contribution made by metaplasticity. In support of this, the study by Thomson and Sack reviewed studies utilizing transcranial magnetic stimulation (TMS, a form of NIBS involving magnetic pulses applied over the scalp) to study and modulate metaplasticity, with specific interest in clinical applications. In particular, they focused on the use of repetitive TMS (rTMS) with intermittent theta burst stimulation (iTBS) and continuous TBS, as these are two of the most known and applied stimulation paradigms within research and clinical settings. After reviewing the relevant literature, the authors concluded that there is indeed a great potential to develop metaplasticity-based treatments to induce or restore a desired level of synaptic plasticity. They further identified accelerated iTBS at longer intervals (60 min) as being of particular

interest, as it seems to maximize metaplasticity effects and clinical outcomes.

While TMS was the original NIBS technique to be used for the investigation of metaplasticity, the more recently developed transcranial direct (tDCS) and alternating (tACS) current stimulation, both of which involve low intensity electrical stimulation to the scalp, have also become widely applied within this field. In the study by Korai et al., the neurobiological mechanisms underlying the after-effects of tDCS and tACS was therefore reviewed. The authors discuss that, in contrast to TMS, these forms of NIBS do not produce action potentials in affected tissues. Instead, they modulate membrane potential within a sub-threshold range, and this leads to consequent changes in synaptic transmission. The role of meta-plasticity in mediating these effects is further discussed by the authors. In particular, the way in which synaptic efficacy is effectively modulated only when concurrent neuronal discharge take place (6, 7). This opens new insights on rehabilitation protocols based on concomitant NIBS and training-induced neuronal activation.

Although applied broadly across many clinical domains, there has been a preponderance of NIBS-based research in the area of stroke. In particular, the development of interventions able to promote functionally beneficial patterns of brain activity in stroke patients has been common, and this approach likely involves metaplastic mechanisms. In an alternative take on this goal, the study by Hamaguchi et al. instead aimed to identify if it is possible to predict participants that will benefit from a combination of NIBS and occupational therapy (OT) (i.e., “responders”) based on pre-treatment functional scores. In 1,254 patients with upper extremity post-stroke paralysis, the authors therefore assessed if the response to low frequency (i.e., 1 Hz) rTMS applied to the contralesional primary motor cortex (M1) immediately prior to OT could be predicted by pretreatment Fugl-Meyer Motor Assessment (FMA) scores of the upper limb. The intervention showed a facilitation of muscle movements by the rTMS-modulation of M1 excitability. Moreover, the probability of being non-responders was 59.2% when the initial FMA score was 48.9, whereas when the initial score was 38.8 the incidence of responders and hyper-responders was 45.5 and 16.0%, respectively. Notably, ~45% of the patients with FMA scores from 30 to 40 before treatment improved, and even >25% of those with more severe initial values. Overall, these results suggest that pretreatment assessment can estimate the possibility of a patient's recovery in the chronic phase, with relevant implications for therapists and patient's compliance and cooperation.

Using a slightly different approach that nonetheless highlights the utility of combining NIBS with functional interventions in stroke patients, the study by Zhong et al. tested how the site of stimulation influences recovery from dysphagia in subacute stroke patients. Specifically, the benefit of 5 Hz-rTMS combined with standard sensory-motor rehabilitation of dysphagia was compared when applying stimulation to the M1 and cerebellum of both affected and unaffected hemispheres. They reported that, relative to a non-stimulated control group, 2-weeks of combined stimulation and training resulted in improved recovery, and this was consistent across all sites of stimulation.

This implies that rTMS may have stimulated the training-induced plasticity involved in swallowing control, possibly by acting on different circuits, although the specific pathomechanisms need to be clarified.

While a large amount of the literature utilizing NIBS in stroke has been focused on improving motor symptoms, the interesting study by Fray et al. instead evaluated the use of intense rTMS to treat post-stroke depression (PSD). In six subacute stroke patients, high-frequency (20 Hz) rTMS was applied over the left dorsolateral prefrontal cortex (DLPFC) during five sessions per day and over 4 consecutive days (20 sessions in total). At the end of the procedure and after 3 months, scores of depression significantly decreased, without any procedure-related adverse event. The authors concluded that, despite the small sample size of this pilot study, intense rTMS may be a safe and effective alternative or adjunctive therapy for PSD patients.

In further support of the cognitive benefits that are achievable when applying NIBS in the clinic, the elegant study by Sumiyoshi et al. determined whether tDCS improves semantic memory in schizophrenia patients, assessed using text-mining analyses of category fluency data. Indeed, semantic memory deficits have been previously reported in schizophrenia and associated with negative symptoms and quality of life. In 28 schizophrenia patients, cognitive assessment was carried out at baseline and 1 month after tDCS, which was performed twice per day for five consecutive days, with the anode electrode over the left DLPFC and cathode electrode over the right supraorbital area. After multi-session tDCS, the authors observed a normalization of semantic associations. The left prefrontal region is assumed to be related to the ability of tDCS to improve the organization of information and retrieval of clustered words, thus supporting the role of neuromodulation in improving cognitive functions in psychiatric disorders.

The third review within this edition also serves to demonstrate the cognitive benefits that can be derived from utilizing NIBS as an adjunctive therapy within a clinical population. Accordingly, the mini-review by Suarez-Garcia et al. sought to characterize the current evidence supporting the use of tDCS for treating cognitive impairment in Parkinson's disease (PD). A systematic review was used to identify 8 studies, the data from which was subsequently entered in to a meta-analysis. Although the results of this analysis were limited by the low number of studies and the heterogeneity of stimulation protocols and clinical features, they nonetheless identified strong benefits to executive functions in patients. In particular, anodal tDCS appears to improve problem solving and planning, verbal fluency, and cognitive flexibility.

Finally, an example of metaplastic modulation in clinical practice has been described in the case reported by Serrano-Castro et al. Despite an invasive neurostimulation approach, they opened the way to a customized neuroplastic-guided rehabilitation protocol, which allowed a previously inoperable tumor to be successfully removed and subsequently help treat the patient's refractory epilepsy.

In conclusion, this Research Topic includes a number of remarkable advances that further our understanding of the complex phenomena underlying metaplasticity, demonstrate how aberrant metaplasticity can contribute to

pathophysiology, and show that modifying metaplasticity with NIBS can be an effective avenue for treating network disorders of the brain. Translationally, this will encourage future clinical and neurophysiological studies and open novel therapeutic perspectives in this fascinating topic.

REFERENCES

1. Müller-Dahlhaus F, Ziemann U. Metaplasticity in human cortex. *Neuroscientist*. (2015) 21:185–202. doi: 10.1177/1073858414526645
2. Cantone M, Catalano MA, Lanza G, La Delfa G, Ferri R, Pennisi M, et al. Motor and perceptual recovery in adult patients with mild intellectual disability. *Neural Plast*. (2018) 2018:3273246. doi: 10.1155/2018/3273246
3. Cantone M, Lanza G, Fisicaro F, Pennisi M, Bella R, Di Lazzaro V, et al. Evaluation and treatment of vascular cognitive impairment by transcranial magnetic stimulation. *Neural Plast*. (2020) 2020:8820881. doi: 10.1155/2020/8820881
4. Hulme SR, Jones OD, Abraham WC. Emerging roles of metaplasticity in behaviour and disease. *Trends Neurosci*. (2013) 36:353–62. doi: 10.1016/j.tins.2013.03.007
5. Vinciguerra L, Lanza G, Puglisi V, Fisicaro F, Pennisi M, Bella R, et al. Update on the neurobiology of vascular cognitive impairment: from lab to clinic. *Int J Mol Sci*. (2020) 21:2977. doi: 10.3390/ijms21082977
6. Fritsch B, Reis J, Martinowich K, Schambra HM, Ji Y, Cohen LG, et al. Direct current stimulation promotes BDNF-dependent synaptic plasticity: potential implications for motor learning. *Neuron*. (2010) 66:198–204. doi: 10.1016/j.neuron.2010.03.035
7. Ranieri F, Podda MV, Riccardi E, Frisullo G, Dileone M, Profice P, et al. Modulation of LTP at rat hippocampal CA3-CA1 synapses by direct current stimulation. *J Neurophysiol*. (2012) 107:1868–80. doi: 10.1152/jn.00319.2011

AUTHOR CONTRIBUTIONS

MC, FR, and CT draft the manuscript. MC, GL, and CT conduct the analysis of data. GL and GO revise the manuscript critically for important intellectual content. All the authors approve the version of the manuscript to be published.

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2021 Cantone, Lanza, Ranieri, Opie and Terranova. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.



Neuroplasticity and Epilepsy Surgery in Brain Eloquent Areas: Case Report

Pedro Jesus Serrano-Castro^{1,2*}, Bienvenido Ros-López¹, Victoria Eugenia Fernández-Sánchez^{1,2}, Natalia García-Casares^{2,3}, Luis Muñoz-Becerra², Pablo Cabezudo-García^{1,2}, María José Aguilar-Castillo¹, María Vidal-Denis¹, Esperanza Cruz-Andreotti¹, María Jose Postigo-Pozo¹, Guillermo Estivill-Torrús² and Guillermo Ibañez-Botella¹

¹ Neuroscience Unit, Regional University Hospital of Malaga, Málaga, Spain, ² Biomedical Research Institute of Malaga, University of Málaga, Málaga, Spain, ³ Department of Medicine, University of Malaga, Málaga, Spain

OPEN ACCESS

Edited by:

Mariagiovanna Cantone,
Sant'Elia Hospital, Italy

Reviewed by:

Maurizio Elia,
Oasi Research Institute (IRCCS), Italy
Raffaele Falsaperla,
University Hospital Polyclinic Vittorio
Emanuele, Italy

*Correspondence:

Pedro Jesus Serrano-Castro
pedro.serrano.c@gmail.com

Specialty section:

This article was submitted to
Neurorehabilitation,
a section of the journal
Frontiers in Neurology

Received: 05 April 2020

Accepted: 09 June 2020

Published: 29 July 2020

Citation:

Serrano-Castro PJ, Ros-López B, Fernández-Sánchez VE, García-Casares N, Muñoz-Becerra L, Cabezudo-García P, Aguilar-Castillo MJ, Vidal-Denis M, Cruz-Andreotti E, Postigo-Pozo MJ, Estivill-Torrús G and Ibañez-Botella G (2020) Neuroplasticity and Epilepsy Surgery in Brain Eloquent Areas: Case Report. *Front. Neurol.* 11:698. doi: 10.3389/fneur.2020.00698

Introduction: Neuronal plasticity includes changes in any component of the central nervous system in response to intrinsic or extrinsic stimuli. Brain functions that depend on the epileptogenic cortex pose a challenge in epilepsy surgery because many patients are excluded from pre-surgical evaluation for fear of the possible sequelae. Some of these patients may be rescued by enhancing neuronal plasticity with brain neuromodulation techniques.

Case Report: We describe a 6-year-old child with refractory focal motor seizures symptomatic to a neuroepithelial dysembryoblastic tumor in the left temporo-parietal region. He underwent limited resection of the lesion in order to avoid sequelae in his language function. A functional study at age of 17 years revealed an overlap of Wernicke's area with the tumor and areas of incipient language reorganization in the contralateral hemisphere. An invasive neuromodulation procedure was designed to enhance neuroplasticity. After craniotomy, he underwent language training and simultaneous electrical inhibition of language using an electrode grid placed over the lesion. The intensity of the language inhibitory stimulus was increased every day to force the use of accessory language areas in the right hemisphere by neuroplasticity.

Results: The language of the patient improved for six consecutive days until he was able to speak and understand while undergoing maximum electrical inhibition. The tumor was resected using a cortical mapping guide.

Discussion: Application of direct cortical stimulation techniques and language pre-habilitation before epilepsy surgery can be useful to rescue patients excluded from resective surgery, especially young patients with long-term lesions.

Keywords: language prehabilitation, epilepsy surgery, eloquent area surgery, neuropsychological measures, Boston test, language functional MRI

INTRODUCTION

In 1894, Santiago Ramón y Cajal was the first to apply the term “plasticity” to the central nervous system at the International Medical Congress held in Rome (1), where he described dynamism or adaptation related to structural neuronal changes in response to external stimuli. Neuronal plasticity is now considered to refer to changes in any component of the central nervous system produced by intrinsic or extrinsic stimuli (2).

Knowledge of neuronal plasticity has expanded over recent decades, through the application of non-invasive electrical or magnetic stimulation procedures to complement conventional cognitive rehabilitation techniques after acquired brain damage (3–6). The main challenges are the evanescence of induced changes due to the distance between application and brain tissue and the interposition of the skull. These limitations may be overcome by using more invasive techniques, such as cortical stimulation mapping (CSM). CSM has long been used to identify eloquent areas in the presurgical study and to demarcate epileptogenic sites. CSM has also confirmed the plastic potential of brains in childhood and adolescence (7, 8). There has been abundant research on the application of CSM in animal models of neuronal plasticity modification (9–11). Functional magnetic resonance imaging (fMRI) studies of humans have also shown that long-term lesions in eloquent areas can permanently modify functional circuits by innate plasticity processes (6, 12, 13).

The prognosis of patients undergoing brain neurosurgery is influenced by the extent of resection, which is limited by the presence of brain functions dependent on the cerebral cortex. This causes many patients to be excluded from functional epilepsy surgery. Some of these patients might be rescued for the only curative treatment currently available if brain neuromodulation techniques could develop their neuronal plasticity. The number of patients who could benefit from such techniques is probably high given that the prevalence of active epilepsy in the world is 6.38/1,000 people (95% CI 5.57–7.30) (14), and of these, ~20–40% behave as refractory to medical treatments (15). Although there are no reliable data in the literature on the percentage of these patients with lesions in eloquent areas, it is known that this is a major clinical problem that has forced the development of various therapeutic strategies in these patients (16).

We report a case in which the neuronal plasticity of language was induced before epilepsy surgery.

CASE REPORT

We describe the case of a right-handed 6-years-old child with focal motor seizures of the right lower limb and sudden aphasia, without awareness impairment secondary to a space-occupying lesion in the left temporoparietal region. He underwent partial resection of the lesion, which was limited by the need to avoid sequelae in his language function. The pathological study reported WHO grade I neuroepithelial dysembryoplastic tumor (Ki-67 cell proliferation index < 1%). After the surgery, the patient continued with daily epileptic seizures refractory to medical treatment.

At the age of 17 years, a follow-up neuroimaging study showed an increase in the volume of the lesion, and an fMRI scan revealed an overlap of the area of Wernicke with the tumor and areas of incipient functional language reorganization in the homologous contralateral hemisphere.

Since the beginning of his illness, the patient has undergone multiple drug regimens, including oxcarbazepine, valproic acid, lamotrigine, eslicarbazepine acetate, lacosamide clobazam, and brivaracetam, in different rational combinations, without achieving the goal of freedom from seizures.

Methods

An invasive neuromodulation procedure was designed to enhance neuroplasticity.

Step 1: First awake intraoperative CSM: As the preoperative fMRI showed some transferred language areas to the right hemisphere, a first CSM was performed intraoperatively to confidently assess whether there was or not residual and functional language located over or nearby the tumor. Awake CSM followed left parietotemporal-wide craniectomy over the lesion. Phase-reversal of N20 was first tested. Once Rolandic sulcus was accurately showed up, the motor strip was stimulated while performing electrocorticography, with a monopolar handheld stimulating probe rectangular, monophasic, anodal multipulse ($N = 7$ ISI = 4 [250 Hz]) stimulus, with a duration of 0.2–0.5 ms and up to 25 mA of intensity using a 16-channel neurophysiological intraoperative monitoring device (Protektor by Xtelk®). With the motor threshold, we started the language direct cortical stimulation mapping using the Penfield technique with a handheld bipolar probe with 5 mm between the tips of the probe (biphasic starting positive, at 60 Hz, duration of 1 ms) with an intensity between 2 and 20 mA during 4 sg ($N = 240$ stimuli) using a cortical stimulator (Nimbus®), while the patient was performing motor language tasks: counting numbers days of the week; comprehension tasks: pictures descriptions, and repetition, reading, and witting tasks. As we found, there was some residual language just over the tumor that should be resected. To minimize language deficits, we decided to continue the procedure of prehabilitation and proceed with the placement of 20 subdural grid electrodes.

Figure 1 depicts the location of functional areas of language; electrode 17 is located on the sensitive area (Wernicke's) and electrode 4 on the motor area (Broca).

Step 2: Language Prehabilitation: 1 day after this surgery, in the patient's room, we performed a cortical stimulation through the 20 subdural grid electrodes, detecting the electrodes that were over Broca and Wernicke areas. Once the target electrodes were identified, we connected them (electrodes 4 and 17) to an external stimulator (Medtronic 3625, Medtronic Ibérica SA) to perform continuous electrical stimulation. The stimulation of these electrodes generated language dysfunction. The parameters used were 130 Hz, 1 ms, and intensity up to 10 V, which was increased daily in steps of 2 V to reach the limit of language function inhibition.

This stimulation was continuously active for 6 days, with increases or changes between the stimulating electrodes when

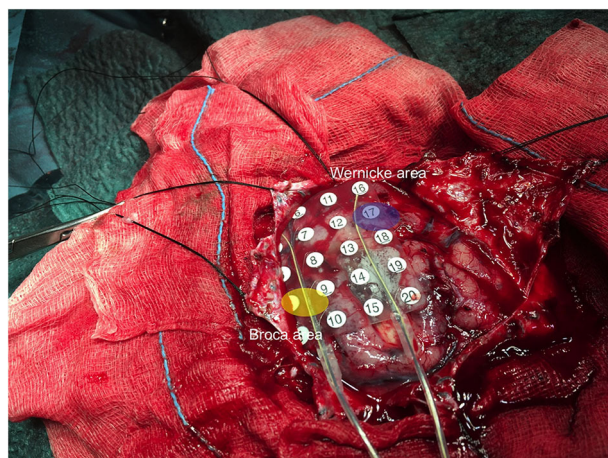


FIGURE 1 | Surgical image during CSM, indicating the sensitive and motor language eloquent areas identified.



FIGURE 2 | Detail of the position of the external stimulator during step 2.

necessary to reach the inhibition of the language again, as a habituation phenomenon was present (**Figure 2**).

During this procedure, every day, after adjustment of the intensity of the continuous electrical stimulation, during at least 6 h a day, the neuropsychologist performed an intensive work on the specific deficits of the patient using material from the Spanish version of Psycholinguistic Assessments of Language Processing in Aphasia (PALPA) for the training of spontaneous language and denomination (17) and material from the Barcelona test for the training of understanding, denomination, repetition, reading, and writing (18).

Step 3. Second Awake Cortical Mapping and Definitive Surgery: On the 7th day after the first awake craniotomy, the definitive surgery was done. The second CSM revealed that there was no residual language over the tumor that was completely resected with no further functional deficits.

At 1 month before surgery and again at 3 months post-surgery, the patient underwent a neuropsychological evaluation of language and an fMRI with language paradigms.

The Boston Naming Test is one of the most widely used visual confrontation naming tests to evaluate the lexical and semantic system in aphasic patients (19). We used this test for language evaluation, as it is a widely used test in the preoperative evaluation of epileptic patients and with which our group has extensive experience.

A 3.0 Teslas Philips Intera® MRI (release 2.6) system was used for blood-oxygen-level-dependent (BOLD) signal fMRI acquisition. The scanning session included one T1 structural image for precise anatomical localization of language areas and T2-weighted fast field echo, echo planar imaging (repetition time 3,000 ms, echo time 35 ms, field-of-view 230 mm, and matrix size 80/128 r). Auditory and block design fMRI paradigms (verbal fluency, semantic decision, verb generation, and passive story listening) were performed to determine the eloquent areas of language Broca and Wernicke.

Ethics

The patient and his parent signed informed consent in the hospital. The study was conducted following the principles of the Declaration of Helsinki (20), with Spanish regulations on biomedical research and with European personal data protection regulations. It was approved (code 0698-N-20) by the institutional ethical committee of our hospital (Comité de Ética de la Investigación provincial de Málaga).

RESULTS

Prehabilitation Procedure

The patient improved his linguistic ability for 6 consecutive days after the start of language prehabilitation. On the day before the second surgery, he was able to speak and understand without major deficits despite the application of maximum electrical inhibition to the Wernicke area of the left hemisphere. The tumor was then completely resected with cortical mapping in the awake patient.

Outcome

The patient has been seizure-free for more than 1 year after the surgery and has returned to his usual academic and social activities. He is currently receiving brivaracetam and eslicarbazepine acetate in descending doses.

Neuropsychological Evaluations

Neuropsychological language evaluations in our patient showed a progressive deterioration over the 2 years preceding surgical intervention in listening, fluency, denomination, and writing. More severe impairment was observed in some categories explored by the Boston test, including those related to category denomination and especially, written vocabulary and narrative writing, which deteriorated from normal results for his age at 2 years presurgery to very low scores at 1 month presurgery (**Table 1**).

These deficits were recovered after the language prehabilitation process and tumor resection. The most important recoveries were observed in areas of auditory understanding, denomination, basic vocabulary, and narrative writing.

TABLE 1 | Results of the regulated neuropsychological evaluation at 2 years before surgery, 1 month before surgery, and 3 months after surgery.

		Boston test				
		2 years before surgery (Percentile)	1 month before surgery (Percentile)	Language evolution previous to surgery	3 months after surgery (Percentile)	Language Evolution after surgery
Auditive understanding	Word discrimination	60	40	↓	50	↑
	Orders	100	70	↓	100	↑↑
	Complex Ideation material	60	40	↓	70	↑↑
Fluency	Phrase length	100	70	↓	100	↓
	Melodic line	100	70	↓	60	↓
	Grammatical form	100	70	↓	70	=
Recitation		100	100	=	100	=
Repetition	Words	100	100	=	100	=
	Sentences	100	100	=	100	=
Denomination	Naming response	100	70	↓	100	↑↑
	Boston vocabulary test	40	60	↓	70	↑
	Category Denomination	100	50	↓↓	100	↑↑
Reading	Match writing types	100	100	=	100	=
	Match numbers	100	100	=	100	=
	Match drawing-word	40	30	↓	40	↑
	Reading words aloud	100	100	=	100	=
	Reading sentences aloud	100	100	=	100	=
	Understanding sentences spoken aloud	100	100	=	100	=
	Understanding sentences and paragraphs spoken aloud	100	60	↓	100	↑↑
Writing	Shape	100	100	=	100	=
	Choice of letters	100	100	=	100	=
	Motor facility	100	40	↓↓	50	↑
	Basic vocabulary	100	30	↓↓↓	100	↓↓↓
	Regular phonetics	100	100	=	100	=
	Common irregular words	100	100	=	100	=
	Written designation of drawings	100	100	=	100	=
	Narrative writing	100	40	↓↓	80	↑↑

Percentile values. Red shading: <50 percentile. Orange shading: 50–90 percentile range. Green shading: >90 percentile. ↓↓↓, very important worsening; ↓↓, important worsening; ↓, worsening; =, no changes; ↑, moderate improvement; ↑↑, significant improvement; ↑↑↑, very important improvement.

From a practical viewpoint, this improvement opened up the possibility of the patient returning to his normal academic life after surgery.

improvements in language comprehension in this patient (see **Figures 3, 4**).

fMRI

When comparing fMRI before and after the tumor resection and the stimulation protocol, postsurgical images show decreased activity in the left hemisphere areas and greater activation in the right temporal areas, including the right homologous area of the left Wernicke's. This suggests neuroplasticity in these right eloquent auditory and language areas and could explain the

DISCUSSION

To our best knowledge, we present the first report of modulation of cerebral plasticity in a patient undergoing epilepsy surgery in language-eloquent areas. In 2016, Rivera et al. (21) described a series of five patients with WHO grade II or III glial lesions in language-eloquent areas who underwent a similar procedure, reporting that it induced an acceleration of neuroplasticity

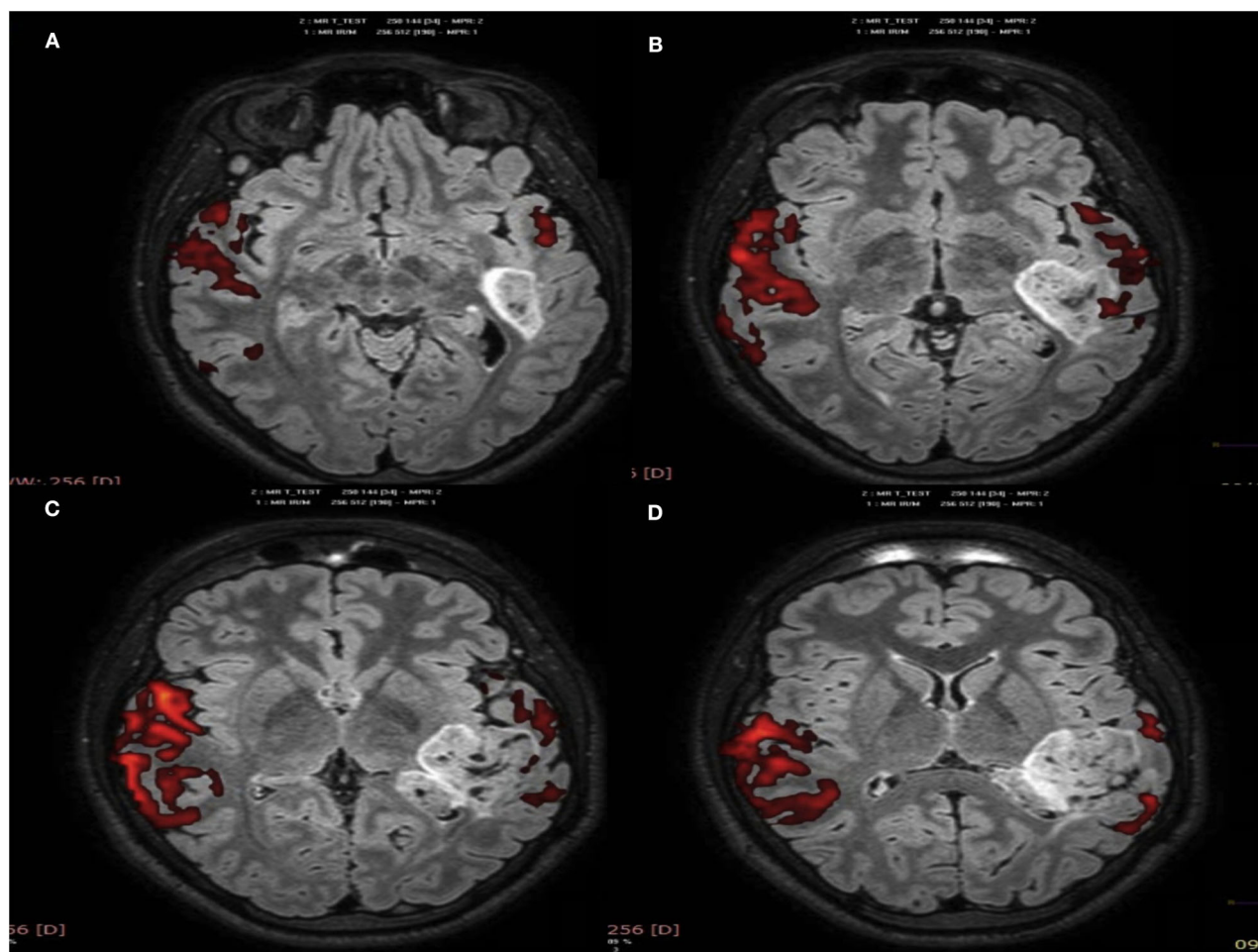


FIGURE 3 | Presurgical fMRI, story passive listening paradigm (A–D): axial brain planes show, in red color, the activation of left temporoparietal areas corresponding to Wernicke's, associative language, and the auditory areas within and surrounding the lesion. Greater activation is shown in the right temporoparietal hemisphere (homologous areas), probably due to neuroplasticity.

processes. They were older than the present patient, and their lesions were more recent, circumstances that do not favor neuroplasticity. Besides his younger age, our patient had a very long-term lesion, and an intrinsic neuroplasticity process was already underway (see **Figure 3**). Chronic lesions in the eloquent cortex are known to cause neuroplasticity that results in the cortical reorganization of functional areas (22–24). In this way, patients can develop language-eloquent areas in other parts of the brain, usually in contralateral homologous areas.

We consider that the improvement found between 1 month presurgery and 2 months post-surgery in our patient indicates the implementation of the incipient functional areas of language developed by neuroplasticity in the right hemisphere over the years, similar to previous observations in patients with long-standing lesions in eloquent areas (23, 24).

We propose that functional inhibition of the Wernicke area of the left hemisphere, together with the simultaneous

intensive language training, enabled this process. In this line, good outcomes have previously been described for the rehabilitation of stroke sequelae through the inhibition of functional areas and the simultaneous rehabilitation of damaged areas (25, 26).

These results suggest that the prehabilitation of language with this type of procedure can help in the implementation of areas developed by intrinsic neuroplasticity in patients with long-term lesions in language-eloquent areas. The prehabilitation process is probably not capable of transferring functions, as claimed by Rivera et al., but it can implement an area previously developed by intrinsic neuroplasticity.

Many molecular adjustments have been found and may constitute the substrate of neuroplasticity changes induced by electrical neuromodulation. According to recent molecular studies, direct current stimulation produces significant changes in neurotrophic factors, especially on brain-derived neurotrophic factor (BDNF). Thus, variations in BDNF secretion correlated

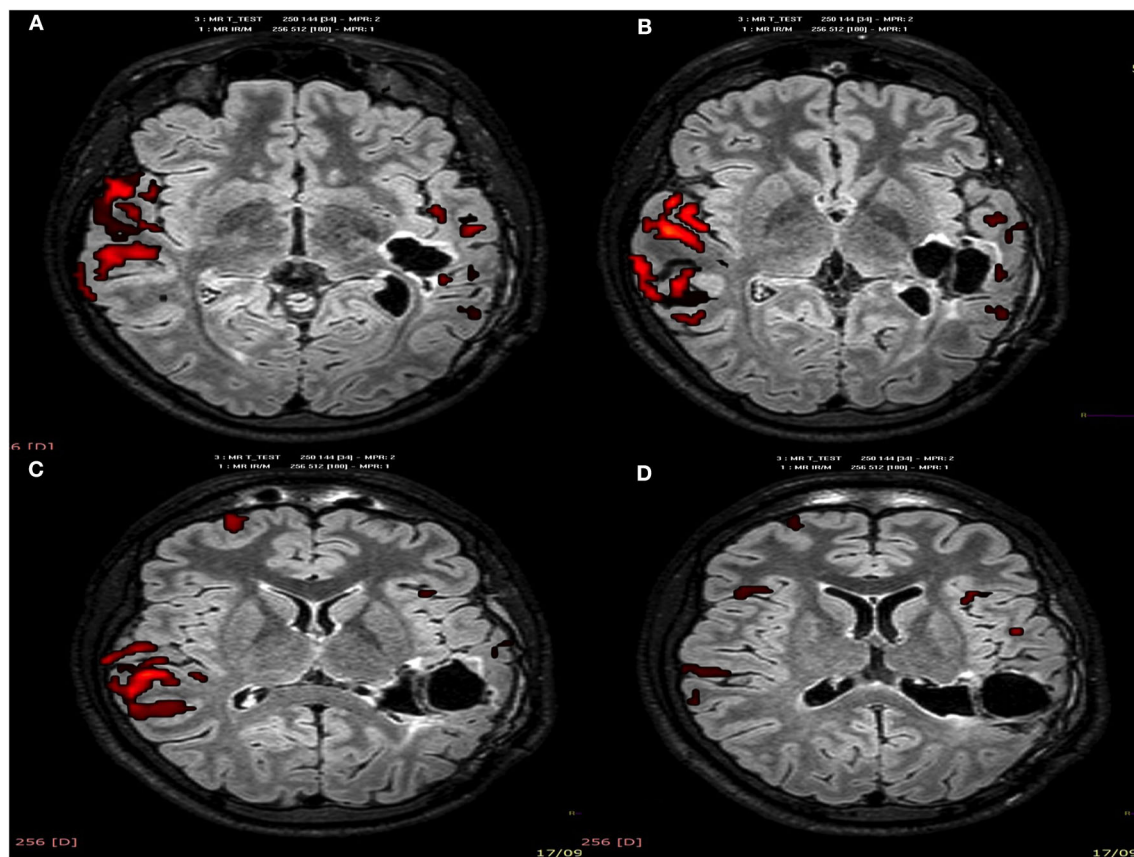


FIGURE 4 | Postsurgical fMRI, story passive listening paradigm (A–D): axial brain planes show, in red color, a decrease activation after surgery of the left temporoparietal areas corresponding to Wernicke's, associative language, and the auditory areas. Greater activation is shown in the right temporoparietal hemisphere (homologous areas), probably due to neuroplasticity.

to recovery after direct stimulation in preoperative treatment of pain control and Parkinson's disease (27, 28). Similarly, elevated nerve growth factor (NGF) serum levels in patients with depression have been suggested as adaptive neuroplasticity and associated with cognitive improvement after direct current stimulation (29). Recent works in experimental models have demonstrated that direct current stimulation in the CA1 region of rat hippocampus mediates elevated levels of BDNF in the hippocampus and priming of N-methyl-D-aspartate receptor-dependent long-term potentiation, eliciting metaplastic aftereffects on hippocampal synaptic plasticity. Induced enhancement of long-term potentiation was completely blocked with an antagonist of TrkB, demonstrating the role of BDNF/TrkB signaling in these effects (30). More recent, circulatory microRNAs (miRNAs) have also been involved in neuronal plasticity response in neuropathological conditions, and they may represent a fine-tuning mechanism able to integrate multiple inputs and outputs. In this sense, a very recent analysis from serum profiles and exosomal miRNAs showed genetic pathways involved in neuronal cell proliferation and differentiation significantly enriched with

miRNA targets and identified epilepsy-induced peripheral downregulation of miR-15a-5p, miR-34a, miR-106b-5p, and miR-146 (31). Furthermore, electric stimulation of the ventral hippocampal commissure delays the development of epilepsy in a rat model and produces a highly specific regulation of a set of miRNAs implicated in the shape of dendritic spines (32).

The most important study weakness is that it addresses an isolated case, limiting the conclusions that can be drawn. Nevertheless, it opens the way for investigation of an application that could have a major impact on patients with refractory epilepsy who experience a progressive deterioration but cannot currently access epilepsy surgery. On the other hand, there is no established protocol for prehabilitation, and it is possible that outcomes could be improved by applying different parameters.

In conclusion, direct cortical stimulation techniques and simultaneous language prehabilitation may be a useful approach in epilepsy surgery, especially in young patients with long-term lesions who have demonstrated the beginning of function remodeling through intrinsic neuroplasticity.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the Institutional Ethics Committees of our hospital (Comité de Ética de la Investigación provincial de Málaga, code 0698-N-20). Written informed consent was obtained from the minor(s)' legal guardian/next of kin for the publication of this case report, including any potentially identifiable images or data included in this article.

REFERENCES

- Serrano-Castro PJ, Garcia-Torrecillas JM. Cajal's first steps in scientific research. *Neuroscience*. (2012) 217:1–5. doi: 10.1016/j.neuroscience.2012.05.008
- Cramer SC, Sur M, Dobkin BH, O'Brien C, Sanger TD, Trojanowski JQ, et al. Harnessing neuroplasticity for clinical applications. *Brain*. (2011) 134:1591–609. doi: 10.1093/brain/awh517
- He W, Fong PY, Leung TWH, Huang YZ. Protocols of non-invasive brain stimulation for neuroplasticity induction. *Neurosci Lett*. (2020) 719:133437. doi: 10.1016/j.neulet.2018.02.045
- D'Agata F, Peila E, Cicerale A, Caglio MM, Caroppo P, Vighetti S, et al. Cognitive and neurophysiological effects of non-invasive brain stimulation in stroke patients after motor rehabilitation. *Front Behav Neurosci*. (2016) 10:135. doi: 10.3389/fnbeh.2016.00135
- Crinion J, Price CJ. Right anterior superior temporal activation predicts auditory sentence comprehension following aphasic stroke. *Brain*. (2005) 128:2858–71. doi: 10.1093/brain/awh659
- Sampaio-Baptista C, Sanders ZB, Johansen-Berg H. Structural plasticity in adulthood with motor learning and stroke rehabilitation. *Annu Rev Neurosci*. (2018) 41:25–40. doi: 10.1146/annurev-neuro-080317-062015
- Chou N, Serafini S, Muh CR. Cortical language areas and plasticity in pediatric patients with epilepsy: a review. *Pediatr Neurol*. (2018) 78:3–12. doi: 10.1016/j.pediatrneurol.2017.10.001
- Serafini S, Komisarow JM, Gallentine W, Mikati MA, Bonner MJ, Kranz PG, et al. Reorganization and stability for motor and language areas using cortical stimulation: case example and review of the literature. *Brain Sci*. (2013) 3:15971614. doi: 10.3390/brainsci3041597
- Zheng J, Liu L, Xue X, Li H, Wang S, Cao Y, et al. Cortical electrical stimulation promotes neuronal plasticity in the peri-ischemic cortex and contralateral anterior horn of cervical spinal cord in a rat model of focal cerebral ischemia. *Brain Res*. (2013) 1504:25–34. doi: 10.1016/j.brainres.2013.01.015
- Cecatto RB, Maximino JR, Chadi G. Motor recovery and cortical plasticity after functional electrical stimulation in a rat model of focal stroke. *Am J Phys Med Rehabil*. (2014) 93:791–800. doi: 10.1097/PHM.0000000000000104
- Adkins DL. Cortical stimulation-induced structural plasticity and functional recovery after brain damage. In: Kobeissy FH, editor. *Brain Neurotrauma: Molecular, Neuropsychological, and Rehabilitation Aspects. Chapter 43*. Frontiers in Neuroengineering. Boca Raton, FL: CRC Press/Taylor & Francis (2015). p. 1–38.
- Rutten GJ, Ramsey NF, van Rijen PC, Alpherts WC, van Veelen CW. FMRI-determined language lateralization in patients with unilateral or mixed language dominance according to the Wada test. *Neuroimage*. (2002) 17:447–60. doi: 10.1006/nimg.2002.1196
- Deng X, Xu L, Zhang Y, Wang B, Wang S, Zhao Y, et al. Difference of language cortex reorganization between cerebral arteriovenous malformations, cavernous malformations, and gliomas: a functional MRI study. *Neurosurg Rev*. (2016) 39:241–9. doi: 10.1007/s10143-015-0682-7
- Fiest KM, Sauro KM, Wiebe S, Patten SB, Kwon C-S, Dykeman J, et al. Prevalence and incidence of epilepsy. A systematic review

AUTHOR CONTRIBUTIONS

PS-C, BR-L, VF-S, LM-B, GE-T, and GI-B contributed in the conception and design of the procedure. PS-C, BR-L, VF-S, LM-B, GI-B, PC-G, MA-C, MV-D, EC-A, and MP-P participated in the implementation of the procedure. PS-C wrote the first draft of the manuscript. All authors contributed to manuscript revision, read, and approved the submitted version.

- and metaanalysis of international studies. *Neurology*. (2017) 88:296–303. doi: 10.1212/WNL.0000000000003509
- Brodie MJ. Diagnosing and predicting refractory epilepsy. *Acta Neurol Scand Suppl*. (2005) 181:36–9. doi: 10.1111/j.1600-0404.2005.00507.x
- Maesawa S, Nakatsubo D, Fujii M, Iijima K, Kato S, Ishizaki T, et al. Application of awake surgery for epilepsy in clinical practice. *Neurol Med Chir*. (2018) 58:442–52. doi: 10.2176/nmc.2018-0122
- Kay J, Lesser R, Coltheart M. *PALPA: Psycholinguistic Assessments of Language Processing in Aphasia*. Hove: Lawrence Erlbaum Associates (1992).
- Peña-Casanova J. *Normalidad, semiología y patología neuropsicológicas. Programa integrado de exploración neuropsicológica. "Test Barcelona."* Barcelona: Masson (1991).
- Kaplan E, Goodglass H, Weintraub S. *The Boston naming test: Experimental edition (1978)*. 2nd ed. Boston: Kapan & Goodglass; Philadelphia: Lea & Febiger (1978).
- World Medical Association. World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects. *JAMA*. (2013) 310:2191–4. doi: 10.1001/jama.2013.281053
- Rivera-Rivera P, Rios-lago M, Sanchez-Casarrubios S, Salazar O, Yus M, Sanz A, et al. Cortical plasticity catalyzed by prehabilitation enables extensive resection of brain tumors in eloquent areas. *J Neurosurg*. (2017) 126:1323–33. doi: 10.3171/2016.2.JNS152485
- Bonnetblanc F, Desmurget M, Duffau H. Low grade gliomas and cerebral plasticity: fundamental and clinical implications. *Med Sci*. (2006) 22:389–94. doi: 10.1051/medsci/2006224389
- Kornfeld S, Delgado Rodríguez JA, Everts R, Kaelin-Lang A, Wiest R, Weisstanner C, et al. Cortical reorganisation of cerebral networks after childhood stroke: impact on outcome. *BMC Neurol*. (2015) 15:90. doi: 10.1186/s12883-015-0309-1
- Mikellidou K, Arrighi R, Aghakhanyan G, Tinelli F, Frijia F, Crespi S, et al. Plasticity of the human visual brain after an early cortical lesion. *Neuropsychologia*. (2019) 128:166–77. doi: 10.1016/j.neuropsychologia.2017.10.033
- Aşkin A, Tosun A, Demirdal ÜS. Effects of low-frequency repetitive transcranial magnetic stimulation on upper extremity motor recovery and functional outcomes in chronic stroke patients: a randomized controlled trial. *Somatosens Mot Res*. (2017) 34:102–7. doi: 10.1080/08990220.2017.1316254
- Paquette C, Thiel A. Rehabilitation interventions for chronic motor deficits with repetitive transcranial magnetic stimulation. *J Neurosurg Sci*. (2012) 56:299–306.
- Ribeiro H, Sesterhenn RB, Souza A, Souza AC, Alves M, Machado JC, et al. Preoperative transcranial direct current stimulation: exploration of a novel strategy to enhance neuroplasticity before surgery to control postoperative pain. A randomized sham-controlled study. *PLoS ONE*. (2017) 12:e0187013. doi: 10.1371/journal.pone.0187013.eCollection.2017
- Hadoush H, Banihani SA, Khalil H, Al-Qaisi Y, Al-Sharman A, Al-Jarrah M. Dopamine, BDNF and motor function postbilateral anodal transcranial direct current stimulation in Parkinson's disease. *Neurodegener Dis Manag*. (2018) 8:171–9. doi: 10.2217/nmt-2017-0048
- Brunoni AR, Padberg F, Vieira ELM, Teixeira AL, Carvalho AF, Lotufo PA, et al. Plasma biomarkers in a placebo-controlled trial comparing tDCS and escitalopram efficacy in major depression. *Prog Neuropsychopharmacol*

- Biol Psychiatry.* (2018) 86:211–7. doi: 10.1016/j.pnpbp.2018.06.003
30. Yu TH, Wu YJ, Chien ME, Hsu KS. Transcranial direct current stimulation induces hippocampal metaplasticity mediated by brain-derived neurotrophic factor. *Neuropharmacology.* (2019) 144:358–67. doi: 10.1016/j.neuropharm.2018.11.012
 31. Cava C, Manna I, Gambardella A, Bertoli G, Castiglioni I. Potential role of miRNAs as theranostic biomarkers of epilepsy. *Mol Ther Nucleic Acids.* (2018) 13:275–90. doi: 10.1016/j.omtn.2018.09.008
 32. Costard LS, Neubert V, Venø MT, Su J, Kjems J, Connolly NMC, et al. Electrical stimulation of the ventral hippocampal commissure delays experimental epilepsy and is associated with altered microRNA expression. *Brain Stimul.* (2019) 12:1390–401. doi: 10.1016/j.brs.2019.06.009

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2020 Serrano-Castro, Ros-López, Fernández-Sánchez, García-Casares, Muñoz-Becerra, Cabezudo-García, Aguilar-Castillo, Vidal-Denis, Cruz-Andreotti, Postigo-Pozo, Estivill-Torrús and Ibañez-Botella. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.



Novel TMS for Stroke and Depression (NoTSAD): Accelerated Repetitive Transcranial Magnetic Stimulation as a Safe and Effective Treatment for Post-stroke Depression

Jessica Frey¹, Umer Najib¹, Christa Lilly² and Amelia Adcock^{1*}

¹ Department of Neurology, West Virginia University, Morgantown, WV, United States, ² Department of Biostatistics, West Virginia University, Morgantown, WV, United States

OPEN ACCESS

Edited by:

Carmen Terranova,
University of Messina, Italy

Reviewed by:

Antonino Naro,
Centro Neurolesi Bonino Pulejo
(IRCCS), Italy
Iris Charlotte Brunner,
Aarhus University, Denmark

*Correspondence:

Amelia Adcock
akadcock@hsc.wvu.edu

Specialty section:

This article was submitted to
Neurorehabilitation,
a section of the journal
Frontiers in Neurology

Received: 03 April 2020

Accepted: 25 June 2020

Published: 11 August 2020

Citation:

Frey J, Najib U, Lilly C and Adcock A
(2020) Novel TMS for Stroke and
Depression (NoTSAD): Accelerated
Repetitive Transcranial Magnetic
Stimulation as a Safe and Effective
Treatment for Post-stroke Depression.
Front. Neurol. 11:788.
doi: 10.3389/fneur.2020.00788

Background: Post-stroke depression (PSD) affects up to 50% of stroke survivors, reducing quality of life, and increasing adverse outcomes. Conventional therapies to treat PSD may not be effective for some patients. Repetitive transcranial magnetic stimulation (rTMS) is well-established as an effective treatment for Major Depressive Disorder (MDD) and some small trials have shown that rTMS may be effective for chronic PSD; however, no trials have evaluated an accelerated rTMS protocol in a subacute stroke population. We hypothesized that an accelerated rTMS protocol will be a safe and viable option to treat PSD symptoms.

Methods: Patients ($N = 6$) with radiographic evidence of ischemic stroke within the last 2 weeks to 6 months with Hamilton Depression Rating Scale (HAMD-17) scores > 7 were recruited for an open label study using an accelerated rTMS protocol as follows: High-frequency (20-Hz) rTMS at 110% resting motor threshold (RMT) was applied to the left dorsolateral prefrontal cortex (DLPFC) during five sessions per day over four consecutive days for a total of 20 sessions. Safety assessment and adverse events were documented based on the patients' responses following each day of stimulation. Before and after the 4-days neurostimulation protocol, outcome measures were obtained for the HAMD, modified Rankin Scale (mRS), functional independence measures (FIM), and National Institutes of Health Stroke Scales (NIHSS). These same measures were obtained at 3-months follow up.

Results: HAMD significantly decreased (Wilcoxon $p = 0.03$) from $M = 15.5$ (2.81)–4.17 (0.98) following rTMS, a difference which persisted at the 3-months follow-up ($p = 0.03$). No statistically significant difference in FIM, mRS, or NIHSS were observed. No significant adverse events related to the treatment were observed and patients tolerated the stimulation protocol well overall.

Conclusions: This pilot study indicates that an accelerated rTMS protocol is a safe and viable option, and may be an effective alternative or adjunctive therapy for patients suffering from PSD. Future randomized, controlled studies are needed to confirm these preliminary findings.

Clinical Trial Registration: <https://clinicaltrials.gov/ct2/show/NCT04093843>.

Keywords: post-stroke depression, transcranial magnetic stimulation, stroke recovery, neurostimulation, ischemic stroke, neurorehabilitation, accelerated TMS

INTRODUCTION

The interplay between depression and cerebrovascular disease is complex and clinically important. Post-stroke depression (PSD) is the most common neuropsychological complication of stroke, with a prevalence of ~33% (1) in stroke survivors. PSD adversely influences outcomes by reducing quality of life, increasing caregiver burden, and increasing early mortality as much as ten-fold (2–4). As acute stroke interventions continue to improve, stroke survivorship and associated morbidity will also increase, making the need to explore innovative treatments for PSD even more urgent.

Despite the significant clinical burden of PSD, there are limited treatment options to prevent or reduce its severity. Psychotherapy and pharmacotherapy are well-established as treatments of choice in major depression, however a subset of patients do not respond to either of these first-line therapies (5). Selective Serotonin Reuptake Inhibitor (SSRI) use has been associated with increased risk of hemorrhagic complications as well as increased risk of falls in the elderly, while other studies have shown that SSRIs are actually associated with increased risk for stroke, myocardial infarction, and all-cause mortality (6). A recent meta-analysis for stroke patients concluded that antidepressants did not significantly improve patients' general recovery, achieved varied response rates, and were not tolerated due to adverse effects (7). Compliance, communication problems, and lack of access to psychiatric care are further challenges to treating PSD.

Repetitive transcranial magnetic stimulation (rTMS) may represent an effective treatment option that mitigates the issues associated with the standard PSD interventions. The FDA approved rTMS for patients with Major Depressive Disorder (MDD) in 2008 (8). The typical rTMS protocol that has been used effectively for major depression is 5 days per week for 4–6 weeks. Conventional rTMS paradigms have been studied in the PSD population, and many studies including a meta-analysis have shown that conventional rTMS is likely effective for chronic, refractory PSD (9, 10). However, these conventional paradigms may be inconvenient for patients with limited transportation access and may limit compliancy of patients. Therefore, an accelerated protocol which minimizes the number of days needed to complete the full treatment may be more accessible to patients and may increase compliancy. While there have been some accelerated rTMS paradigms that have been designed to treat conditions such as alcohol withdrawal and treatment-resistant depression (11–14), similar accelerated protocols have not been

studied in patients suffering from PSD. Applying accelerated rTMS to the PSD population comes with unique and complex factors. For example, the theoretical risk of seizure using an accelerated protocol may be higher, and this risk may increase even further in patients in the acute to subacute stroke period. Therefore, it is important to study the safety of an accelerated protocol in this population. In addition, the period immediately following cerebrovascular ischemia potentially represents a biologically unique phase amenable to intervention given that both neuroplasticity as well as recurrent stroke risk are highest during this time (15, 16).

There is a clear medical need to further address the impact of rTMS for PSD and to optimize stimulation parameters. We hypothesized that an accelerated 4-days rTMS protocol would be a safe and viable method for treating PSD and would help ameliorate depressive symptoms.

METHODS

This prospective open label study was approved by our Institutional Review Board (IRB # 1804090922) and the Food and Drug Administration granted this study an Investigational Device Exemption (IDE) Number: G180102. The raw data supporting the conclusions of this article will be made available upon request, without undue reservation.

Participants

All patients admitted to the inpatient stroke service at our tertiary comprehensive stroke center are routinely screened for depression. Patients were screened for depression with the Hamilton Depression Rating Scale (HAMD-17). Study patients were identified either during their acute hospitalization or their follow up clinic visit. Patients who met the inclusion criteria and were otherwise free from the exclusion criteria were eligible to enroll (Table 1). Patients were eligible if the stimulation protocol could be applied between 2 weeks to 6 months following their acute stroke. Between November 2018 and March 2019, 62 of the 98 screened patients fulfilled the inclusion criteria. Although 62 patients were eligible, several patients had logistical issues unique to their own family or social situation and were unable to participate. Six patients were successfully enrolled and completed the stimulation protocol.

Stimulation

Neurostimulation was performed using the Neurostar system 2.0 figure of eight coil (Neuronetics, Malvern, PA). Prior to

TABLE 1 | List of inclusion and exclusion criteria.**Inclusion criteria**

1. Aged 22–85 years old
2. Radiographic evidence of ischemic stroke
3. Stroke within 2 weeks to 6 months
4. HAMD score ≥ 8

Exclusion criteria

1. Metallic objects or neurostimulators implanted intracranially
2. Stroke in the area of stimulation (L DLPFC)
3. Known history of epilepsy or seizure disorder
4. A woman who is pregnant or breastfeeding
5. History of psychiatric hospitalization unrelated to current PSD
6. Current suicidal ideation or MINI suicide scale > 8
7. ASRM score > 6
8. Current illicit drug use
9. History of head trauma resulting in loss of memory > 5 min or requiring hospitalization
10. Evidence of hemorrhage in the brain at the time of study
11. Clinically significant EKG abnormalities including QTC prolongation > 450 msec in men or > 480 msec in women
12. Any other mental or physical conditions that are inappropriate for study participation at the PI's discretion

stimulation sessions, patients that were successfully enrolled had additional survey tools administered for baseline assessments in the following categories: modified Rankin Scale (mRS) to assess level of independence, Functional Independence Measures (FIM) to assess quality of independent lifestyle, and HAMD to assess level of depression. Patients were also assessed with the National Institutes of Health (NIH) Stroke Scale to determine physical disabilities resulting from their stroke. All functional scales were performed by trained study personnel and the same rater for each patient was used to minimize variability and inter-rater bias. Vital signs including an electrocardiogram (EKG) were performed before and after each stimulation session. Patients were surveyed about adverse events following each stimulation day.

On the first day, patients underwent a mapping procedure to determine the patient's individualized and optimal Resting Motor Threshold (RMT) over the left motor cortex. The RMT was defined as the minimum stimulation intensity required for visual muscle twitch of the right abductor pollicis brevis (APB) muscle in five out of 10 consecutive single pulse stimulations. After establishing RMT, the coil was moved 5.5 cm anteriorly to the patient's left dorsolateral prefrontal cortex (DLPFC). Patients underwent repeat mapping if necessary. The NeuroStar system has a method for saving each patient's measurements in the system to ensure that the coil is positioned in the same place for each new session. Earplugs were used to prevent any hearing injury. All mapping and treatment sessions were performed by TMS-certified nurses and physicians at our Behavioral Medicine facility where emergency equipment was readily available.

Patients sat in the NeuroStar system chair for all treatment sessions, which has mechanisms to keep the patient properly positioned for mapping and stimulation sessions. The treatment protocol was adapted from other accelerated rTMS protocols in the literature for other indications (11, 12). The protocol included high frequency (20 Hz) rTMS applied over the left DLPFC at

110% RMT for five sessions per day, over four consecutive days for a total of 20 sessions. Forty trains of two second duration were applied with a 12 second intertrain interval for a total of 1,560 pulses per session. Patients were given the opportunity to rest for 10–15 min in between sessions. The treatment sessions lasted for about an hour and a half each day. Variations on the accelerated paradigm we used in this study using different frequencies and different trains may be possible to test in future studies.

At the end of the 4 days of stimulation, patients were once again surveyed with the HAMD, mRS, and FIM. Post-treatment NIH was also performed. The patients were also surveyed at the end of each stimulation day as well as at the end of all 4 days regarding any adverse events they may have experienced. These same measures were once again repeated at the patient's 3-months follow-up.

The primary outcome of this study was safety and viability as defined as the successful recruitment and treatment of participants using the outlined accelerated protocol with no significant adverse effects observed. The secondary outcome was any effect on depressive symptoms as measured by the HAMD. We defined a meaningful response as remission of depression to non-depressed range (HAMD < 8) or at least a 50% reduction in overall score.

Statistical Analysis

All analyses were conducted in SAS 9.4. Categorical variables are described with frequencies and valid percentages, continuous variables with means and standard deviations. Alpha was set to 0.05 unless otherwise noted. Differences were explored using Wilcoxon signed rank tests on the differences between pre- and post- for continuous variables. Symmetry tests and McNemar's exact tests were run on the ordinal and binary outcome data. Finally, associations were examined between continuous data using Pearson correlations, and with categorical data using Wilcoxon two-sample tests with two-sided t-approximation.

RESULTS

Demographically, five of the study participants were male and the average age was 66.33 (range 57–71). Stroke etiology included two large artery atherosclerosis (LAA), one small vessel disease (SVD), two cardioembolic (CE), and one embolic source of unknown significance (ESUS). Half of the patients were taking SSRIs at the time of the study (**Table 2**).

No significant adverse events related to the treatment were observed. All participants tolerated the stimulation well. One subject described a headache that was milder than his usual chronic headaches and another subject experienced transient facial sensitivity ipsilateral to the coil at the beginning of the first day of stimulation. Neither of these observations were rated as bothersome by the participants and both were self-limited.

HAMD significantly decreased (Wilcoxon $p = 0.03$) from $M = 15.5$ (2.81) to 4.17 (0.98) following rTMS, a difference which persisted at the 3-months follow-up ($p = 0.03$). There was no statistically significant difference in FIM, mRS, or NIH (**Table 3**).

In terms of number of patients going from “depressed” (HAMD ≥ 8) to “non-depressed” (HAMD < 8), four participants

(66.67%) had moderate depression (HAMD 14–18) and 2 (33.33%) had severe depression (19–22) at baseline. At post-assessment, all scores dropped below the cut-off for non-depressed. At 3-months follow-up, 5 of 6 patients remained non-depressed, and one patient scored eight at the lowest end of mild depression (Figure 1).

DISCUSSION

Our results demonstrate that the use of an accelerated rTMS protocol in patients with PSD during the subacute period

following stroke is a safe and viable option for stroke patients. None of the participating patients reported any significant adverse effects. This high degree of tolerability is similar to the previous published experience with accelerated protocols (11–18). All treated patients experienced a significant improvement in depressive symptoms, with a remission rate of 100% directly following TMS. Remission status persisted in five of the six patients at 3-months follow-up, with one patient scoring borderline mild depressed but still maintaining a 47% reduction in her depression score from baseline.

There have been a few other small studies that have looked at rTMS for chronic PSD (9, 19–21) as well as a recent meta-analysis of 22 randomized controlled trials comparing active rTMS stimulation to sham stimulation (10). These trials indicated that rTMS is an effective tool to treat chronic PSD. Other forms of non-invasive brain stimulation such as electroconvulsive therapy (ECT) and transcranial direct current stimulation have limited data for the treatment of PSD. ECT is largely regarded as the most powerful tool to treat severe depression, however, it is limited by side effects of amnesia (22, 23). Within the PSD population, these findings with ECT are echoed with respective response and remission rates of 60 and 50% (24). In spite of this, rTMS is still the best at controlling frequency and location of stimulation, which offers certain advantages (23). Our data demonstrates that an accelerated version of rTMS may be an effective treatment for PSD as well.

The mechanism underlying rTMS efficacy is still largely unknown. It is hypothesized that low frequency TMS stimulates inhibitory neurons while high frequency TMS stimulates excitatory projection neurons, thus mimicking neuroplasticity through long-term potentiation (23). Thus we chose high-frequency stimulation of the left DLPFC given that this area is associated with depression. However, the translation of cortical excitation to clinical response with rTMS is incompletely characterized (25). Therapeutic benefit is likely achieved through multiple mechanisms enhancing neuroplasticity, increasing available concentrations of critical neurotransmitters, and reinforcing emotionally positive connectivity networks

TABLE 2 | Baseline characteristics of the six participants.

Variable	Mean or N	SD or %
Age		
Years	66.33	4.97
Gender		
Male	5	83.33%
HLD*		
Yes	5	83.33%
DM†		
Yes	2	33.33%
AF‡		
Yes	1	16.67%
Tobacco		
Yes	3	50.00%
SSRI§		
Yes	3	50.00%
Family history		
Yes	1	16.67%

*HLD, hiperlipidemia.

†DM, diabetes mellitus.

‡AF, atrial fibrillation.

§SSRI, serotonin selective reuptake inhibitor.

TABLE 3 | Participant outcome measures (N = 6).

Variable	<i>N</i>	Pre mean (SD)	Post mean (SD)	3 month Mean (SD)	Diff (pre to post)	<i>p</i> -value*	Diff (pre to 3 month)	<i>p</i> -value*
HAMD	6	15.50 (2.81)	4.17 (0.98)	3.50 (2.66)	11.33 (2.94)	0.03	12.00 (3.63)	0.03
FIM	6	115.33 (8.12)	122.17 (6.97)	–	–6.83 (4.17)	0.063	–	–
NIHSS	6	1.83 (2.99)	1.00 (1.67)	–	0.83 (2.04)	1.00	–	–

Variable	Category	Pre <i>N</i>	(%)	Post <i>N</i>	(%)	<i>p</i> -value**
mRS						0.80
	0	1	(16.67%)	2	(33.33%)	
	1	4	(66.67%)	3	(50.00%)	
	2	1	(16.67%)	1	(16.67%)	
NIHSS < 4	Yes	4	(66.67%)	5	(83.33%)	0.32

*Wilcoxon signed rank test.

**Symmetry test, McNemar's exact test.

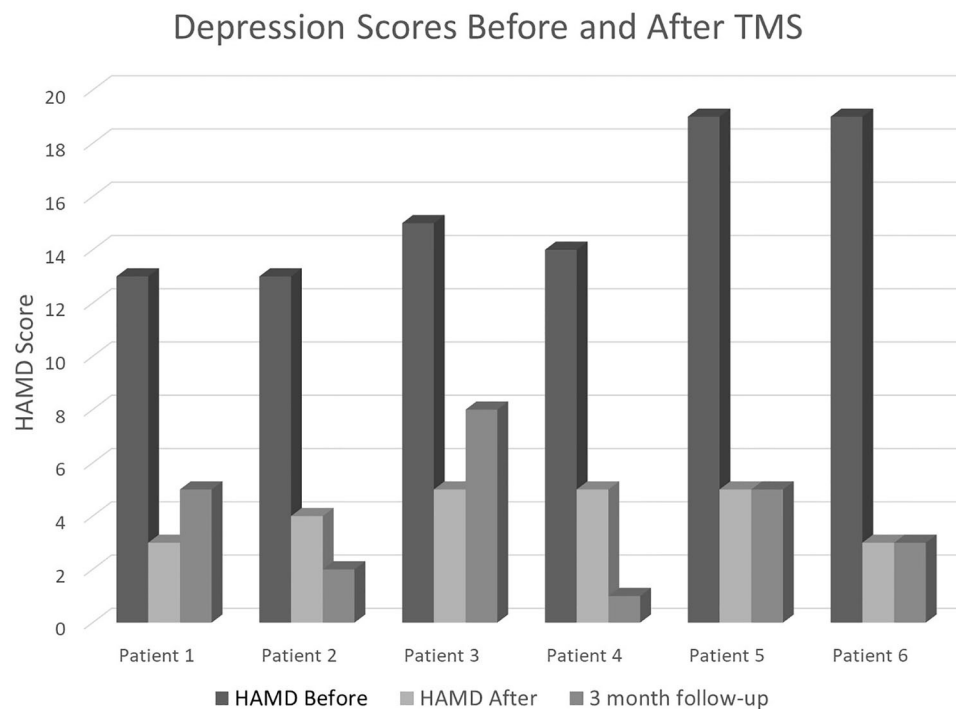


FIGURE 1 | HAMD scores in our six patients before the TMS sessions (baseline), immediately after completing the full 4-days neurostimulation protocol, and at their 3-months follow-up appointment.

while diminishing connectivity in emotionally negative loops (26–28). Low levels of peripheral and central brain derived neurotrophic factor (BDNF) have been observed in depressed individuals as well as those who develop PSD (29–33). Glutamate is emerging as another biomarker for treatment response with increased radiolabeled activity in the DLPFC following stimulation (34). rTMS treatment has also increased dopamine concentrations (35–37), and increased activity within mood networks on functional imaging (38). Exactly how rTMS exerts its influence, however, remains a critical question. Understanding its underlying mechanism will potentially increase our understanding of PSD itself and help identify therapeutic targets.

The novelty of this rTMS paradigm is the accelerated protocol as well as the stimulation in the acute to subacute stroke period. Similar accelerated protocols have been used in other populations (12, 13, 17, 18) (treatment resistant depression and alcohol withdrawal craving) and there have also been studies conducted of rTMS in the acute stroke setting for complications unrelated to depression (39–42); however, a similar paradigm has not yet been employed in a PSD population. A major barrier of current rTMS protocols is the 4–6 weeks timeline before clinical benefit is achieved, so an accelerated protocol is an important potential solution to this problem. The accelerated protocol that was used in this study enabled patients to receive 20 total stimulation sessions, which is the typical minimum number of sessions that patients receive in a conventional rTMS protocol (20

sessions spread out over 4 weeks, receiving one session per day Monday through Friday). Condensing these 20 sessions into four consecutive days allowed patients to participate who otherwise may have faced logistical challenges to obtaining this treatment.

Although this study was underpowered to demonstrate efficacy, the significant remission rate is promising. Larger, randomized studies are needed to confirm these results. There are several limitations in this study. The open label design of this study allows for patients to know they are receiving active stimulation, and the placebo effect could very well have influenced the robust improvement in depression following rTMS. It is important to conduct future trials with a control group and appropriate blinding to truly determine if the rTMS itself is causing a meaningful response in depressive symptoms. Another major limitation is the small sample size. The patients enrolled in the study all had high functional levels according to their FIM, NIHSS, and mRS scores, which may indicate a self-selection bias. It is unclear if patients with a higher functional status were more interested in the study, if these patients were more likely to be aware of their depressive symptoms and want to participate for this reason, or if these patients were more capable of driving themselves to the appointments and thus more willing to participate. In addition, the fact that such a small proportion of eligible patients ultimately enrolled in this study underscores the complexities of treating this patient population and the explicit barriers to enrollment deserve dedicated further study. Regardless, a larger sample size

with a group representative of the whole spectrum of post-stroke functionality would allow the results to be applicable to a broader population. In addition, half of our patients were already taking an anti-depressant at the time of enrollment. We chose not to exclude patients on SSRIs since the main goal of this study was to first establish safety and tolerability of using accelerated rTMS in this population, however we did ensure that all patients continued concurrent pharmacologic treatment throughout the duration of the study. Future studies would benefit from excluding patients on SSRIs, and larger studies would also benefit from comparing patients receiving rTMS alone vs. rTMS plus SSRIs to determine if there is a synergistic effect in this population. Similar to major depression, some studies have shown synergism between rTMS and pharmacologic therapy as opposed to either alone (43). However, a meta-analysis of all rTMS in PSD trials published found an rTMS effect size greater among those not on any pharmacologic treatment (0.96) compared to combination therapy (0.51) (22). Future studies may also benefit from the use of neuronavigation to confirm coil position as well as EEG compatible TMS to assess for subclinical seizure activity in a population with a theoretically increased risk of seizure (44). Given the subjective nature of depressive symptom reporting and known placebo effect among depressed patient populations, it is imperative to confirm our findings in larger, randomized studies with a sham stimulation arm as a control group.

CONCLUSION

Our results indicate that accelerated rTMS is a safe and viable treatment option for PSD in the subacute stroke population. Depressive symptoms significantly improved in all treated patients. Confirming these results in larger randomized settings has the potential to establish accelerated rTMS as a potent therapy for PSD. Further studies regarding mechanism of action, subgroups particularly responsive to the treatment, and durability of rTMS for PSD are warranted. We are currently

conducting a larger randomized controlled study in efforts to answer these questions.

DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by West Virginia University Institutional Review Board (IRB). The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

JF was responsible for the conceptual design of this study, writing of the first draft, all major revisions, patient recruitment, data collection, and data analysis. AA was responsible for writing the first draft, major revisions, collaboration of the experimental design, patient recruitment, data collection, and data analysis. CL was responsible for the statistical analysis and revisions to the manuscript. UN was responsible for revisions to the manuscript and contribution to conceptual design. All authors approved the final manuscript.

FUNDING

This project described was supported by the National Institute of General Medical Sciences, U54GM104942.

ACKNOWLEDGMENTS

We would like to acknowledge Jay Sherman, Louise Moore, Padma Tirumalai, Ashley Petrone, Shelley Welch, Christian Casingal, Lea Colantonio, and Callista Clairmont.

REFERENCES

- Villa RF, Ferrari F, Moretti A. Post-stroke depression: mechanisms and pharmacological treatment. *Pharmacol Ther.* (2018) 184:131–44. doi: 10.1016/j.pharmthera.2017.11.005
- Bartoli F, Lillia N, Lax A, Crocamo C, Mantero V, Carrà G, et al. Depression after stroke and risk of mortality: a systematic review and meta-analysis. *Stroke Res Treat.* (2013) 2013:862978. doi: 10.1155/2013/862978
- Paolucci S. Epidemiology and treatment of post-stroke depression. *Neuropsychiatr Dis Treat.* (2008) 4:145–54. doi: 10.2147/NDT.S2017
- Espárrago Llorca G, Castilla-Guerra L, Fernández Moreno MC, Ruiz Doblado S, Jiménez Hernández MD. Post-stroke depression: an update. *Neurologia.* (2015) 30:23–31. doi: 10.1016/j.nrleng.2012.06.006
- Al-Harbi KS. Treatment-resistant depression: therapeutic trends, challenges, and future directions. *Patient Prefer Adherence.* (2012) 6:369–88. doi: 10.2147/PPA.S29716
- Robinson RG, Jorge RE. Post-stroke depression: a review. *Am J Psychiatry.* (2016) 173:221–31. doi: 10.1176/appi.ajp.2015.15030363
- Xu XM, Zou DZ, Shen LY, Liu Y, Zhou XY, Pu JC, et al. Efficacy and feasibility of antidepressant treatment in patients with post-stroke depression. *Medicine (Baltimore).* (2016) 95:e5349. doi: 10.1097/MD.00000000000005349
- O'Reardon JP, Solvason HB, Janicak PG, Sampson S, Isenberg KE, Nahas Z, et al. Efficacy and safety of transcranial magnetic stimulation in the acute treatment of major depression: a multisite randomized controlled trial. *Biol Psychiatry.* (2007) 62:1208–16. doi: 10.1016/j.biopsych.2007.01.018
- Jorge RE, Robinson RG, Tateno A, Narushima K, Acion L, Moser D, et al. Repetitive transcranial magnetic stimulation as treatment of poststroke depression: a preliminary study. *Biol Psych.* (2004) 55:398–405. doi: 10.1016/j.biopsych.2003.08.017
- Shen X, Liu M, Cheng Y, Jia C, Pan X, Gou Q, et al. Repetitive transcranial magnetic stimulation for the treatment of post-stroke depression: a systematic review and meta-analysis of randomized controlled clinical trials. *J Affect Disord.* (2017) 211:65–74. doi: 10.1016/j.jad.2016.12.058
- Baeken C, Marinazzo D, Wu GR, Van Schuerbeek P, De Mey J, Marchetti I, et al. Accelerated HF-rTMS in treatment-resistant unipolar depression: insights from subgenual anterior cingulate functional connectivity. *World J Biol Psych.* (2014) 12:2014. doi: 10.3109/15622975.2013.872295
- Herremans SC, Van Schuerbeek P, De Raedt R, Matthys F, Buyl R, De Mey J, et al. The impact of accelerated prefrontal high-frequency repetitive

- transcranial magnetic stimulation (rTMS) on cue-reactivity: an fMRI study on craving in recently detoxified alcohol-dependent patients. *PLoS ONE*. (2015) 10:2015. doi: 10.1371/journal.pone.0136182
13. Holtzheimer PE, McDonald WM, Mufti M, Kelley ME, Quinn S, Corso G, et al. Accelerated repetitive transcranial magnetic stimulation (aTMS) for treatment-resistant depression. *Depr Anx*. (2010). 27:960–3. doi: 10.1002/da.20731
 14. Fitzgerald PB, Hoy KE, Elliot D, Susan McQueen RN, Wambeck LE, Daskalakis ZJ. Accelerated repetitive transcranial magnetic stimulation in the treatment of depression. *Neuropsychopharmacology*. (2018) 43:1565–72. doi: 10.1038/s41386-018-0009-9
 15. Coleman ER, Moudgal R, Lang K, Hyacinth HI, Awosika OO, Kissela BM, et al. Early rehabilitation after stroke: a narrative review. *Curr Atheroscler Rep*. (2017) 19:59. doi: 10.1007/s11883-017-0686-6
 16. Wu CM, McLaughlin K, Lorenzetti DL, Hill MD, Manns BJ, Ghali WA. Early risk of stroke after transient ischemic attack: a systematic review and meta-analysis. *JAMA Internal Med*. (2007) 167:2417–22. doi: 10.1001/archinte.167.22.2417
 17. McGirr A, Van den Eynde F, Tovar-Perdomo S, Fleck MP, Berlim MT. Effectiveness and acceptability of accelerated repetitive transcranial magnetic stimulation (rTMS) for treatment-resistant major depressive disorder: an open label trial. *J Affect Disord*. (2015) 173:216–20. doi: 10.1016/j.jad.2014.10.068
 18. Baeken C, Vanderhasselt MA, Remue J, Herremans S, Vanderbruggen N, Zeeuws D, et al. Intensive HF-rTMS treatment in refractory medication-resistant unipolar depressed patients. *J Affect Disord*. (2013) 151:625–31. doi: 10.1016/j.jad.2013.07.008
 19. Gu SY, Chang MC. The effects of 10-Hz repetitive transcranial magnetic stimulation on depression in chronic stroke patients. *Brain Stimul*. (2017) 10:270–4. doi: 10.1016/j.brs.2016.10.010
 20. Kim BR, Kim DY, Chun MH, Yi JH, Kwon JS. Effect of repetitive transcranial magnetic stimulation on cognition and mood in stroke patients: a double-blind, sham-controlled trial. *Am J Phys Med Rehabil*. (2018) 89:362–8. doi: 10.1097/PHM.0b013e3181d8a5b1
 21. Kim KU, Kim SH, An TG. The effects of repetitive transcranial magnetic stimulation (rTMS) on depression, visual perception, and activities of daily living in stroke patients. *J Physical Ther Sci*. (2017) 29:1036–9. doi: 10.1589/jpts.29.1036
 22. Slotema CW, Blom JD, Hoek HW, Sommer IE. Should we expand the toolbox of psychiatric treatment methods to include Repetitive Transcranial Magnetic Stimulation (rTMS)? A meta-analysis of the efficacy of rTMS in psychiatric disorders. *J Clin Psychiatry*. (2010) 71:873–84. doi: 10.4088/JCP.08m04872gre
 23. Duan X, Yao G, Liu Z, Cui R, Yang W. Mechanisms of transcranial magnetic stimulation treating on post-stroke depression. *Front Neurol*. (2018) 12:215. doi: 10.3389/fnhum.2018.00215
 24. Currier MB, Murray GB, Welch CC. Electroconvulsive therapy for post-stroke depressed geriatric patients. *J Neuropsychiatry Clin Neurosci*. (1992) 4:140–4. doi: 10.1176/jnp.4.2.140
 25. Wassermann EM, Zimmermann T. Transcranial magnetic brain stimulation: therapeutic promises and scientific gaps. *Pharmacol Ther*. (2012) 133:98–107. doi: 10.1016/j.pharmthera.2011.09.003
 26. Paus T, Castro-Alamancos MA, Petrides M. Cortico-cortical connectivity of the human mid-dorsolateral frontal cortex and its modulation by repetitive transcranial magnetic stimulation. *Eur J Neurosci*. (2001) 14:1405–11. doi: 10.1046/j.0953-816x.2001.01757.x
 27. Dubin MJ, Mao X, Banerjee S, Goodman Z, Lapidus KA, Kang G, et al. Elevated prefrontal cortex GABA in patients with major depressive disorder after TMS treatment measured with proton magnetic resonance spectroscopy. *J Psychiatry Neurosci*. (2016) 41:E37–45. doi: 10.1503/jpn.150223
 28. Nordmann G, Azorina V, Langguth B, Sackmann M. A systematic review of non-motor rTMS induced motor cortex plasticity. *Front Hum Neurosci*. (2015) 9:416. doi: 10.3389/fnhum.2015.00416
 29. Autry AE, Monteggia LM. Brain-derived neurotrophic factor and neuropsychiatric disorders. *Pharmacol Rev*. (2012) 64:238–58. doi: 10.1124/pr.111.005108
 30. Chang WH, Shin MA, Lee A, Kim H, Kim YH. Relationship between serum BDNF levels and depressive mood in subacute stroke patients: a preliminary study. *Int J Mol Sci*. (2018) 19:3131. doi: 10.3390/ijms19103131
 31. Li Y, Peng C, Guo X, You JJ, Yadav HP. Expression of brain-derived neurotrophic factor and tyrosine kinase b in cerebellum of poststroke depression rat model. *Chin Med J*. (2015) 128:2926–31. doi: 10.4103/0366-6999.168058
 32. Na KS, Won E, Kang J, Chang HS, Yoon HK, Tae WS, et al. Brain-derived neurotrophic factor promoter methylation and cortical thickness in recurrent major depressive disorder. *Sci Rep*. (2016) 6:21089. doi: 10.1038/srep21089
 33. Phillips C. Brain-derived neurotrophic factor, depression, and physical activity: making the neuroplastic connection. *Neural Plast*. (2017) 2017:7260130. doi: 10.1155/2017/7260130
 34. Yang XR, Kirton A, Wilkes TC, Pradhan S, Liu I, Jaworska N, et al. Glutamate alterations associated with transcranial magnetic stimulation in youth depression: a case series. *J ECT*. (2014) 30:242–7. doi: 10.1097/YCT.0000000000000094
 35. Strafella AP, Paus T, Fraraccio M, Dagher A. Striatal dopamine release induced by repetitive transcranial magnetic stimulation of the human motor cortex. *Brain*. (2003) 126:2609–15. doi: 10.1093/brain/awg268
 36. Cho SS, Strafella AP. rTMS of the left dorsolateral prefrontal cortex modulates dopamine release in the ipsilateral anterior cingulate cortex and orbitofrontal cortex. *PLoS ONE*. (2009) 4:e6725–25. doi: 10.1371/journal.pone.0006725
 37. Étévant A, Manta S, Latapy C, Magno LA, Fecteau S, Beaulieu JM. Repetitive transcranial magnetic stimulation induces long-lasting changes in protein expression and histone acetylation. *Scient Rep*. (2015) 5:16873. doi: 10.1038/srep16873
 38. Chervyakov AV, Chernyavsky AY, Sinitsyn DO, Piradov MA. Possible mechanisms underlying the therapeutic effects of transcranial magnetic stimulation. *Front Hum Neurosci*. (2015) 9:303. doi: 10.3389/fnhum.2015.00303
 39. Hosomi K, Morris S, Sakamoto T, Taguchi J, Maruo T, Kageyama Y, et al. Daily repetitive transcranial magnetic stimulation for poststroke upper limb paresis in the subacute period. *J Stroke Cerebrovasc Dis*. (2016) 25:1655–64. doi: 10.1016/j.jstrokecerebrovasdis.2016.02.024
 40. Guo Z, Jin Y, Peng H, Xing G, Liao X, Wang Y, et al. Ipsilesional high frequency repetitive transcranial magnetic stimulation add-on therapy improved diffusion parameters of stroke patients with motor dysfunction: a preliminary DTI study. *Neural Plasticity*. (2016) 2016:11. doi: 10.1155/2016/6238575
 41. Guan YZ, Li J, Zhang XW, Wu S, Du H, Cui LY, et al. Effectiveness of repetitive transcranial magnetic stimulation (rTMS) after acute stroke: a one year longitudinal randomized trial. *CNS Neurosci Therap*. (2017) 23:940–6. doi: 10.1111/cns.12762
 42. Conforto AB, Anjos SM, Saposnik G, Mello EA, Nagaya EM, Santos W, et al. Transcranial magnetic stimulation in mild to severe hemiparesis early after stroke: a proof of principle and novel approach to improve motor function. *J Neurol*. (2012) 259:1399–405. doi: 10.1007/s00415-011-6364-7
 43. Rossini D, Magri L, Lucca A, Giordani S, Smeraldi E, Zanardi R. Does rTMS hasten the response to escitalopram, sertraline, or venlafaxine in patients with major depressive disorder? A double-blind, randomized, sham-controlled trial. *J Clin Psychiatry*. (2005) 66:1569–75. doi: 10.4088/JCP.v66n1212
 44. Taylor R, Galvez V, Loo C. Transcranial magnetic stimulation (TMS) safety considerations and recommendations. *Transcran Magn Stimul*. (2014) 89:15–30. doi: 10.1007/978-1-4939-0879-0_2

Disclaimer: The content is solely the responsibility of the authors and does not necessarily represent the official views of the NIH.

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2020 Frey, Najib, Lilly and Adcock. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.



Prediction of Motor Recovery in the Upper Extremity for Repetitive Transcranial Magnetic Stimulation and Occupational Therapy Goal Setting in Patients With Chronic Stroke: A Retrospective Analysis of Prospectively Collected Data

Toyohiro Hamaguchi^{1,2}, Naoki Yamada¹, Takuya Hada¹ and Masahiro Abo^{1*}

¹ Department of Rehabilitation Medicine, The Jikei University School of Medicine, Tokyo, Japan, ² Department of Rehabilitation, Graduate School of Health Sciences, Saitama Prefectural University, Koshigaya, Japan

OPEN ACCESS

Edited by:

Carmen Terranova,
University of Messina, Italy

Reviewed by:

Luca Sebastianelli,
Hospital of Vipiteno, Italy
Andrew C. Smith,
Regis University, United States

*Correspondence:

Masahiro Abo
abo@jikei.ac.jp

Specialty section:

This article was submitted to
Neurorehabilitation,
a section of the journal
Frontiers in Neurology

Received: 08 July 2020

Accepted: 03 September 2020

Published: 20 October 2020

Citation:

Hamaguchi T, Yamada N, Hada T and Abo M (2020) Prediction of Motor Recovery in the Upper Extremity for Repetitive Transcranial Magnetic Stimulation and Occupational Therapy Goal Setting in Patients With Chronic Stroke: A Retrospective Analysis of Prospectively Collected Data. *Front. Neurol.* 11:581186. doi: 10.3389/fneur.2020.581186

Recovery from motor paralysis is facilitated by affected patients' recognition of the need for and practice of their own exercise goals. Neurorehabilitation has been proposed and used for the treatment of motor paralysis in stroke, and its effect has been verified. If an expected score for the neurorehabilitation effect can be calculated using the Fugl-Meyer Motor Assessment (FMA), a global assessment index, before neurorehabilitation, such a score will be useful for optimizing the treatment application criteria and for setting a goal to enhance the treatment effect. Therefore, this study verified whether the responsiveness to a treatment method, the Neuro intervention using repetitive transcranial magnetic stimulation and occupational therapy (NEURO), in patients with post-stroke upper extremity (UE) motor paralysis could be predicted by the pretreatment FMA score. No control group was established in this study for NEURO treatment. To analyze the recovery of the motor function in the UE, delta-FMA was calculated from the pre- and post-FMA scores obtained during NEURO treatment. The probability of three levels of treatment responsiveness was evaluated in association with delta-FMA score (<5 , $5 \leq \text{delta-FMA} < 10$, and ≥ 10 as non-responders; responders; and hyper-responders, respectively) according to the reported minimal clinically important difference (MCID). The association of the initial FMA scores with post-FMA scores, from the status of the treatment responsiveness, was determined by multinomial logistic regression analysis. Finally, 1,254 patients with stroke, stratified by FMA scores were analyzed. About 45% of the patients who had FMA scores ranging from 30 to 40 before treatment showed improvement over the MCID by NEURO treatment (odds ratio = 0.93, 95% CI = 0.92–0.95). Furthermore, more than 25% of the patients with more severe initial values, ranging from 26 to 30, improved beyond the MCID calculated in the acute phase

(odds ratio = 0.87, 95% CI = 0.85–0.89). These results suggest that the evaluated motor function score of the UE before NEURO treatment can be used to estimate the possibility of a patient recovering beyond MCID in the chronic phase. This study provided clinical data to estimate the effect of NEURO treatment by the pretreatment FMA-UE score.

Keywords: transcranial magnetic stimulation, occupational therapy, stroke, motor paralysis, prediction

INTRODUCTION

Motor paralysis due to the aftereffects of stroke impairs the activities of daily living (ADL) and quality of life (QOL) of patients; it also affects their individual or social activities (1, 2). In particular, motor paralysis of the upper extremity has a large impact on ADL (3). Recovery from motor paralysis is facilitated by patients recognizing the need for and practicing their own exercise goals (4). The type of goals that patients set are related to their goal satisfaction scores, with impairment-based goals being rated significantly higher than activity-based and participation-based goals (5). It is known that patients' level of knowledge of their rehabilitation goals leads to effective treatment results (6). Thus, clinicians and patients are active partners in setting goals within stroke rehabilitation (5). In previous studies, some prognosis prediction systems were developed for motor paralysis (7–9), and they have been used to set goals for rehabilitation in patients with stroke.

Neurorehabilitation has been proposed and used for the treatment of motor paralysis in stroke, and its effect has been verified (10–14). One of the treatment methods, the NovEl intervention Using Repetitive transcranial magnetic stimulation and Occupational therapy (NEURO), facilitates peripheral muscle movement by controlling the excitability of the motor cortices by repetitive transcranial magnetic stimulation (rTMS). It also promotes peripheral muscle exercise and practice, for the active use of the paralyzed upper extremity (15, 16). NEURO's efficacy has been proved in a randomized controlled study (17). To date, many patients have been treated by using NEURO; however, the prediction regarding whether patients' recovery from motor paralysis after treatments can be predicted before treatment, has not been verified. If the Fugl-Meyer Motor Assessment (FMA) score before treatment can be used to predict NEURO treatment response, the score can be used as an effective goal for rehabilitation, by patients and therapists.

The minimal clinically important difference (MCID) of motor paralysis in the upper extremity has been investigated (18–20). If the expected value of an effect exceeding MCID can be calculated using FMA score measured before NEURO treatment, such a value will be useful for optimizing the treatment application criteria and setting a goal to enhance the treatment effect.

Abbreviations: ADL, activities of daily living; AIC, Akaike Information Criterion; FMA, Fugl-Meyer Motor Assessment; HF, high frequency; LF, low frequency; MCID, minimal clinically important difference; NEURO, NovEl intervention Using Repetitive transcranial magnetic stimulation and Occupational therapy; OT, occupational therapy; QOL, quality of life; rTMS, repetitive transcranial magnetic stimulation; SD, standard deviation; TMS, transcranial magnetic stimulation; UE, upper extremity.

For that purpose, it is sufficient to retroactively analyze the band of the FMA score before NEURO for a patient who is significantly improved. Therefore, this study verified whether the responsiveness of NEURO treatment for patients with post-stroke upper extremity motor paralysis could be predicted by the pre-treatment FMA score.

METHODS

Participants

This is a multi-institutional open-label study without control patients. In January 2019, we surveyed the medical records of all patients with post-stroke muscle paralysis who had been admitted to six participating institutions (Jikei University Hospital, Jikei Third Hospital, Tokyo General Hospital, Kyoto Ohara Memorial Hospital, Nishi-Hiroshima Rehabilitation Hospital, Shimizu Hospital) between March 2010 and December 2018 for NEURO. For patients who had been treated with NEURO, the inclusion criteria were based on the TMS guidelines (21, 22) as follows: (1) upper limb hemiparesis categorized as cerebral infarction or cerebral hemorrhage; (2) age >20 years; (3) ≥4 months since stroke; (4) history of a single stroke only (no bilateral cerebrovascular lesions); (5) no cognitive deficits (a Mini Mental State Examination score ≥26); (6) no active physical or mental illness requiring medical management; (7) no history of convulsion for ≥1 year; (8) no intracranial metal clips or intracardiac pacemaker; and (9) no history of neurolytic nerve block (phenol or botulinum toxin) to the affected upper limb.

To verify if the upper extremity function was maintained after NEURO, patients were excluded: (1) if they did not have at least one FMA score before and after treatment, (2) if they had an initial FMA for upper extremity (FMA-UE) score <26/66, with severe motor impairment (15, 23), and those with a diagnosis of subarachnoid hemorrhage were excluded.

NEURO and Occupational Therapy (OT) Sessions

OT was provided in addition to conducting NEURO sessions; therapy was planned to suit the needs of each patient. All the patients were hospitalized for 15 days to receive rTMS (15) and OT (24). During hospitalization, each patient received a 40-min rTMS session and an OT session every day, except on Sundays and the day of admission/discharge. All OT sessions were started within 10 min of rTMS.

Focal 1 Hz rTMS was applied to the contralesional hemisphere over the primary motor area, as described in previous studies (15, 23). A 70-mm figure-8 coil, attached to a MagPro R100 stimulator (MagVenture Company, Farum, Denmark) was used

for rTMS application; for this, 2,400 pulses lasting for 40 min were applied. The stimulation intensity, set to 90% of the resting motor threshold for the first dorsal interosseous muscle on the unaffected side, was defined as the lowest intensity of the stimulation that could activate the motor-evoked potentials (MEP) of the muscle.

OT was performed twice daily, 6 days a week (excluding Sundays), and involved 60-min individual training sessions. The main goal of the OT sessions was to help the patients avoid focusing on the functional training and to encourage them to use their affected upper limbs again in daily activities. Treatment strategy included: (1) daily physical activities (e.g., eating), which included repetitive movements of the arm during flexion and extension; (2) individualized functional training tasks, which enabled the patients to improve on their movements, such as washing their hands and grasping small items with their paralyzed fingers; (3) elements involved in gross motor function, fine motor function, and multitasking; (4) clear demonstrations of the position of the upper limb to draw attention to this position during training; (5) staged interventions; (6) ADLs and unsupervised training tasks that could be continued after discharge; and (7) the provision of action feedback by passive intervention with verbal instructions.

Sample Size Calculation for Analysis

Based on multivariate linear regression (*F*-tests), an effect size f^2 of 0.03, power ($1-\beta$) of 0.95, α of 0.05, and 6 explained predictors, the minimum sample size of each group was 674 patients (derived using G*Power 3.1) (25). Furthermore, with an expected dropout rate of 30%, we planned to recruit in total a minimum of 963 patients with stroke treated with NEURO. To examine whether detectable logistical separations in upper extremity motor function owing to NEURO could occur, about 1,000 patients with stroke were included in the analysis.

Outcomes

The primary outcome was the FMA score. To predict the responsiveness to NEURO treatment from the initial score of FMA-UE, FMA scores (before and after treatment), age, sex, diagnosis (cerebral infarction or intracerebral hemorrhage), the dominant hand, and the time it took to recover motor function after the onset of stroke were investigated (Figure 1).

Clinical Evaluation of the Motor Function

The motor function of the affected upper extremity was evaluated on both the day of the admission and discharge using FMA score. The FMA was devised in 1975 (26), and is a global assessment index used to quantitatively evaluate the recovery of post-stroke hemiparetic limbs. The FMA has high interrater and test-retest reliability, as described previously (27). The FMA is a performance-based quantitative measure made up of 33 items used to evaluate the upper limb motor function. Each item is rated on a 3-point ordinal scale (0 = cannot perform, 1 = can perform partially, and 2 = can perform fully), with a maximum score of 66 points. The severity of paralysis according to the FMA score is distributed as follows: ≤ 25 , 26–45, and 46–66 for severe, moderate, and mild paralysis, respectively (28–30). The MCID

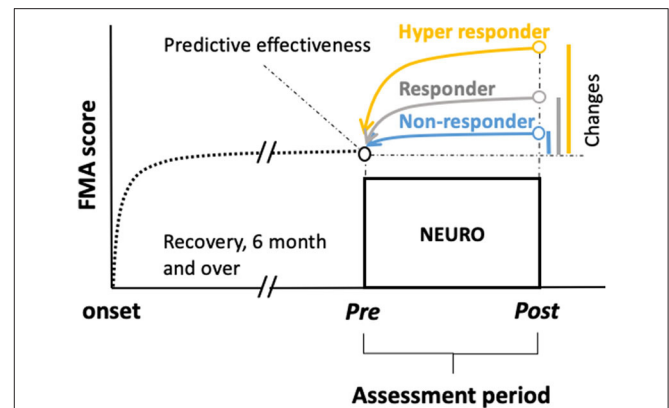


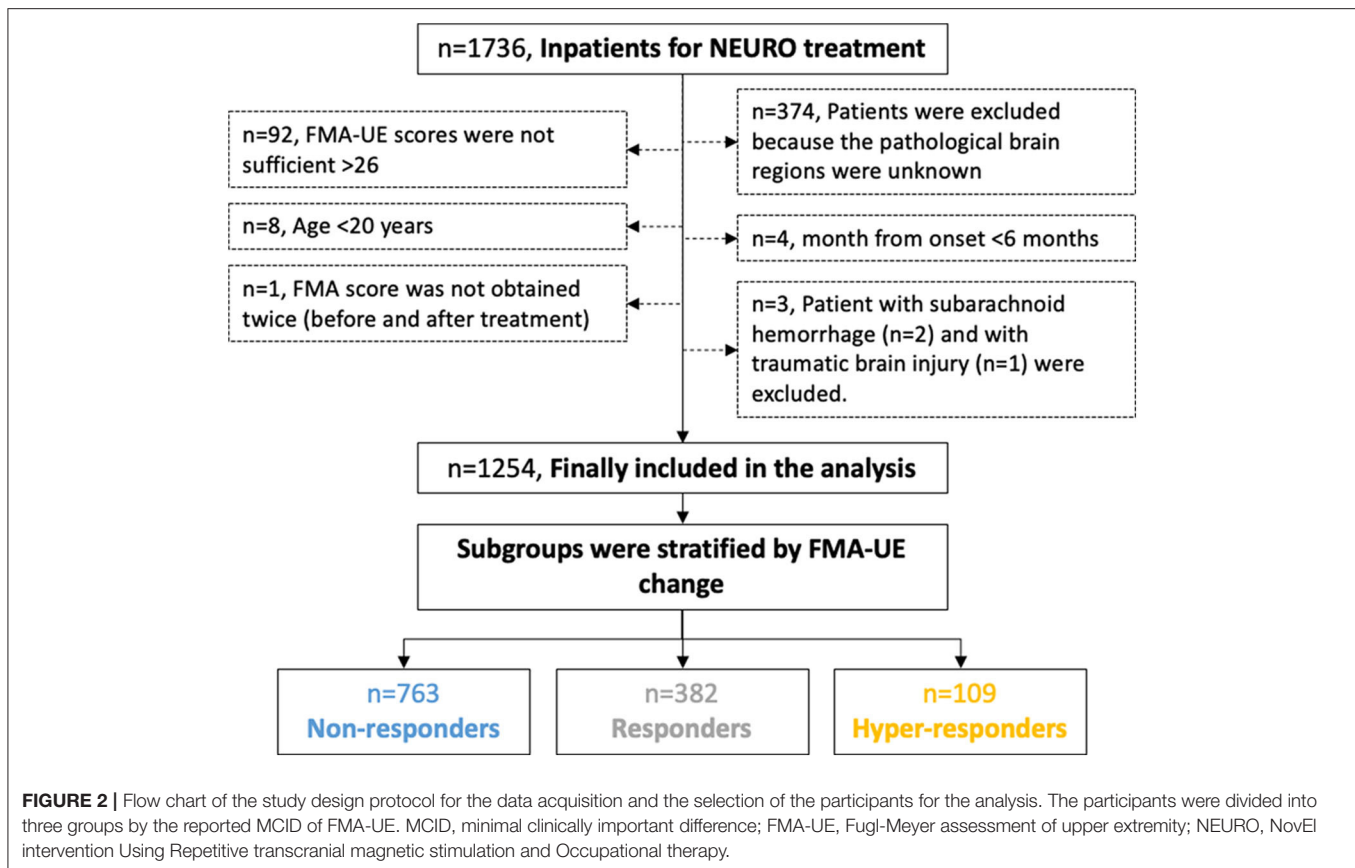
FIGURE 1 | Chart showing schemes of retrospective prediction of the motor recovery of the upper extremities to determine the goals before treatment in patients with chronic stroke undergoing NEURO. To examine the hypothesis that being a responder, non-responder, or hyper-responder resulted in NEURO treatment can be discriminated using multinomial logistic regression to determine the association of FMA score between initial and delta scores in patients with post-stroke hemiparesis. Delta FMA-UE scores were calculated by subtracting the post- from the pre-NEURO score. The black dotted line drawn from the onset indicates the recovery curve from the acute to the chronic phase. The blue, gray, and yellow lines indicate the non-responders, responders, and hyper-responders of NEURO, respectively, regarding the recovery of motor function of the upper extremity. FMA-UE, Fugl-Meyer assessment of upper extremity; NEURO, NovEi intervention Using Repetitive transcranial magnetic stimulation and Occupational therapy.

of FMA for the upper extremity in a population of patients with stroke is 4–10 points in the acute or subacute phase (19, 20), and 5 points in the chronic phase (31).

Statistical Analyses

To analyze the recovery of the motor function in the upper extremity, delta-FMA was calculated from the pre- and post-FMA scores obtained during NEURO treatment. In this study, the probability of the three levels of treatment responsiveness was evaluated in association with the delta-FMA score (<5 , $5 \leq \text{delta-FMA} < 10$, and ≥ 10 as non-responders; responders; and hyper-responders, respectively) according to previous studies (19, 20, 31). The association of the initial FMA scores with post-FMA scores, from the status of the treatment responsiveness, was determined by multinomial logistic regression analysis. The principle of multinomial logistic regression analysis requires that the probability (p) of the three levels (non-responders, responders, and hyper-responders) of the dependent variable, delta-FMA score, be fitted. The probability for the non-responders was the reference level; then the regression models were developed as follows:

$$g(\mathbf{x}_{\text{nonresponders}}) = \frac{1}{1 + e^{f(\mathbf{x}_{\text{responders}})} + e^{f(\mathbf{x}_{\text{hyper-responders}})}} \quad (1 : \text{non-responders})$$



$$f(x_{\text{responders}}) = \text{intercept}_{\text{responders} | \text{nonresponders}} + \beta_{\text{responders} | \text{nonresponders}} x_i$$

$$f(x_{\text{hyper-responders}}) = \text{intercept}_{\text{hyper-responders} | \text{nonresponders}} + \beta_{\text{hyper-responders} | \text{nonresponders}} x_i$$

$$g(x_{\text{responders}}) = \frac{e^{f(x_{\text{responders}})}}{1 + e^{f(x_{\text{responders}})} + e^{f(x_{\text{hyper-responders}})}} \quad (2 : \text{responders})$$

$$g(x_{\text{hyper-responders}}) = \frac{e^{f(x_{\text{hyper-responders}})}}{1 + e^{f(x_{\text{responders}})} + e^{f(x_{\text{hyper-responders}})}} \quad (3 : \text{hyper-responders})$$

where x_i , the initial-FMA-UE score, was the explanatory variable, β_i and **intercept_i** is the partial regression coefficient in each group, and e is Napier's constant. Therefore, for the multilevel responses, the cumulative probability was calculated at each level to generate a simple regression coefficient. The covariates influencing the recovery of the upper limb motor paralysis after treatment were: (1) age, (2) gender, (3) time from stroke onset to NEURO initiation, and 4) the dominant hand. To identify the model, the Akaike Information Criterion (AIC) was used (32). Applicability of the predictive model was assessed using McFadden's coefficient of determination, R^2 , between the initial score and the delta-FMA scores for all 1,254 patients (33). All

statistical analyses were performed using R 3.6.0 software (R Foundation for Statistical Computing, Vienna, Austria).

RESULTS

Figure 2 shows the flow chart of the study design and patients selection based on the diagnosis. The median age and interquartile range of all patients were 63 and 56–70 years respectively. **Table 1** summarizes the clinical characteristics of the patients; the distributions of the characteristics were comparable across groups. Right-handed patients accounted for 95%, which is approximately equal to the same proportion for all Japanese. There were about twice as many males as females.

The multinomial logistic regression model fitted showed statistically significant valid logistic probability between delta- and the initial FMA score, adjusted for covariates, age, sex, time from onset, diagnosis, and dominant hand (McFadden's $R^2 = 0.103$, AIC = 1,999, $\chi^2 = 227$, $p < 0.001$) (**Table 2**). Time-series plots of the FMA scores are shown in **Figure 3**. The logistic curves discriminating between the probability of being responders ($5 \leq \text{delta-FMA} < 10$) from non-responders ($\text{delta-FMA} < 5$) showed a significant model fit ($z = 5.31$; $p < 0.001$; odds ratio = 15.5, 95% CI = 5.7–42.9). Similarly, hyper-responders ($\text{delta-FMA} \geq 10$) and non-responders ($\text{delta-FMA} < 5$) were differentiated according to the initial-FMA score ($z = 6.38$; $p < 0.001$; odds ratio = 166.8, 95% CI = 34.6–803.5).

TABLE 1 | Patient characteristics among groups at baseline.

Characteristic	Non-responders	Responders	Hyper-responders
Participants (n)	763 (61%)	382 (26%)	109 (13%)
Age (years)	63 (56–70)	63 (55–70)	64 (56–69)
Sex (n)			
Female	247 (32%)	123 (32%)	41 (38%)
Male	516 (68%)	259 (68%)	68 (62%)
Paralysis side (n)			
Left	315 (41%)	173 (45%)	55 (50%)
Right	448 (59%)	209 (55%)	54 (50%)
Dominant hand (n)			
Left	35 (5%)	23 (6%)	5 (5%)
Right	728 (95%)	359 (94%)	104 (95%)
Diagnosis			
CI	387 (51%)	194 (51%)	51 (47%)
ICH	376 (49%)	188 (49%)	58 (53%)
Time from onset (months)	41 (23–74)	41 (24–75)	37 (21–58)
FMA-UE (in charge)	54 (46–60)	47 (39–52)	40 (33–45)

Values are n (%) or median (interquartile range). CI, cerebral infarction; ICH, intracranial hemorrhage; FMA-UE, Fugl-Meyer Assessment score.

TABLE 2 | Model coefficients of treatment responsiveness and initial FMA-UE score.

Separated responsiveness	Predictor	Estimate	95% CI	z	p
Responders Non-responders	Intercept	2.75	1.73, 3.76	5.31	<0.001
	Initial FMA-UE	−0.07	−0.08, −0.06	−10.01	<0.001
	Age	−0.00	−0.01, 0.01	−0.17	0.863
	Sex	0.02	−0.25, 0.30	0.16	0.871
	Month from onset	−0.00	−0.00, 0.00	−0.25	0.806
	Diagnosis	0.04	−0.22, 0.30	0.30	0.764
	Handedness	0.35	−0.21, 0.92	1.22	0.224
Hyper-responders Non-responders	Intercept	5.11	3.54, 6.69	6.38	<0.001
	Initial FMA-UE	−0.14	−0.16, −0.12	−11.64	<0.001
	Age	−0.01	−0.03, 0.01	−1.03	0.302
	Sex	−0.24	−0.69, 0.22	−1.02	0.306
	Month from onset	−0.00	−0.01, −0.00	−0.74	0.458
	Diagnosis	0.28	−0.16, 0.72	1.25	0.213
	Handedness	0.18	−0.85, 1.2	0.34	0.729

FMA-UE, motor function score of upper extremity by Fugl-Meyer Assessment; N-R, non-responders; R, responders; H-R, hyper-responders.

According to the multinomial logistic regression models, the probability of being a non-responders was 59.2% when the initial FMA score was 48.9. Similarly, when the initial FMA score was 38.8, the incidence of responders and hyper-responders was 45.5 and 16.0%, respectively (Table 3).

DISCUSSION

Recently, the maximum recovery state of motor function of the upper extremity in patients with stroke hemiparesis,

including spontaneous recovery, has been estimated, based on the measured acute phase value (7, 34–36). Subsequent studies have also shown that NEURO treatment may restore motor function in the upper extremities during the chronic phase (15, 17). In this study, motor function of the upper extremities, based on values measured prior to NEURO treatment, was used to estimate post-treatment recovery rates based on previously reported acute and chronic MCID levels (19, 20, 31). The results of this study showed that about 45% of patients in the chronic stage who had FMA scores ranging from 30 to 40 before treatment showed improvement over the MCID by NEURO treatment. Furthermore, more than 25% of the patients with more severe initial values ranging from 26 to 30 improved beyond the MCID calculated in the acute phase. These results suggest that the evaluated motor function scores of the upper extremities before NEURO treatment can be used to estimate the occurrence of patients recovering beyond MCID among the patients in the chronic phase.

It is known that the effect of rehabilitation is enhanced when patients recognize the need to achieve their own goals and actively engage in pursuing them (37, 38). In addition, patients who practice self-efficacy affect the recovery of the upper limb motor function (4). Patients' recognition of the need to have their own behavioral goals and practice upper limb exercises display enhanced performance (4). Therefore, prediction of the treatment effect on the patient is important for the therapist and can facilitate patients' consent and cooperation with the treatment (39). To judge from the results of this study, the extent of recovery by NEURO treatment can be predicted, to some extent, from the patients' pre-treatment upper extremity functional evaluation, and this is useful information for the attending physician to provide the patient.

In this NEURO treatment, low frequency (LF)-rTMS was used. Ferbert et al. discovered that stimulation of the contralateral motor cortex immediately after stimulation of the motor area reduces the potential of stimulation of the contralateral hemisphere to evoke finger muscles (40). Moreover, Wards et al. reported that in the case of unilateral brain injury, the activity of the contralateral hemisphere was increased, and hyperactivity of the non-lesional hemisphere excessively induced the interhemispheric inhibition on the lesional side (41). In other words, unbalanced excitement of the cerebrum on the non-lesioned hemisphere adversely affects functional improvement. Since nervous activity is suppressed by LF-rTMS, the activity of the non-lesional hemisphere can be suppressed by applying LF-rTMS to the motor cortex of the non-lesional hemisphere (42), and suppression of interhemispheric inhibition of the non-lesional hemisphere indirectly increases the activity of the lesional side (43). On the other hand, high-frequency (HF)-rTMS evokes nervous activity and stimulates the motor cortex of the lesional hemisphere to enhance activity at the lesional site directly (44). Intensive upper-limb exercises are performed immediately after rTMS while the neurological activity of patients with stroke is adjusted, thus facilitating motor function (10, 45). The stimulation method corresponding to the effects of the neuromodulation in patients with various levels of disability will hopefully

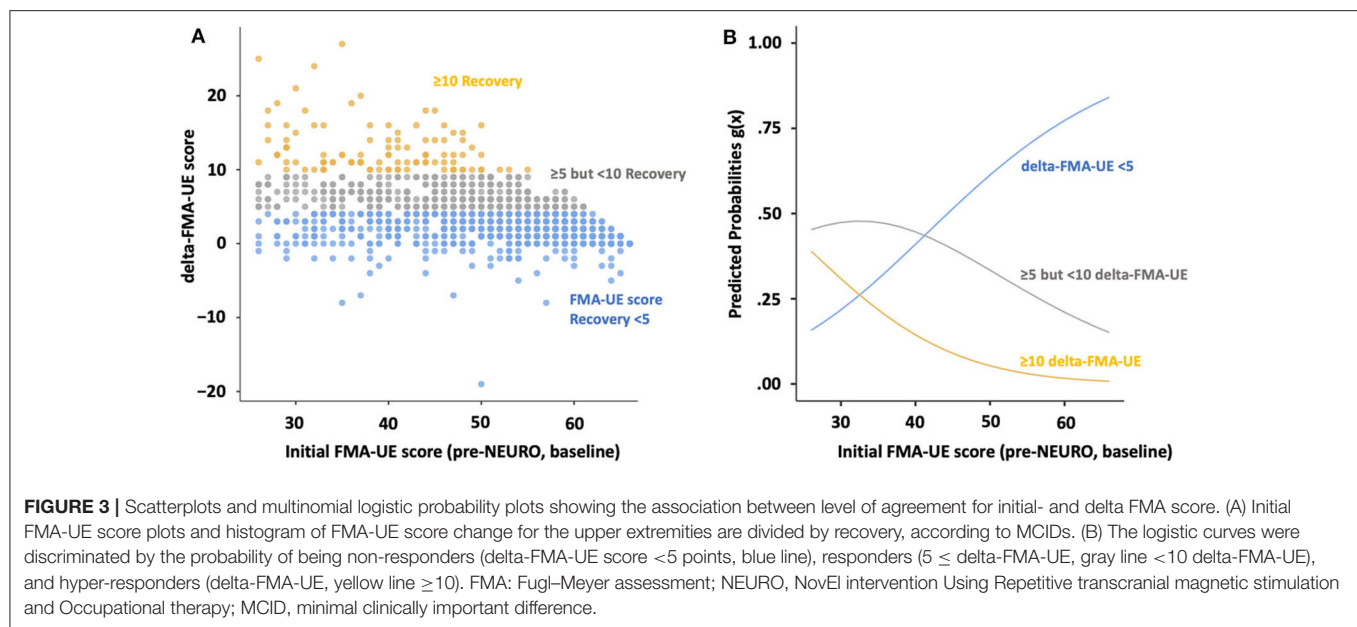


FIGURE 3 | Scatterplots and multinomial logistic probability plots showing the association between level of agreement for initial- and delta FMA score. (A) Initial FMA-UE score plots and histogram of FMA-UE score change for the upper extremities are divided by recovery, according to MCIDs. (B) The logistic curves were discriminated by the probability of being non-responders (delta-FMA-UE score <5 points, blue line), responders ($5 \leq$ delta-FMA-UE, gray line <10 delta-FMA-UE), and hyper-responders (delta-FMA-UE, yellow line ≥ 10). FMA: Fugl-Meyer assessment; NEURO, NovEl intervention Using Repetitive transcranial magnetic stimulation and Occupational therapy; MCID, minimal clinically important difference.

TABLE 3 | Estimated marginal means of Fugl-Meyer Assessment score in upper extremity, compared with responsiveness of treatment.

Initial FMA-UE	Responsiveness	Probability	SE	95% Confidence Interval	
				Lower	Upper
38.8 ⁻	N-R	0.384	0.036	0.307	0.462
	R	0.455	0.039	0.372	0.539
	H-R	0.160	0.034	0.087	0.234
48.9 ^μ	N-R	0.592	0.035	0.518	0.667
	R	0.347	0.033	0.277	0.419
	H-R	0.060	0.016	0.027	0.093
59.0 ⁺	N-R	0.760	0.030	0.695	0.825
	R	0.221	0.029	0.158	0.284
	H-R	0.019	0.006	0.005	0.032

CI, cerebral infarction; ICH, intracerebral hemorrhage; N-R, nonresponders; R, responders; H-R, hyper-responders. ⁻, mean - 1SD; ^μ, mean; ⁺, mean + 1SD; FMA-UE, Fugl-Meyer Assessment score; SD, standard deviation; SE, standard error.

be of use in the clinical setting after further validation of its effectiveness.

In this study, recovery from motor paralysis in the upper extremities with NEURO treatment tended to occur more frequently in patients with moderate paralysis. In the chronic phase of stroke, the most widely accepted explanation for the efficacy of the 1-Hz stimulation of the unaffected hemisphere is the reduction in the abnormally high transcallosal inhibition toward the affected hemisphere (46, 47). In the acute phase, Wang et al. reported that HF-rTMS and exercise therapy could improve motor recovery at about a 10 FMA-UE score in patients with severe hemiplegic stroke (48). Similarly, Watanabe et al. reported that patients in the acute phase had reduced muscle spasticity and recovery of motor function with rTMS (49). Even when motor paralysis was severe, improvement

of motor function in the upper extremities was observed by adjusting the excitability of the motor cortex in this study. In addition, the FMA-UE assesses the patients, post-stroke, per the sequential recovery stages (26). The FMA items are hierarchically organized from synergistic to voluntary movements. Synergistic movements exhibit abnormally stereotyped behavior that does not allow the combination of different movement patterns. For example, an attempt to raise the arm results in elbow flexion, shoulder abduction, and internal rotation. The flexor and extensor synergy components were tested before the movements combining the synergies with the movements out of synergy. Yayun et al. reported that the increase in FMA-UE score reflects the improvement of the proximal upper extremity movement (50). It is considered that the rTMS treatment improved the FMA score, and the patients with more severe motor paralysis had improved proximal upper limb movements. Schambra et al. reported that there was no difference in FME-UE score recovery with or without MEP in patients in the acute phase, but there was less improvement in patients with high FMA scores than in those with low FMA scores, and FMA recovery curves plateaued below the reported normal levels for both the arm and hand (51). The lower response of patients with high motor function compared to moderately paretic patients in our study might be because the treatment-recovery values were low in patients with high motor function. Furthermore, Veldema et al. reported that in patients with stroke, severe hand dysfunction was associated with a strong suppression of the ipsilesional cortico-spinal excitability and a shift in excitability toward the contralesional hemisphere (52). In the same study, mild hand movement impairment was associated with a shift in cortico-spinal excitability toward the ipsilesional hemisphere. Therefore, ipsilesional HF-rTMS may be effective in mild paralysis. As the upper extremities become more active, patients may be willing to actively use it. The results of this study clinically suggested that even in more affected moderate cases of motor paralysis in the chronic phase, the effect of

rehabilitation can be obtained in about 20% of patients, as in the acute phase.

Clinically (although not shown by the data in this study) and frequently, after the treatment there are highly psychologically satisfied patients, because they could use their own extremities and hands due to decreased finger clawing and because objects could be held by the paralyzed hands, even if the FMA score did not significantly change. Therefore, clinicians are required to explain to the patients how much they can improve and motivate them to participate in the treatment. To this end, further research should be conducted on the relationship between patients' motor function and their level of satisfaction, as well as the evaluation of gross and fine movement improvements.

There were some limitations to this study. Although the study did not include treatment data other than for NEURO, the patients included in the analysis may have received other treatments simultaneously, such as exercise therapy or OT. In addition, since the upper extremities are often used in ADL, the amount of functional recovery of the upper extremities is generally increased. The effects of the difference on non-NEURO treatments can be identified by comparing the recovery prediction accuracy between a non-NEURO-treated group and others treated with NEURO. There were more than 1,200 subjects in this study, and performing the stratified analysis described above requires larger samples.

CONCLUSION

This study provided clinical data to estimate the effect of NEURO treatment by pre-treatment FMA-UE score. Further verification is required regarding the need for both the patients and therapists to undergo rehabilitation with the goal of recovery before and after treatment, which has a favorable effect on treatment outcomes.

DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

REFERENCES

- Choi-Kwon S, Choi JM, Kwon SU, Kang DW, Kim JS. Factors that affect the quality of life at 3 years post-stroke. *J Clin Neurol.* (2006) 2:34–41. doi: 10.3988/jcn.2006.2.1.34
- van Mierlo ML, van Heugten CM, Post MW, Hajos TR, Kappelle LJ, Visser-Meily JM. Quality of life during the first two years post stroke: the restore4Stroke cohort study. *Cerebrovasc Dis.* (2016) 41:19–26. doi: 10.1159/000441197
- Fugl-Meyer AR, Jaasko L. Post-stroke hemiplegia and ADL-performance. *Scand J Rehabil Med Suppl.* (1980) 7:140–52.
- Stewart JC, Lewthwaite R, Rocktaschel J, Winstein CJ. Self-efficacy and reach performance in individuals with mild motor impairment due to stroke. *Neurorehabil Neural Repair.* (2019) 33:319–28. doi: 10.1177/1545968319836231
- Rice DB, McIntyre A, Mirkowski M, Janzen S, Viana R, Britt E, et al. Patient-centered goal setting in a hospital-based outpatient stroke rehabilitation center. *PM R.* (2017) 9:856–65. doi: 10.1016/j.pmrj.2016.12.004

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by The ethics committee of the Tokyo Jikei University School of Medicine and included an opt-out consent method (No. 20-041-5231). The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

THam: analysis of data, data interpretation, and writing of the manuscript. NY: data interpretation and revisions. THad: data interpretation and revisions. MA: conception/design of the study, acquisition and analysis of data, data interpretation, writing of the manuscript, and revisions. All authors approved the submitted version and have agreed both to be personally accountable for the authors' contributions and to ensure that questions related to the accuracy or integrity of any part of the work, even ones in which the author was not personally involved, have been appropriately investigated, resolved, and the resolution documented in the literature.

FUNDING

This study was supported by Grants-in-Aid for Scientific Research (c) No. 18K10691.

ACKNOWLEDGMENTS

The authors would like to thank Dr. Hisashi Tatsuno, Department of Rehabilitation Medicine, The Jikei University School of Medicine; Dr. Jinichi Sasanuma and Shin-Yurigaoka, General Hospital; Dr. Kiyohito Kakita and Kyoto Ohara Memorial Hospital; Dr. Takatsugu Okamoto and Nishi-Hiroshima Rehabilitation Hospital; and Dr. Masato Shimizu, Shimizu Hospital, for their clinical advice and material assistance for the successful completion of this study.

- Turner-Stokes L, Ashford S, Esquenazi A, Wissel J, Ward AB, Francisco G, et al. A comprehensive person-centered approach to adult spastic paresis: a consensus-based framework. *Eur J Phys Rehabil Med.* (2018) 54:605–17. doi: 10.23736/S1973-9087.17.04808-0
- Prabhakaran S, Zarahn E, Riley C, Speizer A, Chong JY, Lazar RM, et al. Inter-individual variability in the capacity for motor recovery after ischemic stroke. *Neurorehabil Neural Repair.* (2008) 22:64–71. doi: 10.1177/1545968307305302
- Stinear CM, Barber PA, Petoe M, Anwar S, Byblow WD. The PREP algorithm predicts potential for upper limb recovery after stroke. *Brain.* (2012) 135(Pt. 8):2527–35. doi: 10.1093/brain/awt146
- Stinear CM, Byblow WD, Ackerley SJ, Smith MC, Borges VM, Barber PA. PREP2: a biomarker-based algorithm for predicting upper limb function after stroke. *Ann Clin Transl Neurol.* (2017) 4:811–20. doi: 10.1002/actn.3.488
- Lefaucheur JP, Aleman A, Baeken C, Benninger DH, Brunelin J, Di Lazzaro V, et al. Evidence-based guidelines on the therapeutic use of repetitive transcranial magnetic stimulation (rTMS): an update (2014–2018). *Clin Neurophysiol.* (2020) 131:474–528. doi: 10.1016/j.clinph.2020.02.003

11. Pan W, Wang P, Song X, Sun X, Xie Q. The effects of combined low frequency repetitive transcranial magnetic stimulation and motor imagery on upper extremity motor recovery following stroke. *Front Neurol.* (2019) 10:96. doi: 10.3389/fneur.2019.00096
12. Maier M, Rubio Ballester B, Duff A, Duarte Oller E, Verschure P. Effect of specific over nonspecific VR-based rehabilitation on poststroke motor recovery: a systematic meta-analysis. *Neurorehabil Neural Repair.* (2019) 33:112–29. doi: 10.1177/1545968318820169
13. Jin M, Zhang Z, Bai Z, Fong KNK. Timing-dependent interaction effects of tDCS with mirror therapy on upper extremity motor recovery in patients with chronic stroke: a randomized controlled pilot study. *J Neurol Sci.* (2019) 405:116436. doi: 10.1016/j.jns.2019.116436
14. Kawakami M, Fujiwara T, Ushiba J, Nishimoto A, Abe K, Honaga K, et al. A new therapeutic application of brain-machine interface (BMI) training followed by hybrid assistive neuromuscular dynamic stimulation (HANDS) therapy for patients with severe hemiparetic stroke: a proof of concept study. *Restor Neurol Neurosci.* (2016) 34:789–97. doi: 10.3233/RNN-160652
15. Kakuda W, Abo M, Shimizu M, Sasanuma J, Okamoto T, Yokoi A, et al. A multi-center study on low-frequency rTMS combined with intensive occupational therapy for upper limb hemiparesis in post-stroke patients. *J Neuroeng Rehabil.* (2012) 9:4. doi: 10.1186/1743-0003-9-4
16. Kakuda W, Abo M, Kobayashi K, Momosaki R, Yokoi A, Ito H, et al. Low-frequency rTMS combined with intensive occupational therapy for upper limb hemiparesis after brain tumour resection. *Brain Inj.* (2010) 24:1505–10. doi: 10.3109/02699052.2010.523040
17. Abo M, Kakuda W, Momosaki R, Harashima H, Kojima M, Watanabe S, et al. Randomized, multicenter, comparative study of NEURO versus CIMT in poststroke patients with upper limb hemiparesis: the NEURO-VERIFY study. *Int J Stroke.* (2014) 9:607–12. doi: 10.1111/ijfs.12100
18. van der Lee JH, Wagenaar RC, Lankhorst GJ, Vogelaar TW, Deville WL, Bouter LM. Forced use of the upper extremity in chronic stroke patients: results from a single-blind randomized clinical trial. *Stroke.* (1999) 30:2369–75. doi: 10.1161/01.STR.30.11.2369
19. Arya KN, Verma R, Garg RK. Estimating the minimal clinically important difference of an upper extremity recovery measure in subacute stroke patients. *Top Stroke Rehabil.* (2011) 18(Suppl. 1):599–610. doi: 10.1310/tsr18s01-599
20. Lundquist CB, Maribo T. The Fugl-Meyer assessment of the upper extremity: reliability, responsiveness and validity of the Danish version. *Disabil Rehabil.* (2017) 39:934–9. doi: 10.3109/09638288.2016.1163422
21. Wassermann EM. Risk and safety of repetitive transcranial magnetic stimulation: report and suggested guidelines from the international workshop on the safety of repetitive transcranial magnetic stimulation, June 5–7, 1996. *Electroencephalogr Clin Neurophysiol.* (1998) 108:1–16. doi: 10.1016/S0168-5597(97)00096-8
22. Rossi S, Hallett M, Rossini PM, Pascual-Leone A. Safety of TMS/SCG. Safety, ethical considerations, and application guidelines for the use of transcranial magnetic stimulation in clinical practice and research. *Clin Neurophysiol.* (2009) 120:2008–39. doi: 10.1016/j.clinph.2009.08.016
23. Kondo T, Yamada N, Momosaki R, Shimizu M, Abo M. Comparison of the effect of low-frequency repetitive transcranial magnetic stimulation with that of theta burst stimulation on upper limb motor function in poststroke patients. *Biomed Res Int.* (2017) 2017:4269435. doi: 10.1155/2017/4269435
24. Kondo T, Kakuda W, Yamada N, Shimizu M, Hagino H, Abo M. Effect of low-frequency rTMS on motor neuron excitability after stroke. *Acta Neurol Scand.* (2013) 127:26–30. doi: 10.1111/j.1600-0404.2012.01669.x
25. Faul F, Erdfelder E, Buchner A, Lang AG. Statistical power analyses using G*power 3.1: tests for correlation and regression analyses. *Behav Res Methods.* (2009) 41:1149–60. doi: 10.3758/BRM.41.4.1149
26. Fugl-Meyer AR, Jaasko L, Leyman I, Olsson S, Steglind S. The post-stroke hemiplegic patient. 1. A method for evaluation of physical performance. *Scand J Rehabil Med.* (1975) 7:13–31.
27. Platz T, Pinkowski C, van Wijck F, Kim IH, di Bella P, Johnson G. Reliability and validity of arm function assessment with standardized guidelines for the fugl-meyer test, action research arm test and box and block test: a multicentre study. *Clin Rehabil.* (2005) 19:404–11. doi: 10.1191/0269215505cr832oa
28. Woytowicz EJ, Rietschel JC, Goodman RN, Conroy SS, Sorkin JD, Whittall J, et al. Determining levels of upper extremity movement impairment by applying a cluster analysis to the Fugl-Meyer assessment of the upper extremity in chronic stroke. *Arch Phys Med Rehabil.* (2017) 98:456–62. doi: 10.1016/j.apmr.2016.06.023
29. Duncan PW, Lai SM, Keighley J. Defining post-stroke recovery: implications for design and interpretation of drug trials. *Neuropharmacology.* (2000) 39:835–41. doi: 10.1016/S0028-3908(00)00003-4
30. Gladstone DJ, Danells CJ, Black SE. The fugl-meyer assessment of motor recovery after stroke: a critical review of its measurement properties. *Neurorehabil Neural Repair.* (2002) 16:232–40. doi: 10.1177/154596802401105171
31. Rocha S, Silva E, Foerster A, Wiesiolek C, Chagas AP, Machado G, et al. The impact of transcranial direct current stimulation (tDCS) combined with modified constraint-induced movement therapy (mCIMT) on upper limb function in chronic stroke: a double-blind randomized controlled trial. *Disabil Rehabil.* (2016) 38:653–60. doi: 10.3109/09638288.2015.1055382
32. Shibata R. Selection of the order of an autoregressive model by Akaike's information criterion. *Biometrika.* (1976) 63:117–26. doi: 10.1093/biomet/63.1.117
33. McFadden D. Conditional logit analysis of qualitative choice behavior. In: Zarembka P, editor. *Economic Theory and Mathematical Economics.* New York, NY: Academic Press (1974) 105–45.
34. Duncan PW, Goldstein LB, Horner RD, Landsman PB, Samsa GP, Matchar DB. Similar motor recovery of upper and lower extremities after stroke. *Stroke.* (1994) 25:1181–8. doi: 10.1161/01.STR.25.6.1181
35. Katrak P, Bowring G, Conroy P, Chilvers M, Poulos R, McNeil D. Predicting upper limb recovery after stroke: the place of early shoulder and hand movement. *Arch Phys Med Rehabil.* (1998) 79:758–61. doi: 10.1016/S0003-9993(98)90352-5
36. Stinear CM, Byblow WD, Ackerley SJ, Barber PA, Smith MC. Predicting recovery potential for individual stroke patients increases rehabilitation efficiency. *Stroke.* (2017) 48:1011–9. doi: 10.1161/STROKEAHA.116.015790
37. Ekstrand E, Alt Murphy M, Persson HC, Lundgren-Nilsson A, Sunnerhagen KS. Which clinical and sociodemographic determinants are associated with self-perceived manual ability at one year after stroke? *Disabil Rehabil.* (2020) 42:2279–86. doi: 10.1080/09638288.2018.1557265
38. McEwen SE, Donald M, Jutzi K, Allen KA, Avery L, Dawson DR, et al. Implementing a function-based cognitive strategy intervention within inter-professional stroke rehabilitation teams: changes in provider knowledge, self-efficacy and practice. *PLoS ONE.* (2019) 14:e0212988. doi: 10.1371/journal.pone.0212988
39. Parsons JGM, Plant SE, Slark J, Tyson SF. How active are patients in setting goals during rehabilitation after stroke? A qualitative study of clinician perceptions. *Disabil Rehabil.* (2018) 40:309–16. doi: 10.1080/09638288.2016.1253115
40. Ferbert A, Priori A, Rothwell JC, Day BL, Colebatch JG, Marsden CD. Interhemispheric inhibition of the human motor cortex. *J Physiol.* (1992) 453:525–46. doi: 10.1113/jphysiol.1992.sp019243
41. Ward NS, Brown MM, Thompson AJ, Frackowiak RS. Neural correlates of motor recovery after stroke: a longitudinal fMRI study. *Brain.* (2003) 126:2476–96. doi: 10.1093/brain/awg245
42. Takarada Y, Mima T, Abe M, Nakatsuka M, Taira M. Inhibition of the primary motor cortex can alter one's "sense of effort": effects of low-frequency rTMS. *Neurosci Res.* (2014) 89:54–60. doi: 10.1016/j.neures.2014.09.005
43. Mello EA, Cohen LG, Monteiro Dos Anjos S, Conti J, Andrade KN, Tovar Moll F, et al. Increase in short-interval intracortical facilitation of the motor cortex after low-frequency repetitive magnetic stimulation of the unaffected hemisphere in the subacute phase after stroke. *Neural Plast.* (2015) 2015:407320. doi: 10.1155/2015/407320
44. Guo Z, Jin Y, Peng H, Xing G, Liao X, Wang Y, et al. Ipsilesional High frequency repetitive transcranial magnetic stimulation add-on therapy improved diffusion parameters of stroke patients with motor dysfunction: a preliminary DTI study. *Neural Plast.* (2016) 2016:6238575. doi: 10.1155/2016/6238575
45. Du J, Yang F, Hu J, Hu J, Xu Q, Cong N, et al. Effects of high- and low-frequency repetitive transcranial magnetic stimulation on motor recovery in early stroke patients: evidence from a randomized controlled trial with clinical, neurophysiological and functional imaging assessments. *Neuroimage Clin.* (2019) 21:101620. doi: 10.1016/j.nicl.2018.101620

46. Xu AH, Sun YX. Research hotspots and effectiveness of repetitive transcranial magnetic stimulation in stroke rehabilitation. *Neural Regen Res.* (2020) 15:2089–97. doi: 10.4103/1673-5374.282269
47. McDonnell MN, Stinear CM. TMS measures of motor cortex function after stroke: a meta-analysis. *Brain Stimul.* (2017) 10:721–34. doi: 10.1016/j.brs.2017.03.008
48. Wang Q, Zhang D, Zhao YY, Hai H, Ma YW. Effects of high-frequency repetitive transcranial magnetic stimulation over the contralesional motor cortex on motor recovery in severe hemiplegic stroke: a randomized clinical trial. *Brain Stimul.* (2020) 13:979–86. doi: 10.1016/j.brs.2020.03.020
49. Watanabe K, Kudo Y, Sugawara E, Nakamizo T, Amari K, Takahashi K, et al. Comparative study of ipsilesional and contralesional repetitive transcranial magnetic stimulations for acute infarction. *J Neurol Sci.* (2018) 384:10–4. doi: 10.1016/j.jns.2017.11.001
50. Lee YY, Hsieh YW, Wu CY, Lin KC, Chen CK. Proximal Fugl-Meyer assessment scores predict clinically important upper limb improvement after 3 stroke rehabilitative interventions. *Arch Phys Med Rehabil.* (2015) 96:2137–44. doi: 10.1016/j.apmr.2015.07.019
51. Schambra HM, Xu J, Branscheidt M, Lindquist M, Uddin J, Steiner L, et al. Differential poststroke motor recovery in an arm versus hand muscle in the absence of motor evoked potentials. *Neurorehabil Neural Repair.* (2019) 33:568–80. doi: 10.1177/1545968319850138
52. Veldema J, Bosl K, Nowak DA. Cortico-spinal excitability and hand motor recovery in stroke: a longitudinal study. *J Neurol.* (2018) 265:1071–8. doi: 10.1007/s00415-018-8802-2

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2020 Hamaguchi, Yamada, Hada and Abo. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.



How to Design Optimal Accelerated rTMS Protocols Capable of Promoting Therapeutically Beneficial Metaplasticity

Alix C. Thomson^{1,2,3} and Alexander T. Sack^{1,2,3*}

¹ Department of Cognitive Neuroscience, Faculty of Psychology and Neuroscience, Maastricht University, Maastricht, Netherlands, ² Department of Psychiatry and Neuropsychology, School for Mental Health and Neuroscience (MHeNS), Maastricht, Netherlands, ³ Centre for Integrative Neuroscience, Faculty of Psychology and Neuroscience, Faculty of Health, Medicine and Life Sciences, Maastricht University, Maastricht, Netherlands

Keywords: metaplasticity, homeostatic plasticity, hebbian plasticity, transcranial magnetic stimulation (TMS), accelerated rTMS

INTRODUCTION

Our brain is comprised of billions of neurons, which can connect via synapses that rely on electrical signaling and the release of chemical messengers to communicate and propagate signals through neural networks. By forming such networks, neurons are capable of monitoring previous firing activity, and using this information to adapt subsequent firing rate. This so-called activity-dependent plasticity is critical for the encoding of new information, and the tuning of (low activity) connections (1–3). The physiological mechanisms of synaptic plasticity have largely been attributed to Long-Term Potentiation (LTP) (4, 5), and Long-Term Depression (LTD) (6–8), which result from molecular processes such as receptor trafficking or synaptic scaling (3). Both LTP and LTD are induced by postsynaptic NMDA receptor activation, which lead to an influx of calcium into the postsynaptic dendrites (8–10). This triggers a complex series of intracellular signaling cascades, resulting in synaptic modifications such as AMPA receptor trafficking (11, 12). The pattern of stimuli delivered to the post synapse determines whether LTP or LTD will occur; low frequency stimulation induces LTD, whereas high frequency stimulation induces LTP (8, 13). These processes underlie much of our knowledge on the molecular mechanisms of learning and memory.

However, if the principles of Hebbian synaptic plasticity (LTP, LTD) alone were to drive the strengthening and weakening of synaptic connections, activity would, over time, be driven toward destabilization. This is because continuously firing synapses could only become stronger (driven to saturation) and unused synapses quiescent (until completely lost) (14). Consider a synapse that is strengthened by LTP; meaning the presynaptic neuron becomes more effective at depolarizing the postsynaptic neuron. With each continued stimulation, the postsynaptic neuron will be more easily depolarized, in a positive feedback loop, resulting in a hyperexcitable postsynaptic neuron. Over time, not only will the original presynaptic connection be strengthened, but other unrelated presynaptic inputs could cause a depolarization of the hyperexcitable postsynaptic neuron, resulting in unregulated synaptic transmission (15). Therefore, other mechanisms must exist, which regulate synaptic plasticity on a global network level to maintain stability of synapses and maintain specificity of neural activity (16, 17).

Metaplasticity refers to any change in the direction or degree of synaptic plasticity (ex. LTP, LTD) based on prior neural activity (18). While both synaptic and metaplasticity are dependent on previous neural activity, metaplasticity does not *directly* alter the efficacy of synaptic transmission

OPEN ACCESS

Edited by:

Mariagiovanna Cantone,
Sant'Elia Hospital, Italy

Reviewed by:

Paolo Maria Rossini,
Catholic University of the Sacred
Heart, Italy
Wickliffe C. Abraham,
University of Otago, New Zealand

*Correspondence:

Alexander T. Sack
a.sack@maastrichtuniversity.nl

Specialty section:

This article was submitted to
Neurorehabilitation,
a section of the journal
Frontiers in Neurology

Received: 28 August 2020

Accepted: 22 October 2020

Published: 05 November 2020

Citation:

Thomson AC and Sack AT (2020)
How to Design Optimal Accelerated
rTMS Protocols Capable of Promoting
Therapeutically Beneficial
Metaplasticity.
Front. Neurol. 11:599918.
doi: 10.3389/fneur.2020.599918

(as LTP/LTD), but it adjusts the neurons' ability to induce LTP/LTD with *subsequent* neural activity. Metaplasticity in some sense can be considered as the plasticity of synaptic plasticity, e.g., maintaining the dynamic nature of a neuron's firing threshold, when this neuron reaches a certain firing rate (16, 18, 19). Metaplasticity works through similar synaptic modifications as LTP/LTD, such as NMDA receptor activation and modification (20), and changes in calcium signaling triggering complex signaling cascades (18). Metaplastic modifications, for example at NMDA receptors, can occur either at specific synapses or across the whole neuron, and on time scales from minutes to weeks (19). Depending on the temporal pattern and strength of previous neural activity, metaplastic mechanisms can be additive; for example promoting increased synaptic strengthening through repeated excitatory (LTP-inducing) stimulation. Metaplasticity can also be stabilizing; for example acting against subsequent synaptic strengthening when repeating excitatory (LTP-inducing) stimulation (19, 21). This stabilizing form of metaplasticity is often referred to as *homeostatic* metaplasticity, as it specifically regulates the dynamic threshold of synaptic plasticity to maintain equilibrium, or homeostasis (16, 17). We hypothesize, based on research from human and animal studies, that the timing between excitatory stimulations are what differentiate between promoting additive or homeostatic metaplasticity.

We focus on the role of metaplasticity in Transcranial Magnetic Stimulation (TMS). We describe the recent use of accelerated (repeated) stimulation protocols, both in research and clinical applications, and the molecular mechanisms required to promote either homeostatic or additive metaplastic effects. Finally, we showcase the therapeutic potential of accelerated stimulation, and hypothesize that increasing the currently practiced stimulation intervals may be more efficacious in promoting additive metaplastic effects in various clinical applications of rTMS in rehabilitation, neurology, psychiatry, and cognitive decline.

METAPLASTICITY IN TMS

TMS is a widespread and increasingly popular non-invasive brain stimulation technique, where electromagnetic pulses allow stimulation to pass non-invasively through the skull (22). When pulses are applied in a certain pattern, as repetitive TMS (rTMS), protocols can have lasting excitatory or inhibitory effects (23–25). Two commonly used stimulation protocols are *intermittent* Theta Burst Stimulation (iTBS), requiring only 3 min of stimulation time, resulting in a lasting increase of cortical excitability, and *continuous* Theta Burst Stimulation (cTBS), requiring only 40 s of stimulation for a lasting decrease in cortical excitability (26). The after effects of these protocols have been shown for up to 1 h following stimulation (26, 27).

While iTBS is normally an excitatory protocol, causing an increase in cortical excitability of the stimulated brain region, it has been shown that when applied twice in quick succession iTBS effects switch from excitatory to inhibitory (28). Conversely,

when cTBS (an inhibitory protocol) is applied for double the normal duration, its effects switch from inhibitory to excitatory (28). Several studies have reported similar effects of repeating iTBS or cTBS stimulation protocols, with the timing between protocols being an important factor in the magnitude and direction of aftereffects (19, 29, 30). For example, using a “priming” iTBS protocol which does not induce plasticity, followed by a “test” iTBS protocol has shown that short intervals of 5 min between priming and test resulted in homeostatic-like changes in excitability, i.e., an opposite effect. Interestingly, longer breaks of 15 min resulted in an increase in MEP amplitude after the test iTBS (30). However, 15 min between priming and test iTBS/cTBS has also been shown to induce in homeostatic-like metaplastic effects (29). While the timing between repeated TBS sessions is clearly important, the optimal interval is less clear. 15 min between iTBS sessions has been shown to promote both homeostatic (29) and MEP enhancement after the second iTBS (30), while 10 min between priming and test iTBS has shown enhancement of MEP amplitude (31), but 5 and 20 min between iTBS sessions did not (32). Therefore, when 2 iTBS sessions are repeated with short (<30 min) between, conflicting effects on MEP amplitude have been reported.

“Accelerated” protocols, which consist of multiple stimulation sessions on a single day, have recently been introduced for the treatment of depression (33–37). Due to their short duration, the TBS protocols, in particular iTBS, have been promising candidates for accelerated protocols (38). Also, a large trial recently found that iTBS was not-inferior to the classical 10 Hz rTMS protocol, confirming the clinical potential of this shorter stimulation protocol to treat depression (39). Indeed, several studies have shown additional benefits for accelerated iTBS protocols in the treatment of severe, treatment resistant depression (40, 41). In the clinic, an interval of 15 min is often used between iTBS sessions, with these sessions repeated up to 5 times on a single treatment day (40, 41).

We recently conducted a study investigating the effects of accelerated iTBS over motor cortex, consisting of 5 repeated iTBS sessions in a single day. iTBS with 8- or 15-min time interval between sessions were delivered to healthy participants in a fully within subject design; where participants received 4 different conditions (accelerated iTBS with 8-min intervals, accelerated iTBS with 15-min intervals, single iTBS and sham) (42). We compared change in Motor Evoked Potential (MEP) amplitude up to 90 min following stimulation, across the stimulation conditions.

We found that there was no difference in the effects of accelerated iTBS on MEP amplitude, also when compared to sham stimulation, and thus no additive metaplasticity induced by five stimulation sessions applied successively in 8- or 15-min intervals. We argue that such intervals between iTBS protocols are likely too short to avoid processes of homeostatic plasticity. With only 8 or 15 min between sessions, homeostatic mechanisms may be working against additive metaplastic effects to maintain network stability and therefore result in a net effect of no change in excitability following these accelerated protocols (42).

TIMING-DEPENDENT METAPLASTICITY

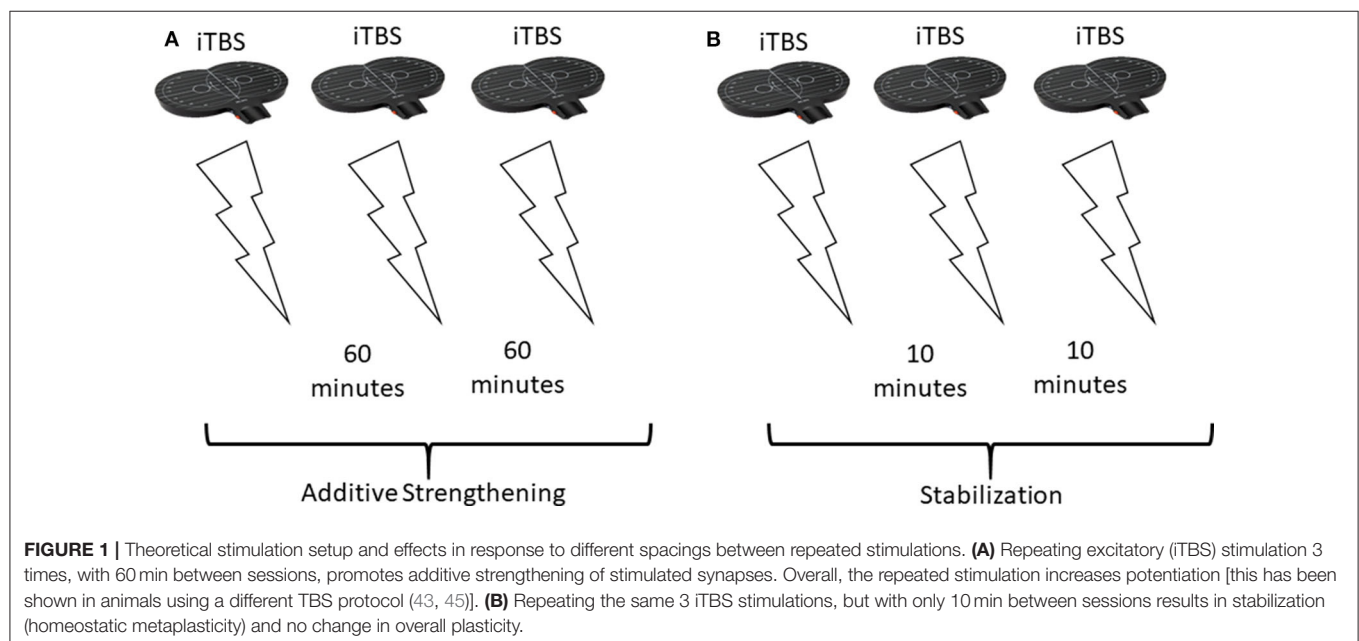
In agreement with this notion, animal studies in rats, and rat hippocampal slices have shown that a sufficiently long pause between excitatory stimulation sessions was necessary for additive (LTP) plasticity effects to occur (43–45). This may have to do with the time required for metaplasticity mechanisms, for example synapse strengthening with AMPA receptor trafficking (15).

It has been well-established in animal studies, that a single round of TBS (a 4-pulse burst at 100 Hz, repeated at 5 Hz for 10 bursts) is effective at inducing LTP in CA1 hippocampal pyramidal neurons (46, 47). TBS has since then been used extensively to reliably induce LTP *in vitro* (48). Interestingly, repeating this single TBS protocol with a time interval of >40 min, was capable of almost doubling the potentiation compared to the first TBS alone (43). This additional potentiation is thought to work through strengthening the smaller synapses which weren't strengthened by the first TBS protocol (43). This may have to do with the number of AMPA receptors; smaller synapses contain fewer AMPA receptors and therefore don't generate a response to trigger a depolarization following a single TBS (43). Several other studies have provided evidence for increased potentiation by spaced TBS, however the magnitude and duration of the effects depended on a series of factors such as rat strain, rat age, and the time interval. In adult Wistar rats, adult Long-Evans (LE) rats, and young LE rats, 4 h was required between TBS to induce additional potentiation (44, 45). However, in young Sprague Dawley (SD) rats, a single TBS repeated at 1-h intervals could induce further potentiation, following up to 3 repeated TBS stimulations (4 did not produce additional potentiation) (43, 45). These different studies used different stimulation intensities; Frey

et al. (44) found that reducing stimulation intensity in the second stimulation was effective for promoting potentiation 4-h later, while Cao and Harris (45) and Kramár et al. (43) kept stimulation intensities constant. However, these studies consistently show that additional potentiation following repeated TBS in animal slices is possible. Enhanced, additive LTP-like plasticity may be promoted when repeating TBS with 50–60 min between sessions (43, 45). After 3 TBS protocols, spaced 60 min apart, potentiation had been raised to 150% baseline, which is about three times higher than if just one protocol was given (43, 48). This suggests that 3 TBS protocols repeated at 60 min-intervals may be effective at promoting maximal, additive metaplasticity effects (**Figure 1A**). If there is less time between TBS protocols, for example 10 min, homeostatic metaplasticity mechanisms may dominate, promoting a stabilizing rather than additive plasticity response (**Figure 1B**).

DISCUSSION

Activity-dependent metaplasticity is considered to be *homeostatic* if the first stimulation protocol alters the threshold for subsequent LTP/LTD in the opposite direction, thereby stabilizing (network) brain activity (49). Interestingly, this reversal of aftereffects has been shown specifically when stimulation protocols were given with a short (0–5 min) interval (28, 30), providing support for homeostatic metaplasticity mechanisms in rTMS protocols (19). While homeostatic metaplasticity mechanisms are important for stabilizing network activity, they can be counteractive when promoting plasticity effects through rTMS. In fact, when applying rTMS protocols, the explicit goal is not stabilization but promotion of additive, increased plasticity effects.



Animal studies have shown that timing is important in the molecular mechanisms underlying metaplasticity. While there is overlap between the mechanisms of additive and homeostatic metaplasticity, there are temporal differences which may differentiate between both principles at the molecular level. Based on evidence from animal models, leaving 60 min between excitatory stimulation protocols may promote additive rather than homeostatic metaplastic effects in accelerated TMS treatment protocols.

Clinical Implications

If longer intervals between iTBS sessions are capable of promoting additive metaplasticity, as has been shown in animal studies (43) as well as improving clinical outcomes in the treatment of depression (50), longer spaced intervals between iTBS sessions will likely be beneficial for other therapeutic applications of iTBS. iTBS is increasingly being used as a treatment in a range of clinical applications such as rehabilitation, as well as neurological and psychiatric disorders. For example, to promote motor recovery after stroke (51), for managing spasticity associated with Multiple Sclerosis (MS) (52), and decreasing obsessive symptomatology associated with Obsessive Compulsive Disorder (OCD) (53), just to name a few. These protocols all must adhere to the established safety guidelines (54), and recommendations for clinical TMS use (55, 56). These include total pulse number, interval between TBS session, intensity of stimulation, and cumulative weekly applications (54). Accelerated iTBS has been successfully and safely used in the treatment of depression (38, 40, 41), with patients receiving a total of 32,400 pulses at 110% resting motor threshold, over 20 sessions (5 sessions per day, 15 min between sessions) in 4 days (41). Therefore, while following the established safety guidelines is the upmost priority, and local health authorities should always approve each stimulation protocol (54), delivering three iTBS sessions on a single day with 1 h between sessions should theoretically be safe and tolerable for most patients.

rTMS is also used as a treatment for the cognitive decline associated with neurodegenerative disorders such as dementia, and Alzheimer's Disease (AD) (57–61). However, there are ethical implications of using rTMS for cognitive enhancement, in particular in healthy participants (62). It is important to maintain the consensus ethical requirements that (1) participants/patients provide informed consent, (2) the benefit of the research outweigh the risks, and (3) there is equal distribution of burdens and benefits across patients (this is violated if a particular group of patients with different economic, physical or social conditions) (54).

Importantly, the here described principles of additive and homeostatic metaplasticity not only apply to the here discussed accelerated TMS treatments and the question of optimal time interval between its repeated stimulation sessions, but likewise can be used to explain and optimize other forms of plasticity-inducing TMS protocols such as Paired Associated-Stimulation (PAS) or paired-coil TMS (pcTMS).

In humans, neural excitability and synaptic plasticity can be probed by TMS to peripheral nerves and motor cortex (63, 64).

In such a transcortical loop, timings of afferent (muscle/nerve to brain), cortical, and efferent (brain to muscle) responses can be used to quantify central motor excitability (63). For example, delivering a conditioning TMS pulse to an afferent tract (ex. the wrist), followed (10–48 ms) by stimulation of the efferent tract (motor cortex), will alter Motor Evoked Potentials (MEPs) measured from thumb flexor muscles (63). It has been shown that wrist stimulation 20–22 msec preceding motor cortex stimulation elicits a facilitated MEP, with a latency of about 1 ms, compared to MEPs given without the conditioning wrist stimulation (63). Repeating this afferent (wrist) efferent (motor cortex) stimulation, in Paired Associated Stimulation (PAS), can induce lasting effects on motor cortex excitability (64, 65), providing evidence for synaptic plasticity. Interestingly, evidence of homeostatic and additive metaplastic responses have also been recorded using PAS stimulation (66, 67). When two LTP-inducing PAS protocols were separated by 30 min, a decrease in MEP amplitude was measured, indicating a homeostatic (stabilizing) metaplastic responses (66). Similarly, LTD-inducing PAS immediately preceding a motor-learning task facilitated motor-learning (67), again providing support for homeostatic plasticity mechanisms dominating at early time points following stimulation.

Additionally, the effects of brain stimulation are not only localized to the site of stimulation, but can also spread to different areas through complex cortical networks. Similarly to PAS, this has been shown using paired-coil TMS (pcTMS), where multiple coils are used to probe different cortical areas and assess connectivity (68, 69). For example, a single TMS pulse to motor cortex can cause a depression of the MEP measured following a subsequent (6–30 ms) TMS pulse to contralateral motor cortex (70). Therefore, TMS can also be used to assess connectivity between brain areas (68). In other words, TMS stimulation can propagate to different cortical regions, having both local and remote effects on (meta) plasticity. This has valuable clinical implications, where inducing plasticity effects in a cortical network are important (69). In stroke patients for example, localized damage can disrupt connectivity and can have functional consequences (69), therefore stimulation effects should promote network plasticity, rather than localized plasticity. Similarly, in the treatment of depression, superficial stimulation uses cortical connectivity to influence deeper cortical structures, resulting in improvement of clinical symptoms (71, 72). Therefore, it is important to use TMS to strengthen connectivity, and to promote additive, metaplastic changes also on the network activity level.

With the increasing and widespread application of rTMS protocols in the clinic, it is important to optimize protocols to maximize their effects, while remaining within established safety and ethical guidelines for use in the clinic (54, 56). Single iTBS has proven promising, but accelerated iTBS at longer time intervals (60 min) between sessions could maximize clinical outcomes through additive metaplasticity, preventing homeostatic metaplasticity from stabilizing stimulation effects. Clinical efficacy of PAS and pcTMS protocols may be similarly increased by optimizing the timing between stimulations according to these principles of metaplasticity.

AUTHOR CONTRIBUTIONS

AS: conceptualization, writing-review and editing, supervision, and funding acquisition. AT: investigation, writing-original draft preparation, and visualization. All authors contributed to the article and approved the submitted version.

REFERENCES

- Hebb DO. *The Organization of Behaviour: A Neurophysiological Theory*. New York, NY: Wiley. (1949).
- Stent GS. A Physiological mechanism for hebb's postulate of learning. *Proc Natl Acad Sci USA*. (1973) 70:997–1001. doi: 10.1073/pnas.70.4.997
- Turrigiano GG, Leslie KR, Desai NS, Rutherford LC, Nelson SB. Activity-dependent scaling of quantal amplitude in neocortical neurons. *Nature*. (1998) 391:892–6. doi: 10.1038/36103
- Madison DV, Malenka RC, Nicoll RA. Mechanisms underlying long-term potentiation of synaptic transmission. *Annu Rev Neurosci*. (1991) 14:379–97. doi: 10.1146/annurev.ne.14.030191.002115
- Bliss TV, Lomo T. Long-lasting potentiation of synaptic transmission in the dentate area of the anaesthetized rabbit following stimulation of the perforant path. *J Physiol*. (1973) 232:331–56. doi: 10.1113/jphysiol.1973.sp010273
- Linden DJ, Connor JA. Long-term synaptic depression. *Annu Rev Neurosci*. (1995) 18:319–57. doi: 10.1146/annurev.ne.18.030195.001535
- Ito M, Kano M. Long-lasting depression of parallel fiber-Purkinje cell transmission induced by conjunctive stimulation of parallel fibers and climbing fibers in the cerebellar cortex. *Neurosci Lett*. (1982) 33:253–8. doi: 10.1016/0304-3940(82)90380-9
- Dudek SM, Bear MF. Homosynaptic long-term depression in area CA1 of hippocampus and effects of N-methyl-D-aspartate receptor blockade. *Proc Natl Acad Sci USA*. (1992) 89:4363–7. doi: 10.1073/pnas.89.10.4363
- Bliss TV, Collingridge GL. A synaptic model of memory: long-term potentiation in the hippocampus. *Nature*. (1993) 361:31–9. doi: 10.1038/361031a0
- Dudek SM, Bear MF. Bidirectional long-term modification of synaptic effectiveness in the adult and immature hippocampus. *J Neurosci*. (1993) 13:2910–8. doi: 10.1523/JNEUROSCI.13-07-02910.1993
- Malenka RC, Bear MF. LTP and LTD: an embarrassment of riches. *Neuron*. (2004) 44:5–21. doi: 10.1016/j.neuron.2004.09.012
- Malenka RC, Nicoll RA. Long-term potentiation—a decade of progress? *Science*. (1999) 285:1870–4. doi: 10.1126/science.285.5435.1870
- Huang YY, Colino A, Selig DK, Malenka RC. The influence of prior synaptic activity on the induction of long-term potentiation. *Science*. (1992) 255:730–3. doi: 10.1126/science.1346729
- Abbott LF, Nelson SB. Synaptic plasticity: taming the beast. *Nat Neurosci*. (2000) 3(Suppl:11):78–83. doi: 10.1038/81453
- Turrigiano GG. The self-tuning neuron: synaptic scaling of excitatory synapses. *Cell*. (2008) 135:422–35. doi: 10.1016/j.cell.2008.10.008
- Bienenstock EL, Cooper LN, Munro PW. Theory for the development of neuron selectivity: orientation specificity and binocular interaction in visual cortex. *J Neurosci*. (1982) 2:32–48. doi: 10.1523/JNEUROSCI.02-01-00032.1982
- Turrigiano GG, Nelson SB. Homeostatic plasticity in the developing nervous system. *Nat Rev Neurosci*. (2004) 5:97–107. doi: 10.1038/nrn1327
- Abraham WC, Bear MF. Metaplasticity: the plasticity of synaptic plasticity. *Trends Neurosci*. (1996) 19:126–30. doi: 10.1016/S0166-2236(96)80018-X
- Muller-Dahlhaus F, Ziemann U. Metaplasticity in human cortex. *Neuroscientist*. (2015) 21:185–202. doi: 10.1177/1073858414526645
- Philpot BD, Cho KK, Bear MF. Obligatory role of NR2A for metaplasticity in visual cortex. *Neuron*. (2007) 53:495–502. doi: 10.1016/j.neuron.2007.01.027
- Li J, Park E, Zhong LR, Chen L. Homeostatic synaptic plasticity as a metaplasticity mechanism - a molecular and cellular perspective. *Curr Opin Neurobiol*. (2019) 54:44–53. doi: 10.1016/j.conb.2018.08.010
- Barker AT, Jalinous R, Freeston IL. Non-invasive magnetic stimulation of human motor cortex. *Lancet*. (1985) 1:1106–7. doi: 10.1016/S0140-6736(85)92413-4
- Pascual-Leone A, Valls-Sole J, Wassermann EM, Hallett M. Responses to rapid-rate transcranial magnetic stimulation of the human motor cortex. *Brain: a J Neurol*. (1994) 117(Pt 4):847–58. doi: 10.1093/brain/117.4.847
- George MS, Wassermann EM, Williams WA, Callahan A, Ketter TA, Basser P, et al. Daily repetitive transcranial magnetic stimulation. (rTMS) improves mood in depression. *Neuroreport*. (1995) 6:1853–6. doi: 10.1097/00001756-199510020-00008
- Hallett M. Transcranial magnetic stimulation: a primer. *Neuron*. (2007) 55:187–99. doi: 10.1016/j.neuron.2007.06.026
- Huang YZ, Edwards MJ, Rounis E, Bhatia KP, Rothwell JC. Theta burst stimulation of the human motor cortex. *Neuron*. (2005) 45:201–6. doi: 10.1016/j.neuron.2004.12.033
- Suppa A, Huang YZ, Funke K, Ridding MC, Cheeran B, Di Lazzaro V, et al. Ten years of theta burst stimulation in humans: established knowledge, unknowns and prospects. *Brain Stimul*. (2016) 9:323–35. doi: 10.1016/j.brs.2016.01.006
- Gamboa OL, Antal A, Moliadze V, Paulus W. Simply longer is not better: reversal of theta burst after-effect with prolonged stimulation. *Exp Brain Res*. (2010) 204:181–7. doi: 10.1007/s00221-010-2293-4
- Murakami T, Muller-Dahlhaus F, Lu MK, Ziemann U. Homeostatic metaplasticity of corticospinal excitatory and intracortical inhibitory neural circuits in human motor cortex. *J Physiol*. (2012) 590:5765–81. doi: 10.1113/jphysiol.2012.238519
- Tse NY, Goldsworthy MR, Ridding MC, Coxon JP, Fitzgerald PB, Fornito A, et al. The effect of stimulation interval on plasticity following repeated blocks of intermittent theta burst stimulation. *Sci Rep*. (2018) 8:8526. doi: 10.1038/s41598-018-26791-w
- Opie GM, Vosnakis E, Ridding MC, Ziemann U, Semmler JG. Priming theta burst stimulation enhances motor cortex plasticity in young but not old adults. *Brain Stimulation*. (2017) 10:298–304. doi: 10.1016/j.brs.2017.01.003
- Gamboa OL, Antal A, Laczó B, Moliadze V, Nitsche MA, Paulus W. Impact of repetitive theta burst stimulation on motor cortex excitability. *Brain Stimul*. (2011) 4:145–51. doi: 10.1016/j.brs.2010.09.008
- Baeken C, Marinazzo D, Wu GR, Van Schuerbeek P, De Mey J, Marchetti I, et al. Accelerated HF-rTMS in treatment-resistant unipolar depression: insights from subgenual anterior cingulate functional connectivity. *World J Biol Psychiatry*. (2014) 15:286–97. doi: 10.3109/15622975.2013.872295
- Herremans SC, Van Schuerbeek P, De Raedt R, Matthys F, Buyl R, De Mey J, et al. The impact of accelerated right prefrontal high-frequency repetitive transcranial magnetic stimulation (rTMS) on cue-reactivity: an fMRI study on craving in recently detoxified alcohol-dependent patients. *PLoS ONE*. (2015) 10:e0136182. doi: 10.1371/journal.pone.0136182
- Baeken C, Marinazzo D, Everaert H, Wu GR, Van Hove C, Audenaert K, et al. The impact of accelerated HF-rTMS on the subgenual anterior cingulate cortex in refractory unipolar major depression: insights from 18FDG PET brain imaging. *Brain Stimul*. (2015) 8:808–15. doi: 10.1016/j.brs.2015.01.415
- McGirr A, Van den Eynde F, Tovar-Perdomo S, Fleck MP, Berlim MT. Effectiveness and acceptability of accelerated repetitive transcranial magnetic stimulation. (rTMS) for treatment-resistant major depressive disorder: an open label trial. *J Affect Disord*. (2015) 173:216–20. doi: 10.1016/j.jad.2014.10.068
- Herremans SC, De Raedt R, Van Schuerbeek P, Marinazzo D, Matthys F, De Mey J, et al. Accelerated HF-rTMS protocol has a rate-dependent effect

FUNDING

This work was supported by the Netherlands Organization for Scientific Research (NWO, Vici to A.T.S 453-15-008), and an internal grant from the Centre for Integrative Neuroscience at Maastricht University.

- on dACC activation in alcohol-dependent patients: an open-label feasibility study. *Alcohol Clin Exp Res.* (2016) 40:196–205. doi: 10.1111/acer.12937
38. Baeken C. Accelerated rTMS: a potential treatment to alleviate refractory depression. *Front Psychol.* (2018) 9:2017. doi: 10.3389/fpsyg.2018.02017
 39. Blumberger DM, Vila-Rodriguez F, Thorpe KE, Feffer K, Noda Y, Giacobbe P, et al. Effectiveness of theta burst versus high-frequency repetitive transcranial magnetic stimulation in patients with depression. (THREE-D): a randomised non-inferiority trial. *Lancet.* (2018) 391:1683–92. doi: 10.1016/S0140-6736(18)30295-2
 40. Desmyter S, Duprat R, Baeken C, Van Autreuve S, Audenaert K, van Heeringen K. Accelerated intermittent theta burst stimulation for suicide risk in therapy-resistant depressed patients: a randomized, sham-controlled trial. *Front Human Neurosci.* (2016) 10:480. doi: 10.3389/fnhum.2016.00480
 41. Duprat R, Desmyter S, Rudi DR, van Heeringen K, Van den Abbeele D, Tandt H, et al. Accelerated intermittent theta burst stimulation treatment in medication-resistant major depression: a fast road to remission? *J Affective Disord.* (2016) 200:6–14. doi: 10.1016/j.jad.2016.04.015
 42. Thomson AC, de Graaf TA, Kenis G, Rutten BPF, Schuhmann T, Sack AT. No additive meta plasticity effects of accelerated iTBS with short inter-session intervals. *Brain Stimul.* (2019) 12:1301–3. doi: 10.1016/j.brs.2019.05.012
 43. Kramár EA, Babayan AH, Gavin CF, Cox CD, Jafari M, Gall CM, et al. Synaptic evidence for the efficacy of spaced learning. *Proc Natl Acad Sci USA.* (2012) 109:5121–6. doi: 10.1073/pnas.1120700109
 44. Frey U, Schollmeier K, Reymann KG, Seidenbecher T. Asymptotic hippocampal long-term potentiation in rats does not preclude additional potentiation at later phases. *Neuroscience.* (1995) 67:799–807. doi: 10.1016/0306-4522(95)00117-2
 45. Cao G, Harris KM. Augmenting saturated LTP by broadly spaced episodes of theta-burst stimulation in hippocampal area CA1 of adult rats and mice. *J Neurophysiol.* (2014) 112:1916–24. doi: 10.1152/jn.00297.2014
 46. Larson J, Lynch G. Induction of synaptic potentiation in hippocampus by patterned stimulation involves two events. *Science.* (1986) 232:985–8. doi: 10.1126/science.3704635
 47. Larson J, Munkácsy E. Theta-burst LTP. *Brain Res.* (2015) 1621:38–50. doi: 10.1016/j.brainres.2014.10.034
 48. Lynch G, Kramar EA, Babayan AH, Rumbaugh G, Gall CM. Differences between synaptic plasticity thresholds result in new timing rules for maximizing long-term potentiation. *Neuropharmacology.* (2013) 64:27–36. doi: 10.1016/j.neuropharm.2012.07.006
 49. Karabanov A, Ziemann U, Hamada M, George MS, Quartarone A, Classen J, et al. Consensus paper: probing homeostatic plasticity of human cortex with non-invasive transcranial brain stimulation. *Brain Stimul.* (2015) 8:442–54. doi: 10.1016/j.brs.2015.01.404
 50. Williams NR, Sudheimer KD, Bentzley BS, Pannu J, Stimpson KH, Duvio D, et al. High-dose spaced theta-burst TMS as a rapid-acting antidepressant in highly refractory depression. *Brain.* (2018) 141:e18–e. doi: 10.1093/brain/awx379
 51. Hensel L, Grefkes C, Tschepel C, Ringmaier C, Kraus D, Hamacher S, et al. Intermittent theta burst stimulation applied during early rehabilitation after stroke: study protocol for a randomised controlled trial. *BMJ Open.* (2019) 9:e034088. doi: 10.1136/bmjopen-2019-034088
 52. Korzhova J, Bakulin I, Sinitsyn D, Poydasheva A, Suponeva N, Zakharova M, et al. High-frequency repetitive transcranial magnetic stimulation and intermittent theta-burst stimulation for spasticity management in secondary progressive multiple sclerosis. *Eur J Neurol.* (2019) 26:680–e44. doi: 10.1111/ene.13877
 53. Naro A, Billeri L, Cannavo A, De Luca R, Portaro S, Bramanti P, et al. Theta burst stimulation for the treatment of obsessive-compulsive disorder: a pilot study. *J Neural Transm.* (2019) 126:1667–77. doi: 10.1007/s00702-019-02098-6
 54. Rossi S, Hallett M, Rossini PM, Pascual-Leone A. Safety, ethical considerations, and application guidelines for the use of transcranial magnetic stimulation in clinical practice and research. *Clin Neurophysiol.* (2009) 120:2008–39. doi: 10.1016/j.clinph.2009.08.016
 55. Lefaucheur JP, Andre-Obadia N, Antal A, Ayache SS, Baeken C, Benninger DH, et al. Evidence-based guidelines on the therapeutic use of repetitive transcranial magnetic stimulation. (rTMS). *Clin Neurophysiol.* (2014) 125:2150–206. doi: 10.1016/j.clinph.2014.05.021
 56. Lefaucheur J-P, Aleman A, Baeken C, Benninger DH, Brunelin J, Di Lazzaro V, et al. Evidence-based guidelines on the therapeutic use of repetitive transcranial magnetic stimulation. (rTMS): an update. (2014–2018). *Clin Neurophysiol.* (2020) 131:474–528. doi: 10.1016/j.clinph.2020.02.003
 57. Weiler M, Stieger KC, Long JM, Rapp PR. Transcranial magnetic stimulation in alzheimer's disease: are we ready? *eNeuro.* (2020) 7:ENEURO.0235-19.2019. doi: 10.1523/ENEURO.0235-19.2019
 58. Cotelli M, Calabria M, Manenti R, Rosini S, Zanetti O, Cappa SF, et al. Improved language performance in Alzheimer disease following brain stimulation. *J Neurol Neurosurg Psychiatry.* (2011) 82:794–7. doi: 10.1136/jnnp.2009.197848
 59. Del Giacco L, Pistocchi A, Cotelli F, Fortunato AE, Sordino P. A peek inside the neurosecretory brain through Orthopedia lenses. *Dev Dyn.* (2008) 237:2295–303. doi: 10.1002/dvdy.21668
 60. Cotelli M, Manenti R, Cappa SF, Geroldi C, Zanetti O, Rossini PM, et al. Effect of transcranial magnetic stimulation on action naming in patients with Alzheimer disease. *Arch Neurol.* (2006) 63:1602–4. doi: 10.1001/archneur.63.11.1602
 61. Rutherford G, Gole R, Moussavi Z. rTMS as a treatment of alzheimer's disease with and without comorbidity of depression: a review. *Neurosci J.* (2013) 2013:679389. doi: 10.1155/2013/679389
 62. Kim TD, Hong G, Kim J, Yoon S. Cognitive enhancement in neurological and psychiatric disorders using transcranial magnetic stimulation (TMS): a review of modalities, potential mechanisms and future implications. *Exp Neurobiol.* (2019) 28:1–16. doi: 10.5607/en.2019.28.1.1
 63. Mariorenzi R, Zarola F, Caramia MD, Paradiso C, Rossini PM. Non-invasive evaluation of central motor tract excitability changes following peripheral nerve stimulation in healthy humans. *Electroencephalogr Clin Neurophysiol.* (1991) 81:90–101. doi: 10.1016/0168-5597(91)90002-F
 64. Stefan K, Kunesch E, Cohen LG, Benecke R, Classen J. Induction of plasticity in the human motor cortex by paired associative stimulation. *Brain.* (2000) 123(Pt 3):572–84. doi: 10.1093/brain/123.3.572
 65. Ziemann U. LTP-like plasticity in human motor cortex. *Suppl Clin Neurophysiol.* (2004) 57:702–7. doi: 10.1016/S1567-424X(09)70410-6
 66. Muller JF, Orekhov Y, Liu Y, Ziemann U. Homeostatic plasticity in human motor cortex demonstrated by two consecutive sessions of paired associative stimulation. *Eur J Neurosci.* (2007) 25:3461–8. doi: 10.1111/j.1460-9568.2007.05603.x
 67. Jung P, Ziemann U. Homeostatic and nonhomeostatic modulation of learning in human motor cortex. *J Neurosci.* (2009) 29:5597–604. doi: 10.1523/JNEUROSCI.0222-09.2009
 68. Rossini PM, Di Iorio R, Bentivoglio M, Bertini G, Ferreri F, Gerloff C, et al. Methods for analysis of brain connectivity: An IFCN-sponsored review. *Clin Neurophysiol.* (2019) 130:1833–58. doi: 10.1016/j.clinph.2019.06.006
 69. Hallett M, de Haan W, Deco G, Dengler R, Di Iorio R, Gallea C, et al. Human brain connectivity: Clinical applications for clinical neurophysiology. *Clin Neurophysiol.* (2020) 131:1621–51. doi: 10.1016/j.clinph.2020.03.031
 70. Ferbert A, Priori A, Rothwell JC, Day BL, Colebatch JG, Marsden CD. Interhemispheric inhibition of the human motor cortex. *J Physiol.* (1992) 453:525–46. doi: 10.1113/jphysiol.1992.sp019243
 71. Fox MD, Buckner RL, White MP, Greicius MD, Pascual-Leone A. Efficacy of TMS targets for depression is related to intrinsic functional connectivity with the subgenual cingulate. *Biol Psychiatry.* (2012) 72:595–603. doi: 10.1016/j.biopsych.2012.04.028
 72. Liston C, Chen AC, Zebley BD, Drysdale AT, Gordon R, Leuchter B, et al. Default mode network mechanisms of transcranial magnetic stimulation in depression. *Biol Psychiatry.* (2014) 76:517–26. doi: 10.1016/j.biopsych.2014.01.023

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2020 Thomson and Sack. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.



Transcranial Direct Current Stimulation to Enhance Cognitive Impairment in Parkinson's Disease: A Systematic Review and Meta-Analysis

Diana M. A. Suarez-García¹, Johan S. Grisales-Cárdenas¹, Máximo Zimmerman² and Juan F. Cardona^{1*}

¹ Instituto de Psicología, Universidad del Valle, Santiago de Cali, Colombia, ² Institute of Cognitive and Translational Neuroscience (INCYT), INECO Foundation, Favaloro University, Buenos Aires, Argentina

OPEN ACCESS

Edited by:

Mariagiovanna Cantone,
Sant'Elia Hospital, Italy

Reviewed by:

Marco Iosa,
Santa Lucia Foundation (IRCCS), Italy
Antonino Naro,
Centro Neurolesi Bonino Pulejo
(IRCCS), Italy

*Correspondence:

Juan F. Cardona
felipe.cardona@correounivalle.edu.co

Specialty section:

This article was submitted to
Neurorehabilitation,
a section of the journal
Frontiers in Neurology

Received: 23 August 2020

Accepted: 09 November 2020

Published: 30 November 2020

Citation:

Suarez-García DMA,
Grisales-Cárdenas JS, Zimmerman M
and Cardona JF (2020) Transcranial
Direct Current Stimulation to Enhance
Cognitive Impairment in Parkinson's
Disease: A Systematic Review and
Meta-Analysis.
Front. Neurol. 11:597955.
doi: 10.3389/fneur.2020.597955

Cognitive deficits are increasingly being recognized as a common trait in Parkinson's disease (PD). Recently, transcranial direct current stimulation (tDCS) has been shown to exert positive effects as an adjunctive therapy on motor and non-motor symptoms in PD. This systematic review and meta-analysis aims to provide an overview of reported evidence on the efficacy of tDCS interventions in the treatment of cognitive impairments in PD. A systematic literature review was conducted to examine articles that were published in the past 10 years and that study the effects of tDCS on cognitive deficits in PD patients. The PubMed, Scopus and Scielo databases were searched. Eight tDCS studies involving 168 participants were included for the analysis. Our meta-analysis results showed that anodal tDCS (atDCS) had various levels or no evidence of effectiveness. In the pre-post stimulation analysis, a strong effect was reported for executive functions (pre-post: $g = 1.51$, $Z = 2.41$, $p = 0.016$); non-significant effects were reported for visuospatial skills (pre-post: $g = 0.27$, $Z = 0.69$, $p = 0.490$); attention (pre-post: $g = 0.02$, $Z = 0.08$, $p = 0.934$), memory (pre-post: $g = 0.01$, $Z = 0.03$, $p = 0.972$) and language (pre-post: $g = 0.07$, $Z = 0.21$, $p = 0.832$). However, in the pre-follow-up stimulation analysis, the duration of the effect was not clear. This study highlights the potential effectiveness of atDCS to improve cognitive performance in PD patients but failed to establish a cause-effect relationship between tDCS intervention and cognitive improvement in PD. Future directions and recommendations for methodological improvements are outlined.

Keywords: Parkinson's disease, transcranial electric stimulation, neuroplasticity, executive functions, cognition

INTRODUCTION

There is growing interest in the potential efficacy of transcranial direct current stimulation (tDCS) for treating neurodegenerative conditions such as Parkinson's disease (PD). Previous systematic reviews on PD have supported the efficacy of tDCS for improving motor functions, including balance, gait, and bradykinesia (1–5). However, evidence is not clear regarding its efficacy for PD patients' cognitive symptoms.

Cognitive impairment is frequent in PD, though it can be heterogeneous in its presentation and progression, as it varies regarding clinical features, severity, and progression to dementia. It has been suggested that interventions for cognitive symptoms may be essential in preventing and delaying the onset of cognitive decline and Parkinson's disease dementia (PDD) (6, 7). Approximately 25% of PD patients have mild cognitive impairment (MCI) and an increased risk of developing PDD (8). Most commonly, reported cognitive disorders in PD include executive deficits (9), visuospatial impairments (10), memory deficits (11), action verb, and action conceptualization impairments (12, 13). These can be progressive and make patients more vulnerable to the onset of affective symptoms, behavioral disorders, and other neuropsychiatric symptoms (14).

tDCS is a non-invasive brain stimulation technique modulating cortical activity that acts by inducing a low-frequency electric current (15), usually between 1 and 2 milliamperes (mA), to activate the potential of the resting neuronal membrane (16, 17). The current transmission modifies the membrane's polarity (18), producing a facilitating effect when the positive electric current (anodal) is administered or hyperpolarization when the negative electric current (cathodal) is administered (19).

Given the increasing use of tDCS in neurodegenerative diseases such as PD, the present study aimed to systematically review and analyze studies evaluating the effects of tDCS on PD patients' cognitive alterations.

MATERIALS AND METHODS

A systematic literature search was conducted for articles on the effect of tDCS interventions on PD patients' cognitive symptoms. PubMed, Scopus, and Scielo databases were searched for articles published between 2000 and 2020, without language restrictions, combining the following terms: "tDCS," "transcranial direct current stimulation," "non-invasive brain stimulation," and "Parkinson's disease." We also conducted cross-reference searches of original articles and reviews to identify additional studies that could not be retrieved from electronic databases.

Inclusion Criteria

This study follows the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) guidelines (20).

Eligibility Criteria

We used the following PICOT criteria (population, intervention, comparison outcome, and study type) to define eligibility criteria (see **Supplementary Material**):

- Population: PD and MCI PD patients without dementia diagnosed following UKBB criteria in levodopa on/off stage;
- Intervention: studies evaluating tDCS effects on cognitive functions;
- Comparison outcome: scores obtained on cognitive measures and standard deviation/error.
- Study type: randomized studies with double/single-blind design.

Studies in which data from pre-defined outcomes could not be extracted were excluded (see **Figure 1**). The following studies were also excluded: (a) animal studies, (b) studies combining tDCS and transcranial magnetic stimulation (21), (c) case studies (22), and (d) non-cortical stimulation studies (23, 24).

Data Analysis

Several meta-analyses of tDCS vs. sham on cognitive processing was performed following the procedures outlined by Borenstein et al. (25). Interventions' effect sizes were estimated through mean, standard deviation, and sample size. When it was not possible to extract the data, a web calculator was used (26). Because Cohen's "d" overestimates the effect size with small samples, Hedges' "g" was used to correct this bias (27), discriminating between small (0–0.20), medium (0.50–0.80), and large (>0.80) effect sizes (28). Additionally, a random effects approach was used, given its usefulness when there are different designs and response variables. For each analysis, a z-test was performed to derive a summary p-value. Lau et al.'s (29) study was excluded since needed data for effect-size calculation could not be extracted, while a social cognition meta-analysis could not be performed because Adenzato et al.'s (30) study was the only one to provide such measure.

The cognitive domains were defined according to the characteristics of each study as follows:

- Report of an index or subscale.
- If there were several tasks associated with the same domain, the tasks most used in clinical practice and research were included.
- In the case of a single task/subtest, its effect size was used as the index of the domain to which it was associated.

Meta-analyses were performed at two time points: (a) pre-stimulation to post-stimulation, and (b) pre-stimulation to follow-up. Additionally, as many studies combined tDCS with cognitive training (CT), task scores in interventions that combined stimulation with standard (non-tailored) CT were preferred over task scores in tDCS-only interventions.

Outcome Variables

As primary outcomes we considered: (1) Measures of executive functions: Problem-solving strategies: The Stockings of Cambridge (SOC) subtest of CANTAB (31); Task-Switching: the Wisconsin Card Sorting Test (WCST), the Trail Making Test B (TMT-B) (32, 33); working memory: the Three-back letter task (34), Visual working memory (VWM), the change detection task (29), the working memory test (WM) (33); inhibition: Stroop Test (Color-word interference) (31–33); verbal and phonological fluency: the Verbal Fluency Test (32), the Controlled Oral Word Association Task (COWAT) (31) and tasks of semantic and phonological fluency (35); (2) Measures of visual attention: the TMT-A (32), the number-letter sequence (LNS) (31); (3) Measures of memory: the Hopkins Verbal Learning Test-Revised (HVLT-R) Immediate recall test, the Paragraph Recall Test (31); (4) Measures of visuospatial skills: the Line Orientation Judgment Test (JLO) and Hooper's Visual Organization Test (HVOT) (31); (5) Measures of language: the Boston Naming



FIGURE 1 | Meta-analyses performed in different cognitive domains for two time points showing both each study effect size and their relative weight within the summary effect size. Effect sizes are expressed in Hedges' g , and the forest plots represent the weight of the studies by the size of the squares, their effect size by their position relative to the x-axis and Hedges' g 95% CI by the squares' lateral bars.

Test-Short Form (BNT), the similarity test (31); (6) Measures of theory of mind: the Reading the Mind in the Eyes task, the Attribution of Intentions (AI) task (30); (8) Measures of

procedural learning: Probabilistic Classification Learning (PCL) (33); (9) Measures of the inhibition of emotional response: the emotional go/no-go paradigm (29).

TABLE 1 | Effect of transcranial direct current stimulation on cognition in Parkinson's disease.

Study	Cognitive abilities	Test	Total sample (n)	Mean age	Evolution of diagnosis	On/off state	Stimulation parameters					Results
							Active electrode	Reference electrode	Intensity (mA)	Duration (min)	Number of sessions	
Adenzato et al. (30)	Theory of mind (ToM)	Reading the mind in the eyes (RME) task Attribution of intentions (AI) task	(n = 20) atDCS (n = 20) stDCS	65.6 (8.4)	N/R (MCI)	N/R	MFC (FPz)	Between Inion and Oz	1.5	6	1 atDCS session 1 stDCS session	atDCS over the MFC enhances ToM in patients with PD-MCI.
Biundo et al. (36)	Cognitive functions	MoCA, RBANS Tot., list learning, story learning, complex figure copy, orientation line, naming, semantic fluency, digit span, written coding test, list recall, list recognition, story recall, figure recall	(n = 24) (n = 12) atDCS (n = 12) stDCS	69.1_7.6	N/R (MCI)	N/R	L-DLPFC	Contralateral supraorbital region	2	20	4 sessions	atDCS over the PFC increased performance in immediate memory skills (story learning test) enhancing declarative and long term memory consolidation.
Boggio et al. (34)	Working memory	Three-back letter working memory paradigm	(n = 18) (n = 9) atDCS 2mA (n = 9) atDCS 1mA	45	Experiment 1 13.7 (8.2) Experiment 2 12.7 (8.1)	OFF	L-DLPFC M1	Contralateral right orbit	Different intensities 1–2	20	2 sessions	2mA of atDCS of the LDLPFC may improve working memory. Beneficial effect on working memory in PD patients depends on the intensity and site of stimulation.
Brandão et al. (32)	Speed processing, executive function, working memory, attention, verbal fluency, inhibitory control	Trail Making Test (TMT), Verbal Fluency test, Stroop test, Timed Up and Go test and video gait analysis.	(n = 20) (n = 10) atDCS (n = 10) stDCS	64.45 ± 8.98	7.80 ± 5.32	N/R	L-DLPFC	Right orbital frontal cortex (Fp2)	2	20	1 session	After a single session of tDCS over the DLPFC there is improvements on cognitive tests. Cognitive areas improved the performance in the Stroop test and in the Verbal Fluency.
Doruk et al. (33)	Cognitive functions, depressive symptoms and motor functions	Trail making tests A and B (TMTA and B), Wisconsin card sorting test (WCST), probabilistic classification learning (PCL), working memory test (WM) and stroop test.	(n = 18) (n = 10) atDCS R-DLPFC (n = 6) atDCS L-DLPFC (n = 7) stDCS	40_71	S/R	ON	L-DLPFC R-DLPFC	Right supraorbital region	2	20	10 sessions	Active stimulation over RDLPCF and LDLPFC resulted in prolonged improvements on executive function (TMT-B test).
Lau et al. (29)	Working memory	Visual working memory task and emotional go/no-go paradigm	(n = 10)	56–78	7.8 ± 3.6	ON	L-DLPFC	Contralateral (right) supraorbital area	2	20	1 atDCS session 1 stDCS session	Single-session of atDCS over the L-DLPFC did not significantly improve cognitive tasks in PD
Lawrence et al. (31)	Cognitive function and functional outcomes	Tockings of Cambridge (SOC) subtest from CANTAB and the controlled oral word association task (COWAT), letter-number sequencing (LNS) and the stroop (color-word interference) test, Hopkins verbal learning test-revised (HVL-R) immediate recall subtest (20) and the paragraph recall test, judgment of line orientation (JLO) test and the Hooper visual organization test (HVOT), y Boston naming test-short form (BNT) and the similarities test.	(n = 42) SCT (n = 7) TCT (n = 7) tDCS (n = 7) SCT + tDCS (n = 7) TCT + tDCS (n = 7) Control (n = 7)	SCT: 68.14 (8.69) TCT: 65.57 (5.20) tDCS: 72.6.45 SCT + tDCS: 63.57 (15.68) TCT + tDCS: 67.43 (6.37) Control: 72.29 (6.21)	SCT: 5.29 TCT: 5.79 tDCS: 5.50 SCT + tDCS: 6.79 TCT + tDCS: 4.43 Control: 5.36	ON	L-DLPFC	Above the left eye	1.5	20	4 sessions	The intervention groups demonstrated variable statistically significant improvements across executive function, attention/working memory, memory, language, activities of daily living, and quality of life.

(Continued)

TABLE 1 | Continued

Study	Cognitive abilities	Test	Total sample (n)	Mean age	Evolution of diagnosis	On/off state	Stimulation parameters				Results	
							Active electrode	Reference electrode	Intensity (mA)	Duration (min)		Number of sessions
Pereira et al. (35)	Phonemic and semantic fluency	Phonemic and semantic fluency tasks	(n = 16)	61.5_9.9	S/R	N/R	L-DLPFC L-TPC	Right supraorbital area	2	20	1 session	Functional connectivity in verbal fluency and deactivation task-related networks was significantly more enhanced by tDCS to DLPFC than to TPC. atDCS over L-DLPFC increased performance on the phonemic fluency task.

L-DLPFC, Left dorsolateral prefrontal cortex; R-DLPFC, Right dorsolateral prefrontal cortex; M1, Primary motor cortex; L-TPC, Left temporoparietal cortex; MFC, Medial Frontal Cortex; MCI, Mild Cognitive Impairment; SCT, Standard Cognitive Training; TCT, Tailored Cognitive Training; tDCS, Transcranial direct current stimulation; atDCS, Anodal transcranial direct current stimulation; cDCS, Cathodal transcranial direct current stimulation; sDCS, Sham transcranial direct current stimulation; N/R, Not reported.

RESULTS

From the initial 248 search results, 32 relevant publications were identified from databases. Of these, eight articles met the inclusion criteria (see **Supplementary Material**). The participants' mean age in these studies was 64.2 ± 3.1 years (min 61–max 69). With a total of 168 subjects, the average size of the groups was 21 (10 min and 42 max). The average disease duration and the L-dopa effect were not reported in all the studies.

Overall, 87.5% of the studies reported better cognitive performance after atDCS (see **Table 1**). Boggio et al. (34) administered 1 and 2 mA atDCS in the left motor cortex (anodal L-M1) or in the left prefrontal dorsolateral cortex (L-DLPFC) with the cathode located in the contralateral supraorbital area (SOAC). They reported high accuracy on the WM, with 2 mA over the L-DLPFC.

Pereira et al. (35) used 2 mA atDCS in the L-DLPFC and left temporoparietal cortex (L-TPC) and cathode in the SOAC. The results showed improvement in phonological verbal fluency after atDCS over L-DLPFC compared to the L-TPC. Additionally, fMRI verified an increase in functional connectivity between the frontal, parietal, and fusiform areas.

Doruk et al. (33) administered 2 mA in the R-DLPFC and L-DLPFC in 18 subjects with PD and located the cathode in the SOAC. The study reports improvement in the TMT-B after bilateral atDCS in the DLPFC.

Biundo et al. (36) used atDCS in the L-DLPFC with 2 mA and placed the cathode in the SOAC in 24 subjects with PD with mild cognitive impairment (MCI-PD). The researchers reported increased immediate memory skills and long-term consolidation of declarative memory.

Lawrence et al. (31) applied atDCS with 1.5 mA in the L-DLPFC and placed the cathode over the left eye in 42 subjects with MCI-PD. The authors implemented various intervention schemes combined with atDCS to assess the impact on cognitive and functional performance. Evidence suggests improvement in executive function, attention/WM, memory, language, daily living activities, and quality of life compared to the control group when combining CT and atDCS.

Adenzato et al. (30) administered 1.5 mA atDCS to the medial frontal cortex (MFC) and placed the cathode between the Inion and Sickle in 20 MCI-PD patients. The authors report a significant correlation between the reaction time (RT) of the Attribution of Intentions (AI) task and the Frontal Assessment Battery (FAB) score and the effect of interference in time and Stroop error. Findings are limited to improvement in RT; no significant improvement in response precision was observed. Researchers suggest that atDCS in MFCs improves deficits in the Theory of Mind (ToM) in MCI-PD.

Brandão et al. (32) investigated the effect of atDCS on executive functions, verbal fluency, and inhibitory control in 20 subjects with PD when administering 2 mA for 20 min in the L-DLPFC. The cathode was placed in the SOAC. The study reports improvement in the performance of cognitive tests STROOP—inhibition and interference—and verbal fluency in the group that received atDCS. The authors do not report a significant difference in the TMT-B or motor measurements.

Lau et al. (29) applied 2 mA to the L-DLPFC in 10 subjects with PD without cognitive compromise, locating the cathode in SOAC. The researchers evaluated VWM and emotional inhibitory control using experimental paradigms. The study suggests that performing a single session of atDCS is insufficient to generate significant VWM and emotional inhibition processes in subjects with PD. However, the authors also highlighted the small sample size.

We ran 2 meta-analyses per cognitive domain: (a) one analyzing the pre-post stimulation period and (b) one analyzing the pre-follow-up stimulation period. Regarding executive functions, the results showed large effects of improvement in performance in the pre-post period and small and non-significant effects in the pre-follow up [pre-post: $g = 1.51$, 95% CI = (0.28, 2.74), $Z = 2.41$, $p = 0.016$; pre-follow up: $g = -0.15$, 95% CI = (-0.75, 0.45), $Z = -0.50$, $p = 0.619$], see **Figure 1**, analysis 1.1 y 1.2. In memory, there was a medium effect for the pre-follow-up period of improvement in cognitive performance, although it was not significant, while for the other period, the effect was small and non-significant [pre-post: $g = 0.01$, 95% CI = (-0.60, 0.63), $Z = 0.03$, $p = 0.972$; pre-follow-up: $g = 0.46$, 95% CI = (-0.24, 1.15), $Z = 1.28$, $p = 0.199$] (**Figure 1**, analysis 1.3 y 1.4). The analyses in visuospatial skills showed medium effects with improvement in the pre-post and decrease in performance in the pre-follow up, although neither was significant [pre-post: $g = 0.27$, 95% CI = (-0.50, 1.04), $Z = 0.69$, $p = 0.490$; pre-follow up: $g = -0.25$, 95% CI = (-0.98, 0.49), $Z = -0.66$, $p = 0.511$], **Figure 1**, analysis 1.5 y 1.6. In language, a small and non-significant effect was observed for both time points [pre-post: $g = 0.07$, 95% CI = (-0.55, 0.68), $Z = 0.21$, $p = 0.832$; pre-follow up: $g = 0.09$, 95% CI = (-1.48, 1.65), $Z = 0.11$, $p = 0.915$], **Figure 1**, analysis 1.7 y 1.8. Finally, for visual attention, a small and non-significant effect was observed [pre-post: $g = 0.02$, 95% CI = (-0.35, 0.38), $Z = 0.08$, $p = 0.934$], see **Figure 1**, analysis 1.9.

DISCUSSION

This systematic review has highlighted that there are a limited number of studies examining the effects of tDCS on cognitive outcome measures in PD. The few studies available, suggest that atDCS has a positive effect mainly in executive functions. In this regard, studies have shown better performance in problem-solving tests (31), verbal fluency (35, 36), cognitive flexibility (33), planning, and WM (33, 34). Additionally, two studies highlight greater precision and retention of information in memory tests and procedural learning (35, 36). The meta-analysis converges, highlighting positive effects on executive performance; however, these analyses are small (2–5 studies) and subject to considerable variability, so they should only be taken as exploratory. Similarly, while most results were non-significant, uncertainty around the point estimates was underscored by the wide confidence intervals calculated, further stretching the need for studies to clarify and improve the effect-sizes estimations. Interestingly, variations in the detected effects may arise depending on the time point chosen for assessment, i.e., an effect may remain or disappear in the follow-up, or even appear in the follow-up after

not having been detected in the post-treatment measure, which would suggest that some effects are only detected after potential learning effects, masking those that could be attributed to tDCS, have vanished. These findings suggest both the need to control for practice effects and to perform at least one follow-up assessment. Consequently, it is important to fine-tune and standardize the time points for follow-up assessments.

Only one study focused and reported positive effect on electrical activity and functional connectivity circuits in PD (35). It could be speculated that, due to action mechanisms and diffuse effects of tDCS, when applied in frontal areas, this technique increases the electrical activity and functional connectivity of cortico-striatal and thalamocortical circuits (37) affected in PD (38). However, it would be hasty to make this statement without clarity on some methodological aspects and more evidence to support this hypothesis.

Although most studies have used atDCS in the L-DLPFC, some studies do not clarify the neuroanatomical coordinate system used to locate the anode. Thus, it is suggested that future studies verify the correct electrodes' position through mathematical simulation of the electric fields generated by the assembly (39). Moreover, there is variability in current intensity (1–2 mA) and the period of exposure to tDCS, which prevents identifying if effects hold over time. Performing a current stimulation process for a few seconds can generate changes in cortical excitability. However, these are insufficient to consider them significant. Indeed, when stimulation is prolonged or repetitive, effects can last for hours (16, 40) and even days (19). The most widely used stimulation parameters to establish the use of tDCS in PD are 6–20 min per session, and no more than twice per day (41).

Our review and meta-analysis suggest that tDCS has been shown to exert positive effects as an adjunctive therapy on non-motor symptoms in PD. It is not sufficiently evidenced to establish a cause-effect relationship between tDCS intervention, cognitive improvement, electrical activity modulation and functional connectivity increase in PD. Thus, it is essential to (a) explore the potential of tDCS to ameliorate another kind of cognitive symptom reported in PD, such as action verb processing impairment (12, 13, 42–45); to date, there is no evidence about it, and it is feasible to stimulate networks involving cortico-cortical fibers and cortico-subcortical circuits (37) primarily affected in PD (43). It is also essential to (b) perform longitudinal studies to determine whether changes in cognition persist over time. Limited number of sessions and periodicity of the process currently impedes testing whether the effect is transitory and experimentally relevant or if it could go beyond therapeutic and clinical applicability.

Limitations and Suggestions for Further Research

Several factors limit interpretations of these studies' results and the understanding of tDCS effects on cognitive impairments in PD patients. As mentioned by Borenstein et al. (25), including studies with independent and related groups in the same meta-analysis introduces a source of error to be considered. However,

the decision was made due to the limited number of studies; therefore, results should be taken carefully and in an exploratory way. An heterogeneity analysis was not conducted since, as reported previously, for such small analyses this type of test has low statistical power (46, 47).

The lack of standardization of the outcome measures used to assess changes in cognitive performance in different domains, has led to a considerable variability in the analyses performed. This should be addressed in the future by establishing a set of measures that can sensibly evaluate tDCS-related changes. Although results are promising and tDCS is positioning itself as a new adjuvant therapy in PD treatment, sample groups are small and heterogeneous; therefore, it is necessary to conduct studies with larger cohorts. Likewise, it is recommended to combine (a) intervention schemes involving pharmacological treatment and physical and CT programs to determine under what conditions the modulating effect of tDCS is enhanced, and (b) further research should employ neurophysiology measurements to characterize and explore the potential cause-effect relationship between tDCS intervention, cognitive improvement, and neural correlates -as connectivity signatures- in PD.

CONCLUSION

This systematic review and meta-analysis highlight potential effectiveness of atDCS to improve executive (including inhibition of prepotent responses, shifting mental sets, monitoring and regulating performance, goal maintenance, planning, working memory, and cognitive flexibility) and mnemonic performance

in PD patients but failed to establish a cause-effect relationship between tDCS intervention and cognitive enhancement in PD.

Considering the potential value of this safe and low-cost technique, it is imperative that well-designed, high-quality, and sufficiently powered randomized studies assess the efficacy of tDCS to treat cognitive impairments in PD and draw new pathways to include it in clinical practice. Evidence from the effects of tDCS on cognitive symptoms in PD patients is sparse, and we suggest that further research is required.

AUTHOR CONTRIBUTIONS

DS-G, JG-C, MZ, and JC developed the review concept. DS-G, JG-C, and JC drafted the manuscript. MZ provided critical revisions. DS-G and JG-C performed the data collection, analysis, and interpretation under the supervision of JC and MZ. All authors approved the final version of the manuscript for submission.

FUNDING

This work was partially supported by Instituto de Psicología, Universidad del Valle; COLCIENCIAS [1106-744-55314], Sistema General de Regalías (BPIN2018000100059), Universidad del Valle [Research Grant Scheme: CI 5316, CI 5292].

SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fneur.2020.597955/full#supplementary-material>

REFERENCES

- Broeder S, Nackaerts E, Heremans E, Vervoort G, Meesen R, Verheyden G, et al. Transcranial direct current stimulation in Parkinson's disease: neurophysiological mechanisms and behavioral effects. *Neurosci Biobehav Rev.* (2015) 57:105–17. doi: 10.1016/j.neubiorev.2015.08.010
- Buch E, Santarnecchi E, Antal A, Born J, Celnik P, Classen J, et al. Effects of tDCS on motor learning and memory formation: a consensus and critical position paper. *Clin Neurophysiol.* (2017) 128:589–603. doi: 10.1016/j.clinph.2017.01.004
- Elsner B, Kugler J, Pohl M, Mehrholz J. Transcranial direct current stimulation (tDCS) for idiopathic Parkinson's disease. *Cochrane Database Syst Rev.* (2016) 7:CD010916. doi: 10.1002/14651858.CD010916.pub2
- Ferrucci R, Mameli F, Ruggiero F, Priori A. Transcranial direct current stimulation as treatment for Parkinson's disease and other movement disorders. *Basal Ganglia.* (2016) 6:53–61. doi: 10.1016/j.baga.2015.12.002
- Tahtis V, Kaski D. Parkinson and #39;s disease treatments: focus on transcranial direct current stimulation (tDCS). *J Parkinsonism Restless Legs Syndr.* (2017) 2017:55–70. doi: 10.2147/JPRLS.S128146
- Bloem BR, Henderson EJ, Dorsey ER, Okun MS, Okubadejo N, Chan P, et al. Integrated and patient-centred management of Parkinson's disease: a network model for reshaping chronic neurological care. *Lancet Neurol.* (2020) 19:623–34. doi: 10.1016/S1474-4422(20)30064-8
- Cardona JF. Embodied cognition: a challenging road for clinical neuropsychology. *Front Aging Neurosci.* (2017) 9:388. doi: 10.3389/fnagi.2017.00388
- Armstrong MJ, Okun MS. Diagnosis and treatment of Parkinson disease: a review. *JAMA.* (2020) 323:548–60. doi: 10.1001/jama.2019.22360
- Halliday G, Leverenz J, Schneider J, Adler C. The neurobiological basis of cognitive impairment in Parkinson's disease. *Mov Disord.* (2014) 29:634–50. doi: 10.1002/mds.25857
- Levin E, Llabre M, Reisman S, Weiner J, Sanchez-Ramos C, Singer M, et al. Visuospatial impairment in Parkinson's disease. *Neurology.* (1991) 41:365–9. doi: 10.1212/WNL.41.3.365
- Rodríguez-Oroz MC, Jahanshahi M, Krack P, Litvan I, Macias R, Bezard E, et al. Initial clinical manifestations of Parkinson's disease: features and pathophysiological mechanisms. *Lancet Neurol.* (2009) 8:1128–39. doi: 10.1016/S1474-4422(09)70293-5
- Ibáñez A, Cardona JF, Dos Santos YV, Blenkmann A, Aravena P, Roca M, et al. Motor-language coupling: direct evidence from early Parkinson's disease and intracranial cortical recordings. *Cortex.* (2013) 49:968–84. doi: 10.1016/j.cortex.2012.02.014
- Birba A, García-Cordero I, Kozono G, Legaz A, Ibáñez A, Sedeño L, et al. Losing ground: frontostriatal atrophy disrupts language embodiment in Parkinson's and Huntington's disease. *Neurosci Biobehav Rev.* 2017 80:673–87. doi: 10.1016/j.neubiorev.2017.07.011
- Giladi N, Treves T, Paleacu D, Shabtai H, Orlov Y, Kandinov B, et al. Risk factors for dementia, depression and psychosis in long-standing Parkinson's disease. *J Neural Transm.* (2000) 107:59–71. doi: 10.1007/s007020050005
- Lefaucheur J, Antal A, Ayache S, Benninger D, Brunelin J, Cogiamanian F, et al. Evidence-based guidelines on the therapeutic use of transcranial direct current stimulation (tDCS). *Clin Neurophysiol.* (2017) 128:56–92. doi: 10.1016/j.clinph.2016.10.087
- Nitsche M, Paulus W. Sustained excitability elevations induced by transcranial DC motor cortex stimulation in humans. *Neurology.* (2001) 57:1899–901. doi: 10.1212/WNL.57.10.1899

17. Stagg C, Antal A, Nitsche M. Physiology of transcranial direct current stimulation. *J ECT*. (2018) 34:144–52. doi: 10.1097/YCT.0000000000000510
18. Nitsche M, Cohen L, Wassermann E, Priori A, Lang N, Antal A, et al. Transcranial direct current stimulation: state of the art 2008. *Brain Stimul*. (2008) 1:206–23. doi: 10.1016/j.brs.2008.06.004
19. Paulus W, Rothwell J. Membrane resistance and shunting inhibition: where biophysics meets state-dependent human neurophysiology. *J Physiol*. (2016) 594:2719–28. doi: 10.1113/JP271452
20. Moher D, Liberati A, Tetzlaff J, Altman D, PRISMA Group. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *Ann Intern Med*. (2013) 151:264–70. doi: 10.7326/0003-4819-151-4-200908180-00135
21. Von Pape M, Fisse M, Sarfeld AS, Fink GR, Nowak DA. The effects of 1 Hz rTMS preconditioned by tDCS on gait kinematics in Parkinson's disease. *J Neural Transm*. (2014) 121:743–54. doi: 10.1007/s00702-014-1178-2
22. Kaski D, Allum JH, Bronstein AM, Dominguez RO. Applying anodal tDCS during tango dancing in a patient with Parkinson's disease. *Neurosci Lett*. (2014) 568:39–43. doi: 10.1016/j.neulet.2014.03.043
23. Baijens LWJ, Speyer R, Passos VL, Pilz W, Roodenburg N, Clavé P. The effect of surface electrical stimulation on swallowing in dysphagic Parkinson patients. *Dysphagia*. (2012) 27:528–37. doi: 10.1007/s00455-011-9387-4
24. Ferrucci R, Cortese F, Bianchi M, Pittera D, Turrone R, Bocci T, et al. Cerebellar and motor cortical transcranial stimulation decrease levodopa-induced dyskinesias in Parkinson's disease. *Cerebellum*. (2016) 15:43–7. doi: 10.1007/s12311-015-0737-x
25. Borenstein M, Hedges L, Higgins J, Rothstein H. *Introduction to Meta-Analysis*. Hoboken, NJ: John Wiley and Sons (2009).
26. Lenhard W, Lenhard A. Calculation of effect sizes. *Psychometrika*. (2016). Retrieved from: https://www.psychometrika.de/effect_size.html
27. Hedges L. Distribution theory for Glass's estimator of effect size and related estimators. *J Edu Stat*. (1981) 6:107–128. doi: 10.2307/1164588
28. Cohen J. *Statistical Power Analysis for the Behavioral Sciences*. 2nd ed. New York, NY: Lawrence Erlbaum Associate (1988).
29. Lau C, Liu M, Chang K, Chang A, Bai C, Tseng C, et al. Effect of single-session transcranial direct current stimulation on cognition in Parkinson's disease. *CNS Neurosci Ther*. (2019) 25:1237–43. doi: 10.1111/cns.13210
30. Adenzato M, Manenti R, Enrici I, Gobbi E, Brambilla M, Alberici A, et al. Transcranial direct current stimulation enhances theory of mind in Parkinson's disease patients with mild cognitive impairment: a randomized, double-blind, sham-controlled study. *Transl Neurodegener*. (2019) 8:1–13. doi: 10.1186/s40035-018-0141-9
31. Lawrence B, Gasson N, Johnson A, Booth L, Loftus A. Cognitive training and transcranial direct current stimulation for mild cognitive impairment in Parkinson's disease: a randomized controlled trial. *Park Dis*. (2018) 2018:4318475. doi: 10.1155/2018/4318475
32. Brandão M, do Nascimento Neto L, Terra M, Barboza N, Okano A, Smaili S. Effectiveness of acute transcranial direct current stimulation on non-motor and motor symptoms in Parkinson's disease. *Neurosci Lett*. (2019). 696:46–51. doi: 10.1016/j.neulet.2018.12.017
33. Doruk D, Gray Z, Bravo GL, Pascual-Leone A, Fregni F. Effects of tDCS on executive function in Parkinson's disease. *Neurosci Lett*. (2014) 582:27–31. doi: 10.1016/j.neulet.2014.08.043
34. Boggio P, Ferrucci R, Rigonatti S, Covre P, Nitsche M, Pascual-Leone A, et al. Effects of transcranial direct current stimulation on working memory in patients with Parkinson's disease. *J Neurol Sci*. (2006) 249:31–8. doi: 10.1016/j.jns.2006.05.062
35. Pereira JB, Junqué C, Bartrés-Faz D, Martí MJ, Sala-Llloch R, Compta Y, et al. Modulation of verbal fluency networks by transcranial direct current stimulation (tDCS) in Parkinson's disease. *Brain Stimul*. (2013) 6:16–24. doi: 10.1016/j.brs.2012.01.006
36. Biundo R, Weis L, Fiorenzato E, Gentile G, Giglio M, Schifano R, et al. Double-blind randomized trial of t-DCS versus sham in Parkinson patients with mild cognitive impairment receiving cognitive training. *Brain Stimul*. (2015) 8:1223–5. doi: 10.1016/j.brs.2015.07.043
37. Polanía R, Paulus W, Nitsche M. Modulating cortico-striatal and thalamo-cortical functional connectivity with transcranial direct current stimulation. *Hum Brain Mapp*. (2012) 33:2499–508. doi: 10.1002/hbm.21380
38. Gao L-L, Wu T. The study of brain functional connectivity in Parkinson's disease. *Transl Neurodegener*. (2016) 5:18. doi: 10.1186/s40035-016-0066-0
39. Ruffini G, Fox MD, Ripolles O, Miranda PC, Pascual-Leone A. Optimization of multifocal transcranial current stimulation for weighted cortical pattern targeting from realistic modeling of electric fields. *NeuroImage*. (2014) 89:216–25. doi: 10.1016/j.neuroimage.2013.12.002
40. Nitsche MA, Paulus W. Excitability changes induced in the human motor cortex by weak transcranial direct current stimulation. *J Physiol*. (2000) 527:633–9. doi: 10.1111/j.1469-7793.2000.t01-1-00633.x
41. Fregni F, Nitsche MA, Loo CK, Brunoni AR, Marangolo P, Leite J, et al. Regulatory considerations for the clinical and research use of transcranial direct current stimulation (tDCS): review and recommendations from an expert panel. *Clin Res Regul Aff*. (2014) 1333:1060–333. doi: 10.3109/10601333.2015.980944
42. Bocanegra Y, García AM, Lopera F, Pineda D, Baena A, Ospina P, et al. Unspeakable motion: selective action-verb impairments in Parkinson's disease patients without mild cognitive impairment. *Brain Lang*. (2017) 168:37–46. doi: 10.1016/j.bandl.2017.01.005
43. Cardona JF, Gershanik O, Gelormini-Lezama C, Houck AL, Cardona S, Kargieman L, et al. Action-verb processing in Parkinson's disease: new pathways for motor-language coupling. *Brain Struct Funct*. (2013) 218:1355–73. doi: 10.1007/s00429-013-0510-1
44. García A, Bocanegra Y, Herrera E, Moreno L, Carmona J, Baena A, et al. Parkinson's disease compromises the appraisal of action meanings evoked by naturalistic texts. *Cortex*. (2018) 100:111–26. doi: 10.1016/j.cortex.2017.07.003
45. Melloni M, Sedeño L, Hesse E, García-Cordero I, Mikulan E, Plastino A, et al. Cortical dynamics and subcortical signatures of motor-language coupling in Parkinson's disease. *Sci Rep*. (2015) 5:11899. doi: 10.1038/srep11899
46. Goh J, Hall J, Rosenthal R. Mini meta-analysis of your own studies: some arguments on why and a primer on how. *Soc Pers Psychol Compass*. (2016) 10:535–49. doi: 10.1111/spc3.12267
47. Huedo-Medina T, Sánchez-Meca J, Marín-Martínez F, Botella J. Assessing heterogeneity in meta-analysis: Q statistic or I² index? *Psychol Methods*. (2006) 11:193–206. doi: 10.1037/1082-989X.11.2.193

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2020 Suarez-García, Grisales-Cárdenas, Zimmerman and Cardona. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.



Facilitative Effects of Transcranial Direct Current Stimulation on Semantic Memory Examined by Text-Mining Analysis in Patients With Schizophrenia

Chika Sumiyoshi^{1,2*}, Zui Narita³, Takuma Inagawa⁴, Yuji Yamada⁴, Kazuki Sueyoshi², Yumi Hasegawa², Aya Shirama², Ryota Hashimoto^{5,6} and Tomiki Sumiyoshi²

¹ Faculty of Human Development and Culture, Fukushima University, Fukushima, Japan, ² Department of Preventive Intervention for Psychiatric Disorders, National Center of Neurology and Psychiatry, Kodaira, Japan, ³ Department of Psychiatry and Behavioral Sciences, Stanford University, Palo Alto, CA, United States, ⁴ Department of Psychiatry, National Center Hospital, National Center of Neurology and Psychiatry, Kodaira, Japan, ⁵ Department of Pathology of Mental Diseases, National Institute of Mental Health, National Center of Neurology and Psychiatry, Kodaira, Japan, ⁶ Department of Psychiatry, Graduate School of Medicine, Osaka University, Osaka, Japan

OPEN ACCESS

Edited by:

Carmen Terranova,
University of Messina, Italy

Reviewed by:

Anushree Bose,
National Institute of Mental Health and
Neurosciences, India
Antonino Naro,
Centro Neurolesi Bonino Pulejo
(IRCCS), Italy

*Correspondence:

Chika Sumiyoshi
sumiyoshi@educ.fukushima-u.ac.jp

Specialty section:

This article was submitted to
Neurorehabilitation,
a section of the journal
Frontiers in Neurology

Received: 14 July 2020

Accepted: 04 January 2021

Published: 11 February 2021

Citation:

Sumiyoshi C, Narita Z, Inagawa T, Yamada Y, Sueyoshi K, Hasegawa Y, Shirama A, Hashimoto R and Sumiyoshi T (2021) Facilitative Effects of Transcranial Direct Current Stimulation on Semantic Memory Examined by Text-Mining Analysis in Patients With Schizophrenia. *Front. Neurol.* 12:583027. doi: 10.3389/fneur.2021.583027

Background: Beneficial effects of transcranial direct current stimulation (tDCS) are relevant to cognition and functional capacity, in addition to psychiatric symptoms in patients with schizophrenia. However, whether tDCS would improve higher-order cognition, e.g., semantic memory organization, has remained unclear. Recently, text-mining analyses have been shown to reveal semantic memory. The purpose of the current study was to determine whether tDCS would improve semantic memory, as evaluated by text-mining analyses of category fluency data, in patients with schizophrenia.

Methods: Twenty-eight patients entered the study. Cognitive assessment including the category fluency task was conducted at baseline (before tDCS treatment) and 1 month after t administration of tDCS (2 mA × 20 min, twice per day) for 5 days, according to our previous study. The category fluency data were also obtained from 335 healthy control subjects. The verbal outputs (i.e., animal names) from the category fluency task were submitted to singular valued decomposition (SVD) analysis. Semantic memory structures were estimated by calculating inter-item cosines (i.e., similarities) among animal names frequently produced in the category fluency task. Data were analyzed longitudinally and cross-sectionally to compare the semantic structure within the patient group (i.e., baseline vs. follow-up) and between groups (patients vs. healthy controls). In the former, semantic associations for frequent items were compared in the form of cosine profiles, while in the latter, the difference in the magnitude of the correlations for inter-item cosines between healthy controls and patients (baseline, follow-up) was examined.

Results: Cosine profiles in the patient group became more cluster-based (i.e., pet, carnivores, and herbivores) at follow-up compared to those at baseline, yielding higher cosines within subcategories. The correlational coefficient of inter-item cosines

between healthy controls and patients was significantly greater at follow-up compared to baseline; semantic associations in patients approached the normality status after multi-session tDCS.

Conclusions: To our knowledge, this is the first study to demonstrate the facilitative effect of tDCS on semantic memory organization in patients with schizophrenia. Text-mining analysis was indicated to effectively evaluate semantic memory structures in patients with psychiatric disorders.

Keywords: schizophrenia, tDCS, semantic memory, category fluency, text-mining analysis

INTRODUCTION

Several domains of cognitive function, specifically, verbal fluency, working memory, and processing speed, are impaired in patients with schizophrenia (1, 2). The cognitive decline compared to healthy adults is in a range of 0.5–2.5 SD (3, 4), hindering functional recovery (5).

Cognitive profiles specific to schizophrenia have been evaluated comprehensively by cognitive batteries, including the Brief Assessment of Cognition in Schizophrenia [BACS; Keefe et al. (6)] and MATRICS Consensus Cognitive Battery [MCCB; Nuechterlein and Green (7)]. Most subtests in these neuropsychological batteries are designed to evaluate executive aspects of cognition (i.e., attention, processing speed, and visual/verbal working memory). Therefore, additional methods are required to assess higher-order cognitive functions, such as semantic memory.

Semantic memory represents a long-term storage of information (8, 9), and semantic structure is defined based on its cohesiveness, i.e., semantic association between items (10). Typically, the semantic structure is represented in the form of clusters, spatial constellations, or networks.

Previous studies have demonstrated aberrant structures of semantic memory in patients with schizophrenia (11–15). Importantly, the disturbance of semantic memory is related with negative symptoms (e.g., alogia) (15) and quality of life (16). These observations indicate the need for the development of effective methods to assess semantic memory in patients with schizophrenia.

Semantic memory is estimated by using data from several cognitive tasks. Specifically, the category fluency task has been used in the study of schizophrenia (11–15). In this task, subjects are instructed to freely recall as many items in a given category (e.g., animal) as possible in a designated time (typically 1 min.). The task is not demanding, and is included in major neurocognitive test batteries, e.g., the MCCB and BACS.

The recent application of text-mining techniques to data from the category fluency task provides objective indices of semantic structures in clinical subjects. For example, network analysis found several parameters, i.e., diameter, average shortest path, and network density, which effectively identify cognitive impairment (17). For the same purpose, latent semantic analysis [LSA; Landauer and Dumais (18)] and singular value decomposition analysis [SVD; Sung et al. (19)] have also been

used (19–21). Generally, these methods use a cosine value and vector length to evaluate semantic memory structure (19, 20, 22). The former represents cohesiveness while the latter indicates unusualness of items composing semantic memory. Assuming that disorganization of semantic memory is one of the intermediate cognitive phenotypes of schizophrenia, Nicodemus et al. (20) examined candidate genes related with semantic memory formation by using LSA of category fluency data. They found that average vector length of items was associated with DISC1 in men with schizophrenia. Meanwhile, Sung et al. (19) and Sumiyoshi et al. (21) used SVD analysis, and reported cosine profiles of patients with schizophrenia were deviated from those of healthy controls, revealing unusual structure of semantic memory.

To ameliorate cognitive impairments in schizophrenia, pharmacological, psychosocial, and neuromodulatory approaches have been attempted. Specifically, some types of brain stimulation, particularly non-invasive methods, e.g., transcranial magnetic stimulation and transcranial direct current stimulation (tDCS) have been drawing attention (23, 24). tDCS modulates neural activities in the brain with weak electrical currents (23, 24). The beneficial effects of tDCS are relevant to cognition as well as psychiatric symptoms, functional capacity, and depression in patients with schizophrenia (25, 26).

Although evidence has been accumulated regarding the efficacy of tDCS on cognitive impairment of schizophrenia (26), only a few studies have been conducted to determine whether tDCS would improve higher-order cognition. For example, Vannorsdall et al. (27) reported that tDCS facilitated retrieval of semantically related words in healthy adults. Also, the facilitative effect of tDCS has been found to be more pronounced in category, rather than letter fluency performance (28). These observations suggest that the cognitive enhancement with tDCS is not limited to attention and executive functions, but is also beneficial for a higher level cognitive function, e.g., organization of semantic memory. Thus, it was hypothesized that tDCS would be effective to improve semantic memory structure in patients with schizophrenia.

The aim of the current study was to determine whether tDCS would improve semantic structure, as evaluated by text-mining analyses of category fluency data, in patients with schizophrenia. For this purpose, data were analyzed to compare the semantic structure longitudinally (within the patient group: data at baseline vs. those after tDCS administration) and

cross-sectionally (between groups: patients vs. healthy controls), as demonstrated in **Figure 1D**.

METHODS

Participants

A total of 28 participants were inpatients ($n = 22$) or outpatients ($n = 6$) treated at National Center Hospital, National Center of Neurology and Psychiatry (25). They met DSM-5 criteria for schizophrenia. Patients with alcohol or substance disorder, traumatic brain injury, or epilepsy were excluded. The patients received antipsychotic drugs (25), which were not changed throughout the sessions. Healthy volunteers ($N = 335$) were recruited from the community through local advertisements

at Osaka University as participants in a general cognitive assessment (29, 30). They were evaluated using the non-patient version of the Structured Clinical Interview for DSM-IV (SCID) to exclude individuals who had current or past contact with psychiatric services or had received psychiatric medication (31, 32). Data was extracted from our previous study of the effect of tDCS on cognitive function in patients with schizophrenia (25), and from text-mining study using healthy adults (21).

This study was approved by Ethical Committee of National Center of Neurology and Psychiatry, Research Ethics Committee of Fukushima University, and Ethical Committee of Osaka University. The procedures were conducted according to the Declaration of Helsinki and all subjects gave written informed consents.

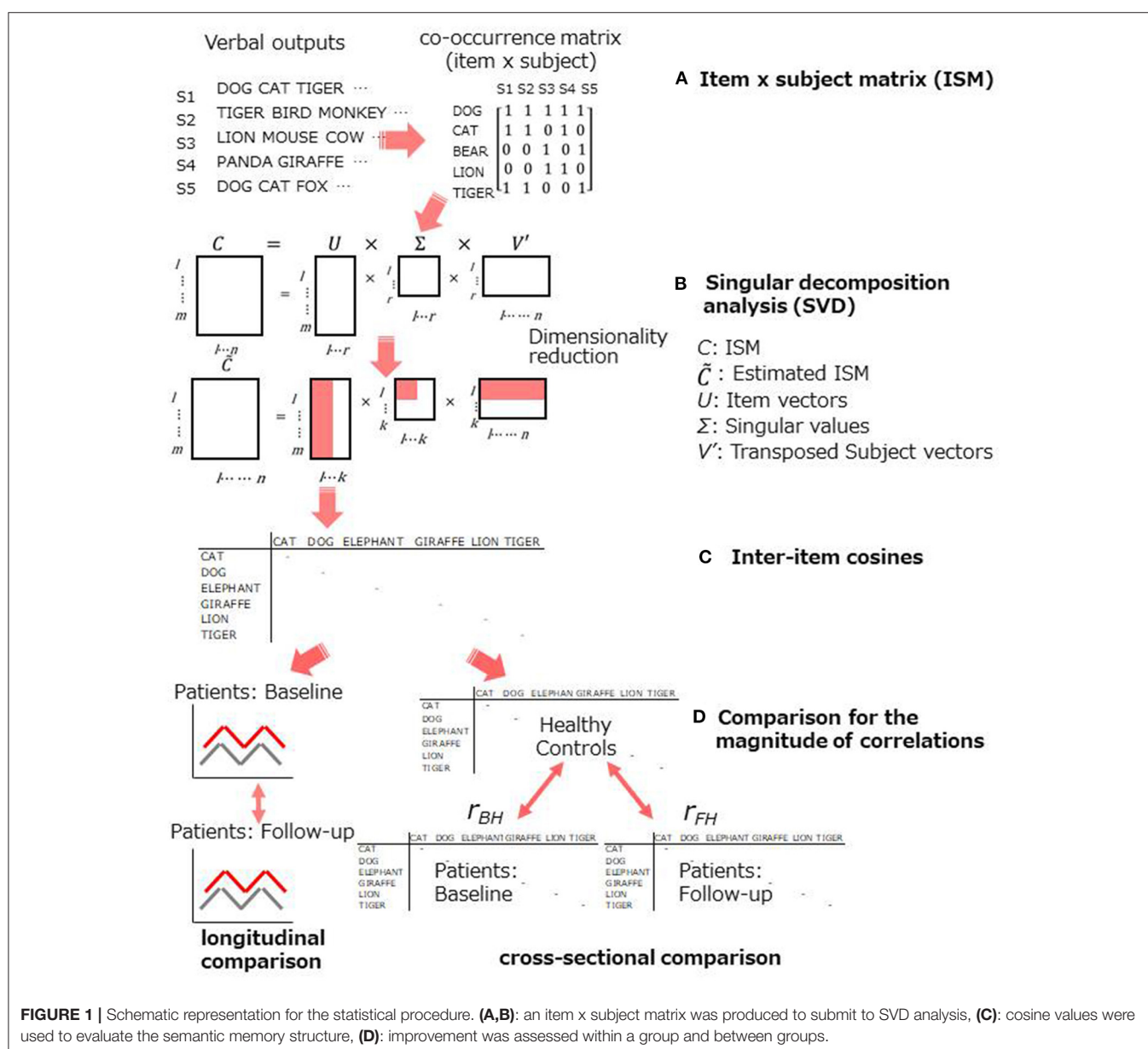


FIGURE 1 | Schematic representation for the statistical procedure. **(A,B)**: an item x subject matrix was produced to submit to SVD analysis, **(C)**: cosine values were used to evaluate the semantic memory structure, **(D)**: improvement was assessed within a group and between groups.

Intervention

tDCS was administered according to a method previously reported (33) in line with a previous study of tDCS on cognition in patients with Schizophrenia (34). Participants underwent 10 active tDCS sessions in 5 consecutive days, twice per day. On each day, tDCS intervention was performed approximately at 10 a.m. and 2 p.m. Patients received no additional behavioral treatment or therapeutic adjustment other than tDCS.

Possible adverse effects related to tDCS, including itching, tingling, headache, burning sensation and discomfort, were monitored using semi-structured checklist (35) after each intervention.

A Soterix Medical 1×1 Transcranial Direct Current Low-Intensity Stimulator Model 1,300 A was used for the tDCS through two 35 cm² electrodes. We usually soaked 4 ml of saline per side (8 ml into each sponge). For each session, direct current of 2 mA for 20 min was applied. The tDCS montage comprised placement of the anode over the left dorsolateral prefrontal cortex (DLPFC) and the cathode over the right supraorbital area (corresponding to F3 and FP2, according to the International 10–20 electroencephalography system).

Assessment for Cognition and Psychiatric Symptoms

Cognitive function was assessed at baseline and 1-month after the last tDCS administration using the BACS. Verbal outputs of the category fluency task were obtained from the BACS. Category fluency is a free recall task, asking subjects to produce as many animal names as possible in 1 min. According to the normative method (36), errors (i.e., repetitions, proper nouns, and intrusions [e.g., *APPLE* for an animal cue]) were removed from the analysis. Premorbid intelligence was estimated at baseline using the Japanese version of the Adult Reading Test [JART, Matsuoka et al. (37)]. As for healthy controls, category fluency task and the JART were conducted in a general cognitive assessment (29, 30).

Psychiatric symptoms were assessed at baseline and follow-up using the Positive and Negative Syndrome Scale [PANSS; Kay et al. (38)].

Statistical Analysis

Demographic variables and category fluency scores were compared between patients and healthy controls using *t*-test. Comparisons between baseline and follow-up in patients were conducted based on our previous report (25). Inequality of variance between the groups was examined using Levene test. Welch method was applied if inequality was significant.

To evaluate the semantic structure, SVD analysis was conducted for verbal outputs of the category fluency task. **Figure 1** demonstrates schematic representation of the procedure. First, an item \times subject matrix (ISM) was created. Rows of the ISM contained animal items (e.g., *DOG CAT*, etc.), while columns contained subjects, and each cell contained a co-occurrence of items (**Figure 1A**). Then, SVD analysis was applied to the matrices obtained from patients and healthy controls (**Figure 1B**). SVD is a general matrix factorization technique based on eigenvalue decomposition [for further

information, see Supplementary Materials in Sung et al. (19, 22, 39)]. Each row (i.e., item) is treated as a vector in the space produced by SVD.

A key component of the structure of semantic memory is cosine values in reduced (i.e., higher) dimensions (**Figure 1C**). A cosine close to 1.0 indicates that two items are highly similar (two words frequently co-occur across subjects).

To assess the improvement on semantic memory structure, cosines between the highly frequent items were contrasted longitudinally and cross-sectionally. In the former, cosine profiles of the 6 most frequent items were produced for patients at baseline and at follow-up and compared (**Figure 1D**, left). As for the latter, the improvement was evaluated as follows: (1) inter-item cosines were obtained between the 6 most frequent items; (2) Pearson's correlational coefficients for those cosines were calculated between healthy controls and patients at baseline (r_{BH}) and follow-up (r_{FH}); (3) The difference in the magnitude of the two correlational coefficients were tested by the Meng's method (40) (**Figure 1D**, right). The method was employed because the healthy control group was used as a "reference," and therefore, it was "overlapped" in testing the magnitude of the difference. The significance level was set for $p < 0.05$ with one-tailed (i.e., $r_{BH} < r_{FH}$), hypothesizing that the tDCS treatment could improve higher, as well as lower, level of cognition.

R version 3.2.2 (41) and its LSA package (42) were used for conducting SVD analysis and producing inter-item cosines. For testing correlations, R based software cocor (43) was used. Other statistical analyses were conducted by SPSS ver. 22.

RESULTS

Demographic and Cognitive Variables

Table 1 presents demographic and clinical variables at baseline and category fluency performance. Inequality of variances was significant only in Estimated premorbid IQ ($F = 12.22$, $p < 0.001$) to which Welch method was applied. Healthy controls were significantly younger, more educated, and showed higher premorbid IQ compared to patients. The former group also produced more words in the category fluency task.

SVD Analysis

Table 2 presents 20 items most frequently produced by patients and healthy controls. Out of them, 12 items, i.e., BEAR, BIRD, CAT, DOG, ELEPHANT, GIRAFFE, LION, MONKEY, MOUSE, PANDA, RABBIT, TIGER, were chosen for SVD analysis. They commonly appeared at baseline and follow-up, with the frequency more than 10 (**Table 2**, in bold).

There are no definite rules for choosing an appropriate number of singular values (dimensions) for the dimensionality reduction (44). Therefore, a six-dimensional solution (6D) was used where the sum of the singular values reached 70% to the entire sum. Accordingly, inter-item cosines were calculated in the 6D space.

TABLE 1 | Characteristics of participants^a.

Variables	Healthy controls <i>N</i> = 335		Patients <i>N</i> = 28		<i>x</i> ² / <i>t</i>	<i>df</i>	<i>p</i>
	<i>M</i>	<i>SD</i>	<i>M</i>	<i>SD</i>			
M/F	154/181		16/12		1.295	1	0.255
Age (year)	35.8	11.9	40.9	9.8	−2.205	361	0.028
Education (year)	15.2	2.2	13.8	1.7	3.164	361	0.002
Estimated premorbid IQ (JART ^b)	109.3	12.2	99.6	12.0	3.262	29	0.003
Category fluency (Baseline)	20.9	4.5	16.4	5.1	5.071	361	0.000
Category fluency (Follow-up ^c)	—	—	16.9	5.5	4.475	361	0.000
Age at onset (year)	—	—	23.6	6.7	—	—	—
Duration of illness (year)	—	—	17.4	9.9	—	—	—
Neuroleptics (CPZ)	—	—	889.0	587.2	—	—	—
PANSS ^d Positive syndrome	—	—	15.7	5.7	—	—	—
PANSS Negative syndrome	—	—	14.9	8.0	—	—	—
PANSS General psychopathology	—	—	32.0	8.1	—	—	—

^aDemographic variables and PANSS are baseline scores. For the follow-up PANSS scores, see Narita et al. (25) for details.

^bJART, Japanese Adult Reading Test.

^cScores at Baseline and Follow-up were not statistically different (*t* = 0.56, *df* = 27 *p* = 0.58). See Narita et al. (25) for details.

^dPANSS, the Positive and Negative Syndrome Scale.

TABLE 2 | Frequencies of animal items.

Rank	Healthy controls (<i>N</i> = 335)		Patients (<i>N</i> = 28)			
			Base	Follow-up		
1	DOG	309	DOG	24	CAT	24
2	CAT	305	LION	23	DOG	24
3	LION	250	CAT	22	LION	23
4	GIRAFFE	244	ELEPHANT	21	ELEPHANT	19
5	TIGER	239	GIRAFFE	21	MONKEY	15
6	ELEPHANT	235	MOUSE	17	TIGER	15
7	MONKEY	234	TIGER	17	BIRD	13
8	HORSE	171	HORSE	13	GIRAFFE	12
9	SHEEP	163	MONKEY	13	BEAR	11
10	COW	155	BEAR	10	GORILLA	11
11	MOUSE	152	BIRD	10	MOUSE	11
12	RABBIT	148	PANDA	10	PANDA	11
13	HIPPOTAMUS	143	RABBIT	10	RABBIT	11
14	BEAR	122	RACCOON_DOG	8	COW	9
15	RHINOCEROS	116	SHEEP	8	SHEEP	9
16	BIRD	115	HAMSTER	7	HIPPOTAMUSE	8
17	PANDA	110	LEOPARD	7	HORSE	8
18	CHEETAH	102	RHINOCEROS	7	CHEETA	7
19	SNAKE	102	SPARROW	7	CHIMPANZEE	7
20	ZEBRA	102	ZEBRA	7	RACCOON_DOG	7

Cosine Profiles

Each line represents 6D cosine values between one of the top 6 items (e.g., CAT) and the other most frequent 12 items (Table 2, in bold). Overall, cosine values uniformly fluctuated at baseline (Figure 2, top) indicating the lack of distinct clusters (i.e., subcategories). The profiles became more cluster-based at

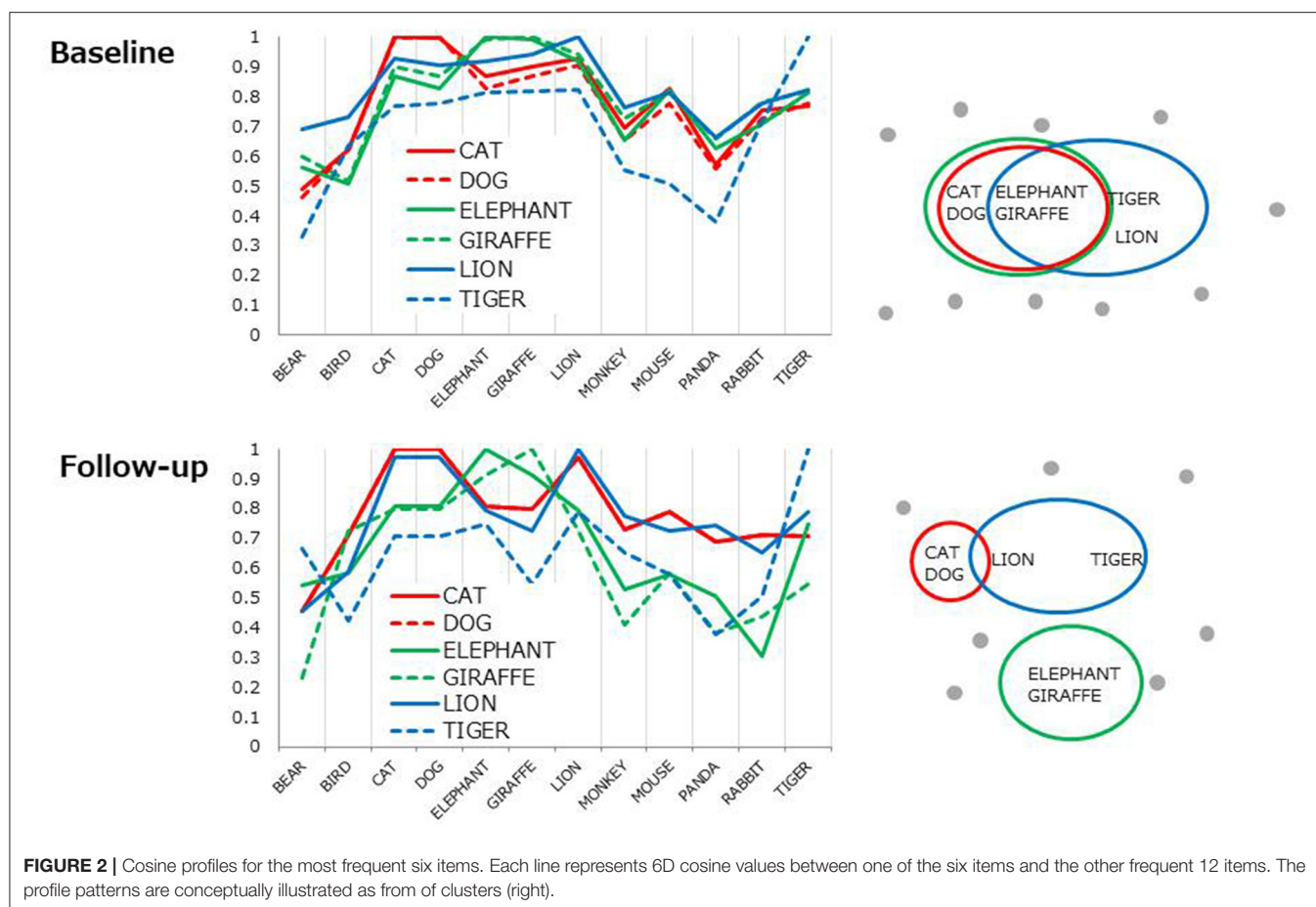
follow-up, yielding a higher cosine within a pair (e.g., CAT-DOG) but lower cosines between pairs (e.g., [CAT-DOG]-[GIRAFFE-ELEPHANT], Figure 2, the bottom) as conceptually shown in Figure 2, right.

Difference in Magnitude of Correlations

The top six items in healthy controls (DOG, CAT, ELEPHANT, GIRAFFE, LION, and TIGER, Table 2) were used for the comparison between r_{BH} and r_{FH} to examine how semantic memory in patients became close to that in healthy controls. Table 3 summarizes correlational coefficients and the difference of the magnitude of correlations. The correlation was considerably higher in follow-up ($r_{FH} = 0.75$) than baseline ($r_{BH} = 0.41$), and the difference was significant ($z = -1.90$, $p = 0.03$, 95% CI = -1.06 , 0.02). Figure 3 schematically illustrates the cognitive process of the result. For example, LION is more easily and quickly accessed than other items (e.g., ELEPHANT or CAT) when TIGER is recalled.

DISCUSSION

Multi-session tDCS was found to improve semantic memory organization, as evaluated by text-mining analyses of category fluency data, in patients with schizophrenia. The longitudinal comparison of cosine profiles suggests that the semantic association among typical items (animal names) was more cluster-based, as in healthy adults (21) at follow-up compared to baseline (Figure 2). Also, the correlation of cosine values between healthy controls and patients was greater at follow-up than at baseline, indicating that semantic structures of patients approached the normality status after administration of tDCS (Figure 3). Probably, patients at follow-up recalled animal names in a similar manner as did healthy people, referring to subcategory (i.e., pet, carnivorous, herbivorous



items, **Figure 3**) to access items more easily and quickly. Associational memory of this kind would be important in real world settings where meaningful conversations and discourses are taking place. Furthermore, it is possible that impairment of associating information in semantic memory may negatively affect competent linguistic behaviors. In fact, adults who later developed psychosis were found to produce discourses similar to those of children, with presentations of repetitions and a limited scope of vocabulary (45). Likewise, schizophrenia patients with severe formal thought disorder exhibited utterances that are syntactically less complex (e.g., reduction of embedded or dependent clauses) compared to those of first-degree relatives or healthy adults (46). Difficulties in associating information in semantic memory may underlie such restricted linguistic behavior in patients with schizophrenia.

There are several hypotheses to explain deterioration of semantic memory structure in patients with psychiatric conditions [(19), for review]. Some assume structural distortions of memory (47) while others claim poor memory activation (19). In both cases, associational retrieval of stored information would be compromised. Although the current study did not directly address this issue, it is worth pursuing the basis for the impairment to understand higher-order cognition in schizophrenia in further studies.

TABLE 3 | Tests for differences in correlational coefficients^a.

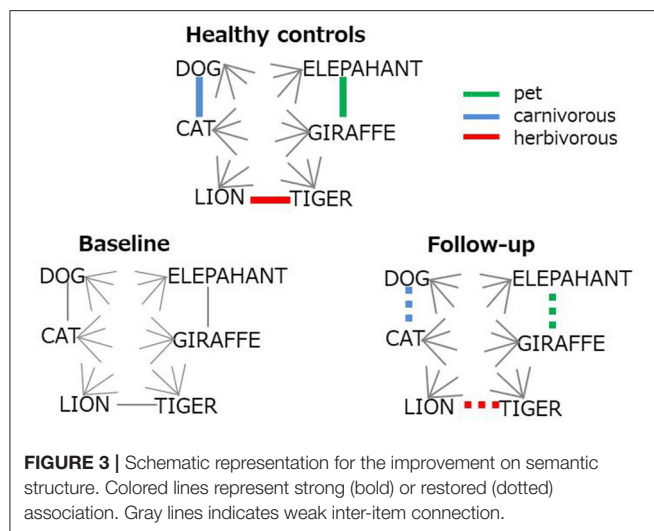
	SCZ Baseline	SCZ Follow-up	Healthy controls
SCZ Baseline	–	0.68	0.41
SCZ Follow-up		–	0.75 ^{ab}
Healthy controls			–

^aSample size: SCZ = 28; HC = 335.

^b* $r_{FH} = 0.75 > r_{BH} = 0.41$, $z = -1.90$, $p = 0.028$ (one-tailed), 95% CI = -1.06 , 0.02 .

The number of word outputs itself in the category fluency task was not increased significantly after administration of tDCS (**Table 1**). This may be partly due to the relatively short duration assessment span (1 month). Possibly, patients tended to repeat a limited variety of items. In fact, type token ratios (TTR), a measure of variety of words, showed only a slight increase in follow-up (baseline: TTR = 0.26, follow-up: TTR = 0.27). Despite, co-occurrences of typical items came to closer to those in healthy adults, as was indicated by the significantly higher correlation in follow-up than baseline (**Table 3**).

Previous studies support our results with providing the neurophysiological substrate. The left prefrontal region is



assumed to be related to the ability of tDCS to improve organizing of information. For example, a previous study (27) found tDCS over the left DLPFC facilitated retrieval of clustered words. A functional imaging study also found that activation in the left frontal region was correlated with categorical clustering in the recall of a verbal learning task (48). These findings are in accord with our result indicating improvement of semantic association in patients with schizophrenia after tDCS treatment over the left prefrontal region.

Although the number of words in the category fluency task was not significantly changed after administration of tDCS, letter fluency was found to be improved in our previous study with the same protocol (25). Meta-analysis results indicate that tDCS over the left ventral inferior frontal gyrus (49) or the left prefrontal cortex (50) increased the number of words produced in the category fluency task.

Results of the current study based on SVD analysis of the category fluency task may add to the usefulness of text-mining analysis in psychiatry, as has been discussed (51–53). Possibly, novel computational linguistic techniques herein reported, i.e., SVD, LSA, and network analysis may contribute to the advance of the National Institute of Mental Health's Research Domain Criteria (RDoC) initiative (54). For example, these techniques may help evaluate the language or declarative memory construct in the RDoC (53).

Several limitations should be mentioned. First, the current study used the data obtained in a previous one-armed open label study (25, 33) that did not adopt sham comparisons. Second, sample size was considerably larger in healthy controls compared to patients. Inequality happened because the former was used as a reference group to estimate normative semantic structure, requiring relatively large sample size. Finally, healthy control subjects were younger, more educated, and in a higher intellectual status compared with patients. However, this demographic bias may not have affected the comparisons of semantic memory structures, because the knowledge about animals is acquired in the early stage of the development

(55). Furthermore, the primitive structures, e.g., clustering, are already present in early childhood (56–58); basic semantic structures should be relatively invariant across ages and educational backgrounds.

In conclusion, the current study demonstrated the facilitative effect of tDCS on semantic memory organization in patients with schizophrenia. Semantic associations in these patients approached the normality status after multi-session tDCS. Text-mining analysis was indicated to effectively evaluate semantic memory structures in patients with psychiatric disorders.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/supplementary material, further inquiries can be directed to the corresponding author/s.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by Ethical Committee of National Center of Neurology and Psychiatry, Research Ethics Committee of Fukushima University, and Ethical Committee of Osaka University. The patients/participants provided their written informed consent to participate in this study.

AUTHOR CONTRIBUTIONS

CS and TS designed the study in collaboration with ZN and RH. ZN, TI, YY, KS, YH, and AS collected and prepared the data. CS conducted the analyses and wrote the initial draft. TS, ZN, and RH critically revised the draft for important intellectual contents. All authors contributed to the manuscript writing.

FUNDING

This work was partially supported by the following funding: the Japan Society for the Promotion of Science (JSPS) KAKENHI Grant Number 20K03433 to CS; JSPS KAKENHI Grant No. 20H03610, Health and Labor Sciences Research Grants for Comprehensive Research on Persons with Disabilities, AMED (18dk0307081 and 20dk0307099), and Intramural Research Grants (30-1, 30-8, 2-3) for Neurological and Psychiatric Disorders of NCNP and JH 2020-B-08 to TS; JSPS KAKENHI Grant Numbers JP18KT0022, JP19H05467, JP20H03611, and JP19H05467, AMED under Grant Number JP20dk0307081 and Brain/MINDS & beyond studies (Grant Number JP20dm0307102) by AMED to RH.

ACKNOWLEDGMENTS

We thank all individuals who participated in this study.

REFERENCES

- Reichenberg A, Harvey PD. Neuropsychological impairments in schizophrenia: Integration of performance-based and brain imaging findings. *Schizophr Bull.* (2007) 133:833–58. doi: 10.1037/0033-2909.133.5833
- Gray BE, McMahon RP, Gold JM. General intellectual ability does not explain the general deficit in schizophrenia. *Schizophr Res.* (2013) 147:315–9. doi: 10.1016/j.schres.2013.04016
- Cholet J, Sauvaget A, Vanelle JM, Hommet C, Mondon K, Mamet JP, et al. Using the brief assessment of cognition in schizophrenia (BACS) to assess cognitive impairment in older patients with schizophrenia and bipolar disorder. *Bipolar Disord.* (2014) 16:326–36. doi: 10.1111/bdi12171
- McCleery A, Ventura J, Kern RS, Subotnik KL, Gretchen-Doorly D, Green MF, et al. Cognitive functioning in first-episode schizophrenia: MATRICS consensus cognitive battery (MCCB) profile of impairment. *Schizophr Res.* (2014) 157:33–9. doi: 10.1016/j.schres.2014.04039
- Green MF. Cognitive impairment and functional outcome in schizophrenia and bipolar disorder. *J. Clin. Psychiatry.* (2006) 67(Suppl. 9):3–8. doi: 10.4088/JCP1006e12
- Keefe RS, Goldberg TE, Harvey PD, Gold JM, Poe MP, Coughenour L. The brief assessment of cognition in schizophrenia: reliability, sensitivity, and comparison with a standard neurocognitive battery. *Schizophr Res.* (2004) 68:283–97. doi: 10.1016/j.schres.2003.09011
- Nuechterlein KH, Green MF. *MATRICS Consensus Cognitive Battery Manual*. Los Angeles, CA: MATRICS Assessment Inc (2006).
- Budson AE, Price BH. Memory dysfunction. *N Engl J Med.* (2005) 352:692–9. doi: 10.1056/NEJMr041071
- Patterson K, Nestor PJ, Rogers TT. Where do you know what you know? The representation of semantic knowledge in the human brain. *Nat Rev Neurosci.* (2007) 8:976–87. doi: 10.1038/nrn2277
- Goni J, Arrondo G, Sepulcre J, Martincorena I, Velez de Mendizabal N, Corominas-Murtra B, et al. The semantic organization of the animal category: evidence from semantic verbal fluency and network theory. *Cogn Process.* (2011) 12:183–96. doi: 10.1007/s10339-010-0372-x
- Aloia MS, Gourovitch ML, Weinberger DR, Goldberg TE. An investigation of semantic space in patients with schizophrenia. *J Int Neuropsychol Soc.* (1996) 2:267–73. doi: 10.1017/S1355617700001272
- Paulsen JS, Romero R, Chan A, Davis AV, Heaton RK, Jeste DV. Impairment of the semantic network in schizophrenia. *Psychiatry Res.* (1996) 63:109–21. doi: 10.1016/0165-1781(96)02901-0
- Troyer AK, Moscovitch M, Winocur G. Clustering and switching as two components of verbal fluency: evidence from younger and older healthy adults. *Neuropsychology.* (1997) 11:138–46. doi: 10.1037/0894-4105.11.1.138
- Sumiyoshi C, Matsui M, Sumiyoshi T, Yamashita I, Sumiyoshi S, Kurachi M. Semantic structure in schizophrenia as assessed by the category fluency test: effect of verbal intelligence and age of onset. *Psychiatry Res.* (2001) 105:187–99. doi: 10.1016/S0165-1781(01)00345-6
- Sumiyoshi C, Sumiyoshi T, Nohara S, Yamashita I, Matsui M, Kurachi M, et al. Disorganization of semantic memory underlies alogia in schizophrenia: an analysis of verbal fluency performance in Japanese subjects. *Schizophr Res.* (2005) 74:91–100. doi: 10.1016/j.schres.2004.05011
- Sumiyoshi C, Sumiyoshi T, Roy A, Jayathilake K, Meltzer HY. Atypical antipsychotic drugs and organization of long-term semantic memory: multidimensional scaling and cluster analyses of category fluency performance in schizophrenia. *Int J Neuropsychopharmacol.* (2006) 9:677–83. doi: 10.1017/S1461145705006310
- Bertola L, Mota NB, Copelli M, Rivero T, Diniz BS, Romano-Silva MA, et al. Graph analysis of verbal fluency test discriminate between patients with Alzheimer's disease, mild cognitive impairment and normal elderly controls. *Front Aging Neurosci.* (2014) 6:185. doi: 10.3389/fnagi.201400185
- Landauer TK, Dumais ST. A solution to plato's problem: the latent semantic analysis theory of acquisition, induction, and representation of knowledge. *Psychol Rev.* (1997) 104:211–40. doi: 10.1037/0033-295X.104.2.211
- Sung K, Gordon B, Vannorsdall TD, Ledoux K, Pickett EJ, Pearson GD, et al. Semantic clustering of category fluency in schizophrenia examined with singular value decomposition. *J Int Neuropsychol Soc.* (2012) 18:565–75. doi: 10.1017/S1355617712000136
- Nicodemus KK, Elvevag B, Foltz PW, Rosenstein M, Diaz-Asper C, Weinberger DR. Category fluency, latent semantic analysis and schizophrenia: a candidate gene approach. *Cortex.* (2014) 55:182–91. doi: 10.1016/j.cortex.2013.12004
- Sumiyoshi C, Fujino H, Sumiyoshi T, Yasuda Y, Yamamori H, Fujimoto M, et al. Semantic memory organization in Japanese patients with schizophrenia examined with category fluency. *Front Psychiatry.* (2018) 9:87. doi: 10.3389/fpsy.201800087
- Sung K, Gordon B, Vannorsdall TD, Ledoux K, Schretlen DJ. Impaired retrieval of semantic information in bipolar disorder: a clustering analysis of category-fluency productions. *J Abnorm Psychol.* (2013) 122:624–34. doi: 10.1037/a0033068
- Yokoi Y, Sumiyoshi T. Application of transcranial direct current stimulation to psychiatric disorders: trends and perspective. *Neuropsychiatr Electrophysiol.* (2015) 1:10. doi: 10.1186/s40810-015-0012-x
- Yokoi Y, Narita Z, Sumiyoshi T. Transcranial direct current stimulation in depression and psychosis: a systematic review. *Clin EEG Neurosci.* (2018) 49:93–102. doi: 10.1177/1550059417732247
- Narita Z, Inagawa T, Sueyoshi K, Lin C, Sumiyoshi T. Possible facilitative effects of repeated anodal transcranial direct current stimulation on functional outcome 1 month later in schizophrenia: an open trial. *Front Psychiatry.* (2017) 8:184. doi: 10.3389/fpsy.2017.00184
- Narita Z, Stickley A, DeVlyder J, Yokoi Y, Inagawa T, Yamada Y, et al. Effect of multi-session prefrontal transcranial direct current stimulation on cognition in schizophrenia: a systematic review and meta-analysis. *Schizophr Res.* (2019) 216:367–73. doi: 10.1016/j.schres.2019.11.011
- Vannorsdall TD, Schretlen DJ, Andrejczuk M, Ledoux K, Bosley LV, Weaver JR, et al. Altering automatic verbal processes with transcranial direct current stimulation. *Front Psychiatry.* (2012) 3:73. doi: 10.3389/fpsy.2012.00073
- Cattaneo Z, Pisoni A, Papagno C. Transcranial direct current stimulation over Broca's region improves phonemic and semantic fluency in healthy individuals. *Neuroscience.* (2011) 183:64–70. doi: 10.1016/j.neuroscience.2011.03.058
- Hashimoto R, Ikeda M, Ohi K, Yasuda Y, Yamamori H, Fukumoto M, et al. Genome-wide association study of cognitive decline in schizophrenia. *Am J Psychiatry.* (2013) 170:683–4. doi: 10.1176/appi.ajp.2013.120.9.1228
- Ohi K, Hashimoto R, Ikeda M, Yamamori H, Yasuda Y, Fujimoto M, et al. Glutamate networks implicate cognitive impairments in schizophrenia: genome-wide association studies of 52 cognitive phenotypes. *Schizophr Bull.* (2015) 41:909–18. doi: 10.1093/schbul/sbu171
- Koshiyama D, Fukunaga M, Okada N, Yamashita F, Yamamori H, Yasuda Y, et al. Subcortical association with memory performance in schizophrenia: a structural magnetic resonance imaging study. *Transl Psychiatry.* (2018) 8:20. doi: 10.1038/s41398-017-0069-3
- Yasuda Y, Okada N, Nemoto K, Fukunaga M, Yamamori H, Ohi K, et al. Brain morphological and functional features in cognitive subgroups of schizophrenia. *Psychiatry Clin Neurosci.* (2020) 74:191–203. doi: 10.1111/pcn.12963
- Narita Z, Inagawa T, Maruo K, Sueyoshi K, Sumiyoshi T. Effect of transcranial direct current stimulation on functional capacity in schizophrenia: a study protocol for a randomized controlled trial. *Front Psychiatry.* (2017) 8:233. doi: 10.3389/fpsy.2017.00233
- Smith RC, Boules S, Mattiuz S, Youssef M, Tobe RH, Serhsen H, et al. Effects of transcranial direct current stimulation (tDCS) on cognition, symptoms, and smoking in schizophrenia: a randomized controlled study. *Schizophr Res.* (2015) 168:260–6. doi: 10.1016/j.schres.2015.06.011
- Brunoni AR, Amadera J, Berbel B, Volz MS, Rizzerio BG, Fregni F. A systematic review on reporting and assessment of adverse effects associated with transcranial direct current stimulation. *Int J Neuropsychopharmacol.* (2011) 14:1133–45. doi: 10.1017/S1461145710001690
- Spreen OS, Strauss E. *A Compendium of Neuropsychological Tests*. New York, NY: Oxford University Press (1998).
- Matsuoka K, Uno M, Kasai K, Koyama K, Kim Y. Estimation of premorbid IQ in individuals with Alzheimer's disease using Japanese ideographic script (Kanji) compound words: Japanese version of

- national adult reading test. *Psychiatry Clin Neurosci.* (2006) 60:332–9. doi: 10.1111/j.1440-1819.2006.01510.x
38. Kay SR, Opler LA, Lindenmayer JP. Reliability and validity of the positive and negative syndrome scale for schizophrenics. *Psychiatry Res.* (1988) 23:99–110. doi: 10.1016/0165-1781(88)90038-8
 39. Sung K, Gordon B, Yang S, Schretlen DJ. Evidence of semantic clustering in letter-cued word retrieval. *J Clin Exp Neuropsychol.* (2013) 35:1015–23. doi: 10.1080/13803395.2013.845141
 40. Meng XL, Rosenthal R, Rubin DB. Comparing correlated correlation coefficients. *Psychol Bull.* (1992) 111:172–5. doi: 10.1037/0033-2909.111.1.172
 41. R. R version 3.2.2 (2015-08-14) – “Fire Safety”. The R Foundation for Statistical Computing Platform: x86_64-w64-mingw32/x64 (64-bit) (2006).
 42. Wild F. *Latent Semantic Analysis*. 0.73.1 ed (2015).
 43. Diedenhofen B, Musch J. cocor: a comprehensive solution for the statistical comparison of correlations. *PLoS ONE.* (2015) 10:e0121945. doi: 10.1371/journal.pone.0121945
 44. Quesada J. Creating your own LSA spaces. In: Landauer TK, McNamara DS, Dennis S, Kintsch W, editors. *Handbook of Latent Semantic Analysis*. Mahwah, NJ: LEA (2007). p. 71–88.
 45. Mota NB, Sigman M, Cecchi G, Copelli M, Ribeiro S. The maturation of speech structure in psychosis is resistant to formal education. *NPJ Schizophr.* (2018) 4:25. doi: 10.1038/s41537-018-0067-3
 46. Cokal D, Sevilla G, Jones WS, Zimmerer V, Deamer F, Douglas M, et al. The language profile of formal thought disorder. *NPJ Schizophr.* (2018) 4:18. doi: 10.1038/s41537-018-0061-9
 47. Rossell SL, David AS. Are semantic deficits in schizophrenia due to problems with access or storage? *Schizophr Res.* (2006) 82:121–34. doi: 10.1016/j.schres.2005.11.001
 48. Nohara S, Suzuki M, Kurachi M, Yamashita I, Matsui M, Seto H, et al. Neural correlates of memory organization deficits in schizophrenia. A single photon emission computed tomography study with 99mTc-ethyl-cysteinate dimer during a verbal learning task. *Schizophr Res.* (2000) 42:209–22. doi: 10.1016/S0920-9964(99)00131-0
 49. Joyal M, Fecteau S. Transcranial direct current stimulation effects on semantic processing in healthy individuals. *Brain Stimul.* (2016) 9:682–91. doi: 10.1016/j.brs.2016.05.003
 50. Price AR, McAdams H, Grossman M, Hamilton RH. A meta-analysis of transcranial direct current stimulation studies examining the reliability of effects on language measures. *Brain Stimul.* (2015) 8:1093–100. doi: 10.1016/j.brs.2015.06.013
 51. Elvevag B, Foltz PW, Rosenstein M, Delisi LE. An automated method to analyze language use in patients with schizophrenia and their first-degree relatives. *J Neurolinguistics.* (2010) 23:270–84. doi: 10.1016/j.jneuroling.2009.05.002
 52. Garrard P, Elvevag B. Language, computers and cognitive neuroscience. *Cortex.* (2014) 55:1–4. doi: 10.1016/j.cortex.2014.02.012
 53. Elvevag B, Cohen AS, Wolters MK, Whalley HC, Gountouna VE, Kuznetsova KA, et al. An examination of the language construct in NIMH's research domain criteria: time for reconceptualization! *Am. J Med Genet B Neuropsychiatr Genet.* (2016) 171:904–19. doi: 10.1002/ajmg.b.32438
 54. Cuthbert BN, Insel TR. Toward new approaches to psychotic disorders: the NIMH research domain criteria project. *Schizophr Bull.* (2010) 36:1061–2. doi: 10.1093/schbul/sbq108
 55. Carey S. *Conceptual Change in Childhood*. Cambridge, MA: Bradford Books, MIT Press (1985).
 56. Howard DV, Howard JH. A multidimensional scaling analysis of the development on animal names. *Dev Psychol.* (1977) 13:108–13. doi: 10.1037/0012-1649.13.2.108
 57. Storm C. The semantic structure of animal terms: a developmental study. *Int J Behav Dev.* (1980) 3:381–407. doi: 10.1177/016502548000300403
 58. Crowe SJ, Prescott TJ. Continuity and change in the development of category structure: Insights from the semantic fluency task. *Int J Behav Dev.* (2003) 27:467–79. doi: 10.1080/01650250344000091

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2021 Sumiyoshi, Narita, Inagawa, Yamada, Sueyoshi, Hasegawa, Shirama, Hashimoto and Sumiyoshi. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.



Neurobiological After-Effects of Low Intensity Transcranial Electric Stimulation of the Human Nervous System: From Basic Mechanisms to Metaplasticity

Sohaib Ali Korai¹, Federico Ranieri², Vincenzo Di Lazzaro³, Michele Papa^{1,4} and Giovanni Cirillo^{1,2*}

¹ Division of Human Anatomy – Laboratory of Neuronal Networks, University of Campania “Luigi Vanvitelli”, Naples, Italy,

² Neurology Unit, Department of Neurosciences, Biomedicine and Movement Sciences, University of Verona, Verona, Italy,

³ Neurology, Neurophysiology and Neurobiology Unit, University Campus Bio-Medico, Rome, Italy, ⁴ ISBE Italy, SYSBIO Centre of Systems Biology, Milan, Italy

OPEN ACCESS

Edited by:

Carol Di Perri,
University of Edinburgh,
United Kingdom

Reviewed by:

Antonio Suppa,
Sapienza University of Rome, Italy
Carlo Cavaliere,
Institute of Research and Medical
Care (IRCCS) SDN, Italy

*Correspondence:

Giovanni Cirillo
giovanni.cirillo@unicampania.it

Specialty section:

This article was submitted to
Neurorehabilitation,
a section of the journal
Frontiers in Neurology

Received: 27 July 2020

Accepted: 04 January 2021

Published: 15 February 2021

Citation:

Korai SA, Ranieri F, Di Lazzaro V,
Papa M and Cirillo G (2021)
Neurobiological After-Effects of Low
Intensity Transcranial Electric
Stimulation of the Human Nervous
System: From Basic Mechanisms to
Metaplasticity.
Front. Neurol. 12:587771.
doi: 10.3389/fneur.2021.587771

Non-invasive low-intensity transcranial electrical stimulation (tES) of the brain is an evolving field that has brought remarkable attention in the past few decades for its ability to directly modulate specific brain functions. Neurobiological after-effects of tES seems to be related to changes in neuronal and synaptic excitability and plasticity, however mechanisms are still far from being elucidated. We aim to review recent results from *in vitro* and *in vivo* studies that highlight molecular and cellular mechanisms of transcranial direct (tDCS) and alternating (tACS) current stimulation. Changes in membrane potential and neural synchronization explain the ongoing and short-lasting effects of tES, while changes induced in existing proteins and new protein synthesis is required for long-lasting plastic changes (LTP/LTD). Glial cells, for decades supporting elements, are now considered constitutive part of the synapse and might contribute to the mechanisms of synaptic plasticity. This review brings into focus the neurobiological mechanisms and after-effects of tDCS and tACS from *in vitro* and *in vivo* studies, in both animals and humans, highlighting possible pathways for the development of targeted therapeutic applications.

Keywords: transcranial direct current stimulation, transcranial alternating current stimulation, neurobiological after-effects, synaptic plasticity, non-invasive brain stimulation

INTRODUCTION

In the last two decades, therapeutic efficacy of non-invasive transcranial brain stimulation techniques through low-intensity electrical fields has been demonstrated by a number of works and clinical trials providing promising results for many neurological disorders, including stroke (1) and epilepsy (2, 3), movement disorders/Parkinson's (PD) (4) and Alzheimer's (AD) (5, 6). Due to non-invasiveness and transient side effects (7), transcranial electrical stimulation (tES) has found progressively a wide field of applications. Moreover, acquisition of recent experimental data has extended our knowledge of the cellular and molecular mechanisms involved in the after-effects of tES, thus supporting its therapeutic potential for brain disorders based on impaired synaptic plasticity (2).

The basic principle of tES is very simple and based on the negative (anodal) and positive (cathodal) currents and their flow into the brain (8). However, neurobiological mechanisms and after-effects are not yet fully understood. Experimental evidence has demonstrated that weak low-intensity ES (at an intensity lower than that needed for triggering action potentials) induces polarity-specific changes in spontaneous and evoked neuronal activity (9, 10): anodal polarization increases neuronal activity, whereas cathodal polarization decreases it (11–14). Accordingly, transcranial direct current stimulation (tDCS) has been shown to induce long-lasting and polarity-specific changes of human motor cortex excitability (15–17) related to modifications of synaptic efficacy similar to those underlying long-term potentiation (LTP) and long-term depression (LTD) of synaptic activity (18, 19). Studies of the effects of direct current stimulation (DCS) in slices of mouse primary motor cortex have shown that anodal DCS, in the absence of simultaneous synaptic activation, does not induce LTP/LTD like changes but it can modulate LTP induction (20). In contrast, by coupling DCS with low frequency stimulation (at 0.1 Hz), a long-lasting polarity- (anodal DCS) and frequency- specific LTP is obtained, mainly depending on N-methyl-D- aspartate (NMDA) receptor activation and secretion of brain-derived neurotrophic factor (BDNF) (21). In summary, these studies highlight the complex nature of tDCS effects, characterized by the capability of inducing and modulating LTP/LTD. However, while the immediate effects of tES can be explained by changes in transmembrane potential influencing neuronal firing, it is plausible that the long-term after-effects are likely due to modifications of intracellular calcium dynamics and mechanisms of synaptic plasticity, based on LTP/LTD processes (18, 22, 23) and/or induction of metaplasticity, the activity-dependent physiological changes that modulate neural plasticity (24). Anodal tDCS, for example, induces neurotrophic BDNF-mediated priming after-effects on synaptic plasticity and memory, making synapses susceptible to LTP induction in the rat hippocampus (25).

This work aims to comprehensively summarize the neurobiological mechanisms of tES and discuss future clinical applications. In particular, we first analyzed the technical aspects of electrical stimulation techniques, and then the neurobiological after-effects of tES on the constituents of the synaptic structure, distinguishing those on membrane polarity, neural transmission, synaptic plasticity, neuronal network and connectivity, and finally the effects on glial cells and neuroinflammation.

We believe that understanding the basis of the modulatory effect of tES would be particularly relevant for its clinical application in humans, where it could be used to shape the plastic properties of the brain.

TECHNICAL ASPECTS: TRANSCRANIAL CURRENT AND MAGNETIC STIMULATION

According to whether direct or alternating current is applied to the brain, the method is referred to as either transcranial

direct current stimulation (tDCS) or transcranial alternating current stimulation (tACS). Both techniques produce effects on cortical excitability outlasting the stimulation, up to 3 h with tDCS (26) and up to 1 h with high-frequency tACS (27–29). TDCS acts in a polarity-dependent fashion, with anodal stimulation increasing and cathodal stimulation decreasing neuronal excitability, whereas tACS consists in the application of a sinusoidal waveform current that alternates between the anode and the cathode (*switching polarity*) and modulates the power of oscillatory rhythms in a frequency-dependent manner by synchronizing or desynchronizing neuronal networks (30). For example, in studies that coupled transcranial magnetic stimulation (TMS) with ES, tACS was found to synchronize cortical networks bursting at frequencies higher than 300 Hz (31).

The association between the type of stimulation and neural response depends on many physical properties including the electrode type, length, strength, and frequency of stimulation (32). Low-intensity, constant, or non-constant currents are used for tDCS and delivered in rectangular or sinusoidal waves with pulses of unidirectional current, whilst non-constant current is used for tACS (33). TDCS flows into the brain from a battery-powered generator through a couple of sponge electrodes, with one or both the electrodes fixed over the scalp. It has been demonstrated that current density (i.e., current intensity/electrode size), duration, polarity, and location of stimulating electrodes have important implications in the modulatory outcome of stimulation (34). Generally, tDCS does not involve synaptic effects but polarity changes of the membrane resting potential, does not induce neuronal firing but rather modulates spontaneous neuronal network activity, polarizing brain tissue (35–37). The two types of stimulation, anodal and cathodal, do not contrast each other in terms of after-effects and modulation of their intensity dramatically produces different results. Generally, the cortical excitability is increased by anodal tDCS while it is decreased by the cathodal tDCS over the same area (site specificity).

TACS is a non-constant current which alternates its pulses with the opposite amplitude (38, 39). Despite site specificity, its effects are not site limited as tACS influences other areas of the brain through interneuronal circuits (33) and directly interferes with ongoing brain oscillations (40). TACS shares the same setup of tDCS: it is applied between electrodes placed over the target scalp sites, with intensity in the same range of 1–2 mA. The physiological bases of tACS are less explored than tDCS. The main biophysical (electric field strength and spatial distribution) and polarizing properties of tDCS should also apply to tACS, with the main difference that the polarity (i.e., the direction of current flow) changes of 180° during each cycle of the sinusoidal waveform of tACS and that the maximum current flow is present only at the peak of the alternating current.

The advantage of tACS is that it allows the manipulation of amplitude, frequency, and coherence of intrinsic neuronal oscillations (41, 42). In addition, the effects of tACS could be translated into whole larger brain-network activity through five different neuronal mechanisms (43, 44): (1) *stochastic resonance*, consisting in the stochastic response of tACS-affected neurons to be either polarized or hyperpolarized; (2) *rhythm resonance*,

Abbreviations: tES, transcranial electrical stimulation.

synchronizing tACS frequency with the endogenous oscillations; (3) *temporal biasing of spikes*, a synergistically excitation of the same groups of neurons during each cycle of stimulation; (4) *network entrainment* of an endogenous irregular neuronal activity that necessitates an external current with sufficiently stronger amplitude; (5) *imposed pattern*, tACS overcomes endogenous regular oscillations and introduce a new oscillation. These mechanisms attribute the large-scale effects of tACS to two synergistic phenomena: entrainment and neuroplasticity, respectively. The first takes place when an external rhythmic system affects another one, forcing it to follow its own oscillating frequency and phase; the second, through LTP/LTD phenomena, elicits offline tACS effects by increasing or decreasing neural synchronization, as confirmed by many studies (29, 45–47).

TACS has diverse modes of administration in terms of frequency: the beta (20 Hz), alpha (10–12 Hz), and gamma range (40 Hz), each producing diverse neurobiological effects for modulation of different bands of neural oscillations (42). The effects of alpha and gamma stimulation have been studied on attention with gamma stimulation demonstrating to facilitate endogenous attention (48).

Experimental and clinical applications of transcranial magnetic stimulation (TMS) is widely and progressively increased over the past two decades. In particular, several repetitive TMS (rTMS) protocols have been proved to modulate brain functions (from the molecular to the network scale) and human behavior (49, 50). For example, application of simple rTMS to a target cortical area for several minutes induces after-effects in a frequency- dependent manner (low frequency, ≤ 1 Hz, reduces cortical excitability whereas high-frequency, > 5 Hz, does the opposite) (51) while theta-burst stimulation (TBS), a patterned protocol, induces longer-lasting effects with shorter application time (continuous TBS has primarily an inhibitory effect on corticospinal excitability, while intermittent TBS has an excitatory effect) (52).

TMS shares fundamental similarities with tES as both share neurobiological modulations at similar levels and involve rapid changes in magnetic fields (53). While TMS requires passing of current through coils to generate a magnetic field that in turn generates an electric field and a current density, in tES the electric field and the current density are proportional to injected current (54).

NEUROBIOLOGICAL AFTER-EFFECTS OF CURRENT STIMULATION OF CENTRAL NERVOUS SYSTEM

Effects on Membrane Polarity

Table 1 summarizes the results of the studies that analyzed the effects of tES on membrane polarity. Evidence has demonstrated that tDCS can modify neuronal membrane polarity and therefore the action potential generation (15, 19, 55) through activation of voltage-gated pre and postsynaptic Na^+ and Ca^{2+} channels thus causing increased presynaptic release of excitatory neurotransmitters and postsynaptic calcium influx, respectively

TABLE 1 | tES after-effects on membrane polarity.

References/Study	Methodology tES	Targets	Main results
Nitsche and Paulus (15); Liebetanz (19); Stagg and Nitsche (55)	tDCS	Pre/post synaptic Na^+ and Ca^{2+} channels	tDCS generates action potential via Na^+ and Ca^{2+} channels by increasing presynaptic release of excitatory transmitters and Ca^{2+} influx
Zaghi et al. (33); Bikson et al. (56)	tDCS	Hippocampal neurons	Somatic polarization was obtained with electric field parallel to somato-dendritic axis in hippocampal neurons
Bikson et al. (56); Arlotti et al. (57); Rahman et al. (58); Pelletier and Cicchetti (32); Seo and Jun (59)	tDCS - aDCS - cDCS	Structural components of neurons	Components at the cathode depolarize while those at the anode hyperpolarize
Francis et al. (60); Deans et al. (61); Reato et al. (62)	tACS	Neuronal resonance	tACS can induce cumulative effects over multiple cycles that can shift in spike timing.
Bindman et al. (11); Bikson et al. (56); Antal and Herrmann (63)	tDCS - aDCS - cDCS	Transmembrane potentials	Constant electric field shifts neuronal transmembrane potential to less negative in cDCS and more negative in aDCS which makes it more prone to generate action potential.

tES, transcranial electrical stimulation; **tDCS**, transcranial direct current stimulation; **tACS**, transcranial alternating current stimulation; **a/c tDCS**, anodal/cathodal transcranial direct current stimulation.

(15). Moderate but prolonged intracellular Ca^{2+} increase causes LTD while short but large Ca^{2+} increase causes LTP (64).

The polarity-dependent effect of tDCS is strictly dependent on the orientation of axons and dendrites (33). Specifically, when the effect of polarity was studied *in vitro* on hippocampal neurons (56), somatic polarization was obtained with the electric field parallel to the somato-dendritic axis, while an effect on afferents without somatic polarization was produced by the electric field perpendicular to the apical-dendritic axis. Moreover, the structural components of the cell at the cathode depolarize while the elements facing the anode are subject to hyperpolarization (32, 56–59). On the other hand, tACS, matching resonant neuronal properties, can induce cumulative effects over multiple cycles that may cause shift in spike timing (60–62).

However, these biophysical properties might produce complex modulatory effects when tES is applied to circuits of the human brain with no uniform spatial orientations. Based on experimental studies (11, 56), the applied constant electric field shifts the transmembrane potential of neurons toward less negative (anodal stimulation) or more negative values (cathodal

stimulation), thus increasing or decreasing the likelihood of generation of action potentials (63), thus influencing both spontaneous and evoked neuronal firing.

Effects on Neural Transmissions

Many studies have shown that tACS interferes with several neurotransmitter systems. The balance between cholinergic and adrenergic system after administration of reserpine (an anti-adrenergic drug that irreversibly blocks the H⁺-coupled vesicular monoamine transporters—VMAT) and physostigmine (a parasympathomimetic reversible cholinesterase inhibitor) occurred much faster while applying tACS: it was observed that the quantity of presynaptic vesicles first declined, then increased after 5 min and then returned to baseline levels after tACS (65). Evidence suggested that this type of stimulation might modulate the serotonergic raphe nuclei, the noradrenergic locus coeruleus, the cholinergic latero-dorsal tegmental, and pedunculo pontine nuclei in the brainstem (66). Additionally, tACS was found to modulate the levels of endorphins into the cerebrospinal fluid (67) and naloxone, a pure opioid antagonist, was reported to reduce tACS analgesic effects (67), prompting to hypothesize a tACS-induced modulation of the neurotransmitters' release.

The blockage of serotonin reuptake increases LTP in the motor cortex by anodal tDCS and shifts LTD to LTP after cathodal tDCS (68). In addition, anodal tDCS was demonstrated to reduce γ -aminobutyric acid (GABA) concentration in the stimulated cerebral cortex while cathodal tDCS impaired glutamatergic neuronal activity and reduced GABA concentration (2, 69). Authors argue that these protocols might be used therapeutically to reduce the imbalance between excitatory and inhibitory transmitters (70, 71). These results were also confirmed in humans by magnetic resonance spectroscopy (MRS) studies examining the effects of tDCS on the hand area of the primary motor cortex. Accordingly, authors reported that anodal tDCS causes GABA decrease while cathodal tDCS decreases both the levels of glutamate and GABA (70). Upon administration of GABA antagonists, anodal tDCS produces delayed but enhanced excitability increase in cortical or subcortical areas (72). See **Table 2** for a summary of the studies that analyzed the effects of tES on neural transmissions.

Effects on Synaptic Plasticity

Experimental and human studies suggest that the after-effects of tES might originate from persistent modifications of synaptic efficacy similar to those underlying LTP and LTD of synaptic activity (18, 19, 73). Synaptic plasticity usually involves short- and long-term modifications of existing synapses (formation, removal, and remodeling of synapses and dendritic spines) that in turn modify the activity of brain networks in which they are interposed (50). Mechanisms of synaptic plasticity occur at different levels, from ultrastructural to synapse: calcium dynamics, neurotransmitter release, proteins (receptors, transporters, and ion channels) and gene expression (74). **Table 3** summarizes the main results of the studies that analyzed the tES after-effects on synaptic plasticity.

TABLE 2 | tES after-effects on neural transmission.

References/Study	Methodology tES	Targets	Main results
Kirsch and Nichols (65)	tACS	Cholinergic and adrenergic neural transmission	After administration of reserpine and physostigmine and administration of tACS, the quantity of presynaptic vesicles declines and then increased
Nitsche et al. (68)	tDCS - aDCS - cDCS	Motor cortex	Blockage of serotonin reuptake increases LTP via aDCS and shifts LTD to LTP after cDCS
Stagg et al. (70); Nitsche et al. (72)	tDCS - aDCS - cDCS	GABA and glutamate in cortical and subcortical areas	aDCS reduces GABA while cDCS reduces both glutamate and GABA. With GABA antagonists, aDCS produced enhanced excitability in cortical and subcortical areas

tES, transcranial electrical stimulation; **tDCS**, transcranial direct current stimulation; **tACS**, transcranial alternating current stimulation; **a/c tDCS**, anodal/cathodal transcranial direct current stimulation; **LTP**, long-term potentiation; **LTD**, long-term depression; **GABA**, gamma amino butyric acid.

Experimental evidence using a high frequency pre-synaptic stimulation protocol has showed a polarity-specificity of tDCS in the modulation of LTP induction, with anodal stimulation increasing and cathodal stimulation decreasing the amount of LTP (20). These data suggest that tDCS alone is not capable of changing synaptic strength (i.e., inducing LTP), but rather that tDCS changes the propensity of the synapse to undergo LTP. Accordingly, in the study by Fritsch and colleagues, LTP was obtained after a conditioning anodal tDCS protocol but only in the presence of concomitant synaptic activation by presynaptic inputs (21).

Neurotrophins (BDNF, NGF, NT-3, and NT-4/5) are a large family of complex proteins that regulate several functions, including neuronal survival, differentiation, synaptic function, and plasticity but also neuronal death through interaction with two types of receptors, the tyrosine kinase receptors (TrkA, TrkB, and TrkC) and the common p75NTR receptor (82). Most of neurotrophins, including BDNF, is secreted in an immature form and then converted into the mature, active form by a complex fine-regulated system of proteases (83–85). With this premise, it has been demonstrated that tDCS might increase BDNF concentration when combined with presynaptic stimulation (21) inducing LTP via BDNF/TrkB signaling (25). TrkB stimulation by BDNF also promotes long-lasting synaptic potentiation and late phase LTP requires the conversion of pro-BDNF into mature BDNF in the hippocampus (21). Moreover, enhanced LTP in animals undergoing continuous tDCS can be reduced by TrkB antagonist (86) and anodal tDCS enhances hippocampal LTP and memory via chromatin remodeling of the *Bdnf* gene regulatory

TABLE 3 | tES after-effects on synaptic plasticity.

References/Study	Methodology tES	Targets	Main results
Ranieri et al. (20)	tDCS - cDCS - aDCS	Neuronal LTP	aDCS increased LTP while cDCS decreased LTP
Fritsch et al. (21); Yu et al. (25)	tDCS	BDNF/TrkB signaling	tDCS increases BDNF concentration which induces LTP. TrkB stimulation by BDNF promotes late phase LTP
Lanté et al. (75); Luscher and Malenka (76)	tDCS	NMDA/AMPA receptors	High frequency stimulation induced LTP in active NMDA receptors, expression of AMPA receptors in postsynaptic neuron and Ca^{2+} rise. Low frequency stimulation induces small rise in Ca^{2+} and presynaptic internalization of AMPA by phosphatase activation and LTD generation
Mycielska and Djamgoz (77); McCaig et al. (78)	tDCS	Cellular migration	tDCS modified the speed and direction of cell migration by shifting intracellular Ca^{2+} and modifying expression of EGFR due to electrostatic effects
Monte-Silva et al. (79); Kuo et al. (80)	tDCS - cDCS - aDCS	L-DOPA induced plastic changes	Anodal L-DOPA suppressed plasticity induced by aDCS while prolonged the reduction of excitability by cDCS
Hurley and Machado (6)	tDCS	Neuronal polarity	When synaptic activity is preconditioned by tDCS, continuous tDCS after interval will modulate polarity
Carvalho et al. (81)	tDCS - aDCS - cDCS	Working memory	Continuous aDCS facilitates performance and cDCS enhances working memory
Zaehle et al. (45)	tACS	Rhythmic patterns and natural pattern	tACS modulates neural synchronization by increasing or decreasing it and induces LTP and LTD

tES, transcranial electrical stimulation; **tDCS**, transcranial direct current stimulation; **tACS**, transcranial alternating current stimulation; **a/c tDCS**, anodal/cathodal transcranial direct current stimulation; **LTP**, long-term potentiation; **LTD**, long-term depression; **BDNF**, brain-derived neurotrophic factor; **TrkB**, tyrosine kinase receptor B; **NMDA**, N-methyl-D- aspartate; **AMPA**, alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid.

sequence, increasing the expression of this gene (87). In addition, through TrkB/Fyn signaling, BDNF induces a phosphorylation-dependent enhancement of NMDA receptor activity that further enhances effects of tDCS on LTP (88, 89).

The most prominent phenomena mediating LTP/LTD are the functional state of the synapse, Ca^{2+} signals and activity of NMDA glutamate receptors (74) (**Figure 1**). High-frequency current stimulation, in fact, induces LTP only in active synapses, which express active/open NMDA receptors, rapid expression of alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) receptors in the postsynaptic neuron, and fast intracellular Ca^{2+} increase (90). In contrast, low-frequency, long-lasting stimulation induces small and slow rise in Ca^{2+} concentration, presynaptic internalization of AMPA receptors by phosphatase activation (that reduces glutamate sensitivity), and LTD generation (75, 76).

Studies have showed the tDCS induces changes in the direction and speed of cell migration which may be related to the shift of intracellular Ca^{2+} (77, 78) and to changes in the expression of the epidermal growth factor receptors' (EGFR) due to electrostatic effects of tDCS, ultimately contributing to long-term modulation (78).

The effects of tES on synaptic plasticity are also modulated by concomitant administration of drugs acting on neural transmissions. The dopaminergic, cholinergic, serotonergic systems all affect tDCS-induced plasticity (91) in a dose-dependent manner. For example, low dose administration

of the D2/D3 agonist ropinirole abolishes plasticity (91), medium dosed ropinirole reestablishes facilitatory and inhibitory plasticity, whilst high dosage decreases facilitatory plasticity (92). Administration of low dosage or high dosage of anodal L-DOPA suppressed the plasticity induced by tDCS (79), however L-DOPA prolonged the reduction of excitability induced by cathodal tDCS (80).

Induction of plasticity through tES, however, might also arise from simultaneous stimulation of the different components of the neural circuit, from the excitatory/inhibitory synapses to different brain networks, therefore, as a result, it is important to consider the main excitatory (LTP-like) or inhibitory (LTD-like) effect of the brain stimulation. Early LTP/LTD modifications usually last for 30–60 min after induction and reflect post-transcriptional modifications of pre-existing proteins, such as protein phosphorylation, in contrast late LTP/LTD could last hours, days, and even months and require genes and proteins expression (e.g., glutamate NMDA and metabotropic receptors) (50).

In order to shed light on the pathways leading to the synthesis of new proteins, attention has been focused on the group of immediate early genes (IEGs), that are rapidly induced following neuronal activation and are thought to be involved in the maintenance of LTP (93, 94). Among IEGs, zif268 is likely to be specifically related to LTP, since it is expressed under virtually all LTP-inducing situations and shows a high correlation with the duration of LTP (95). After application of

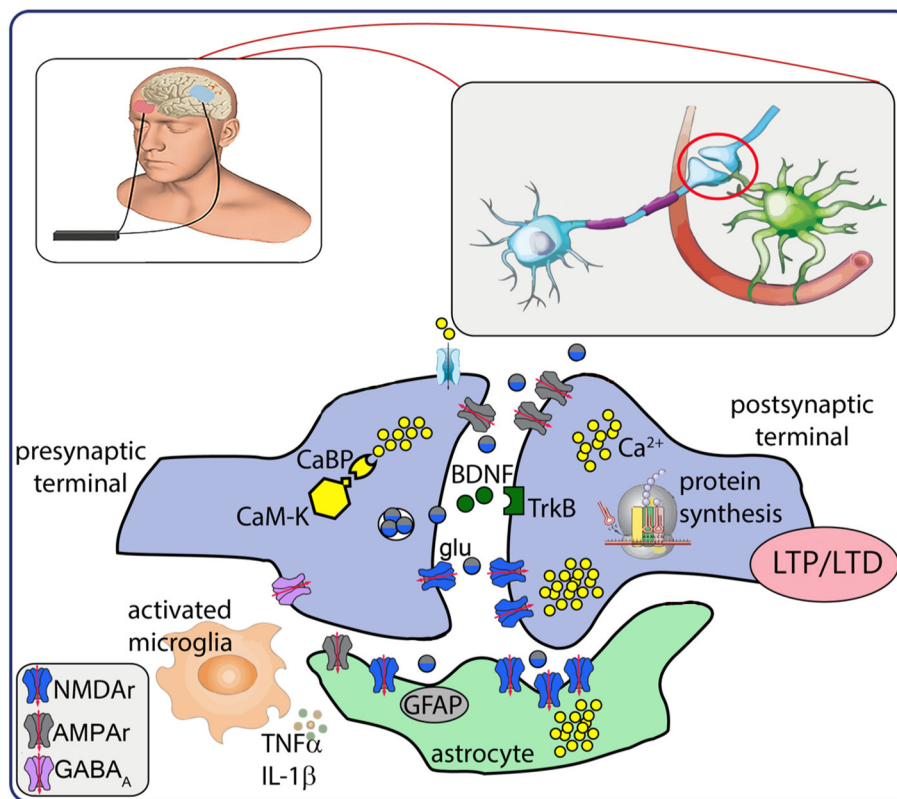


FIGURE 1 | Schematic representation of neurobiological after-effects of transcranial electrical stimulation (tES). tES induces intracellular Ca^{2+} increase and activation of Ca^{2+} -dependent enzymes (CaM-K). Presynaptic mechanisms result in glutamate release that activates AMPA/NMDA receptors, modulates BDNF release and interaction with TrkB receptor, responsible for a cascade of intracellular events that lead to *de novo* protein synthesis. Electrical stimulation also modulates activation of astrocytes and neuroinflammatory response. Altogether, these mechanisms may underlie the establishment of LTP/LTD. **CaBP**, Ca^{2+} binding proteins; **CaM-K**, Ca^{2+} kinases; **glu**, glutamate; **BDNF**, brain-derived neurotrophic factor; **TrkB**, tyrosine kinase receptor B; **LTP/LTD**, long term potentiation/depression; **GFAP**, glial fibrillary acidic protein; **TNF α** , tumor necrosis factor α ; **IL-1 β** , interleukin 1 β ; **NMDAr**, N-methyl-D- aspartate receptor; **AMPAr**, alpha-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptor; **GABA_A**, gamma amino butyric acid A receptor.

both anodal and cathodal DCS to hippocampal rat brain slices, *zif268* expression was increased, pointing to a possible initial role of *zif268* in a cascade of activation of other downstream target genes (20).

Abnormally high activity and hyperexcitability of some subcortical pathways, as in the case of after stroke or during central nervous system (CNS) development, may respond to tES that modulates homeostatic plasticity of the hyperexcitable tissue (96–99). The hyperexcitability is maintained because neurons receive deficient inputs and, in order to compensate, increase excitatory synaptic strength and intrinsic excitability (100, 101).

In addition, metaplastic changes are observed with the administration of tES (6). The term metaplasticity refers to a higher order form of plasticity and reflects the activity-dependent physiological changes that modulate neural plasticity (102). The history of synaptic or cellular activity influences the direction and degree of synaptic plasticity, favoring or inhibiting plasticity induction, synaptic stabilization, and homeostatic regulation of cellular activity (103). Therefore, metaplasticity acts to avoid excessive synaptic strengthening or weakening, to maintain a relatively stable equilibrium of the neural activity in space and time (homeostatic synaptic plasticity), adjusting the balance

between synaptic input and neuronal firing, and to prolong the time-window for associative interactions between neural events (associative plasticity) (6). Basically, any recent neural synaptic activity will affect the ongoing activity. For example, if synaptic activity is preconditioned by applying tDCS, the application of continuous tDCS after an interval will modulate polarity which will affect performance (6). Continuous anodal tDCS has shown to facilitate performances while consecutive sessions of cathodal tDCS have shown to enhance working memory (81). Preconditioning neural networks may induce synaptic homeostatic changes that seems to be related to compensatory upregulation at post-synaptic membrane receptors due to inhibition (104, 105). This has been called as the “rebound effect” where neurons are more excitable due to initial downregulation induced by cathodal tDCS and reversed by conditioning cathodal tDCS (13).

Aberrant plasticity induced by non-invasive brain stimulation techniques has been demonstrated in many neurological and neuropsychiatric disorders including PD (106–108), dystonia (109, 110), multiple sclerosis (111), ischemic stroke (112), migraine (113), AD (114), schizophrenia (115–117), and drug addiction (103, 118).

Regarding tACS, both online and offline effects have reported to generate entertainment and neuroplasticity (45). Entertainment is where external rhythmic pattern imposes itself on the intrinsic natural pattern. Neuroplastic changes have been reported via LTP and LTD as tACS modulates neural synchronization by increasing or decreasing it (45). In summary, tES-induced mechanisms of synaptic plasticity cover different aspects of the neurobiology and neurophysiology of CNS, ranging from gene and protein expression, modulation of neurotrophins activity, and neural transmission and, finally, metaplasticity.

Effects on Neuronal Networks and Connectivity

Polarization of the brain tissue can extend beyond the area under the electrodes (119–121) and it may have a functional effect also on distant interconnected neural networks (122, 123). Anodal tDCS of the premotor cortex, for example, increases the excitability of the ipsilateral motor cortex (124) and stimulation of the primary motor cortex has inhibitory effects on contralateral motor areas (125). EEG studies support these findings, showing that stimulation of frontal areas induces all-brain synchronous changes of the oscillatory activity (126, 127). Altered prefrontal oscillations and brain synchronization have been reported by magnetoencephalography (MEG) and EEG study in AD, showing functional disconnection between prefrontal cortex and hippocampus and changes of network connectivity (128–130).

Functional connectivity of cortical networks increased within motor, premotor, and somatosensory areas after anodal tDCS, inducing significant intra and interhemispheric connectivity changes, as revealed by analysis of EEG frequency bands (131).

Brain areas interact mutually creating a complex network that underlie higher brain functions and neural synchronization represents an essential system to coordinate cortico-cortical and cortico-subcortical areas (132, 133). A combined tDCS-fMRI study revealed that after active stimulation functional connectivity showed an increased synchrony in the anti-correlated network (that includes DLPFC) and reduced in the default mode network (DMN) components, thus suggesting a functional reconfiguration of intrinsic brain networks after tDCS (134). This could represent a putative mechanism for tDCS-induced improvement of cognitive functions (134). In addition, using fMRI, anodal tDCS was also shown to modulate functional connectivity of cortical (70), cortico-striatal and thalamo-cortical motor pathway (135). To better grasp the precision of tES, stochastic resonance should be underlined. The concept of stochastic resonance attempts to highlight the importance of wide range of affects due to TES. The electric field can be considered as noise and when added to non-linear systems may enhance or disrupt the state of signal and the noise introduced (136–138). Since the after-effects are not focal but global, the dynamic interactions will modulate not only particular group of neurons but also induce global effects thus affecting neurons near their discharge threshold, thus facilitating or inhibiting a

TABLE 4 | tES after-effects on neuronal networks and connectivity.

References/Study	Methodology tES	Targets	Main results
Boros et al. (124); Vines et al. (125)	tDCS - aDCS	Motor cortex	aDCS of premotor cortex increases the excitability in ipsilateral motor cortex. Stimulation of primary motor cortex has inhibitory effect on contralateral motor area
Polanía et al. (131)	tDCS	Motor/premotor/ somatosensory areas	Functional connectivity of cortical networks increased with aDCS with intra/interhemispheric connectivity changes
Peña-Gómez et al. (134)	tDCS	Default mode network and DLPFC	tDCS increased synchrony in anti-correlated network and reduced in default mode network
Stagg et al. (55)	tDCS	Cortical/cortico- striatal/thalamo- cortical motor pathways	tDCS modulates functional connectivity of cortical, cortico-striatal and thalamo-cortical motor pathways
Fertonani and Miniussi (138)	tACS/tDCS	–	tES induces stochastic resonance which affects neuronal groups and induces wide range of global effects by facilitating or inhibiting a subthreshold signal

tES, transcranial electrical stimulation; *tDCS*, transcranial direct current stimulation; *tACS*, transcranial alternating current stimulation; *a/c tDCS*, anodal/cathodal transcranial direct current stimulation; *DLPFC*, dorsolateral prefrontal cortex.

subthreshold signal which will produce two different polarized after effects (138).

See Table 4 for a summary of the main tES studies and results on neuronal networks and connectivity.

Effects on Glial Cells and Neuroinflammation

The relevance of glial biology cannot be neglected to understand the complexity of the CNS and the comprehensive mechanisms and effects of tES. The significance is clinically appealing as glial cells create a wide neuro-glial network for rapid inter-cellular long-range signaling (73) and are early affected in many CNS disorders. Although the glial cells have attracted limited interest for decades, it is only recently that studies have focused on their role in maintaining synaptic homeostasis and modulating synaptic plasticity in health and disease (139). Astrocytes and microglial cells are in close proximity with synapses as they directly modulate synapse formation and

elimination (140). The loss of integrity of these supportive cells is the trigger of neurodegenerative disorders (141–143). Initially it was believed that AD was consequentially due to A β oligomers and fibrils that accumulate and inflammation. However, now it has been demonstrated that glial cells drive the synaptic loss in AD (144–147). In addition, glial mediated synapse formation may impair synaptic turnover and homeostasis which disrupts synaptic plasticity. Reactive gliosis is a process of hypertrophy and proliferation of glial cells in response to an insult such as infection/trauma/neurodegenerative disorders (140, 148). This is preceded by release of chemokines, cytokines and neurotrophic factors that have both neuroprotective (M2-like microglia) and neuroinflammatory effect (M1-like microglia) (84). This leads to a simultaneous process of neural damage and synaptic loss with tissue remodeling and phagocytosis.

To our best knowledge, there are no reports regarding the activity of tACS on glial cells. Significant after-effects of tDCS on glial cells function and plasticity are reported by several groups in the last years (see Table 5). This is supported by the fact that astrocytes possess voltage-gated channels and transporters that are sensitive to changes of membrane potential (152, 153). Administration of tDCS has shown to cause a surge in Ca²⁺ in cortical astrocytes that is correlated to an overexpression of the glutamate NMDA receptor (154). Evidence suggests that tES modulates the activity of microglia cells but also the neuroinflammatory response, triggering both pro-inflammatory and anti-inflammatory reaction (149). Cathodal and anodal tDCS produce microglial activation as indicated by the increase of Iba-1, an immunostaining marker of activated microglia (150). High voltage anodal and cathodal tDCS was demonstrated to trigger an inflammatory response in the microglial cell line BV2, showing increase of cyclooxygenase 2 (COX-2) expression, leukocyte transmigration through blood brain barrier (32, 149). On the other hand, there was decrease of tumor necrosis factor- α (TNF- α) in rat hippocampus after anodal tDCS of parietal cortex (151). Modulation of the neuroinflammatory reaction is relevant because microglia activation can be beneficial as well as detrimental for neural tissue depending on the time of activation. This is clinically relevant in the case of ischemic stroke, because tDCS can activate innate immune response and attract neural stem cells. *In vitro* experiments suggest that cathodal tDCS, delivered for 5 days, can induce cell proliferation and attract neural crest stem cells (149), forming a reservoir of neurotrophic factors which improved functional recovery. In addition, tDCS has also been shown to influence astrocytes by aligning them perpendicular to the electrical field in both *vitro* and *in vivo* (155–157).

Due to the remarkable connectivity of astrocytes and their pivotal role in neuronal connectivity, non-invasive brain modulation may have profound neurobiological effects (158).

POTENTIAL CLINICAL APPLICATIONS OF CURRENT STIMULATION

Efficacy of tES in the clinical setting has been supported by many experimental works and clinical reports that has demonstrated

TABLE 5 | tES after-effects on glial cells and inflammation.

References/Study	Methodology tES	Targets	Main results
Rueger et al. (149)	DCS	Microglial cells	tES produces both proinflammatory and anti-inflammatory reactions
Pikhovych et al. (150)	tDCS - cDCS - aDCS	Microglial cells and Iba-1	cDCS and aDCS cause microglial activation with increase in Iba-1 markers
Rueger et al. (149); Pelletier and Cicchetti (32)	High voltage DCS - cDCS - aDCS	Microglial cell BV2	High voltage aDCS and cDCS induces activation of microglial cells BV2 with increased expression of COX-2 (cyclooxygenase 2) and leukocyte transmigration
Spezia Adachi et al. (151)	DCS - aDCS	Hippocampal neurons	aDCS of parietal cortex decreased tumor necrosis factor α (TNF- α) in the rat hippocampus
Rueger et al. (149)	DCS	Neural crest stem cells	5-day cDCS induced cell proliferation and attracted neural stem cells

tES, transcranial electrical stimulation; DCS, direct current stimulation; a/c DCS, anodal/cathodal direct current stimulation.

a long-lasting efficacy in many neurological and psychiatric conditions (5). Despite neurobiological mechanisms have not been yet fully understood, it is supposed that tES-induced modulation of cortical excitability through changes in cell firing rate could pave the way for future therapeutic applications (159).

Application of tACS in the clinical setting is very limited and largely implemented in the psychiatric settings (160, 161). Accordingly, tACS was shown to successfully manipulate auditory hallucinations in schizophrenia by decoupling interhemispheric connectivity and, when administered to schizophrenic patients to the left dorsolateral prefrontal cortex and posterior parietal region in theta frequency (6 Hz), improved working memory tasks (162). Moreover, 40 Hz tACS induced improvement/remission of symptoms in major depression (163) and obsessive compulsive disorder (164) by modulation of EEG-gamma frequency bands. Enhancement of gamma band power connectivity by tACS was also effective in patients with AD and mild cognitive impairment (165, 166).

Experimental and clinical research with tDCS has been widely explored for its ability to suppress neuronal hyperexcitability or by enhancing inhibition (167). While cathodal tDCS reduces cortical excitability due to neuronal hyperpolarization, anodal tDCS causes an increase in cortical excitability and promotes neuronal depolarization (168). These neurobiological effects might be the substrate to counteract the temporoparietal hypoactivity (atrophy, reduced metabolic rate, and perfusion)

reported in AD, suggesting an innovative therapeutic strategy (169).

In an experimental rat model of stroke, tDCS induced a dramatic increase in spine density of cortical neurons at the site of infarct, indicating that it may promote neural plasticity after stroke (170). Accordingly, tDCS was found to down-regulate the elevated hemichannel pannexin-1 mRNA expression after brain ischemia (thus reducing membrane permeability), but also increase the expression of MAP-2 and GAP-43 proteins, allowing axons to regrow at the infarcted site through the glial scar and redevelop their functions (171). Interestingly, tDCS performed within 3 days after stroke did not improve motor function, in contrast when performed 7–14 days after stroke resulted in more pronounced motor function improvement, thus identifying an optimal time-window for tDCS therapy after stroke (171).

In patients with multiple sclerosis (MS) that received tDCS, MRI detected (1) increased cerebral metabolic rate of oxygen (CMRO₂), an indicator of the overall brain/neural activity, and (2) a reduced neuronal reactivity (172).

Seizures are described as a result of an increased excitability and inefficient inhibitory control in foci with altered neuronal homeostasis (72, 173, 174). In the recent years, many works have reported the efficacy of tES in the treatment of drug-resistant seizures. Authors observed an enhanced neuronal plasticity and synaptic reorganization after tES (100). For example, it has been reported that temporal lobe epilepsy responded to tES of hippocampus (101) and low frequency tACS applied over the epileptic foci might reduce interictal and ictal activities in epileptics (175). Moreover, experimental evidence in a rat model of focal epilepsy demonstrated that cathodal tDCS has an anticonvulsant effect through increase of the localized seizure threshold that outlasted the stimulation (176). Similar results were confirmed on a refractory pediatric epileptic patient with focal cortical dysplasia who was treated with cathodal tDCS and experienced marked reduction in the frequency of seizures (177). Along with this, cathodal tDCS was reported to prevent the loss of GABAergic inhibition, which provokes seizures after pentylentetrazol administration, thus proposing a new antiepileptic mechanism (178). These results, therefore, have posed the basis to the clinical combination of the cathodal tDCS with GABA-agonist antiepileptic drugs (AEDs), such as benzodiazepines, valproic acid, felbamate, topiramate, and barbiturates, in order to increase the antiepileptic stimulation effect.

Application of tDCS is not limited to the cerebral cortex and its disorders but also for the modulation of the excitability in the cerebellum and spinal cord. Since pharmacological approaches to treat cerebellar diseases are still lacking, tES might represent a new potential therapeutic approach that is yet to be explored. The mechanisms behind the neurophysiological effects of tDCS applied over cerebellum have not been extensively researched as compared to cerebral cortex. However, it could be inferred that ionic gradient shifts, cellular activation and inhibition, modulation of neurotransmission may occur in the same way (179). Evidence suggests that cerebellar cathodal tDCS decreases the inhibitory tone of cerebellum on primary motor cortex while anodal tDCS increases it, likely through a specific

modulation of dentate-thalamo-cortical connections (21). TDCS also modulates cerebellum-dependent motor learning: anodal tDCS improved the performance in a locomotor adaptation task (180). Mechanisms need to be further explored, however it has been hypothesized that anodal tDCS may broaden the availability of Purkinje cells for learning or increase the dynamic range of these cells, whereas cathodal tDCS may reduce the excitability of Purkinje cells (181). The effects of tDCS on cerebello-motor connectivity were studied in 20 patients with ataxia with administration of cerebello-spinal tDCS (179). Improvement in ataxia was reported and was associated with restoration of motor cortex excitability and cerebellar-brain inhibition.

Application of spinal tDCS is very limited but the preliminary results are extremely interesting. It has been reported that spinal anodal tDCS reduces the amplitude of laser evoked potentials of stimulated A δ fibers (182) and increases cortico-spinal excitability in a polarity-independent manner (183). While spinal anodal tDCS inhibits the ascending pathways and enhances the reflex circuitry, the spinal cathodal tDCS enhances the activity of ascending pathways and suppresses the reflex circuitry in humans (181). Since there is involvement of the ascending and descending pathways, the glutamatergic, GABAergic and glycinergic systems should be involved in modulating the spinal plasticity (181). The effects of this kind of stimulation can vary in response to several factors including intensity, polarity and direction (184) but also through modulation of the voltage-gated Ca²⁺ channels in the spinal motor neuron dendrites (185). Altogether, these preliminary results demonstrate the ability to modulate spinal plasticity with electrical current stimulation, paving the way for new therapeutic strategies in neurological disorders with impaired spinal excitability.

CHALLENGES AND FUTURE DIRECTIONS

To date, despite the undisputed role of tES in experimental settings in humans as a tool to “switch on/off” specific brain regions that are supposed to be involved in several higher brain functions, its translation into clinical settings is still far to be reached due to the difficulty in producing clinically significant effects in the majority of subjects/patients. This is largely due to the lack of a full comprehension of both the neurobiological bases of tES and the specific neuropathological mechanisms of disease. There are still few data on the possible clinical efficacy of prolonged/repeated protocols of stimulation that might produce persistent changes in synaptic efficacy that cannot be achieved by a short-lasting intervention. In this context, successful trials of prolonged tES protocols could eventually be translated into invasive implants of cortical electrodes for chronic stimulation. Finally, tDCS shows lack of selectivity that might influence different cortical circuits and produce side effects that counteract the effects responsible for the therapeutic action. Therefore, optimizing protocols, electrode size and intensity of stimulation should help to overcome these technical limitations that impedes a tailored approach to the patient and disease.

AUTHOR CONTRIBUTIONS

SK: acquisition, analysis, and interpretation of data for the work and drafting the manuscript. FR: supervising and editing the manuscript and final approval of the manuscript to be submitted. VD: conception and design of the work, supervising and editing the manuscript, and final approval of the manuscript to be submitted. MP: critical supervision, manuscript editing, and final approval of the draft to be submitted. GC: conception and design of the work, analysis and interpretation of data, revising

the manuscript, and final approval of the manuscript to be submitted. All authors contributed to the article and approved the submitted version.

FUNDING

This work was supported by grants from the Italian Minister of University and Research (MUR) (PRIN2017-2017XJ38A4_003-to GC and MP).

REFERENCES

- Schlaug G, Renga V, Nair D. Transcranial direct current stimulation in stroke recovery. *Arch Neurol.* (2008) 65:1571–6. doi: 10.1001/archneur.65.12.1571
- Nitsche MA, Paulus W. Noninvasive brain stimulation protocols in the treatment of epilepsy: current state and perspectives. *Neurotherapeutics.* (2009) 6:244–50. doi: 10.1016/j.nurt.2009.01.003
- VanHaerents S, Chang BS, Rotenberg A, Pascual-Leone A, Shafi MM. Noninvasive brain stimulation in epilepsy. *J Clin Neurophysiol.* (2020) 37:118–30. doi: 10.1097/WNP.0000000000000573
- Wu AD, Fregni F, Simon DK, Deblieck C, Pascual-Leone A. Noninvasive Brain stimulation for Parkinson's disease and dystonia. *Neurotherapeutics.* (2008) 5:345–61. doi: 10.1016/j.nurt.2008.02.002
- Lefaucheur JP, Antal A, Ayache SS, Benninger DH, Brunelin J, Cogiamanian F, et al. Evidence-based guidelines on the therapeutic use of transcranial direct current stimulation (tDCS). *Clin Neurophysiol.* (2017) 128:56–92. doi: 10.1016/j.clinph.2016.10.087
- Hurley R, Machado L. Using tDCS priming to improve brain function: can metaplasticity provide the key to boosting outcomes? *Neurosci Biobehav Rev.* (2017) 83:155–9. doi: 10.1016/j.neubiorev.2017.09.029
- Matsumoto H, Ugawa Y. Adverse events of tDCS and tACS: A review. *Clin Neurophysiol Pract.* (2017) 2:19–25. doi: 10.1016/j.cnp.2016.12.003
- Ritaccio AL, Brunner P, Schalk G. Electrical stimulation mapping of the brain: basic principles and emerging alternatives. *J Clin Neurophysiol.* (2018) 35:86–97. doi: 10.1097/WNP.0000000000000440
- Priori A. Brain polarization in humans: a reappraisal of an old tool for prolonged non-invasive modulation of brain excitability. *Clin Neurophysiol.* (2003) 114:589–95. doi: 10.1016/S1388-2457(02)00437-6
- Zago S, Ferrucci R, Fregni F, Priori A. Bartholow, sciamanna, alberti: pioneers in the electrical stimulation of the exposed human cerebral cortex. *Neuroscientist.* (2008) 14:521–8. doi: 10.1177/1073858407311101
- Bindman LJ, Lippold OCJ, Redfearn JWT. Long-lasting changes in the level of the electrical activity of the cerebral cortex produced by polarizing currents. *Nature.* (1962) 196:584–5. doi: 10.1038/196584a0
- Bindman LJ, Lippold OCJ, Redfearn JWT. The action of brief polarizing currents on the cerebral cortex of the rat (1) during flow and (2) in the production of long-lasting effects. *J Physiol.* (1964) 172:369–82. doi: 10.1113/jphysiol.1964.sp007425
- Creutzfeldt OD, Fromm GH, Kapp H. Influence of transcortical d-c currents on cortical neuronal activity. *Exp Neurol.* (1962) 5:436–52. doi: 10.1016/0014-4886(62)90056-0
- Purpura DP, McMurtry JG. Intracellular activities and evoked potential changes during polarization of motor cortex. *J Neurophysiol.* (1965) 28:166–85. doi: 10.1152/jn.1965.28.1.166
- Nitsche MA, Paulus W. Sustained excitability elevations induced by transcranial DC motor cortex stimulation in humans. *Neurology.* (2001) 57:1899–901. doi: 10.1212/WNL.57.10.1899
- Lang N, Nitsche MA, Paulus W, Rothwell JC, Lemon RN. Effects of transcranial direct current stimulation over the human motor cortex on corticospinal and transcallosal excitability. *Exp Brain Res.* (2004) 156:439–43. doi: 10.1007/s00221-003-1800-2
- Lang N, Nitsche MA, Dileone M, Mazzone P, Andr s-Ar s J, Diaz-Jara L, et al. Transcranial direct current stimulation effects on I-wave activity in humans. *J Neurophysiol.* (2011) 105:2802–10. doi: 10.1152/jn.00617.2010
- Islam N, Aftabuddin M, Moriwaki A, Hattori Y, Hori Y. Increase in the calcium level following anodal polarization in the rat brain. *Brain Res.* (1995) 684:206–8. doi: 10.1016/0006-8993(95)00434-R
- Liebetanz D. Pharmacological approach to the mechanisms of transcranial DC-stimulation-induced after-effects of human motor cortex excitability. *Brain.* (2002) 125:2238–47. doi: 10.1093/brain/awf238
- Ranieri F, Podda MV, Riccardi E, Frisullo G, Dileone M, Profice P, et al. Modulation of LTP at rat hippocampal CA3-CA1 synapses by direct current stimulation. *J Neurophysiol.* (2012) 107:1868–80. doi: 10.1152/jn.00319.2011
- Fritsch B, Reis J, Martinowich K, Schambra HM, Ji Y, Cohen LG, et al. Direct current stimulation promotes BDNF-dependent synaptic plasticity: potential implications for motor learning. *Neuron.* (2010) 66:198–204. doi: 10.1016/j.neuron.2010.03.035
- Albensi BC, Oliver DR, Toupin J, Otero G. Electrical stimulation protocols for hippocampal synaptic plasticity and neuronal hyperexcitability: are they effective or relevant? *Exp Neurol.* (2007) 204:1–13. doi: 10.1016/j.expneurol.2006.12.009
- Polan a R, Nitsche MA, Ruff CC. Studying and modifying brain function with non-invasive brain stimulation. *Nat Neurosci.* (2018) 21:174–87. doi: 10.1038/s41593-017-0054-4
- Cooper LN, Bear MF. The BCM theory of synapse modification at 30: Interaction of theory with experiment. *Nat Rev Neurosci.* (2012) 13:798–810. doi: 10.1038/nrn3353
- Yu TH, Wu YJ, Chien ME, Hsu KS. Transcranial direct current stimulation induces hippocampal metaplasticity mediated by brain-derived neurotrophic factor. *Neuropharmacology.* (2019) 144:358–67. doi: 10.1016/j.neuropharm.2018.11.012
- Di Lazzaro V, Manganelli F, Dileone M, Notturmo F, Esposito M, Capasso M, et al. The effects of prolonged cathodal direct current stimulation on the excitatory and inhibitory circuits of the ipsilateral and contralateral motor cortex. *J Neural Transm.* (2012) 119:1499–506. doi: 10.1007/s00702-012-0845-4
- Moliadze V, Antal A, Paulus W. Boosting brain excitability by transcranial high frequency stimulation in the ripple range. *J Physiol.* (2010) 588:4891–904. doi: 10.1113/jphysiol.2010.196998
- Str ber H, Rach S, Neuling T, Herrmann C. On the possible role of stimulation duration for after-effects of transcranial alternating current stimulation. *Front Cell Neurosci.* (2015) 9:311. doi: 10.3389/fncel.2015.00311
- Kasten FH, Dowsett J, Herrmann CS. Sustained aftereffect of α -tACS lasts Up to 70 min after stimulation. *Front Hum Neurosci.* (2016) 10:245. doi: 10.3389/fnhum.2016.00245
- Battleday RM, Muller T, Clayton MS, Cohen Kadosh R. Mapping the mechanisms of transcranial alternating current stimulation: a pathway from network effects to cognition. *Front Psychiatry.* (2014) 5:162. doi: 10.3389/fpsyt.2014.00162
- Guerra A, Ranieri F, Falato E, Musumeci G, Di Santo A, Asci F, et al. Detecting cortical circuits resonant to high-frequency oscillations in the human primary motor cortex: a TMS-tACS study. *Sci Rep.* (2020) 10:7695. doi: 10.1038/s41598-020-64717-7

32. Pelletier SJ, Cicchetti F. Cellular and molecular mechanisms of action of transcranial direct current stimulation: evidence from *in vitro* and *in vivo* models. *Int J Neuropsychopharmacol.* (2015) 18:1–13. doi: 10.1093/ijnp/pyu047
33. Zaghi S, Acar M, Hultgren B, Boggio PS, Fregni F. Noninvasive brain stimulation with low-intensity electrical currents: putative mechanisms of action for direct and alternating current stimulation. *Neuroscientist.* (2010) 16:285–307. doi: 10.1177/1073858409336227
34. Rostami M, Golesorkhi M, Ekhtiari H. Methodological dimensions of transcranial brain stimulation with the electrical current in human. *Basic Clin Neurosci.* (2013) 4:8–26.
35. Priori A, Hallett M, Rothwell JC. Repetitive transcranial magnetic stimulation or transcranial direct current stimulation? *Brain Stimul.* (2009) 2:241–5. doi: 10.1016/j.brs.2009.02.004
36. Nitsche MA, Cohen LG, Wassermann EM, Priori A, Lang N, Antal A, et al. Transcranial direct current stimulation: state of the art 2008. *Brain Stimul.* (2008) 1:206–23. doi: 10.1016/j.brs.2008.06.004
37. Nitsche MA, Kuo M-F, Paulus W, Antal A. Transcranial direct current stimulation: protocols and physiological mechanisms of action. In: *Textbook of Neuromodulation*. New York, NY: Springer. (2014). p. 101–11. doi: 10.1007/978-1-4939-1408-1_9
38. Paulus W. Transcranial electrical stimulation (tES - tDCS; tRNS, tACS) methods. *Neuropsychol Rehabil.* (2011) 21:602–17. doi: 10.1080/09602011.2011.557292
39. Antal A, Paulus W. Transcranial alternating current stimulation (tACS). *Front Hum Neurosci.* (2013) 7:317. doi: 10.3389/fnhum.2013.00317
40. Vosskuhl J, Strüber D, Herrmann CS. Non-invasive brain stimulation: a paradigm shift in understanding brain oscillations. *Front Hum Neurosci.* (2018) 12:211. doi: 10.3389/fnhum.2018.00211
41. Thut G, Schyns PG, Gross J. Entrainment of perceptually relevant brain oscillations by non-invasive rhythmic stimulation of the human brain. *Front Psychol.* (2011) 2:170. doi: 10.3389/fpsyg.2011.00170
42. Tavakoli AV, Yun K. Transcranial alternating current stimulation (tACS) mechanisms and protocols. *Front Cell Neurosci.* (2017) 11:214. doi: 10.3389/fncel.2017.00214
43. Liu A, Vöröslakos M, Kronberg G, Henin S, Krause MR, Huang Y, et al. Immediate neurophysiological effects of transcranial electrical stimulation. *Nat Commun.* (2018) 9:5092. doi: 10.1038/s41467-018-07233-7
44. Elyamany O, Leicht G, Herrmann CS, Mulert C. Transcranial alternating current stimulation (tACS): from basic mechanisms towards first applications in psychiatry. *Eur Arch Psychiatry Clin Neurosci.* (2020) doi: 10.1007/s00406-020-01209-9
45. Zaehle T, Rach S, Herrmann CS. Transcranial alternating current stimulation enhances individual alpha activity in human EEG. *PLoS ONE.* (2010) 5:e13766. doi: 10.1371/journal.pone.0013766
46. Guerra A, Suppa A, Bologna M, D'Onofrio V, Bianchini E, Brown P, et al. Boosting the LTP-like plasticity effect of intermittent theta-burst stimulation using gamma transcranial alternating current stimulation. *Brain Stimul.* (2018) 11:734–42. doi: 10.1016/j.brs.2018.03.015
47. Wischniewski M, Engelhardt M, Salehinejad MA, Schutter DJLG, Kuo MF, Nitsche MA. NMDA receptor-mediated motor cortex plasticity after 20 Hz transcranial alternating current stimulation. *Cereb Cortex.* (2019) 29:2924–31. doi: 10.1093/cercor/bhy160
48. Hopfinger JB, Parsons J, Fröhlich F. Differential effects of 10-Hz and 40-Hz transcranial alternating current stimulation (tACS) on endogenous versus exogenous attention. *Cogn Neurosci.* (2017) 8:102–11. doi: 10.1080/17588928.2016.1194261
49. Hallett M. Transcranial magnetic stimulation: a primer. *Neuron.* (2007) 55:187–99. doi: 10.1016/j.neuron.2007.06.026
50. Cirillo G, Di Pino G, Capone F, Ranieri F, Florio L, Todisco V, et al. Neurobiological after-effects of non-invasive brain stimulation. *Brain Stimul.* (2017) 10:1–18. doi: 10.1016/j.brs.2016.11.009
51. Pascual-Leone A, Grafman J, Hallett M. Modulation of cortical motor output maps during development of implicit and explicit knowledge. *Science.* (1994) 263:1287–9. doi: 10.1126/science.8122113
52. Suppa A, Huang Y-Z, Funke K, Ridding MC, Cheeran B, Di Lazzaro V, et al. Ten years of theta burst stimulation in humans: established knowledge, unknowns and prospects. *Brain Stimul.* (2016) 9:323–35. doi: 10.1016/j.brs.2016.01.006
53. Terao Y, Ugawa Y. Basic mechanisms of TMS. *J Clin Neurophysiol.* (2002) 19:322–43. doi: 10.1097/00004691-200208000-00006
54. Peterchev AV, Wagner TA, Miranda PC, Nitsche MA, Paulus W, Lisanby SH, et al. Fundamentals of transcranial electric and magnetic stimulation dose: definition, selection, and reporting practices. *Brain Stimul.* (2012) 5:435–53. doi: 10.1016/j.brs.2011.10.001
55. Stagg CJ, Nitsche MA. Physiological basis of transcranial direct current stimulation. *Neuroscientist.* (2011) 17:37–53. doi: 10.1177/1073858410386614
56. Bikson M, Inoue M, Akiyama H, Deans JK, Fox JE, Miyakawa H, et al. Effect of uniform extracellular DC electric fields on excitability in rat hippocampal slices *in vitro*. *J Physiol.* (2004) 557:175–90. doi: 10.1113/jphysiol.2003.055772
57. Arlotti M, Rahman A, Minhas P, Bikson M. Axon terminal polarization induced by weak uniform DC electric fields: a modeling study. *Annu Int Conf IEEE Eng Med Biol Soc.* (2012) 2012:4575–8. doi: 10.1109/EMBC.2012.6346985
58. Rahman A, Reato D, Arlotti M, Gasca F, Datta A, Parra LC, et al. Cellular effects of acute direct current stimulation: somatic and synaptic terminal effects. *J Physiol.* (2013) 591:2563–78. doi: 10.1113/jphysiol.2012.247171
59. Seo H, Jun SC. Relation between the electric field and activation of cortical neurons in transcranial electrical stimulation. *Brain Stimul.* (2019) 12:275–89. doi: 10.1016/j.brs.2018.11.004
60. Francis JT, Gluckman BJ, Schiff SJ. Sensitivity of neurons to weak electric fields. *J Neurosci.* (2003) 23:7255–61. doi: 10.1523/JNEUROSCI.23-19-07255.2003
61. Deans JK, Powell AD, Jefferys JGR. Sensitivity of coherent oscillations in rat hippocampus to AC electric fields. *J Physiol.* (2007) 583:555–65. doi: 10.1113/jphysiol.2007.137711
62. Reato D, Rahman A, Bikson M, Parra LC. Low-intensity electrical stimulation affects network dynamics by modulating population rate and spike timing. *J Neurosci.* (2010) 30:15067–79. doi: 10.1523/JNEUROSCI.2059-10.2010
63. Antal A, Herrmann CS. Transcranial alternating current and random noise stimulation: possible mechanisms. *Neural Plast.* (2016) 2016:1–12. doi: 10.1155/2016/3616807
64. Lisman JE. Three Ca²⁺ levels affect plasticity differently: the LTP zone, the LTD zone and no man's land. *J Physiol.* (2001) 532:285. doi: 10.1111/j.1469-7793.2001.0285f.x
65. Kirsch DL, Nichols F. Cranial electrotherapy stimulation for treatment of anxiety, depression, and insomnia. *Psychiatr Clin North Am.* (2013) 36:169–76. doi: 10.1016/j.psc.2013.01.006
66. Kirsch D. *The Science Behind Cranial Electrotherapy Stimulation*. 2nd Ed. Alberta: Medical Scope Publishing Corporation (2002).
67. Limoge A, Robert C, Stanley TH. Transcutaneous cranial electrical stimulation (TCES): a review 1998. *Neurosci Biobehav Rev.* (1999) 23:529–38. doi: 10.1016/S0149-7634(98)00048-7
68. Nitsche MA, Boggio PS, Fregni F, Pascual-Leone A. Treatment of depression with transcranial direct current stimulation (tDCS): a review. *Exp Neurol.* (2009) 219:14–9. doi: 10.1016/j.expneurol.2009.03.038
69. Zhao X, Ding J, Pan H, Zhang S, Pan D, Yu H, et al. Anodal and cathodal tDCS modulate neural activity and selectively affect GABA and glutamate syntheses in the visual cortex of cats. *J Physiol.* (2020) 598:3727–45. doi: 10.1113/JP279340
70. Stagg CJ, O'Shea J, Kincses ZT, Woolrich M, Matthews PM, Johansen-Berg H. Modulation of movement-associated cortical activation by transcranial direct current stimulation. *Eur J Neurosci.* (2009) 30:1412–23. doi: 10.1111/j.1460-9568.2009.06937.x
71. Hansen N. Action mechanisms of transcranial direct current stimulation in Alzheimer's disease and memory loss. *Front Psychiatry.* (2012) 3:48. doi: 10.3389/fpsy.2012.00048
72. Nitsche M, Liebetanz D, Schlitterlau A, Henschke U, Fricke K, Frommann K, et al. GABAergic modulation of DC stimulation-induced motor cortex excitability shifts in humans. *Eur J Neurosci.* (2004) 19:2720–6. doi: 10.1111/j.0953-816X.2004.03398.x
73. Malenka RC, Bear MF. LTP and LTD: an embarrassment of riches. *Neuron.* (2004) 44:5–21. doi: 10.1016/j.neuron.2004.09.012
74. Cooke SF, Bliss TVP. Plasticity in the human central nervous system. *Brain.* (2006) 129:1659–73. doi: 10.1093/brain/awl082

75. Lanté F, de Jésus Ferreira M-C, Guiramand J, Récasens M, Vignes M. Low-frequency stimulation induces a new form of LTP, metabotropic glutamate (mGlu5) receptor- and PKA-dependent, in the CA1 area of the rat hippocampus. *Hippocampus*. (2006) 16:345–60. doi: 10.1002/hipo.20146
76. Luscher C, Malenka RC. NMDA receptor-dependent long-term potentiation and long-term depression (LTP/LTD). *Cold Spring Harb Perspect Biol*. (2012) 4:a005710. doi: 10.1101/cshperspect.a005710
77. Mycielska ME, Djamgoz MBA. Cellular mechanisms of direct-current electric field effects: Galvanotaxis and metastatic disease. *J Cell Sci*. (2004) 117:1631–9. doi: 10.1242/jcs.01125
78. McCaig CD, Rajnicek AM, Song B, Zhao M. Controlling cell behavior electrically: current views and future potential. *Physiol Rev*. (2005) 85:943–78. doi: 10.1152/physrev.00020.2004
79. Monte-Silva K, Liebetanz D, Grundey J, Paulus W, Nitsche MA. Dose-dependent non-linear effect of l-dopa on human motor cortex plasticity. *J Physiol*. (2010) 588:3415–24. doi: 10.1113/jphysiol.2010.190181
80. Kuo MF, Paulus W, Nitsche MA. Boosting focally-induced brain plasticity by dopamine. *Cereb Cortex*. (2008) 18:648–51. doi: 10.1093/cercor/bhm098
81. Carvalho S, Boggio PS, Gonçalves ÓF, Vigário AR, Faria M, Silva S, et al. Transcranial direct current stimulation based metaplasticity protocols in working memory. *Brain Stimul*. (2015) 8:289–94. doi: 10.1016/j.brs.2014.11.011
82. Cirillo G, Colangelo AM, Bianco MR, Cavaliere C, Zaccaro L, Sarmientos P, et al. BB14, a Nerve Growth Factor (NGF)-like peptide shown to be effective in reducing reactive astrogliosis and restoring synaptic homeostasis in a rat model of peripheral nerve injury. *Biotechnol Adv*. (2012) 30:223–32. doi: 10.1016/j.biotechadv.2011.05.008
83. Pang PT. Cleavage of proBDNF by tPA/Plasmin is essential for long-term hippocampal plasticity. *Science*. (2004) 306:487–91. doi: 10.1126/science.1100135
84. Cirillo G, Bianco MR, Colangelo AM, Cavaliere C, Daniele DL, Zaccaro L, et al. Reactive astrogliosis-induced perturbation of synaptic homeostasis is restored by nerve growth factor. *Neurobiol Dis*. (2011) 41:630–9. doi: 10.1016/j.nbd.2010.11.012
85. Nagappan G, Zaitsev E, Senatorov VV, Yang J, Hempstead BL, Lu B. Control of extracellular cleavage of ProBDNF by high frequency neuronal activity. *Proc Natl Acad Sci USA*. (2009) 106:1267–72. doi: 10.1073/pnas.0807322106
86. Grau JW, Russell Huie J, Lee KH, Hoy KC, Huang YJ, Turtle JD, et al. Metaplasticity and behavior: how training and inflammation affect plastic potential within the spinal cord and recovery after injury. *Front Neural Circuits*. (2014) 8:100. doi: 10.3389/fncir.2014.00100
87. Podda MV, Cocco S, Mastrodonato A, Fusco S, Leone L, Barbati SA, et al. Anodal transcranial direct current stimulation boosts synaptic plasticity and memory in mice via epigenetic regulation of Bdnf expression. *Sci Rep*. (2016) 6:22180. doi: 10.1038/srep22180
88. Matsuda N, Lu H, Fukata Y, Noritake J, Gao H, Mukherjee S, et al. Differential activity-dependent secretion of brain-derived neurotrophic factor from axon and dendrite. *J Neurosci*. (2009) 29:14185–98. doi: 10.1523/JNEUROSCI.1863-09.2009
89. Park H, Popescu A, Poo M ming. Essential role of presynaptic NMDA receptors in activity-dependent BDNF secretion corticostriatal LTP. *Neuron*. (2014) 84:1009–22. doi: 10.1016/j.neuron.2014.10.045
90. Sumi T, Harada K. Mechanism underlying hippocampal long-term potentiation and depression based on competition between endocytosis and exocytosis of AMPA receptors. *Sci Rep*. (2020) 10:14711. doi: 10.1038/s41598-020-71528-3
91. Nitsche MA, Paulus W. Transcranial direct current stimulation - Update 2011. *Restor Neurol Neurosci*. (2011) 29:463–92. doi: 10.3233/RNN-2011-0618
92. Thirugnanasambandam N, Paulus W, Kuo M-F, Liebetanz D, Monte-Silva K, Nitsche MA. Dose-dependent inverted u-shaped effect of dopamine (D2-Like) receptor activation on focal and nonfocal plasticity in humans. *J Neurosci*. (2009) 29:6124–31. doi: 10.1523/JNEUROSCI.0728-09.2009
93. Pláteník J, Kuramoto N, Yoneda Y. Molecular mechanisms associated with long-term consolidation of the NMDA signals. *Life Sci*. (2000) 67:335–64. doi: 10.1016/S0024-3205(00)00632-9
94. Guzman-Karlsson MC, Meadows JP, Gavin CE, Hablitz JJ, Sweatt JD. Transcriptional and epigenetic regulation of Hebbian and non-Hebbian plasticity. *Neuropharmacology*. (2014) 80:3–17. doi: 10.1016/j.neuropharm.2014.01.001
95. Davis S, Bozon B, Laroche S. How necessary is the activation of the immediate early gene zif 268 in synaptic plasticity and learning? *Behav Brain Res*. (2003) 142:17–30. doi: 10.1016/S0166-4328(02)00421-7
96. Turrigiano GG, Leslie KR, Desai NS, Rutherford LC, Nelson SB. Activity-dependent scaling of quantal amplitude in neocortical neurons. *Nature*. (1998) 391:892–6. doi: 10.1038/36103
97. Latremoliere A, Woolf CJ. Central sensitization: a generator of pain hypersensitivity by central neural plasticity. *J Pain*. (2009) 10:895–926. doi: 10.1016/j.jpain.2009.06.012
98. Davis GW, Bezprozvanny I. Maintaining the stability of neural function: a homeostatic hypothesis. *Annu Rev Physiol*. (2001) 63:847–69. doi: 10.1146/annurev.physiol.63.1.847
99. Fedorov A, Chibisova Y, Szymaszek A, Alexandrov M, Gall C, Sabel BA. Non-invasive alternating current stimulation induces recovery from stroke. *Restor Neurol Neurosci*. (2010) 28:825–33. doi: 10.3233/RNN-2010-0580
100. Demirtas-Tatlıdide A, Vahabzadeh-Hagh AM, Bernabeu M, Tormos JM, Pascual-Leone A. Noninvasive brain stimulation in traumatic brain injury. *J Head Trauma Rehabil*. (2012) 27:274–92. doi: 10.1097/HTR.0b013e318217df55
101. Han CL, Hu W, Stead M, Zhang T, Zhang JG, Worrell GA, et al. Electrical stimulation of hippocampus for the treatment of refractory temporal lobe epilepsy. *Brain Res Bull*. (2014) 109:13–21. doi: 10.1016/j.brainresbull.2014.08.007
102. Abraham WC. Metaplasticity: tuning synapses and networks for plasticity. *Nat Rev Neurosci*. (2008) 9:387–99. doi: 10.1038/nrn2356
103. Müller-Dahlhaus F, Ziemann U. Metaplasticity in human cortex. *Neurosci*. (2015) 21:185–202. doi: 10.1177/1073858414526645
104. Turrigiano GG, Nelson SB. Homeostatic plasticity in the developing nervous system. *Nat Rev Neurosci*. (2004) 5:97–107. doi: 10.1038/nrn1327
105. Bienenstock EL, Cooper LN, Munro PW. Theory for the development of neuron selectivity: Orientation specificity and binocular interaction in visual cortex. *J Neurosci*. (1982) 2:32–48. doi: 10.1523/JNEUROSCI.02-01-00032.1982
106. Morgante F, Espay AJ, Gunraj C, Lang AE, Chen R. Motor cortex plasticity in Parkinson's disease and levodopa-induced dyskinesias. *Brain*. (2006) 129:1059–69. doi: 10.1093/brain/awl031
107. Udupa K, Chen R. Motor cortical plasticity in Parkinson's disease. *Front Neurol*. (2013) 4:128. doi: 10.3389/fneur.2013.00128
108. Ueki Y, Mima T, Ali Kotb M, Sawada H, Saiki H, Ikeda A, et al. Altered plasticity of the human motor cortex in Parkinson's disease. *Ann Neurol*. (2006) 59:60–71. doi: 10.1002/ana.20692
109. Quartarone A, Siebner HR, Rothwell JC. Task-specific hand dystonia: can too much plasticity be bad for you? *Trends Neurosci*. (2006) 29:192–9. doi: 10.1016/j.tins.2006.02.007
110. Weise D. The two sides of associative plasticity in writer's cramp. *Brain*. (2006) 129:2709–21. doi: 10.1093/brain/awl221
111. Mori F, Rossi S, Piccinin S, Motta C, Mango D, Kusayanagi H, et al. Synaptic plasticity and PDGF signaling defects underlie clinical progression in multiple sclerosis. *J Neurosci*. (2013) 33:19112–9. doi: 10.1523/JNEUROSCI.2536-13.2013
112. Di Lazzaro V, Profice P, Pilato F, Capone F, Ranieri F, Pasqualetti P, et al. Motor cortex plasticity predicts recovery in acute stroke. *Cereb Cortex*. (2010) 20:1523–8. doi: 10.1093/cercor/bhp216
113. Pierelli F, Iacovelli E, Bracaglia M, Serrao M, Coppola G. Abnormal sensorimotor plasticity in migraine without aura patients. *Pain*. (2013) 154:1738–42. doi: 10.1016/j.pain.2013.05.023
114. Koch G, Mori F, Marconi B, Codeca C, Pecchioli C, Salerno S, et al. Changes in intracortical circuits of the human motor cortex following theta burst stimulation of the lateral cerebellum. *Clin Neurophysiol*. (2008) 119:2559–69. doi: 10.1016/j.clinph.2008.08.008

115. Daskalakis ZJ, Christensen BK, Fitzgerald PB, Chen R. Dysfunctional neural plasticity in patients with schizophrenia. *Arch Gen Psychiatry*. (2008) 65:378. doi: 10.1001/archpsyc.65.4.378
116. Frantseva M V., Fitzgerald PB, Chen R, Moller B, Daigle M, Daskalakis ZJ. Evidence for impaired long-term potentiation in schizophrenia and its relationship to motor skill learning. *Cereb Cortex*. (2008) 18:990–996. doi: 10.1093/cercor/bhm151
117. Hasan A, Nitsche MA, Rein B, Schneider-Axmann T, Guse B, Gruber O, et al. Dysfunctional long-term potentiation-like plasticity in schizophrenia revealed by transcranial direct current stimulation. *Behav Brain Res*. (2011) 224:15–22. doi: 10.1016/j.bbr.2011.05.017
118. Barr MS, Farzan F, Wing VC, George TP, Fitzgerald PB, Daskalakis ZJ. Repetitive transcranial magnetic stimulation and drug addiction. *Int Rev Psychiatry*. (2011) 23:454–66. doi: 10.3109/09540261.2011.618827
119. Liebetanz D, Nitsche MA, Lang N, Antal A, Paulus W, Tergau F. Safety criteria for transcranial direct current stimulation (tDCS) in humans. *Clin Neurophysiol*. (2003) 114:2220–2. doi: 10.1016/S1388-2457(03)00235-9
120. Nitsche MA, Niehaus L, Hoffmann KT, Hengst S, Liebetanz D, Paulus W, et al. MRI study of human brain exposed to weak direct current stimulation of the frontal cortex. *Clin Neurophysiol*. (2004) 115:2419–23. doi: 10.1016/j.clinph.2004.05.001
121. Faria P, Hallett M, Miranda PC. A finite element analysis of the effect of electrode area and inter-electrode distance on the spatial distribution of the current density in tDCS. *J Neural Eng*. (2011) 8:066017. doi: 10.1088/1741-2560/8/6/066017
122. Nitsche MA, Seeber A, Frommann K, Klein CC, Rochford C, Nitsche MS, et al. Modulating parameters of excitability during and after transcranial direct current stimulation of the human motor cortex. *J Physiol*. (2005) 568:291–303. doi: 10.1113/jphysiol.2005.092429
123. Lefaucheur JP. Principles of therapeutic use of transcranial and epidural cortical stimulation. *Clin Neurophysiol*. (2008) 119:2179–84. doi: 10.1016/j.clinph.2008.07.007
124. Boros K, Poreisz C, Münchau A, Paulus W, Nitsche MA. Premotor transcranial direct current stimulation (tDCS) affects primary motor excitability in humans. *Eur J Neurosci*. (2008) 27:1292–300. doi: 10.1111/j.1460-9568.2008.06090.x
125. Vines BW, Cerruti C, Schlaug G. Dual-hemisphere tDCS facilitates greater improvements for healthy subjects' non-dominant hand compared to uni-hemisphere stimulation. *BMC Neurosci*. (2008) 9:103. doi: 10.1186/1471-2202-9-103
126. Marshall L. Transcranial direct current stimulation during sleep improves declarative memory. *J Neurosci*. (2004) 24:9985–92. doi: 10.1523/JNEUROSCI.2725-04.2004
127. Ardolino G, Bossi B, Barbieri S, Priori A. Non-synaptic mechanisms underlie the after-effects of cathodal transcutaneous direct current stimulation of the human brain. *J Physiol*. (2005) 568:653–63. doi: 10.1113/jphysiol.2005.088310
128. Montez T, Poil SS, Jones BF, Manshanden I, Verbunt JPA, Van Dijk BW, et al. Altered temporal correlations in parietal alpha and prefrontal theta oscillations in early-stage Alzheimer disease. *Proc Natl Acad Sci USA*. (2009) 106:1614–9. doi: 10.1073/pnas.0811699106
129. Gili T, Cercignani M, Serra L, Perri R, Giove F, Maraviglia B, et al. Regional brain atrophy and functional disconnection across Alzheimer's disease evolution. *J Neurol Neurosurg Psychiatry*. (2011) 82:58–66. doi: 10.1136/jnnp.2009.199935
130. Grady CL, Furey ML, Pietrini P, Horwitz B, Rapoport SI. Altered brain functional connectivity and impaired short-term memory in Alzheimer's disease. *Brain*. (2001) 124:739–56. doi: 10.1093/brain/124.4.739
131. Polanía R, Nitsche MA, Paulus W. Modulating functional connectivity patterns and topological functional organization of the human brain with transcranial direct current stimulation. *Hum Brain Mapp*. (2011) 32:1236–49. doi: 10.1002/hbm.21104
132. Klostermann E, Nikulin V V., Kühn AA, Marzinzik F, Wahl M, Pogosyan A, et al. Task-related differential dynamics of EEG alpha- and beta-band synchronization in cortico-basal motor structures. *Eur J Neurosci*. (2007) 25:1604–15. doi: 10.1111/j.1460-9568.2007.05417.x
133. Uhlhaas PJ, Singer W. Neural synchrony in brain disorders: relevance for cognitive dysfunctions and pathophysiology. *Neuron*. (2006) 52:155–68. doi: 10.1016/j.neuron.2006.09.020
134. Peña-Gómez C, Sala-Lonch R, Junqué C, Clemente IC, Vidal D, Bargalló N, et al. Modulation of large-scale brain networks by transcranial direct current stimulation evidenced by resting-state functional MRI. *Brain Stimul*. (2012) 5:252–63. doi: 10.1016/j.brs.2011.08.006
135. Polanía R, Paulus W, Nitsche MA. Modulating cortico-striatal and thalamo-cortical functional connectivity with transcranial direct current stimulation. *Hum Brain Mapp*. (2012) 33:2499–508. doi: 10.1007/978-3-662-45797-9_7
136. Gammaitoni L, Hänggi P, Jung P, Marchesoni F. Stochastic resonance. *Rev Mod Phys*. (1998) 70:223–87. doi: 10.1103/RevModPhys.70.223
137. Kitajo K, Nozaki D, Ward LM, Yamamoto Y. Behavioral stochastic resonance within the human brain. *Phys Rev Lett*. (2003) 90:218103. doi: 10.1103/PhysRevLett.90.218103
138. Fertonani A, Miniussi C. Transcranial electrical stimulation. *Neurosci*. (2017) 23:109–23. doi: 10.1177/1073858416631966
139. Papa M, De Luca C, Petta F, Alberghina L, Cirillo G. Astrocyte-neuron interplay in maladaptive plasticity. *Neurosci Biobehav Rev*. (2014) 42:35–54. doi: 10.1016/j.neubiorev.2014.01.010
140. Lee E, Chung W-S. Glial control of synapse number in healthy and diseased brain. *Front Cell Neurosci*. (2019) 13:42. doi: 10.3389/fncel.2019.00042
141. Phatnani H, Maniatis T. Astrocytes in neurodegenerative disease: table 1. *Cold Spring Harb Perspect Biol*. (2015) 7:a020628. doi: 10.1101/cshperspect.a020628
142. Liddelow SA, Barres BA. Reactive astrocytes: production, function, and therapeutic potential. *Immunity*. (2017) 46:957–67. doi: 10.1016/j.immuni.2017.06.006
143. Li Q, Barres BA. Microglia and macrophages in brain homeostasis and disease. *Nat Rev Immunol*. (2018) 18:225–42. doi: 10.1038/nri.2017.125
144. Hong S, Beja-Glasser VF, Nfonoyim BM, Frouin A, Li S, Ramakrishnan S, et al. Complement and microglia mediate early synapse loss in Alzheimer mouse models. *Science*. (2016) 352:712–6. doi: 10.1126/science.aad8373
145. Shi Q, Chowdhury S, Ma R, Le KX, Hong S, Caldarone BJ, et al. Complement C3 deficiency protects against neurodegeneration in aged plaque-rich APP/PS1 mice. *Sci Transl Med*. (2017) 9:eaf6295. doi: 10.1126/scitranslmed.aaf6295
146. Kullmann S, Kleinridders A, Small DM, Fritsche A, Häring H-U, Preissl H, et al. Central nervous pathways of insulin action in the control of metabolism and food intake. *Lancet Diabetes Endocrinol*. (2020) 8:524–34. doi: 10.1016/S2213-8587(20)30113-3
147. Srikanth V, Sinclair AJ, Hill-Briggs F, Moran C, Biessels GJ. Type 2 diabetes and cognitive dysfunction—towards effective management of both comorbidities. *Lancet Diabetes Endocrinol*. (2020) 8:535–45. doi: 10.1016/S2213-8587(20)30118-2
148. Colangelo AM, Cirillo G, Lavitrano ML, Alberghina L, Papa M. Targeting reactive astrogliosis by novel biotechnological strategies. *Biotechnol Adv*. (2012) 30:261–71. doi: 10.1016/j.biotechadv.2011.06.016
149. Rueger MA, Keuters MH, Walberer M, Braun R, Klein R, Sparing R, et al. Multi-session transcranial direct current stimulation (tDCS) Elicits inflammatory and regenerative processes in the rat brain. *PLoS ONE*. (2012) 7:e43776. doi: 10.1371/journal.pone.0043776
150. Pikhovych A, Stolberg NP, Jessica Flitsch L, Walter HL, Graf R, Fink GR, et al. Transcranial direct current stimulation modulates neurogenesis and microglia activation in the mouse brain. *Stem Cells Int*. (2016) 2016:1–9. doi: 10.1155/2016/2715196
151. Spezia Adachi LN, Caumo W, Laste G, Fernandes Medeiros L, Ripoll Rozisky J, de Souza A, et al. Reversal of chronic stress-induced pain by transcranial direct current stimulation (tDCS) in an animal model. *Brain Res*. (2012) 1489:17–26. doi: 10.1016/j.brainres.2012.10.009
152. Barbour B, Brew H, Attwell D. Electrogenic glutamate uptake in glial cells is activated by intracellular potassium. *Nature*. (1988) 335:433–5. doi: 10.1038/335433a0
153. Dallérac G, Chever O, Rouach N. How do astrocytes shape synaptic transmission? Insights from electrophysiology. *Front Cell Neurosci*. (2013) 7:159. doi: 10.3389/fncel.2013.00159

154. Monai H, Hirase H. Astrocytic calcium activation in a mouse model of tDCS—Extended discussion. *Neurogenesis*. (2016) 3:e1240055. doi: 10.1080/23262133.2016.1240055
155. Borgens RB, Shi R, Mohr TJ, Jaeger CB. Mammalian cortical astrocytes align themselves in a physiological voltage gradient. *Exp Neurol*. (1994) 128:41–9. doi: 10.1006/exnr.1994.1111
156. Alexander JK, Fuss B, Colello RJ. Electric field-induced astrocyte alignment directs neurite outgrowth. *Neuron Glia Biol*. (2006) 2:93–103. doi: 10.1017/S1740925X0600010X
157. Moriarty LJ, Borgens RB. An oscillating extracellular voltage gradient reduces the density and influences the orientation of astrocytes in injured mammalian spinal cord. *J Neurocytol*. (2001) 30:45–57. doi: 10.1023/A:1011917424450
158. Cicchetti F, Barker RA. The glial response to intracerebrally delivered therapies for neurodegenerative disorders: is this a critical issue? *Front Pharmacol*. (2014) 5:139. doi: 10.3389/fphar.2014.00139
159. Cambiaghi M, Velikova S, Gonzalez-Rosa JJ, Cursi M, Comi G, Leocani L. Brain transcranial direct current stimulation modulates motor excitability in mice. *Eur J Neurosci*. (2010) 31:704–9. doi: 10.1111/j.1460-9568.2010.07092.x
160. Herrmann CS, Rach S, Neuling T, Strüder D. Transcranial alternating current stimulation: a review of the underlying mechanisms and modulation of cognitive processes. *Front Hum Neurosci*. (2013) 7:279. doi: 10.3389/fnhum.2013.00279
161. Klink K, Paßmann S, Kasten FH, Peter J. The modulation of cognitive performance with transcranial alternating current stimulation: a systematic review of frequency-specific effects. *Brain Sci*. (2020) 10:932. doi: 10.3390/brainsci10120932
162. Steinmann S, Leicht G, Mulert C. The interhemispheric miscommunication theory of auditory verbal hallucinations in schizophrenia. *Int J Psychophysiol*. (2019) 145:83–90. doi: 10.1016/j.ijpsycho.2019.02.002
163. Wilkening A, Kurzeck A, Dechantreiter E, Padberg F, Palm U. Transcranial alternating current stimulation for the treatment of major depression during pregnancy. *Psychiatry Res*. (2019) 279:399–400. doi: 10.1016/j.psychres.2019.06.009
164. Klimke A, Nitsche MA, Maurer K, Voss U. Case report: successful treatment of therapy-resistant OCD with application of transcranial alternating current stimulation (tACS). *Brain Stimul*. (2016) 9:463–5. doi: 10.1016/j.brs.2016.03.005
165. van Deursen JA, Vuurman EPPM, Verhey FRJ, van Kranen-Mastenbroek VHJM, Riedel WJ. Increased EEG gamma band activity in Alzheimer's disease and mild cognitive impairment. *J Neural Transm*. (2008) 115:1301–11. doi: 10.1007/s00702-008-0083-y
166. Naro A, Corallo F, De Salvo S, Marra A, Di Lorenzo G, Muscarà N, et al. Promising role of neuromodulation in predicting the progression of mild cognitive impairment to dementia. *J Alzheimer's Dis*. (2016) 53:1375–88. doi: 10.3233/JAD-160305
167. Chai Z, Ma C, Jin X. Cortical stimulation for treatment of neurological disorders of hyperexcitability: A role of homeostatic plasticity. *Neural Regen Res*. (2019) 14:34–8. doi: 10.4103/1673-5374.243696
168. Nitsche MA, Paulus W. Excitability changes induced in the human motor cortex by weak transcranial direct current stimulation. *J Physiol*. (2000) 527:633–9. doi: 10.1111/j.1469-7793.2000.t01-1-00633.x
169. Fernández A, Maestú F, Amo C, Gil P, Fehr T, Wienbruch C, et al. Focal temporoparietal slow activity in Alzheimer's disease revealed by magnetoencephalography. *Biol Psychiatry*. (2002) 52:764–70. doi: 10.1016/S0006-3223(02)01366-5
170. Jiang T, Xu RX, Zhang AW, Di W, Xiao ZJ, Miao JY, et al. Effects of transcranial direct current stimulation on hemichannel pannexin-1 and neural plasticity in rat model of cerebral infarction. *Neuroscience*. (2012) 226:421–6. doi: 10.1016/j.neuroscience.2012.09.035
171. Yoon KJ, Oh BM, Kim DY. Functional improvement and neuroplastic effects of anodal transcranial direct current stimulation (tDCS) delivered 1 day vs. 1 week after cerebral ischemia in rats. *Brain Res*. (2012) 1452:61–72. doi: 10.1016/j.brainres.2012.02.062
172. Shaw M, Pawlak N, Choi C, Khan N, Datta A, Bikson M. Transcranial Direct Current Stimulation (tDCS) induces acute changes in brain metabolism. *Brain Stimul*. (2019) 12:518. doi: 10.1016/j.brs.2018.12.703
173. Sun DA, Sombati S, Blair RE, Delorenzo RJ. Calcium-dependent epileptogenesis in an *in vitro* model of stroke-induced "epilepsy." *Epilepsia*. (2002) 43:1296–305. doi: 10.1046/j.1528-1157.2002.09702.x
174. Pineda E, Shin D, Sankar R, Mazarati AM. Comorbidity between epilepsy and depression: experimental evidence for the involvement of serotonergic, glucocorticoid, and neuroinflammatory mechanisms. *Epilepsia*. (2010) 51:110–4. doi: 10.1111/j.1528-1167.2010.02623.x
175. Yamamoto J, Ikeda A, Kinoshita M, Matsumoto R, Satow T, Takeshita K, et al. Low-frequency electric cortical stimulation decreases interictal and ictal activity in human epilepsy. *Seizure*. (2006) 15:520–7. doi: 10.1016/j.seizure.2006.06.004
176. Liebetanz D, Klinker F, Hering D, Koch R, Nitsche MA, Potschka H, et al. Anticonvulsant effects of transcranial direct-current stimulation (tDCS) in the rat cortical ramp model of focal epilepsy. *Epilepsia*. (2006) 47:1216–24. doi: 10.1111/j.1528-1167.2006.00539.x
177. Yook S-W, Park S-H, Seo J-H, Kim S-J, Ko M-H. Suppression of seizure by cathodal transcranial direct current stimulation in an epileptic patient - a case report -. *Ann Rehabil Med*. (2011) 35:579–82. doi: 10.5535/arm.2011.35.4.579
178. Gschwind M, Seck M. Modern management of seizures and epilepsy. *Swiss Med Wkly*. (2016) 146:w14310. doi: 10.4414/smww.2016.14310
179. Benussi A, Dell'Era V, Cantoni V, Bonetta E, Grasso R, Manenti R, et al. Cerebellar-spinal tDCS in ataxia A randomized, double-blind, sham-controlled, crossover trial. *Neurology*. (2018) 91:E1090–E101. doi: 10.1212/WNL.0000000000006210
180. Jayaram G, Tang B, Pallegadda R, Vasudevan E, Celnik P, Bastian A. Modulating locomotor adaptation with cerebellar stimulation. *J Neurophysiol*. (2012) 107:2950–7. doi: 10.1152/jn.00645.2011
181. Priori A, Ciocca M, Parazzini M, Vergari M, Ferrucci R. Transcranial cerebellar direct current stimulation and transcutaneous spinal cord direct current stimulation as innovative tools for neuroscientists. *J Physiol*. (2014) 592:3345–69. doi: 10.1113/jphysiol.2013.270280
182. Truini A, Romaniello A, Galeotti F, Iannetti GD, Cruccu G. Laser evoked potentials for assessing sensory neuropathy in human patients. *Neurosci Lett*. (2004) 361:25–8. doi: 10.1016/j.neulet.2003.12.008
183. Lim CY, Shin HI. Noninvasive DC stimulation on neck changes MEP. *Neuroreport*. (2011) 22:819–23. doi: 10.1097/WNR.0b013e32834b939d
184. ElBasiouny SM, Mushahwar VK. Suppressing the excitability of spinal motoneurons by extracellularly applied electrical fields: insights from computer simulations. *J Appl Physiol*. (2007) 103:1824–36. doi: 10.1152/jappphysiol.00362.2007
185. Hounsgaard J, Kiehn O. Calcium spikes and calcium plateaux evoked by differential polarization in dendrites of turtle motoneurons *in vitro*. *J Physiol*. (1993) 468:245–59. doi: 10.1113/jphysiol.1993.sp019769

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2021 Korai, Ranieri, Di Lazzaro, Papa and Cirillo. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.



Repetitive Transcranial Magnetic Stimulation at Different Sites for Dysphagia After Stroke: A Randomized, Observer-Blind Clinical Trial

Lida Zhong^{1†}, Jinzhu Rao^{1†}, Jing Wang^{1†}, Fang Li¹, Yang Peng¹, Huiyu Liu^{1*}, Yan Zhang^{2*} and Pu Wang^{3*}

OPEN ACCESS

Edited by:

Federico Ranieri,
University of Verona, Italy

Reviewed by:

Ayodele Sasegbon,
The University of Manchester,
United Kingdom
Fabio Pilato,
Policlinico Universitario Campus
Bio-Medico, Italy

*Correspondence:

Huiyu Liu
liuhuiyudoc@sohu.com
Yan Zhang
zhangyan1981@hust.edu.cn
Pu Wang
wangpu_03@126.com

[†]These authors have contributed
equally to this work

Specialty section:

This article was submitted to
Neurorehabilitation,
a section of the journal
Frontiers in Neurology

Received: 03 November 2020

Accepted: 04 May 2021

Published: 26 May 2021

Citation:

Zhong L, Rao J, Wang J, Li F, Peng Y,
Liu H, Zhang Y and Wang P (2021)
Repetitive Transcranial Magnetic
Stimulation at Different Sites for
Dysphagia After Stroke: A
Randomized, Observer-Blind Clinical
Trial. *Front. Neurol.* 12:625683.
doi: 10.3389/fneur.2021.625683

¹ Department of Rehabilitation Medicine, Yue Bei People's Hospital, Shaoguan, China, ² School of Educational Science, Huazhong University of Science and Technology, Wuhan, China, ³ Department of Rehabilitation Medicine, The Seventh Affiliated Hospital Sun Yat-sen University, Shenzhen, China

Background: The clinical efficacy of repetitive transcranial magnetic stimulation (rTMS) protocols on patients with poststroke dysphagia is still unclear.

Objective: This trial aimed to explore and analyze the effectiveness of 5 Hz rTMS on the unaffected hemisphere, affected hemisphere, and cerebellum in stroke patients with dysphagia.

Methods: This observer-blind and randomized controlled trial included a total of 147 patients with stroke. Patients were divided into four treatment groups: the unaffected hemispheric group, the affected hemispheric group, the cerebellum group and the control group. Each group received traditional dysphagia treatment 5 days a week for 2 weeks. All recruited patients except for those in the control group underwent 10 consecutive rTMS sessions for 2 weeks. For the affected hemispheric group and unaffected hemispheric group, 5 Hz rTMS was applied to the affected mylohyoid cortical region or to the unaffected mylohyoid cortical region. For the cerebellum group, 5 Hz rTMS was applied to the mylohyoid cortical representation of the cerebellum (4.3 cm lateral and 2.4 cm below theinion). The Fiberoptic Endoscopic Dysphagia Severity Scale (FEDSS), Penetration/Aspiration Scale (PAS), Gugging Swallowing Screen (GUSS), and Standardized Swallowing Assessment (SSA) were used to evaluate clinical swallowing function before the intervention (baseline), immediately after the intervention and 2 weeks after the intervention.

Results: There were significant time and intervention interaction effects on the FEDSS, PAS, SSA, and GUSS scores ($p < 0.05$). In a direct comparison of the swallowing parameters of the four groups, the changes in FEDSS, PAS, SSA, and GUSS scores showed a significantly greater improvement in the unaffected hemispheric group, the affected hemispheric group and cerebellum group than in the control group ($p < 0.05$).

Conclusions: Whether stimulating the unaffected hemisphere or the affected hemisphere, 5 Hz high-frequency rTMS on mylohyoid cortical tissue might have a positive

effect on poststroke patients with dysphagia. In addition, cerebellar rTMS is a safe method that represents a potential treatment for poststroke dysphagia, and more clinical trials are needed to develop this technique further.

Clinical Trial Registration: chictr.org.cn, identifier: ChiCTR2000032255.

Keywords: repetitive transcranial magnetic stimulation, dysphagia, stroke, cerebellum, mylohyoid cortical

INTRODUCTION

Dysphagia, affecting 27–64% of stroke patients, is one of the most common poststroke sequelae (1) and is often associated with malnutrition, pneumonia, and dehydration (2). Conventional therapies for dysphagia include postural interventions, swallowing maneuvers, and exercises. Even though the above treatments have been widely applied in clinical practice, there is not enough clinical evidence to prove their efficacy (3–5). Recently, non-invasive cortical stimulation, a new strategy, has been used as a way of promoting neurologic rehabilitation after stroke. For example, transcranial magnetic stimulation is considered a well-tolerated technique that can modulate cortical excitability (6, 7). Moreover, repetitive transcranial magnetic stimulation (rTMS) of the motor cortex area related to swallowing directly induces the excitability of swallowing muscles regulated by corticobulbar projections (8), thereby enhancing swallowing function (9, 10). In patients with dysphagia after stroke, the application of 3 Hz (11) and 10 Hz (12) rTMS on the ipsilateral motor cortex represented by the esophageal or mylohyoid cortex showed significant improvement compared with sham stimulation. Meanwhile, both 1 Hz (13) and 5 Hz (9) rTMS on the contralateral motor cortex represented by the pharyngeal or mylohyoid cortex showed improved swallowing function. According to reports, rTMS showed different efficacies when patients with dysphagia were subjected to different stimulation parameters, such as intensity, frequency, and stimulation position.

It is controversial to stimulate either the ipsilesional or contralesional hemisphere. Previous systematic studies have shown different outcomes regarding the efficacy of non-invasive brain stimulation (NIBS) according to its stimulating point. Specifically, a review reported that no differences were found dependent on the stimulation site (14), whereas another study discovered that contralesional stimulation is better than ipsilesional stimulation (15). The latter study applied a combination of 5 Hz rTMS with pharyngeal electrical stimulation on the contralesional hemisphere (16). In conclusion, previous reviews reported different results because of the various stimulation applications, and it was relatively difficult to confirm whether the effect of contralesional rTMS was better than ipsilesional rTMS in regard to improving swallowing function.

Cerebellar neurostimulation has been considered an unexplored method and a prelude of treatment for dysphagia by modulating swallowing pathways. It has been shown that the cerebellum can be strongly activated during swallowing exercise (17), and stimulation of the cerebellum in the hemispheres or midline can induce different pharyngeal electromyography

responses. For example, Sasegbon et al. (18) demonstrated that rTMS on the cerebellar vermis had inhibitory effects on pharyngeal motor cortical activity and swallowing behavior. Vasant et al. (19) demonstrated that hemispheric cerebellar rTMS increases cortical pharyngeal motor evoked potential (PMEP) amplitudes. Using the advantages of neuronavigation and comparing the latency and amplitude of pharyngeal motor evoked potentials, the authors confirmed the best position to obtain these responses, which was 4.3 cm lateral and 2.4 cm below theinion (19). Recently, some studies (20, 21) have explored the possibility of rTMS on cerebellar tissue in the treatment of dysphagia.

Therefore, this prospective, randomized, observer-blind clinical study focused on the effectiveness and safety of rTMS in stroke patients with dysphagia. Outcomes after stimulation of the unaffected side, the affected side and the cerebellum were compared to determine which area of stimulation is more beneficial for the recovery of patients with dysphagia to guide clinical work in the future.

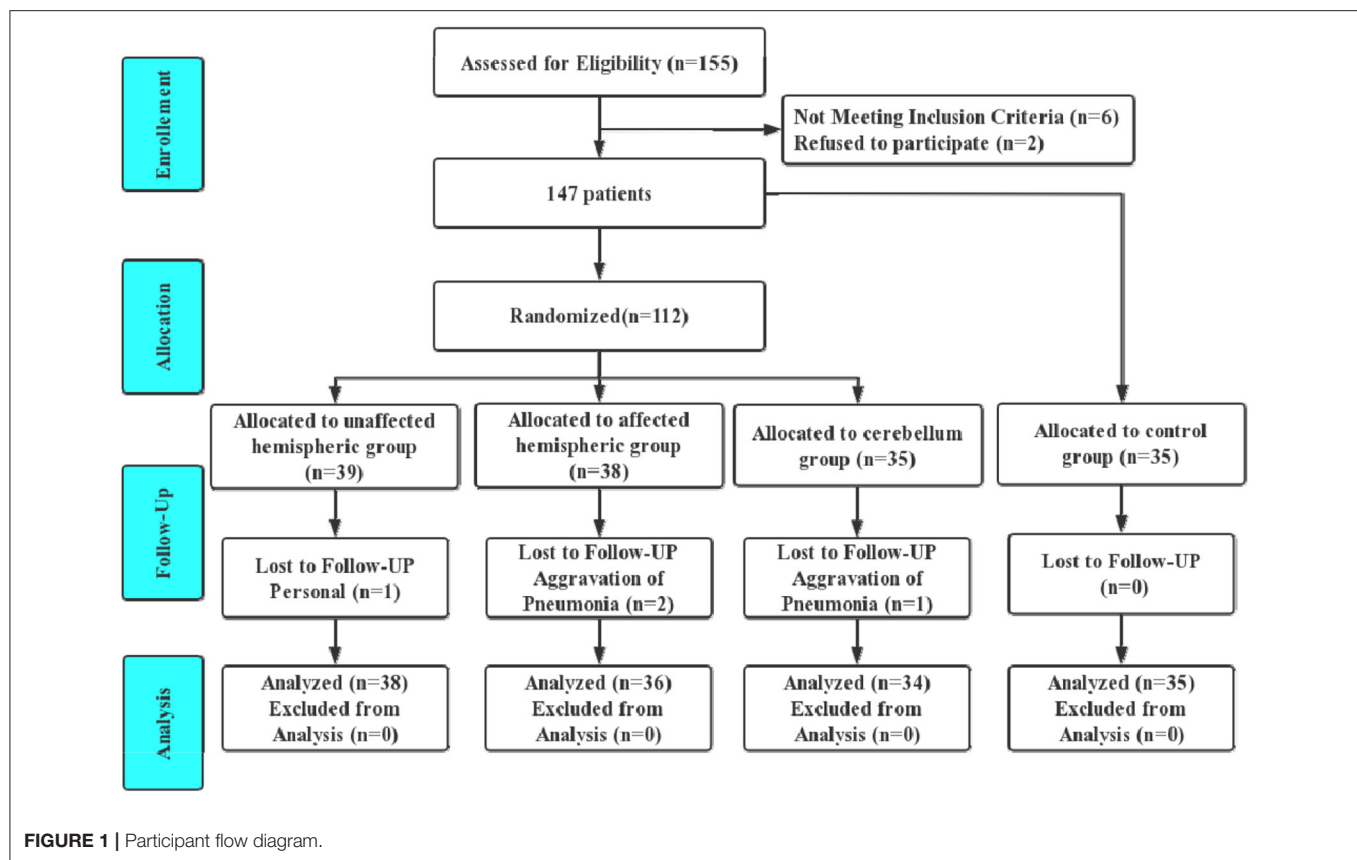
MATERIALS AND METHODS

Subjects

One hundred fifty-five poststroke patients suffering from dysphagia were included from April 2020 to April 2021. All of the patients were hospitalized to the Department of Rehabilitation Medicine, Yue Bei People's Hospital, Guangdong Province, China. The inclusion criteria were as follows: (1) subacute stroke <3 months diagnosed by imaging tests, including computed tomography (CT) or magnetic resonance imaging (MRI), hemorrhagic stroke or unilateral ischemia; (2) dysphagia confirmed by fiberoptic endoscopic evaluation of swallowing (FEES); and (3) no prior dysphagia rehabilitation. The exclusion criteria included history of any other neurogenic disease, epilepsy, tumor; severe cognitive impairment or aphasia; and contraindication to electrical or magnetic stimulation. All patients provided written informed consent before inclusion. The trial protocol was approved by the Ethics Committee of Yue Bei People's Hospital, and this clinical study was carried out and reported according to the Consolidated Standards of Reporting Trials (CONSORT) guidelines (22). Details of trial protocol registration can be seen in chictr.org.cn (chictr.org.cn Identifier: ChiCTR2000032255).

A total of 155 poststroke patients with dysphagia were recruited before assessment for eligibility, and 147 were included after exclusion.

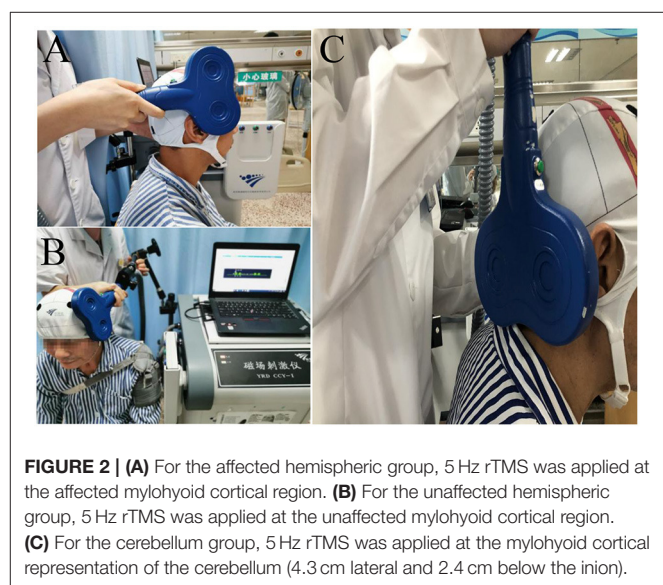
One hundred forty-seven patients were divided into four groups: the unaffected hemispheric group, affected hemispheric



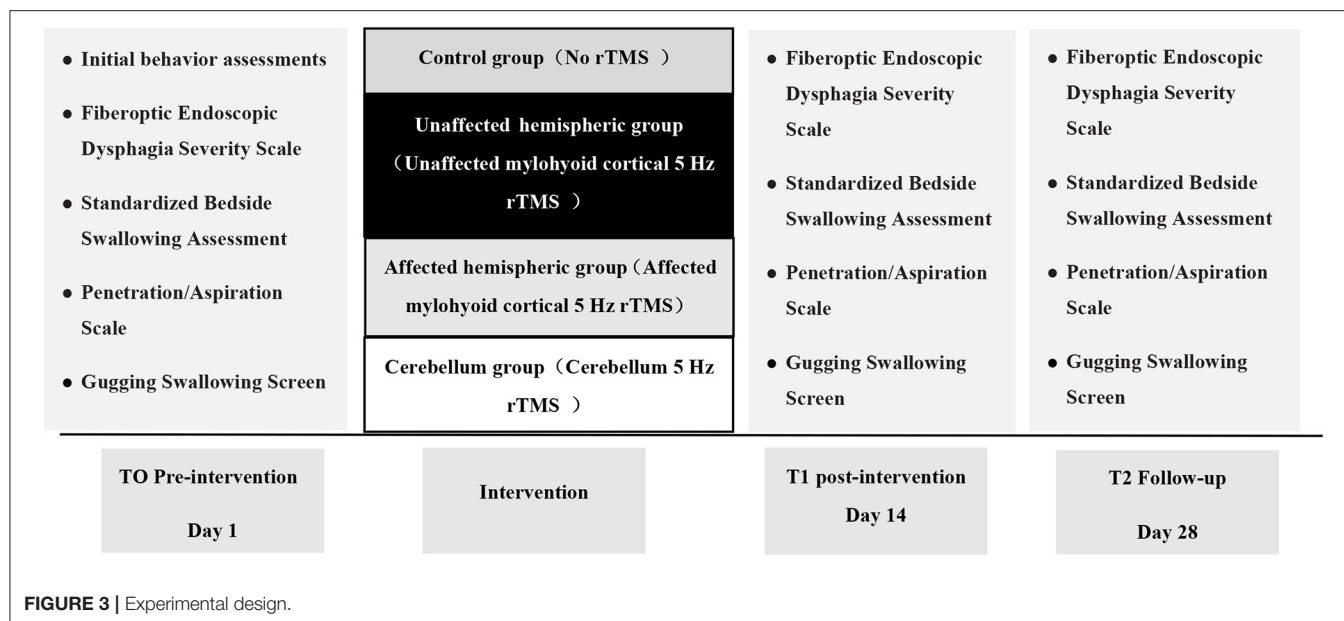
group, cerebellum group and control group. Four included patients withdrew from the trial. One patient in the unaffected hemispheric group withdrew for a personal reason not relevant to the trial. Two patients in the affected hemispheric group and one in the cerebellum group quit the study due to exacerbated pneumonia. Consequently, 143 patients completed the trial (Figure 1).

Experimental Design

This study was an observer-blind and random controlled trial. Patients were randomly divided into three groups by the random number table method. A sealed opaque envelope was opened at patient enrollment to determine whether the patient was to be assigned to the unaffected hemispheric, affected hemispheric or cerebellum group. These three groups of patients received 10 consecutive rTMS sessions for 2 weeks. For the affected hemispheric group and unaffected hemispheric group, 5 Hz rTMS was applied to the affected mylohyoid cortical region (Figure 2A) or to the unaffected mylohyoid cortical region (Figure 2B). For the cerebellum group, 5 Hz rTMS was applied to the mylohyoid cortical representation of the cerebellum (4.3 cm to lateral and 2.4 cm below theinion) (Figure 2C) (19). These three groups of patients received the same amount of traditional dysphagia treatment for 30 min daily after the intervention, such as thermal tactile stimulation, vocal cord exercises, Shaker exercises, Masako maneuvers, oropharyngeal muscle strengthening exercises, and tongue retraction exercises.



These exercises were conducted 5 days a week for 10 days with the guidance of an experienced physical therapist. Meanwhile, patients treated with rTMS were compared with a population of 35 post-stroke patients (control group) suffering from dysphagia who did not receive rTMS. The 35 post-stroke patients only



received traditional dysphagia treatment 5 days a week for 2 weeks.

Determination of the Resting Motor Threshold (RMT)

Unaffected Hemispheric Group and Affected Hemispheric Group

Each patient in the affected hemispheric group and unaffected hemispheric group was seated in a quiet environment and relaxed state. Electromyography (EMG) data representing oral swallowing musculature from mylohyoid muscles were detected using the same methods as Hamdy et al. (23). MagPro CCY-I stimulator (purchased from YIRUIDE Company, Wuhan, China) was used for magnetic stimulations with a 9 cm outer diameter figure-eight coil.

Cortical excitability on both hemispheres separately of each patient, including the motor evoked potential (MEP) and resting motor threshold (RMT) were measured using single-pulse TMS. The coil was moved around in an area within 2–4 cm anteriorly and 4–6 cm laterally of the vertex of the cranium to locate the mylohyoid cortical region of the hemisphere to obtain the maximum MEP recording (23). The maximum MEP recording location was regarded as the “hot spot,” representing magnetic stimulation delivered to the area. Single-pulse TMS was then delivered to the “hot spot” with a 2% reduction in the output of the stimulator. The definition of the rMT is that in 10 consecutive trials of mylohyoid muscles, five trials can induce the minimum stimulus intensity of MEP > 50 μ V. The “hot spot” was defined as an unaffected symmetrical hemisphere if MEPs were absent when the stroke-affected hemisphere was stimulated.

The Cerebellum Group

In previous studies, it has been identified that rTMS stimulation is effective regardless of which side of the cerebellum is stimulated

(19, 24). For the cerebellum group, the coil was fixed at the mylohyoid cortical representation of the cerebellum (4.3 cm to lateral and 2.4 cm below theinion) (19). The rMT was determined by the rMT of the mylohyoid cortical area of the unaffected hemisphere.

Repetitive Transcranial Magnetic Stimulation Application

The same parameters of stimulation were used for each intervention group. For each patient, 20 min rMT intensity with 5 Hz at 110% was applied at the “hot spot” area, which would last for 10 days with a total of 1,800 pulses per day. The protocols of rTMS applied in this study were strictly followed by the clinical safety guidelines for rTMS applications (25).

Outcome Measurements

All included participants were assessed at three different times: baseline (before the treatment), 2 weeks (after the treatment), and follow-up (2 weeks after the treatment) (see **Figure 3**). The primary outcome included the FEDSS scale; secondary outcomes involved assessments of the other dysphagia rating scales, such as the SSA scale, PAS scale, and GUSS scale.

Fiberoptic Endoscopic Dysphagia Severity Scale (FEDSS)

All included patients required FEES. First, the secretion status of patients was measured, and then the patient received standard volumes of semiliquid diet, such as soft solid food, liquids, or puree. Stroke-related dysphagia was divided into a six-point FEDSS with 1 score for the best and 6 scores for the worst based on different consistencies of diet observed in the endoscopic examination and the risk of saliva penetration or aspiration (26).

TABLE 1 | The demographic and clinical characteristics of the included patients.

	Unaffected N = 38	Affected N = 36	Cerebellum N = 34	Control N = 35	P
Sex (F:M)	10: 28	8: 28	14: 20	17: 18	0.063
Age (years)	64.47 ± 13.95	64.67 ± 10.87	63.18 ± 9.92	62.34 ± 11.54	0.814
Type of stroke (Hemorrhage: Ischemia)	18: 20	12: 24	10: 24	14: 21	0.411
Affected hemisphere (Right: Left: infratentorial)	10: 20: 8	10: 14: 12	6: 12: 16	5: 15: 15	0.265
Duration of onset of stroke (days)	30 (15–60)	18 (14–60)	20 (14.25–30)	25 (15–30)	0.433
BADL	28.95 ± 21.91	26.94 ± 22.62	21.47 ± 23.08	23.71 ± 20.66	0.489
MMSE	13.84 ± 6.71	17.43 ± 8.35	15.02 ± 6.43	14.60 ± 7.57	0.182
EAT-10	17.70 ± 8.72	17.84 ± 10.09	18.84 ± 6.76	18.89 ± 8.64	0.890
NRS 2002	3 (2–4)	2.5 (2–4)	3.25 (2.75–3.44)	3 (2–4)	0.412
WST	4 (3–5)	4 (3–5)	4 (4–5)	4 (4–5)	0.089
FEDSS	3.68 ± 0.93	3.69 ± 1.19	4.06 ± 0.95	4.06 ± 0.76	0.168
PAS	5.47 ± 1.64	5.19 ± 1.79	5.91 ± 1.38	5.46 ± 1.54	0.311
SSA	27.79 ± 4.83	27.61 ± 4.99	27.56 ± 4.35	27.71 ± 3.50	0.996
GUSS	6.42 ± 5.52	5.72 ± 4.77	5.59 ± 4.77	5.60 ± 4.91	0.874

Data are described as the mean ± SD or median (interquartile range). FEDSS, Fiberoptic Endoscopic Dysphagia Severity Scale; MMSE, Mini-Mental State Examination; PAS, Penetration/Aspiration Scale; WST, Water Swallow Test; SSA, Standardized Swallowing Assessment; BADL, Basic Activities of Daily Living; GUSS, Gugging Swallowing Screen.

Standardized Bedside Swallowing Assessment (SSA)

The SSA consists of three parts. One section comprises eight indicators, including the responsiveness level, breathing, sound intensity, lip closure, control of trunk and head, voluntary cough and pharyngeal reflex. It is scored vary from 8 to 23 points. In the second section, the patients swallowed 5 mL water three times, and at the same time, salivary management and laryngeal movement were assessed. Repetitive swallowing, stridor, choking, and vocal quality were also evaluated, with a score range of 5–11 points. Once patients completed the first two parts of the assessment, they underwent the third part that entailed swallowing 60 mL water; this activity was scored from 5 to 12 points. The total SSA score varied from 18 to 46 points, and higher scores indicated worse swallowing function (27, 28).

Penetration/Aspiration Scale (PAS)

Dysphagia severity was scored by an 8-point scale named the Penetration/Aspiration Scale (PAS). This scale was widely conducted for semiquantitative assessment of the degree of penetration and aspiration of endoscopic or radiological measurements, with higher scores indicating more severe impairment (29).

Gugging Swallowing Screen (GUSS)

The GUSS is a validated reliable screening test for swallowing with a maximum score of 20. This tool consists of two parts: five indirect questions were used to measure the swallow function of the patient, and four direct questions were conducted to assess the physical condition of patients when ingesting liquid, semisolid and solid food. A higher score suggested a milder condition of dysphagia, but a lower score suggested a more serious dysphagia condition. Fourteen points were deemed passing scores for swallowing, and patients who scored

<14 points were regarded as having a high likelihood of aspiration (30).

Statistical Analysis

In this study, statistical analyses were conducted with SPSS 23.0 software (SPSS Inc., Chicago, IL, USA). Two-way analysis of variance (ANOVA) was used for continuous data among multigroup comparisons (normal distribution), and the chi-squared test was performed for categorical data. To assess the effect of the interaction between intervention and time, repeated measure analysis of variance (ANOVA) was used, in which time was used as a within-subject factor and intervention as a between-subject factor. *Post-hoc* analysis was performed using Bonferroni correction. A Greenhouse-Geisser correction was performed to correct the non-sphericity of the data. A $P < 0.05$ was considered significantly different.

RESULTS

One hundred forty-seven subjects were randomized into four groups. The average ages in the unaffected hemisphere group, the affected hemisphere group, the cerebellum group and the control group were 64.47 ± 13.95 years (28 males and 10 females), 64.67 ± 10.87 years (28 males and 8 females), 63.18 ± 9.92 years (20 males and 14 females), and 62.34 ± 11.54 years (18 males and 17 females), respectively. There were no significant differences between the groups at baseline in clinical and demographic characteristics, Basic Activities of Daily Living (BADL) score, Mini-Mental State Examination (MMSE) score, Eating Assessment Tool-10 (EAT-10) score, Nutrition Risk Screening-2002 (NRS2002) score, Water Swallow Test (WST) score, FEDSS score, PAS score, SSA score, or GUSS score (Table 1).

Compared with baseline, the FEDSS and PAS scores of all groups improved at 4 weeks. The FEDSS scores were significantly

TABLE 2 | Clinical rating scales (FEDSS, PAS, SSA, and GUSS) for the four groups at each time.

	Unaffected	Affected	Cerebellum	Control	P-value	
FEDSS						
Baseline	3.68 ± 0.93	3.69 ± 1.19	4.06 ± 0.95	4.06 ± 0.76	0.168	
2 weeks	3.05 ± 1.16	3.06 ± 1.12	3.59 ± 1.21	3.77 ± 0.81	0.008	
4 weeks	2.53 ± 1.45	2.50 ± 1.32	2.76 ± 1.54	3.66 ± 1.11	0.001	
PAS						
Baseline	5.47 ± 1.64	5.19 ± 1.79	5.91 ± 1.38	5.46 ± 1.54	0.311	
2 weeks	4.03 ± 1.82	4.03 ± 2.16	4.41 ± 2.20	5.23 ± 1.17	0.024	
4 weeks	3.37 ± 2.17	3.53 ± 2.26	3.59 ± 2.56	5.00 ± 1.28	0.005	
SSA						
Baseline	27.79 ± 4.83	27.61 ± 4.99	27.56 ± 4.35	27.71 ± 3.50	0.996	
2 weeks	23.92 ± 4.57	22.86 ± 4.32	23.79 ± 3.83	26.03 ± 3.49	0.012	
4 weeks	21.66 ± 4.58	21.11 ± 3.66	21.79 ± 2.78	24.46 ± 3.27	0.001	
GUSS						
Baseline	6.42 ± 5.52	5.72 ± 4.77	5.59 ± 4.77	5.60 ± 4.91	0.874	
2 weeks	10.37 ± 6.28	8.78 ± 5.14	9.41 ± 6.57	6.23 ± 4.26	0.017	
4 weeks	11.37 ± 6.72	10.94 ± 6.38	11.24 ± 7.32	6.94 ± 3.95	0.008	
	Unaffected vs. affected (P-value)	Unaffected vs. cerebellum (P-value)	Affected vs. cerebellum (P-value)	Unaffected vs. control (P-value)	Affected vs. control (P-value)	Cerebellum vs. control (P-value)
FEDSS						
Baseline	1.000	0.631	0.718	0.625	0.712	1.000
2 weeks	1.000	0.232	0.254	0.033	0.038	1.000
4 weeks	1.000	1.000	1.000	0.003	0.003	0.044
PAS						
Baseline	1.000	1.000	0.375	1.000	1.000	1.000
2 weeks	1.000	1.000	1.000	0.043	0.048	0.442
4 weeks	1.000	1.000	1.000	0.008	0.024	0.039
SSA						
Baseline	1.000	1.000	1.000	1.000	1.000	1.000
2 weeks	1.000	1.000	1.000	0.176	0.008	0.148
4 weeks	1.000	1.000	1.000	0.008	0.001	0.018
GUSS						
Baseline	1.000	1.000	1.000	1.000	1.000	1.000
2 weeks	1.000	1.000	1.000	0.013	0.354	0.123
4 weeks	1.000	1.000	1.000	0.017	0.046	0.029

FEDSS, Fiberoptic Endoscopic Dysphagia Severity Scale; GUSS, Gugging Swallowing Screen. SSA, Standardized Swallowing Assessment; PAS, Penetration/Aspiration Scale.

different at 2 weeks ($P = 0.008$) and 4 weeks ($P = 0.001$). Similarly, there was a significant difference in PAS scores at 2 weeks ($P = 0.024$) and 4 weeks ($P = 0.005$) (Table 2). Figures 4A,B showed FEDSS and PAS scores at each time point in the four groups.

After 2 weeks of rTMS treatment, the improvement of dysphagia in the unaffected hemisphere group, the affected hemisphere group and the cerebellum group was significantly better than that in the control group. For the FEDSS, repeated measure analysis of variance showed a significant main effect of assessment time point ($F = 86.106$, $df = 1.724$, $P < 0.001$) and a significant time–group interaction ($F = 3.889$, $df = 5.173$, $P = 0.002$) (Table 2; Figure 4A).

The SSA and GUSS scores of all patients improved during the follow-up. There were significant differences in SSA scores at 2 weeks ($P = 0.012$) and 4 weeks ($P = 0.001$) (Table 2; Figure 4C). Similarly, at 2 weeks ($P = 0.017$) and 4 weeks ($P = 0.008$), the GUSS scores were significantly different. Repeated measure analysis of variance showed a significant main effect of the assessment time point ($F = 87.728$, $df = 1.416$, $P < 0.001$) and a significant interaction (time–group) for the GUSS ($F = 5.122$, $df = 4.372$, $P < 0.001$; Figure 4D).

Three participants (one unaffected and two affected) suffered transient headache. No participants developed seizures during or after therapy.

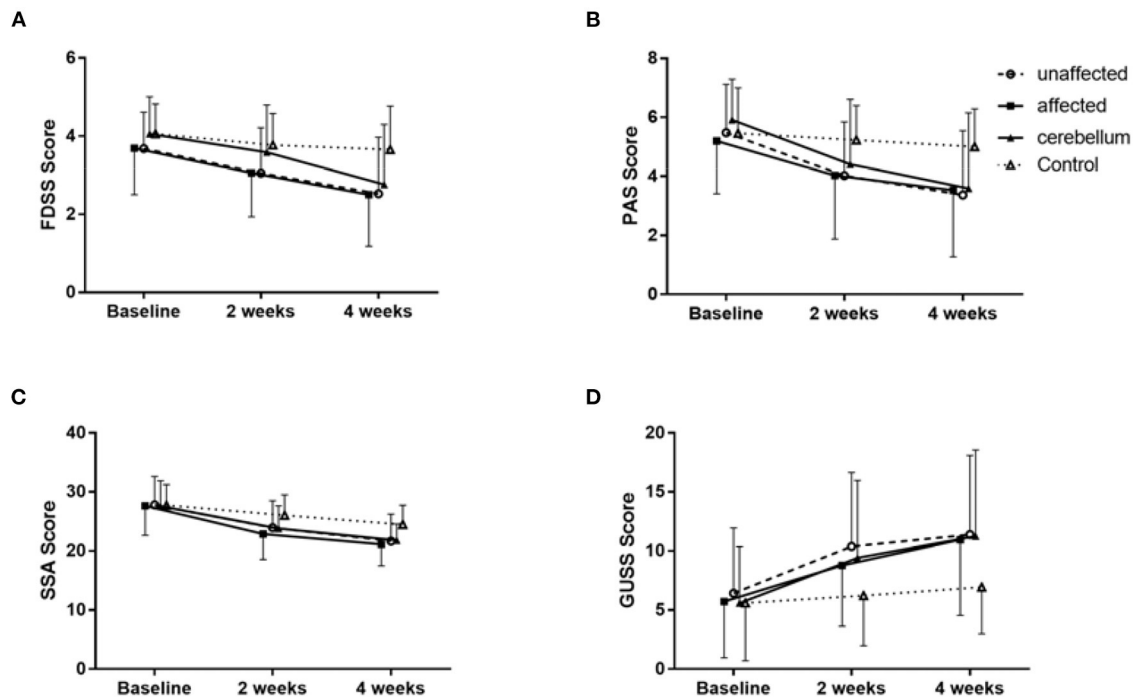


FIGURE 4 | Changes in the mean rating scores of FEDSS (A), PAS (B), SSA (C), and GUSS (D) at the three evaluation points in the four groups of patients. Data are described as the mean \pm SD. Each group showed significant improvement separately.

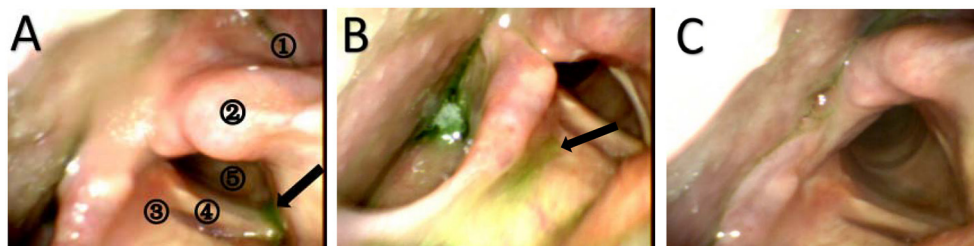


FIGURE 5 | Still images from the FEES examination of a 66-year-old man with dysphagia at three different times. (A) FEES examination before the treatment (baseline). The black arrow represents aspirated puree in the subglottis. The patient does not try to cough and clear the material. Therefore, the FEDSS score is 5 points, and the PAS score is 8 points. A1 = pyriform sinus, A2 = arytenoid, A3 = laryngeal vestibule, A4 = vocal fold, A5 = subglottic. (B) FEES examination after the treatment (2 weeks). Puree is attached to the laryngeal vestibule, and the patient tries to cough but cannot clear it. The FEDSS score was 4 points, and the PAS score was 3 points. (C) FEES examination at the time of follow-up. Food is not inhaled into the laryngeal vestibule or subglottis. The FEDSS score and PAS score were both 1 point.

DISCUSSION

Our study compared the effects of dysphagia intervention based on the stimulation site: the affected mylohyoid cortical area, unaffected mylohyoid cortical area and cerebellum. This study revealed large effect sizes for swallow scores (FEDSS, PAS, SSA, and GUSS) after the end of intervention in the unaffected hemispheric group, the affected hemispheric group and the cerebellum group compared to the control group. These results suggest that rTMS stimulation of the affected hemisphere, unaffected hemisphere and cerebellum was useful in improving swallowing function in patients with dysphagia after stroke. Nevertheless, the effects among these sites were not significantly

different. **Figure 5** shows the changes in FEDSS and PAS scores in a patient treated with rTMS.

The mechanism of rTMS is not fully understood. Some previous studies (13, 31) were based on the hypothesis that the balance of activity between the hemispheres of the brain is perturbed after stroke, leading to impaired neurological function. Neurophysiologically, this interhemispheric imbalance is considered to be caused by altered transcallosal inhibition, with an abnormal increase in excitability in the contralesional hemisphere inhibiting the ipsilesional hemisphere. Therefore, in some previous studies (13, 31), rTMS has been used to restore the balance between the hemispheres of the brain to improve functional outcomes. In recent years, studies have confirmed that

the projection of swallowing function in the human cerebral cortex is bilateral, with a dominant hemisphere that controls swallowing in patients with dysphagia (32, 33). High-frequency stimulation promotes cortical excitability, while low-frequency stimulation lowers excitability (34). rTMS can directly affect the cerebral cortex, effectively adjust the excitability of the cerebral cortex, reconstruct the central nervous system, form neural pathways, regulate swallowing centers, and improve swallowing function. Regarding the effects of cerebellar targeted rTMS, it is potentially interpreted that rTMS activates the cerebellar cortex, resulting in subsequent stimulation of dentate nuclei in each individual cerebellar hemisphere (24) because the functions of the cerebellum, which serves as a sensor and motor regulated organ, are predominantly suppressive (35). Hence, rTMS over the cerebellar cortices may lead to a decrease in inhibitory outflow and an increase in cortical activity. In this study, 5 Hz rTMS stimulation of the affected hemisphere, unaffected hemisphere and cerebellum may have facilitated swallowing function by improving cortical excitability of the mylohyoid cortex.

Previous studies have shown different outcomes in which various stimulation parameters of rTMS could improve the function of dysphagia in patients after stroke. For example, Park et al. (9) showed that high-frequency (5 Hz) rTMS application on the contralesional pharyngeal motor cortex was beneficial for poststroke dysphagic patients. Khedr et al. (11) proved that rTMS with 3 Hz high frequency at the lesional pharyngeal motor cortex resulted in significant improvement in dysphagia compared to a sham-stimulated group. These studies indicate that contralesional and lesional pharyngeal motor rTMS stimulation are both beneficial for reducing poststroke dysphagia. This is consistent with our research showing that rTMS stimulation at a high frequency in the unaffected hemisphere and affected hemisphere could significantly promote dysphagia recovery compared with the control group. The recovery of swallowing function may be related to changes in cortical excitability and neuroplasticity. Increases in cortical excitability by application of 5 Hz rTMS may increase stimulation to the motor neurons in the corticobulbar and corticospinal tracts, which enhances the synaptic innervations that project to the mylohyoid muscles, improves the movement of mylohyoid muscle, and promotes the recovery of swallowing function. Further neuroimaging tests or neurophysiologic evaluation are needed to delineate the underlying neuromechanism. Overall, our study and previous studies indicate that high-frequency rTMS stimulation of mylohyoid cortical tissue benefits poststroke dysphagia.

Recently, a growing number of studies have explored the possibility of rTMS on cerebellar tissue in the treatment of dysphagia. Some studies (19, 36) have shown that hemispheric cerebellar rTMS can increase cortical PMEP amplitudes. Vasant et al. (20) found that active cerebellar rTMS can increase PMEP amplitude, and their results indicated that cerebellar rTMS is a safe method that represents a potential treatment for poststroke dysphagia. Sasegbon et al. (24) demonstrated that high-frequency rTMS on the cerebellum could reverse the disruptive effects of a “virtual lesion.” These findings provide evidence for the development of cerebellar rTMS as a treatment for dysphagia after stroke. Our findings showed that

rTMS stimulation at a high frequency in the cerebellum could significantly promote dysphagia recovery compared with the control group. However, one study (37) showed that, compared with unilateral stimulation, bilateral cerebellar rTMS has a greater promotion effect on corticobulbar motor pathways to the pharynx and may be a more effective clinical therapy. Another study (19) found that 10 Hz rTMS seems to be the best frequency to promote excitement of the pharyngeal motor cortex. At present, the optimal stimulation parameters of rTMS on cerebellar tissue are still uncertain. More clinical trials are needed in the future to further improve the technology.

Recent studies show that compared to unilateral stimulation, bilateral pharyngeal stimulation with 10 Hz rTMS stimulation on “hot spots” has more positive outcomes in both acute and chronic stroke patients (38, 39). However, these trials did not compare the effects of ipsilesional and contralesional rTMS. Furthermore, they did not compare the effects of cerebellar rTMS to cerebral hemispheric rTMS. To the best of our knowledge, our study was the first to directly compare the therapeutic impact of high-frequency rTMS applications on the unaffected hemisphere, affected hemisphere and cerebellum to evaluate the effects on swallowing function applications in stroke patients. Our findings show no difference, based on FEDSS, PAS, SSA, and GUSS outcomes, among the affected hemisphere, unaffected hemisphere and cerebellum. Similarly, there was no statistically significant difference between the groups in the subgroup analysis of a meta-analysis according to intervention site (ipsilesional vs. contralesional site stimulation) (14). However, another meta-analysis reported that contralesional stimulation is better than ipsilesional stimulation (15). The meta-analysis involved interventions that included non-invasive brain stimulation, either rTMS or tDCS. The pooled effect showed high heterogeneity concerning dysphagia evaluations, population, stroke etiology, clinical characteristics of stroke, and intervention time after stroke onset. Therefore, more rigorously designed original studies are necessary to identify the effects of different stimulation sites.

This study may possess the following limitations. First, the difference in swallowing function rehabilitation by stroke type was not analyzed. We were not able to perform cerebellar subgroup analysis according to affected, unaffected and cerebellar stroke lesions on account of the insufficient number of patients with infratentorial stroke lesions. Second, the effect of rTMS in our study was evaluated based on the clinical severity and fiberoptic endoscopic dysphagia severity scale and not on neurophysiologic evaluation, such as MEP amplitude and latency of rTMS. Finally, the effect of rTMS on brain plasticity was not evaluated by neuroimaging tests or neurophysiologic evaluation in our study. In the future, the combination of neuroimaging studies and neurophysiology would be beneficial in exploring the potential mechanism of rTMS in the recovery of dysphagia.

CONCLUSIONS

The present study suggested that 5 Hz rTMS in the affected hemisphere, unaffected hemisphere and cerebellum for 10

days improves swallowing function in poststroke dysphagia patients. However, no difference among the affected hemisphere, unaffected hemisphere and cerebellum was observed. Therefore, regardless of whether the unaffected hemisphere or the affected hemisphere is stimulated, 5 Hz high-frequency rTMS on mylohyoid cortical tissue might have a positive effect on patients with poststroke dysphagia. In addition, cerebellar rTMS is a safe method that represents a potential treatment for poststroke dysphagia, and more clinical trials are needed to further improve this technique.

DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

ETHICS STATEMENT

The studies involving human participants were reviewed and approved by the Ethics Committee of Yue Bei People's Hospital. The patients/participants provided their written informed consent to participate in this study. Written informed consent

was obtained from the individual(s) for the publication of any potentially identifiable images or data included in this article.

AUTHOR CONTRIBUTIONS

HL contributed to the conception of the study, supervised the clinical trial, and performed manuscript writing and editing. LZ, JW, and JR performed data analyses and manuscript writing and editing. PW and YZ contributed to the conception and design of the study. FL and YP performed data collection. All authors have agreed with the submitted version of the manuscript.

FUNDING

This study was funded by grants from Guangdong Science and Technology Department, No. 201803010.

ACKNOWLEDGMENTS

The authors are grateful to the patients, providers, administrative staff, and management staff at Yue Bei People's Hospital who made the trial possible.

REFERENCES

- Rofes L, Vilardell N, Clave P. Post-stroke dysphagia: progress at last. *Neurogastroenterol Motil.* (2013) 25:278–82. doi: 10.1111/nmo.12112
- Clavé P, Rofes L, Carrión S, Ortega O, Cabré M, Serra-Prat M, et al. Pathophysiology, relevance and natural history of oropharyngeal dysphagia among older people. *Nestle Nutr Inst Workshop Ser.* (2012) 72:57–66. doi: 10.1159/000339986
- Vose A, Nonnenmacher J, Singer ML, González-Fernández M. Dysphagia management in acute and sub-acute stroke. *Curr Phys Med Rehabil Rep.* (2014) 2:197–206. doi: 10.1007/s40141-014-0061-2
- Bath PM, Lee HS, Everton LF. Swallowing therapy for dysphagia in acute and subacute stroke. *Cochrane Database Syst Re.* (2018) 10:CD000323. doi: 10.1002/14651858.CD000323.pub3
- Ni X, Lin H, Li H, Liao W, Luo X, Wu D, et al. Evidence-based practice guideline on integrative medicine for stroke 2019. *J Evid Based Med.* (2020) 13:137–52. doi: 10.1111/jebm.12386
- Ziemann U. Improving disability in stroke with RTMS. *Lancet Neurol.* (2005) 4:454–5. doi: 10.1016/S1474-4422(05)70126-5
- Machii K, Cohen D, Ramos-Estebanez C, Pascual-Leone A. Safety of rTMS to non-motor cortical areas in healthy participants and patients. *Clin Neurophysiol.* (2006) 117:455–71. doi: 10.1016/j.clinph.2005.10.014
- Gow D, Rothwell J, Hobson A, Thompson D, Hamdy S. Induction of long-term plasticity in human swallowing motor cortex following repetitive cortical stimulation. *Clin Neurophysiol.* (2004) 115:1044–51. doi: 10.1016/j.clinph.2003.12.001
- Park JW, Oh JC, Lee JW, Yeo JS, Ryu KH. The effect of 5Hz high-frequency rTMS over contralesional pharyngeal motor cortex in post-stroke oropharyngeal dysphagia: a randomized controlled study. *Neurogastroenterol Motil.* (2013) 25:324–e250. doi: 10.1111/nmo.12063
- Khedr EM, Abo-Elfetoh N. Therapeutic role of rTMS on recovery of dysphagia in patients with lateral medullary syndrome and brainstem infarction. *J Neurol Neurosurg Psychiatry.* (2010) 81:495–9. doi: 10.1136/jnnp.2009.188482
- Khedr EM, Abo-Elfetoh N, Rothwell JC. Treatment of post-stroke dysphagia with repetitive transcranial magnetic stimulation. *Acta Neurol Scand.* (2009) 119:155–61. doi: 10.1111/j.1600-0404.2008.01093.x
- Lee JH, Kim SB, Lee KW, Lee SJ, Lee JU. Effect of repetitive transcranial magnetic stimulation according to the stimulation site in stroke patients with dysphagia. *Ann Rehabil Med.* (2015) 39:432–9. doi: 10.5535/arm.2015.39.3.432
- Du J, Yang F, Liu L, Hu J, Cai B, Liu W, et al. Repetitive transcranial magnetic stimulation for rehabilitation of poststroke dysphagia: a randomized, double-blind clinical trial. *Clin Neurophysiol.* (2016) 127:1907–13. doi: 10.1016/j.clinph.2015.11.045
- Yang SN, Pyun SB, Kim HJ, Ahn HS, Rhyu BJ. Effectiveness of non-invasive brain stimulation in dysphagia subsequent to stroke: a systemic review and meta-analysis. *Dysphagia.* (2015) 30:383–91. doi: 10.1007/s00455-015-9619-0
- Pisegna JM, Kaneoka A, Pearson WG Jr, Kumar S, Langmore SE. Effects of non-invasive brain stimulation on post-stroke dysphagia: a systematic review and meta-analysis of randomized controlled trials. *Clin Neurophysiol.* (2016) 127:956–68. doi: 10.1016/j.clinph.2015.04.069
- Michou E, Mistry S, Jefferson S, Tyrrell P, Hamdy S. Characterizing the mechanisms of central and peripheral forms of neurostimulation in chronic dysphagic stroke patients. *Brain Stimul.* (2014) 7:66–73. doi: 10.1016/j.brs.2013.09.005
- Hamdy S, Rothwell JC, Brooks DJ, Bailey D, Aziz Q, Thompson DG. Identification of the cerebral loci processing human swallowing with H2(15)O PET activation. *J Neurophysiol.* (1999) 81:1917–26. doi: 10.1152/jn.1999.81.4.1917
- Sasegbon A, Niziolek N, Zhang M, Smith CJ, Bath PM, Rothwell J, et al. The effects of midline cerebellar rTMS on human pharyngeal cortical activity in the intact swallowing motor system. *Cerebellum.* (2021) 20:101–15. doi: 10.1007/s12311-020-01191-x
- Vasant DH, Michou E, Mistry S, Rothwell JC, Hamdy S. High-frequency focal repetitive cerebellar stimulation induces prolonged increases in human pharyngeal motor cortex excitability. *J Physiol.* (2015) 593:4963–77. doi: 10.1113/jp270817
- Vasant DH, Sasegbon A, Michou E, Smith C, Hamdy S. Rapid improvement in brain and swallowing behavior induced by cerebellar repetitive transcranial magnetic stimulation in poststroke dysphagia: a single patient case-controlled study. *Neurogastroenterol Motil.* (2019) 31:e13609. doi: 10.1111/nmo.13609
- Wilkinson G, Sasegbon A, Smith CJ, Rothwell J, Bath PM, Hamdy S. An exploration of the application of noninvasive cerebellar stimulation in the neuro-rehabilitation of dysphagia after stroke

- (EXCITES) protocol. *J Stroke Cerebrovasc Dis.* (2020) 29:104586. doi: 10.1016/j.jstrokecerebrovasdis.2019.104586
22. Yao X, Florez ID, Zhang P, Zhang C, Zhang Y, Wang C, et al. Clinical research methods for treatment, diagnosis, prognosis, etiology, screening, and prevention: a narrative review. *J Evid Based Med.* (2020) 13:130–6. doi: 10.1111/jebm.12384
 23. Hamdy S, Aziz Q, Rothwell JC, Singh KD, Barlow J, Hughes DG. The cortical topography of human swallowing musculature in health and disease. *Nat Med.* (1996) 2:1217–24. doi: 10.1038/nm1196-1217
 24. Sasegbon A, Watanabe M, Simons A, Michou E, Vasant DH, Magara J, et al. Cerebellar repetitive transcranial magnetic stimulation restores pharyngeal brain activity and swallowing behaviour after disruption by a cortical virtual lesion. *J Physiol.* (2019) 597:2533–46. doi: 10.1113/JP277545
 25. Rossi S, Hallett M, Rossini PM, Pascual-Leone A. Safety, ethical considerations, and application guidelines for the use of transcranial magnetic stimulation in clinical practice and research. *Clin Neurophysiol.* (2009) 120:2008–39. doi: 10.1016/j.clinph.2009.08.016
 26. Warnecke T, Ritter MA, Kroger B, Oelenberg S, Teismann I, Heuschmann PU, et al. Fiberoptic endoscopic Dysphagia severity scale predicts outcome after acute stroke. *Cerebrovasc Dis.* (2009) 28:283–9. doi: 10.1159/000228711
 27. Perry L. Screening swallowing function of patients with acute stroke. Part one: identification, implementation and initial evaluation of a screening tool for use by nurses. *J Clin Nurs.* (2001) 10:463–73. doi: 10.1046/j.1365-2702.2001.00501.x
 28. Perry L. Screening swallowing function of patients with acute stroke. Part two: detailed evaluation of the tool used by nurses. *J Clin Nurs.* (2001) 10:474–81. doi: 10.1046/j.1365-2702.2001.00502.x
 29. Rosenbek JC, Robbins JA, Roecker EB, Coyle JL, Wood JL. A penetration-aspiration scale. *Dysphagia.* (1996) 11:93–8. doi: 10.1007/BF00417897
 30. Trapl M, Enderle P, Nowotny M, Teuschl Y, Matz K, Dachenhausen A, et al. Dysphagia bedside screening for acute-stroke patients: the Gugging Swallowing Screen. *Stroke.* (2007) 38:2948–52. doi: 10.1161/STROKEAHA.107.483933
 31. Ünlüer NÖ, Temuçin ÇM, Demir N, Serel Arslan S, Karaduman AA. Effects of low-frequency repetitive transcranial magnetic stimulation on swallowing function and quality of life of post-stroke patients. *Dysphagia.* (2019) 34:360–71. doi: 10.1007/s00455-018-09965-6
 32. Lowell SY, Reynolds RC, Chen G, Horwitz B, Ludlow CL. Functional connectivity and laterality of the motor and sensory components in the volitional swallowing network. *Exp Brain Res.* (2012) 219:85–96. doi: 10.1007/s00221-012-3069-9
 33. Malandraki GA, Sutton BP, Perlman AL, Karampinos DC, Conway C. Neural activation of swallowing and swallowing-related tasks in healthy young adults: an attempt to separate the components of deglutition. *Hum Brain Mapp.* (2009) 30:3209–26. doi: 10.1002/hbm.20743
 34. Michou E, Raginis-Zborowska A, Watanabe M, Lodhi T, Hamdy S. Repetitive transcranial magnetic stimulation: a novel approach for treating oropharyngeal dysphagia. *Curr Gastroenterol Rep.* (2016) 18:10. doi: 10.1007/s11894-015-0483-8
 35. Roostaei T, Nazeri A, Sahraian MA, Minagar A. The human cerebellum: a review of physiologic neuroanatomy. *Neurol Clin.* (2014) 32:859–69. doi: 10.1016/j.ncl.2014.07.013
 36. Jayasekeran V, Rothwell J, Hamdy S. Non-invasive magnetic stimulation of the human cerebellum facilitates cortico-bulbar projections in the swallowing motor system. *Neurogastroenterol Motil.* (2011) 23:831–e341. doi: 10.1111/j.1365-2982.2011.01747.x
 37. Sasegbon A, Smith CJ, Bath P, Rothwell J, Hamdy S. The effects of unilateral and bilateral cerebellar rTMS on human pharyngeal motor cortical activity and swallowing behavior. *Exp Brain Res.* (2020) 238:1719–33. doi: 10.1007/s00221-020-05787-x
 38. Park E, Kim MS, Chang WH, Oh SM, Kim YK, Lee A, et al. Effects of bilateral repetitive transcranial magnetic stimulation on post-stroke dysphagia. *Brain Stimul.* (2017) 10:75–82. doi: 10.1016/j.brs.2016.08.005
 39. Zhang C, Zheng X, Lu R, Yun W, Yun H, Zhou X. Repetitive transcranial magnetic stimulation in combination with neuromuscular electrical stimulation for treatment of post-stroke dysphagia. *J Int Med Res.* (2019) 47:662–72. doi: 10.1177/0300060518807340

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Copyright © 2021 Zhong, Rao, Wang, Li, Peng, Liu, Zhang and Wang. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.

Advantages of publishing in Frontiers



OPEN ACCESS

Articles are free to read
for greatest visibility
and readership



FAST PUBLICATION

Around 90 days
from submission
to decision



HIGH QUALITY PEER-REVIEW

Rigorous, collaborative,
and constructive
peer-review



TRANSPARENT PEER-REVIEW

Editors and reviewers
acknowledged by name
on published articles

Frontiers

Avenue du Tribunal-Fédéral 34
1005 Lausanne | Switzerland

Visit us: www.frontiersin.org

Contact us: frontiersin.org/about/contact



REPRODUCIBILITY OF RESEARCH

Support open data
and methods to enhance
research reproducibility



DIGITAL PUBLISHING

Articles designed
for optimal readership
across devices



FOLLOW US

@frontiersin



IMPACT METRICS

Advanced article metrics
track visibility across
digital media



EXTENSIVE PROMOTION

Marketing
and promotion
of impactful research



LOOP RESEARCH NETWORK

Our network
increases your
article's readership