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Complex modulation of fingertip forces during precision grasp and lift after theta burst stimulation over the dorsal premotor cortex

Kompleksna modulacija sila tokom preciznog hvata šake primenom ponavljane transkranijalne magnetne stimulacije pražnjenjima u teta frekvenciji iznad dorzalnog premotornog korteksa

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Abstract

Background/Aim. Adaptive control and fingertip force synchronization of precise grasp stability during unimanual manipulation of small objects represents an illustrative example of highly fractionated movements that are foundation of fine motor control. It is assumed that this process is controlled by several motor areas of the frontal lobe, particularly applicable to the primary motor (M-1) and dorsal premotor cortex (PMd). Aiming to examine the role of PMd during fine coordination of fingertip forces we applied theta burst repetitive magnetic stimulation (TBS) to disrupt neural processing in that cortical area. **Methods.** Using a single-blind, randomized, crossover design, 10 healthy subjects (29 ± 3.9 years) received single sessions of continuous TBS (cTBS600), intermittent TBS (iTBS600), or sham stimulation, separate from one another at least one week, over the PMd region of dominant hemisphere. Precision grasp and lift were assessed by instrumented device, recording grip (G) and load (L) forces, during three manipulation tasks (ramp-and-hold, oscillation force producing and simple lifting tasks), with each hand separately, before and after interventions. **Results.** We observed the improvement of task performance related to constant error (CE) in oscillation task with the dominant hand (DH) after the iTBS ($p = 0.009$). On the contrary, the cTBS reduced variable error (VE) for non-dominant hand (NH), $p = 0.005$. Considering force coordination we found that iTBS worsened variables for NH (G/L ratio, $p = 0.017$; cross-correlation of the G and L, $p = 0.047$; Gain, $p = 0.047$). **Conclusion.** These results demonstrate the ability of TBS to modulate fingertip forces during precision grasping and lifting, when applied over PMd. These findings support the role of PMd in human motor control and forces generation required to hold small objects stable in our hands.

Key words:
motor cortex; transcranial magnetic stimulation; hand strength.

Apstrakt

Uvod/Cilj. Adaptivna kontrola i sinhronizacija sila prstiju šake tokom preciznog hvata pri manipulisanju malim predmetima jednom rukom predstavlja ilustrativni primer visoko frakcionisanih pokreta koji predstavljaju temelj motorne kontrole preciznih pokreta. Pretpostavlja se da ovim procesom upravlja nekoliko motornih oblasti frontalnog režnja, i to prvenstveno primarni motorni (M-1) i dorzalni premotorni korteks (PMd). Cilj istraživanja bio je ispitivanje uloge PMd-a tokom vršenja pokreta koji zahtevaju finu koordinaciju sila prstiju šake. U istraživanju smo primenili ponavljanu magnetnu stimulaciju pražnjenjima u teta frekvenciji, kako bi ometali neuralno procesiranje u toj oblasti moždane kore. **Metode.** Primenom jednostrano slepe studije, uz nasumičnu raspodelu i ukršteni dizajn, 10 zdravih ispitanika (29 ± 3,9 godina) bilo je izloženo pojedinačnim sesijama kontinuirane magnetne stimulacije (cTBS600), ili intermitentne ponavljane magnetne stimulacije (iTBS600), pražnjenjima u teta frekvenciji kao i prividnoj stimulaciji iznad PMd regiona dominantne hemisfere, odvojenih međusobno, najkraće nedelju dana. Precizanost hvata šake i podizanja procenjivani su uređajem koji je registrovao silu stiska (G) i silu podizanja (L) prilikom izvođenja tri zadatka (zadatak sa zadatim profilom L, zadatak sa oscilatornim variranjem nivoa L i zadatak sa podizanjem), koji su izvođeni sa obe ruke odvojeno, i to pre i nakon svake intervencije. **Rezultati.** Nakon primene iTBS protokola zabeleženo je poboljšanje izvođenja iskazano konstantnom greškom (CE) u zadatku sa oscilatornim variranjem nivoa L, kada je izvođen dominantnom rukom (DH), $p = 0.009$. Suprotno to-

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me, primena cTBS protokola dovela je do smanjenja promenjive greške (VE) za nedominantnu ruku (NH), $p = 0.005$. Sa aspekta koordinacije sila utvrđeno je da je iTBS protokol doveo do pogoršanja rezultata praćenih pokazatelja za nedominantnu ruku (G/L odnos, $p = 0.017$; korelacija G i L, $p = 0.047$; prirast sile $p = 0.047$). **Zaključak.** Rezultati našeg istraživanja ukazuju na mogućnost modulacije sila prstiju šake tokom preciznog hvata i podizanja, ukoliko se TBS primeni iznad PMd-a. Dobijeni nalazi podržavaju ulogu PMd u motornoj kontroli i generisanju sila neophodnih za stabilno držanje malih predmeta kod ljudi.

Ključne reči:
motorna kora; transkranijalna magnetna stimulacija; ruka, snaga.

Introduction

The development of a skilled and sophisticated grasping technique represents one of the key evolutionary advantages of human beings comparing to subhuman primates [1]. Therefore, grasping is a subject of interest of many researchers, given the importance of precision grasping in the activities of daily life [2].

In order to evaluate these functions, different manipulandums have been developed, that serve to evaluate complex control over precision grip and coordination of grip and load forces applied to the object [3, 4].

Hand grip force and their coordination are controlled by the nervous system, so that a number of receptors (visual, mechanoreceptors, tactile receptors) passed through somatosensory afferents information about the mechanical characteristics of cases [3, 5] as well as change the path of movement, and through feedback [6, 7] and feed-forward mechanisms [8, 9] which regulate the process. However, in addition to afferent mechanisms of motor control, the precise modulation of grip and load force is provided by the activation of primary and non-primary motor areas. Despite the fact that the primary motor cortex (M-1) and its main output projection, the corticospinal tract, are considered as neural basis of hand dexterity, there are several non-primary motor areas (premotor, supplementary motor, and cingulate motor areas). These parts of the frontal lobe play a role in modulation of the output signal at the levels of the M-1 and spinal cord [10]. Most of the findings related to the role of non-primary motor areas are collected on the basis of cell recordings on monkeys [11]. However, the trains of magnetic pulses, repetitive transcranial magnetic stimulation (rTMS) applied over intact scalp, provide the new tool to investigate modulation of motor output with humans awake, on safe and painless way. Because the effects of rTMS extend beyond the period of stimulation, there is a possibility to modulate cortical plasticity. In the case of creation of so-called virtual lesions of restricted brain areas, trains of TMS pulses temporarily interfere with neural processing while the subject is performing behavioral tasks. Through rTMS, there are different possibilities of modulation functions at the very site of stimulation, but also on other distal sites producing a disinhibition through the synaptic connections [12].

Contrary, to extensively study the role of M-1 and the corticospinal projection in control of skilled hand movements, the role of premotor cortex in this function is less known. The success of the skilled manipulation of objects with hand depends on setting hand grip before the object is reached, requiring cooperation of visuo-motor and sensory-motor loops, the kind of transformation that takes place within the parieto-frontal connections, including the M-1, but not least ventral premotor cortex (PMv) [13, 14]. It has been shown that the function of premotor dorsal cortex (PMd) in monkeys refers to the planning and execution of reaching movements. However, in humans, the contribution of PMd in the execution of complex hand grip is reflected through connecting sensory information with motor actions [15], as well as visually guided activities [16], although many aspects remain essentially unknown.

Transcranial magnetic stimulation represents non-invasive, safe and painless method aimed to activate restricted neuronal population at target point, with purpose of modulating activity of certain cortical area. Depending on the stimulation intensity, the cortical interneurons are commonly activated, and only at higher intensities the pyramidal cells could discharge, too. However, in this way, the excitatory and inhibitory neurons are activated at the same time, and related to the stimulation pattern, the net effect of repetitive TMS could be either inhibitory or facilitatory. However, there is an additional differences between M-1 and PMd, because functional imaging studies have revealed the activation of premotor regions in both hemispheres, contrary to primarily M-1 activation on the contralateral side during a variety of motor tasks, including isolated movements of the distal arm (e.g. opening a drawer and retrieving food with the same hand) [17, 18].

The aim of this study was to determine the role and contribution of PMd during precision grip in healthy subjects assessed by kinetic analysis of various static and dynamic manipulation tasks with both hands after rTMS intervention over dominant PMd.

Methods

Subjects

Ten healthy volunteers (6 males) aged 29 ± 3.9 years, without history of any neurological and psychiatric conditions, neurosurgery, or metal or electronic implants participated in the study. Subjects were screened for potential risk of adverse reactions to TMS by using the adult safety screen questionnaire for transcranial magnetic stimulation [19]. None of the subjects did take any CNS-acting medications.

Nine subjects were right-handed and one was left-handed according to the Edinburgh handedness inventory [20]. Considering hand motor with manipulandum applied in the study, none of the subjects had previous experience.

The experimental protocol was approved and monitored by the local ethics committee according to the Declaration of Helsinki (www.wma.net/en/30publications). After an explanation of the treatment procedures, all subjects signed a written informed consent.

Grip-lift tasks

Subjects were seated in a comfortable chair in front of the manipulandum which consisted of the single handle in the form of lever with the grasping surfaces covered by rubber (Figure 1A) and steel stand fixed to the table. A single-axis force transducer (SW-20L, CAS Cor., NY, USA; range 200 N; linearity 0.03%; hysteresis 0.03%) located inside the handle recorded the grip force (G) of the finger and the thumb applied perpendicularly against the opposing grasping surface. Another single-axis transducer (LCM300 FUTEK Advanced Sensor Technology, Inc, CA, USA; range 450 N; non-linearity 0.5%; hysteresis 0.5%), located at the bottom of the handle, recorded the load force (L) exerted tangentially to the grasping surfaces. With its lower part load force transducer is attached to the spherical joint so that the force that transmits to the fixed transducer L when pulling the handle upwards is always projected in the ideal vertical position. By the spherical joint, the handle could be either externally fixed to the steel stand, or attached to additional weights and be free to move. Additional weights in steps of 100 and 200 g of mass served to adjust the total weight of device to the prescribed L_{max}.

Within a single session subjects were tested on three manipulation tasks – two "static" (ramp-and-hold and oscillatory task) and one "dynamic" (lifting task) [21]. Each experimental task was well explained and demonstrated by experimenter. Thereafter, subjects were submitted to a familiarization procedure practicing manipulation tasks unimanualy, with three practice trials performed by each hand. After practicing subjects performed four experimental trials and the last three trials were taken for further analysis. The sequence of tasks, as well as the sequence of hands within each task was pseudo-randomized. During testing subjects were focused on the movement task based on L exertion, since G was never mentioned throughout the entire experiment. All measurements conducted by same experienced investigator. Figure 1B illustrate horizontal projection of subjects' body while performing the task [21].

While performing ramp-and-hold task (R&H-T), manipulandum was externally fixed to the steel stand. Subjects were asked to match a prescribed L_{max} profile by pulling up device corresponding to a gradual increase and, thereafter, a steady L exertion against an externally fixed device. Both, the prescribed L_{max}, as well as the current value of L were displayed on a computer monitor placed in front of the seated subject. The profile had the following three phases: zero L (duration 1 s), gradually increasing L (3 s), and constant L (3 s) (Figure 2A). The initiation of each phase and the termination of the last one were indicated by four consecutive computer-generated auditory beeps.

In the oscillatory task (Osc-T) of subjects were expected to correspond to a rapidly changing L against externally fixed device. They were instructed to exert a sinusoidal L on the computer monitor, by pulling the device vertically (upward-downward) in a way that L minima and maxima corresponded to 0 N and the individually prescribed L_{max} (Figure 2B). The computer monitor displayed those horizontal lines depicting the prescribed minima and the maxima, as well as the current value of L. Frequency of oscillatory variations (1.33 Hz) was set by a metronome, while duration of the trials was 8 s.

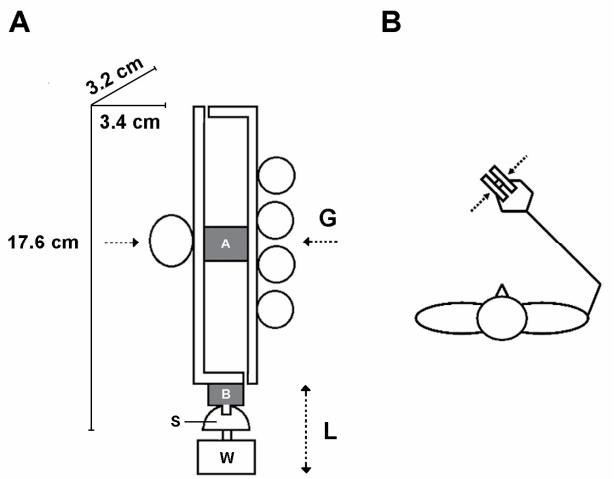

Fig. 1 – (A) Schematic illustration of experimental manipulandum for the assessment of grip and load performance and force coordination. The circles illustrate the position of the tips of the fingers and the thumb of subject's hand applying a precise grasp against the manipulandum. Letters A and B denote grip and load force transducers – load force transducers records grip force (G) and load force (L), respectively. S indices spherical joint, and W – additional weight or, alternatively, fixation of the manipulandum to the steel stand; (B) Position of subject during the task performance - top view.

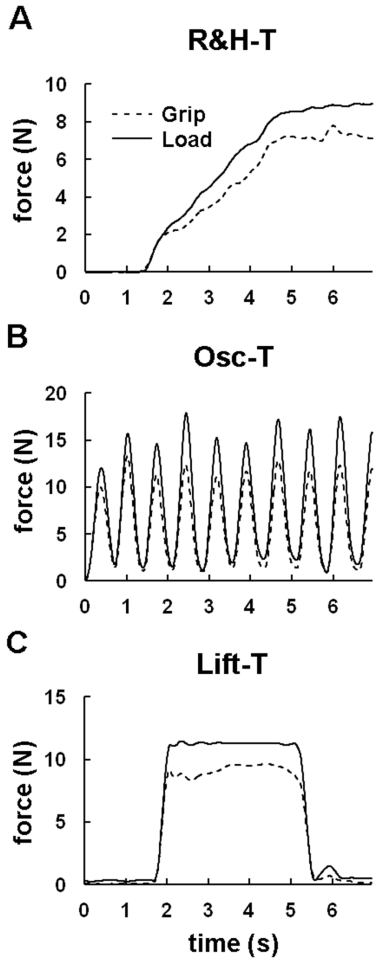

Fig. 2 – Grip and load force exerted against the manipulandum in the: A) ramp-and-hold task (R&H-T), oscillation task (Osc-T) and lifting task (Lift-T) obtained from a healthy subject.

In the lifting task (Lift-T), based on individually prescribed L_{max}, manipulandum was attached with additional weights served to adjust the total weight of device to the prescribed L_{max}. The subjects were instructed to prepare their hand for grasping the device by opening their fingers near the grasping area without touching it. Upon the first computer-generated beep, subject grasped the device, lifted it approximately 3 cm above the table, and held steady until the second beep (3 s later) and, thereafter, place it back on the table and release (Figure 2C).

Transcranial magnetic stimulation

The subjects were seated in a reclining chair that allowed them to keep their arms and hands relaxed during TMS and recording of motor evoked potentials (MEPs).

Single Pulse TMS

Magnetic stimulation was delivered by a 70-mm figure-eight coil and a Magstim Rapid^2 (Magstim Co., Whitland, UK) stimulator for rTMS and a Bistim module (Magstim) for single pulse TMS. MEPs were recorded from the thenar muscle (*abductor pollicis brevis* – APB) using surface electrodes and (Medelec Synergy, VIASYS Healthcare, UK) with a band pass of 20 to 2,000 Hz. Resting motor threshold (RMT) was determined in the contralateral APB muscle, determined with TMS delivered to the optimal scalp site for induction of MEPs in target muscle, according to international standards [22]. The coil was placed tangentially to the scalp, with the handle pointing 45° posterolaterally.

Thirty magnetic pulses were delivered successively (inter-trial interval of 5 ± 1.2 sec), at the intensity optimal to evoke MEPs of 1 mV amplitude (measured from peak to peak). The intensity was approximately between 120-130 % RMT. The time points of MEP measurements were immediately before (PRE) and after (POST) intervention.

Repetitive TMS

Theta burst stimulation (TBS) was performed according to current safety recommendations [23], using original protocols with triplets of very short bursts at 50 Hz repeated at 0.2 s (5 Hz – the range of EEG theta frequency band) for a total of 600 pulses. Therefore, $cTBS_{600}$ protocol lasted for 40 s, while $iTBS_{600}$ protocol includes 10 burst of triplets who were applied every 10 seconds (with pause of 8 s) causing the delivery of 600 pulse over a period of 190 s. Sham TBS was delivered using a matching coil produced by Magstim that delivers only 5% of the stimulator output, but with similar clicking sound produced mechanically by the sham coil with each TMS pulse.

TBS was applied at subthreshold level (80% of RMT), over the PMd of the dominant hemisphere. The stimulation point on the scalp was determined in accordance with the PMd localization at the specific location situated about 2 cm rostral to the representation of hand muscles in the primary motor cortex (half of the distance between Cz and Fz and 15% of the distance from tragus to tragus to the left) [24].

Experimental design

All recordings were conducted in the Laboratory for non-invasive brain stimulation, Military Medical Academy (MMA) in Belgrade. Experiment was carried out in three individual sessions for each individual subject, each separated not shorter than one week. After an evaluation of the excitability of the motor cortex, surface electromiography (EMG) electrodes were removed, and the place where they had been placed was labeled by the marker. Subjects then carried out the hands manipulative tasks (Figure 3).

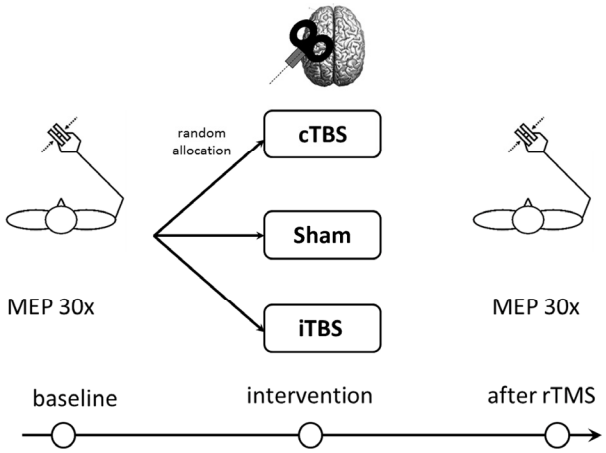

Fig. 3 – Overview of the experimental design. All subjects underwent three different interventional protocols (intermittent theta burst stimulation – iTBS, continous theta burst stimulation – cTBS and Sham), in a crossover study design. At the baseline, immediately before intervention, motor evoked potentials (MEP) amplitudes were collected, and grip performance and force coupling were evaluated by unimanual task on three different tasks: ramp and hold task (R&H-T), oscillation task (Osc-T) and lifting task (Lift-T). The same procedure was performed immediately after intervention, aiming to evaluate each single-session effects.

Before starting the test fingertips were cleaned with alcohol. Since previous results of Jaric et al. [25] have shown that prolonged tasks require L below 15% of the maximum G to avoid fatigue, maximum G exerted by tips of all 5 fingers of each hand was recorded separately. Ten percent of the maximum G of the weaker hand was prescribed as the maximum L (L_{max}) [21] in each of the experimental tasks and was participant specific (range 5-17 N).

After the baseline evaluation, TBS protocols or sham were applied in pseudo-randomized order. Following interventions, all baseline procedure were repeated immediately in the same way.

Data processing

A custom made LabView application (National Instruments, Austin, TX, USA) was used for the data acquisition and processing of data obtained from the grip-and-lift tasks. The signals from both transducers were A/D converted and recorded at the sampling rate of 200 Hz. The raw force data were low-pass filtered at 10 Hz with a fourth order (zero-phase lag) Butterworth filter [21]. In the R&H-T the ramp

phase and the hold phase were separately analyzed [21]. To exclude the initial and final adjustments, in the Osc-T, only the middle 5 s were analyzed [26]. The lift phase (the initiation of lifting, starts when L reaches 8 % of L_{max} and ends with reaching L_{max}) and the hold phase (interval of 2 s, after the period of 0.25 s when L reaches L_{max}) in the Lift-T, were also analyzed separately [21].

Based on directly measured variables (G and L) obtained using LabView application, derived variables were calculated. To assess hand function, two groups of dependent variables were selected. The ability of subject to exert the required pattern of L was assessed by task performance variables (describing how successful subject were regarding performing the instructed task), while the ability of subject to exert the required pattern of L was assessed by root mean square error (RMSE) of L in the R&H-T and coefficient of variation (CV) of L in the Lift-T. Constant error (CE), calculated as a difference of peaks of L and required level of force, and variable error (VE), assessed by standard peak deviations of L, were selected as an indices of task performance in the Osc-T [21].

Force coordination variables describing to what extent G and L were coordinating and assessed the relationship between the temporal profiles of G and L. G-L scaling, assessed by grip-to-load ratio (G/L ratio) evaluated the magnitude of G with respect to the magnitude of L assuming that lower ratio was index of better coordination [27]. It was calculated from the steady holding phases of the R&H-T and the Lift-T, as well as from the averaged G and L of the Osc-T [21]. As an index of G-L coupling, the cross-correlation of the G and L (r) of the R&H-T and the Osc-T were used [25]. Note that maximum correlation coefficient, based on previous studies, should indicate higher force coordination [28,29]. G-L modulation was assessed from G–L diagrams (the slope and intercept were interpreted as Gain and Offset, respectively) of the Osc-T [26,29,30]. Higher force coordination was expected to be revealed by high Gain and low Offset of G [31].

Statistical analysis

For the assessment of normality of distribution the Shapiro-Wilk test was used. To assess the effects of intervention protocols (Sham *vs* iTBS *vs* iTBS) on the global excitability of the motor cortex, the results obtained before (PRE) and after intervention (POST) are normalized (POST/PRE) and ANOVA for repeated measures and *post-hoc* test with Bonferroni correction were used. To assess effects of TMS intervention on hand function, non-parametric statistics was applied. Potential differences between TMS protocols PRE, for the dominant (DH) and the non-dominant hand (NH), separately, were assessed by Friedman's test. Differences between DH and NH PRE and POST, separately, as well as for the results obtained PRE and POST for each of the three interventions, for each hand separately, were assessed by Wilcoxon's signed-rank test. To assess the differences between the interventions, the results obtained PRE and POST were normalized (POST/PRE) and the Friedman's test was applied. When significant differences were found, additional Wilcoxon's signed-rank tests with Bonferroni correction were performed. The level of statistical significance was set to $p < 0.05$. Statistical analysis was performed in SPSS v 20.0 (SPSS Inc., Chicago, USA).

Results

Table 1 shows baseline data of kinetic analysis of static and dynamic manipulation tasks with an instrumented device that recorded the grip and load force, before interventions.

The effects of interventions on motor cortex excitability

The evaluation of motor cortex excitability was performed through comparison of resting motor threshold ($F_{(2,14)} = 0.41$, $p = 0.575$) and MEP modulation (starting from baseline value of 1 mV). Normalized data for MEP modulation have shown significant differences between $iTBS_{600}$ (137.28 ± 27.76) *vs* Sham (99.58 ± 7.79) and $cTBS_{600}$ (87.34 ± 19.54) stimulation ($F_{(2,14)} = 10.80$, $p < 0.05$) (Figure 4). Statistical significance was achieved only for iTBS intervention, that MEP amplitude was increased.

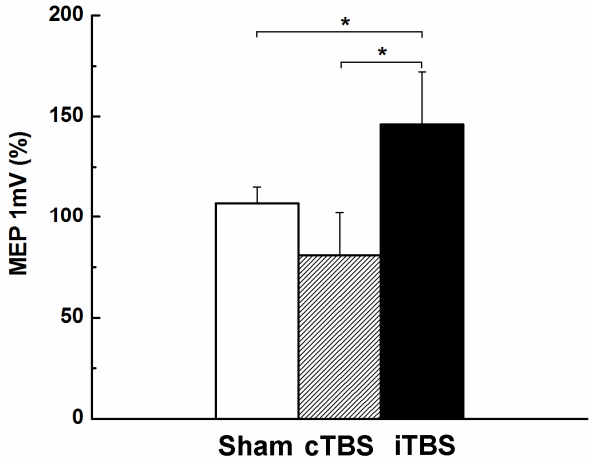

Fig. 4 – Histogram showing normalized data (mean and standard deviation) for three experimental protocols obtained from changes of motor evoked potentials (MEP) amplitudes, after motor cortex stimulation. Data are averaged across the subjects. *$p < 0.05$; cTBS – continous theta burst; iTBS – intermittent theta burst magnetic stimulation.

The effects of interventions on the task performance variables

Our results do not reveal any effects of the interventions on modulation of grip performance and force coordination in the R&H-T and the Lift-T tasks. The performance of R&H-T, assessed by RMSE, was not affected by the intervention, both for DH [Sham (Mdn = 1.05) *vs* cTBS (Mdn = 1.15) *vs* iTBS (1.02), $x^2_{(2)} = 0.60$, $p = 0.741$] and NH [Sham (Mdn = 1.17) *vs* cTBS (Mdn = 1.07) *vs* iTBS (0.96), $x^2_{(2)} = 0.60$, $p = 0.741$]. Coefficient of variation of L, indicator of task performance in the Lift-T, also remained unchanged [DH, Sham (Mdn = 1.00) *vs* cTBS (Mdn = 0.65) *vs* iTBS (Mdn = 0.69), $x^2_{(2)} = 0.20$, $p = 0.905$; NH, Sham (Mdn = 0.87) *vs* cTBS (Mdn = 0.94) *vs* iTBS (Mdn = 0.66), $x^2_{(2)} = 0.60$, $p = 0.670$].

Table 1

Baseline characteristics of healthy subjects pre - interventional

Task	Task permormance		SHAM	cTBS	iTBS	
			Mdn (int)	Mdn (int)	Mdn (int)	*p*
R&H-T	RMSE	DH	2.3 (1.2–3.8)	3.3 (1.4–4.8)	2.9 (1.5–4.6)	0.202
		NH	2.6 (1.7–4.2)	2.8 (2.1–3.5)	2.7 (1.85–3.4)	0.273
Osc-T	CE	DH	2.01 (0.82–6.32)	2.70 (0.34–6.24)	2.22 (0.39–9.57)	0.497
		NH	2.40 (1.16–6.02)	3.56 (0.80–10.69)	2.62 (0.53–11.97)	0.273
	VE	DH	6.72 (5.03–11.38)	6.30 (5.03–8.33)	6.80 (4.78–9.70)	0.326
		NH	5.76 (4.12–11.66)	7.44 (5.13–9.90)	7.14 (3.36–11.88)	0.497
Lift-T	CV	DH	0.81 (0.33–2.24)	0.89 (0.53–3.83)	1.01 (0.22–1.62)	0.905
		NH	1.01 (0.26–1.66)	0.91 (0.60–1.80)	0.96 (0.28–2.09)	0.670
Force coordination						
R&H-T	G/L ratio	DH	1.03 (0.77–1.14)	0.95 (0.59–1.21)	1.01 (0.72–1.24)	0.407
		NH	0.93 (0.68–1.42)	0.89 (0.44–1.73)	0.93 (0.39–1.42)	0.905
	r	DH	0.995 (0.992–0.998)	0.995 (0.992–0.997)	0.996 (0.986–0.997)	0.497
		NH	0.996 (0.985–0.998)	0.995 (0.991–0.998)	0.996 (0.964–0.998)	0.905
Osc-T	G/L ratio	DH	1.18 (0.78–1.29)	1.00 (0.60–1.35)	0.96 (0.73–1.42)	0.497
		NH	1.01 (0.74–1.27)	1.01 (0.56–1.49)	0.95 (0.46–1.38)	0.670
	r	DH	0.980 (0.967–0.993)	0.981 (0.966–0.992)	0.982 (0.972–0.991)	0.741
		NH	0.986 (0.963–0.994)	0.986 (0.963–0.992)	0.986 (0.974–0.993)	0.407
	Gain	DH	0.96 (0.82–1.26)	1.05 (0.87–1.36)	1.07 (0.69–1.41)	0.497
		NH	1.08 (0.79–1.37)	1.09 (0.86–1.82)	1.07 (0.82–2.28)	0.407
	Offset	DH	-0.46 (-1.89–0.36)	-0.56 (-2.12–0.72)	-0.21 (-1.38–0.64)	0.067
		NH	-0.20 (-1.54–0.64)	-0.18 (-1.29–0.68)	-0.28 (-0.79–0.39)	0.895
Lift-T	G/L ratio	DH	1.03 (0.94–1.15)	1.02 (0.77–1.35)	0.94 (0.79–1.23)	0.082
		NH	1.05 (0.76–1.19)	1.05 (0.63–1.52)	0.96 (0.53–1.25)	0.497

SHAM – sham stimulation; R&H-T – ramp and hold task; Osc-T – oscillation task; Lift-T – lifting task; RMSE – root mean square error; CE – constant error; VE – variable error; CV – coefficient of variation; G/L ratio – grip-to-load ratio; DH – dominant hand; NH – non-dominant hand; r – cross correlation of the G and L; *p* – probability; Mdn – median; CTBS – continous theta burst stimulation; iTBS – intermittent theta burst stimulation.

In the Osc-T the ability of subjects to exert required pattern of L was assessed by absolute CE and VE (Figure 5A–B). Our results have shown better task performance regarding to CE for DH *vs* NH after iTBS protocol (z = -2.60, $p = 0.009$) with a large difference between hands (r = 0.58). Median of the results for both hands decreased after the iTBS, but note that differences between PRE and POST was larger for DH (from Mdn = -2.22 pre-intervention, to Mdn = 1.28 after intervention) relative to NH (Mdn = 2.61, PRE *vs* Mdn = 2.23, POST). Between different TBS protocols were no significance differences either for DH [Sham (Mdn = 1.08) *vs* cTBS (Mdn = 0.85) *vs* iTBS (Mdn = 0.95), $\chi^2_{(2)} = 1.40$, $p = 0.497$], as well as for NH [Sham (Mdn = 1.03) *vs* cTBS (Mdn = 0.95) *vs* iTBS (Mdn = 0.98), $\chi^2_{(2)} = 0.60$, $p = 0.741$].

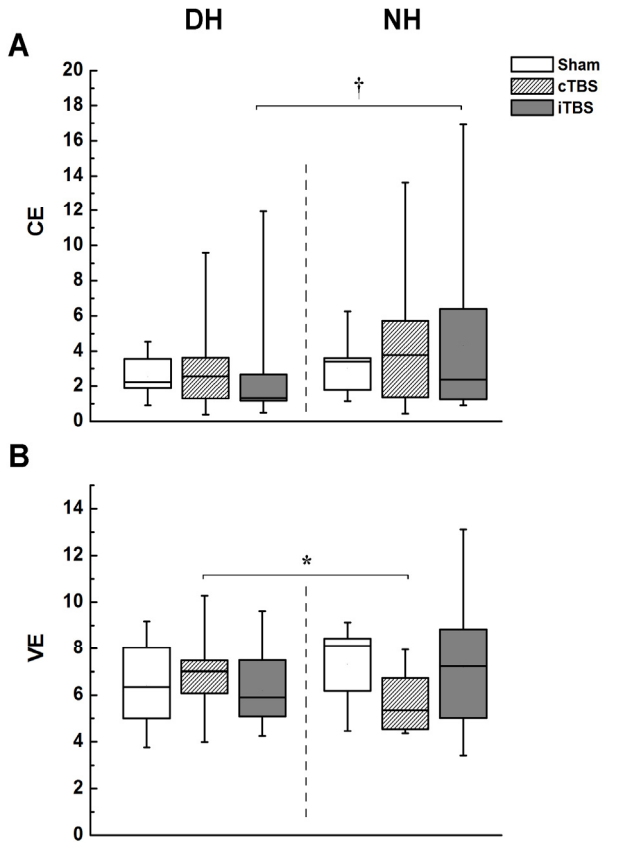

Fig. 5 – Task performance variables in oscillation task. A) Constant error (CE) and variable error (VE) values for dominant (DH) and B) non-dominant hand (NH) are averaged across the subjects for each of three experimental protocols (Sham, continous theta burst stimulation – cTBS and intermittent theta burst stimulation – iTBS). The box plots represent the 25th and 75th percentile of the distribution and the middle line represents the median. †$p < 0.01$ and *$p < 0.05$ between the groups (DH *vs* NH). Note that iTBS improved the CE when the task was performed with DH, while cTBS improved the VE if the task was performed with NH.

Results of Wilxocon's test for VE (the standard deviations of peaks of L) revealed better task performance for NH after the cTBS (z = -2.80, $p = 0.005$) with a large effect size (r = 0.63). Contrary, for DH there were no differences between values of VE pre- and post- the cTBS intervention (z = -0.56, $p = 0.575$). The only significant differences have shown for better task performances for NH relative to DH (z = -2.09, $p = 0.037$, r = 0.47). Using the Friedman's test on normalized set of data aiming to detect potential differences between protocols, we found no significant differences between Sham, cTBS and iTBS for DH ($\chi^2_{(2)} = 4.20$, $p = 0.122$), while results for NH revealed different effects of intervention [Sham (Mdn = 0.81) *vs* cTBS (Mdn = 0.81) *vs* iTBS (Mdn = 0.97), $\chi^2_{(2)} = 6.20$, $p = 0.045$]. Additional Wilcoxon's tests with Bonferroni correction for multiple comparisons revealed significant differences between cTBS i iTBS (z = -2.40, $p < 0.05$) with a large effect size (r = 0.54) in a form of worse task performance for NH using iTBS protocol.

The effects of interventions on the force coordination variables

TBS interventions did not affect the coordination of G and L in the R&H-T and the Lift-T. The adjustment of G and L forces during precision grip (assessed by G/L ratio) was not affected in the R&H-T [DH, Sham (Mdn = 0.93) *vs* cTBS (Mdn = 0.96) *vs* iTBS (Mdn = 1.00), $\chi^2_{(2)} = 0.20$, $p = 0.905$; NH, Sham (Mdn = 1.03) *vs* cTBS (Mdn = 1.14) *vs* iTBS (Mdn = 1.05), $\chi^2_{(2)} = 0.20$, $p = 0.905$], nor in the Lift-T [DH, Sham (Mdn = 0.94) *vs* cTBS (Mdn = 0.99) *vs* iTBS (Mdn = 1.09), $\chi^2_{(2)} = 4.20$, $p = 0.122$; NH, Sham (Mdn = 1.00) *vs* cTBS (Mdn = 1.01) *vs* iTBS (Mdn = 1.10), $\chi^2_{(2)} = 2.60$, $p = 0.273$]. The force coupling between G and L (assessed by *r*) in the R&H-T [DH, Sham (Mdn = 1.000) *vs* cTBS (Mdn = 1.000) *vs* iTBS (Mdn = 1.001), $\chi^2_{(2)} = 0.60$, $p = 0.741$; NH, Sham (Mdn = 1.000) *vs* cTBS (Mdn = 1.000) *vs* iTBS (Mdn = 1.000), $\chi^2_{(2)} = 4.20$, $p = 122$] was unchanged, too.

The results obtained by assessing force coordination variables in the Osc-T (Figure 6A-C) showed impairment of G-L scaling for NH at iTBS protocol, comparing results before (Mdn = 0.95) and after (Mdn = 1.02), z = -2.40, $p = 0.017$; see (Figure 6A), with a large effect of intervention (r = 0.54), while differences were not revealed for the Sham (z = -1.27, $p = 0.203$) neither for cTBS protocol (z = -0-56, $p = 0.57$). Contrary to those findings, no differences were shown, when task was performed with DH [Sham (z = -0.25, $p = 0.799$), cTBS (z = -0.50, $p = 0.646$) i iTBS (z = -1.82, $p = 0.69$).

Friedman's test for repeated measure did not reveal different effects of interventions on a G-L scaling either for DH or for NH (DH, $\chi^2_{(2)} = 0.80$, $p = 0.670$; NH, $\chi^2_{(2)} = 2.40$, $p = 0.301$).

Analyzing the data about G and L coordination through so called force coupling we found the similar impairment performing oscillatory task with NH after iTBS (z = -1.99, $p = 0.047$) (Figure 6B), while at the other two protocols (Sham and cTBS), as well as for DH at all three experimental protocols (Sham, cTBS and iTBS) were not significantly changed by interventions. However, between-group comparison of post-interventional data has shown significant differences for NH ($\chi^2_{(2)} = 13.40$, $p = 0.001$). Additional *post-hoc* tests with Bonferroni correction showed differences between cTBS and iTBS (z = -2.80, $p < 0.05$), with a large effect of intervention (r = 0.63).

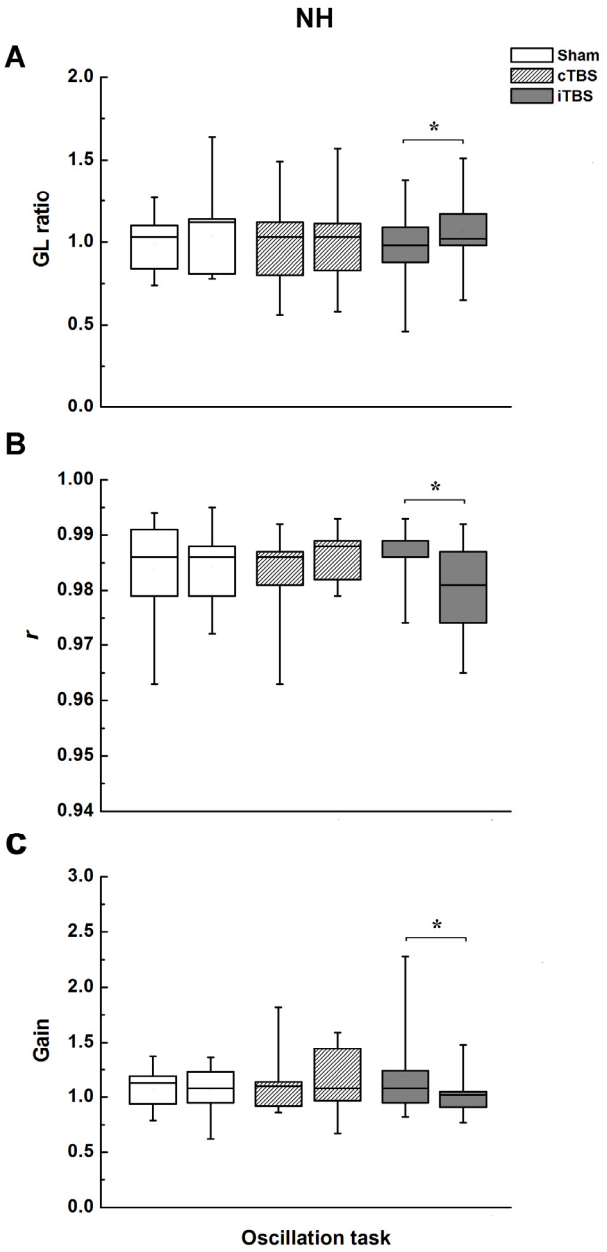

Fig. 6 – Force coordination variables, for non-dominant hand (NH), in oscillation task, before and after interventions. A) Grip-to-load ratio (G/L ratio), B) cross-correlation of the G and L (*r*), and C) gain of G, data are averaged across the subjects for each of three experimental protocols (Sham, continous theta burst stimulation – cTBS and intermittent theta burst stimulation – iTBS). The box plots represent the 25th and 75th percentile of the distribution and the middle line represents the median. Note that iTBS worsened most of the coordination variables for NH.
***p* < 0.05 within the group (PRE *vs* POST).**

Examination of median results for *r*, showed lower G-L coupling POST for NH at iTBS (Mdn = 0.980), relative to cTBS (Mdn = 0.988). Friedman's test used on relativized set of data (POST/PRE) revealed absence of the effects of intervention for DH ($\chi^2_{(2)}$ = 0.60, p = 0.741). Contrary, for NH significant differences between experimental protocols were found ($\chi^2_{(2)}$ = 12.80, p = 0.002) between Sham and iTBS (z = -2.29, p < 0.05, r = 0.51) and cTBS and iTBS (z = -2.80, p < 0.05, r = 0.63) in the form of lower G-L coupling after iTBS (Mdn = 0.997) relative to the Sham (Mdn = 1.000) and cTBS (Mdn = 1.004). The effect of intervention (POST/PRE) was revealed for DH *vs* NH, also at iTBS protocol [DH (Mdn = 1.000) *vs* NH (Mdn = 0.997), z = -1.99, p = 0.47, r = 0.44], while at Sham [DH (Mdn = 1.000) *vs* NH (Mdn = 1.000), z = -0.56, p = 0.575] and cTBS [DH (Mdn = 1.001) *vs* NH (Mdn = 1.004), z = -0.51, p = 0.959] differences as an effect of intervention were not found.

The G-L modulation in the Osc-T was assessed by Gain and Offset, disclosing the presence of significant differences, exclusively for Gain (Figure 6C). Wilcoxon's test revealed difference between PRE and POST at iTBS protocol, as the impairment hand function for NH after intervention (Mdn = 1.07, PRE; Mdn = 1.01, POST), z = -1.99, p = 0.047, r = 0.44. Comparing the results obtained from all three interventional protocols, for the same parameter, we did not find differences between-group for after-intervention effect (DH, $\chi^2_{(2)}$ = 0.60, p = 0.741; NH, $\chi^2_{(2)}$ = 5.00, p = 0.082), as well as within-group effects [POST/PRE; DH ($\chi^2_{(2)}$ = 1.40, p = 0.497), NH – $\chi^2_{(2)}$ = 2.60, p = 0.273].

Considering the Offset, we did not find any within-group, either between-group differences (DH, $\chi^2_{(2)}$ = 0.67, p = 0.717; NH, $\chi^2_{(2)}$ = 0.67, p = 0.717).

Discussion

In the present study we demonstrate that application of facilitatory and inhibitory TBS protocols over the dominant PMdn lead to bi-directional and complex modulation of grip performance and coordination when unimanual tasks were performed in healthy individuals. To our knowledge, this is the first experiment designed specifically to address effects of rTMS intervention on precision grasp including both hemispheres. This is especially important if one bears in mind that previous study has revealed the inhibitory effects of low-frequency rTMS (1 Hz) on MEP amplitudes, as well as differences in cerebral blood flow in multiple brain regions, including motor regions in the frontal cortex as well as more associational regions in the parietal and prefrontal cortices, when it applied over the PMd [32]. However, beyond these basic parameters of cortical excitability, virtual lesions produced by low frequency rTMS over M-1 and PMd of the dominant hemisphere lead to disturbances of anticipatory scaling of force for pinch grip [33]. Results of that study have shown that virtual lesion of M-1 causes disruption of scaling force based on information from a previous attempt, while lesion of PMd disturbs scaling based on arbitrary visual cues. These findings, actually confirm the prominent role of PMd in coupling arbitrary sensory cues to motor acts [34, 35].

In our study, we were using unimanual tasks that were primarily focused on scaling grip and load forces, but also included a visuomotor coordination. However, changes after TBS intervention were detected in only one of three manipulative tasks, the oscillatory task, interfering with the ability of subject to reach the required L peaks (task performance

variables), but also with the ability of grip and load forces coupling (force coordination variables).

Namely, the application of $iTBS_{600}$ or $cTBS_{600}$ over the M-1 in healthy individuals produced a relatively simple effect in terms of increased or decreased global motor system output, respectively [36]. In our study, however, most of the changes are registered after the $iTBS_{600}$ protocol and, as already indicated. As regards the task performance variables, during the oscillatory task, it was shown that application of $iTBS_{600}$ over dominant PMd induce the significant increase of tracking accuracy task, expressed as reduction of CE when task was performed with DH, while precision to follow prescribed peaks was disturbed for the NH performance. The effects of $cTBS_{600}$ protocols were significant only as improved task performance with NH.

In accordance with the contemporary viewpoint, cortical activity which reflects the performance of unimanual voluntary movements (or bimanual with a pronounced asymmetry), is distributed across both hemisphere [37]. Furthermore, communication between the hemispheres is carried out through transcallosal fibers, which transmit both, inhibitory and excitatory signals, although the prevailing opinion is that the inhibitory effects are stronger [38]. However, it is important to note that in addition to the most important interhemispheric communication between two homologous M-1 areas, a couple of non-primary motor areas are also included in the interhemispheric inhibitory network, but with a significantly less impact [39, 40].

Pronounced indirect changes, as we noted in our experiment, can be attributed to changes in the level of interhemispheric inhibition. Namely, according to the hypothesis of interhemispheric competition, two hemispheres behave as opposing systems, so that modulation of cortical excitability can change tonic transcallosal inhibition that is present under normal circumstances [41, 42].

If we apply this model of hemispheric rivalry to our experiment, it would mean that the facilitatory rTMS protocol (iTBS) over dominant PMd, in addition to increase of cortical excitability at the site of stimulation, leads to strengthening of interhemispheric inhibition directed against the homologous area of non-dominant hemisphere which is not under stimulation. By contrast, the use of inhibitory protocol (cTBS) over the dominant hemisphere should result in the weakening of interhemispheric inhibition transmitted *via* transcallosal fibers, so this would facilitate and improve precise grasping and object lifting in oscillatory task force.

Previous studies have shown that the application of iTBS in healthy subjects, leads to post-interventional reduction of MEP amplitudes over contralateral hemisphere [41]. In this case, it is assumed that the iTBS changes transcallosal input and amplifies interhemispheric tonic inhibition, leading to reduced excitability of non-stimulated hemisphere. In contrast, the study in which cTBS was applied over the M-1, showed the weakening of tonic interhemispheric inhibition and subsequent increase of cortical excitability over the hemisphere that was not stimulated [43]. This sequence of events might suggest that amplification of the motor output of the ipsilateral hand could interfere with the precise force gradation or magnification of the error.

Conclusion

This study further explores the relevant parameters involved in precise hand grip, mediated by PMd, including the effects on contralateral and ipsilateral hand. In this way, these results expand the knowledge arising from animal experiments and neuroimaging studies in humans, confirming the pivotal role of the PMd activation for the scaling of forces.

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REFERENCES

1. *Whishaw IQ, Karl JM.* The contribution of the reach and the grasp to shaping brain and behaviour. Can J Exp Psychol 2014; 68(4): 223−35.
2. *Napier JR.* Studies of the hands of living primates. Proc Zool Soc (London) 1960; 134(6): 647−57.
3. *Johansson RS, Westling G.* Roles of glabrous skin receptors and sensorimotor memory in automatic control of precision grip when lifting rougher or more slippery objects. Exp Brain Res 1984; 56(3): 550−64.
4. *Jaric S, Uygur M.* Assessment of hand function through the coordination of contact forces in manipulation tasks. J Hum Kinet 2013; 36: 5−15.
5. *Danion F.* The contribution of non-digital afferent signals to grip force adjustments evoked by brisk unloading of the arm or the held object. Clin Neurophysiol 2007; 118(1): 146−54.
6. *Macefield VG, Johansson RS.* Control of grip force during restraint of an object held between finger and thumb: Responses of muscle and joint afferents from the digits. Exp Brain Res 1996; 108(1): 172−84.
7. *Häger-Ross C, Johansson RS.* Nondigital afferent input in reactive control of fingertip forces during precision grip. Exp Brain Res 1996; 110(1): 131−41.
8. *Johansson RS, Birznieks I.* First spikes in ensembles of human tactile afferents code complex spatial fingertip events. Nat Neurosci 2004; 7(2): 170−7.
9. *Johansson RS, Flanagan J.* Coding and use of tactile signals from the fingertips in object manipulation tasks. Nat Rev Neurosci 2009; 10(5): 345−59.
10. *Geyer S, Matelli M, Luppino G, Zilles K.* Functional neuroanatomy of the primate isocortical motor system. Anat Embryol 2000; 202(6): 443−74.
11. *Hao Y, Zhang Q, Controzzi M, Cipriani C, Li Y, Li J,* et al. Distinct neural patterns enable grasp types decoding in monkey dorsal premotor cortex. J Neural Eng 2014; 11(6): 66011.
12. *Walsh V, Cowey A.* Transcranial magnetic stimulation and cognitive neuroscience. Nat Rev Neurosci 2000; 1(1): 73−9.
13. *Castiello U, Begliomini C.* The cortical control of visually guided grasping. Neuroscientist 2008; 14(2): 157−70.
14. *Rizzolatti G, Camarda R, Fogassi L, Gentilucci M, Luppino G, Matelli M.* Functional organization of inferior area 6 in the macaque monkey. II. Area F5 and the control of distal movements. Exp Brain Res 1988; 71(3): 491−507.

15. *Rizzolatti G, Luppino G.* The cortical motor system. Neuron 2001; 31(6): 889−901.
16. *Rizzolatti G, Luppino G, Matelli M.* The organization of the cortical motor system: New concepts. Electroencephalogr Clin Neurophysiol 1998; 106(4): 283−96.
17. *Kollias SS, Alkadhi H, Jaermann T, Crelier G, Hepp-Reymond MC.* Identification of multiple nonprimary motor cortical areas with simple movements. Brain Res Brain Res Rev 2001; 36(2−3): 185−95.
18. *Nirkko AC, Ozdoba C, Redmond SM, Bürki M, Schroth G, Hess CW*, et al. Different ipsilateral representations for distal and proximal movements in the sensorimotor cortex: Activation and deactivation patterns. Neuroimage 2001; 13(5): 825−35.
19. *Keel JC, Smith MJ, Wassermann EM.* A safety screening questionnaire for transcranial magnetic stimulation. Clin Neurophysiol 2001; 112(4): 720.
20. *Oldfield RC.* The assessment and analysis of handedness: The Edinburgh inventory. Neuropsychologia 1971; 9(1): 97−113.
21. *Krishnan V, Jaric S.* Hand function in multiple sclerosis: Force coordination in manipulation tasks. Clin Neurophysiol 2008; 119(10): 2274−81.
22. *Rossini PM, Burke D, Chen R, Cohen LG, Daskalakis Z, Di Iorio R,* et al. Non-invasive electrical and magnetic stimulation of the brain, spinal cord, roots and peripheral nerves: Basic principles and procedures for routine clinical and research application. An updated report from an I.F.C.N. Committee. Clin Neurophysiol 2015; 126(6): 1071−107.
23. *Rossi S, Hallett M, Rossini PM, Pascual-Leone A*; Safety of TMS Consensus Group. Safety, ethical considerations, and application guidelines for the use of transcranial magnetic stimulation in clinical practice and research. Clin Neurophysiol 2009; 120(12): 2008−39.
24. *Busan P, Barbera C, Semenic M, Monti F, Pizzolato G, Pelamatti G,* et al. Effect of transcranial magnetic stimulation (TMS) on parietal and premotor cortex during planning of reaching movements. PLoS ONE 2009; 4(2): e4621.
25. *Jaric S, Knight CA, Collins JJ, Marwaha R.* Evaluation of a method for bimanual testing coordination of hand grip and load forces under isometric conditions. J Electromyogr Kinesiol 2005; 15(6): 556−63.
26. *Freitas PB, Krishnan V, Jaric S.* Force coordination in static manipulation tasks: Effects of the change in direction and handedness. Exp Brain Res 2007; 183(4): 487−97.
27. *Westling G, Johansson RS.* Factors influencing the force control during precision grip.Exp Brain Res 1984; 53(2): 277−84.
28. *Flanagan JR, Tresilian JR.* Grip-load force coupling: A general control strategy for transporting objects. J Exp Psychol Hum Percept Perform 1994; 20(5): 944−57.
29. *Zatsiorsky VM, Gao F, Latash ML.* Motor control goes beyond physics: differential effects of gravity and inertia on finger forces during manipulation of hand-held objects. Exp Brain Res 2005; 162(3): 300−8.
30. *Flanagan JR, Wing AM.* Modulation of grip force with load force during point-to-point arm movements. Exp Brain Res 1993; 95(1): 131−43.
31. *Flanagan JR, Tresilian J, Wing AM.* Coupling of grip force and load force during arm movements with grasped objects. Neurosci Lett 1993; 152(1−2): 53−6.
32. *Chouinard PA, Van DW, Leonard G, Paus T.* Modulating neural networks with transcranial magnetic stimulation applied over the dorsal premotor and primary motor cortices. J Neurophysiol 2003; 90(2): 1071−83.
33. *Chouinard PA, Leonard G, Paus T.* Role of the primary motor and dorsal premotor cortices in the anticipation of forces during object lifting. J Neurosci 2005; 25(9): 2277−84.
34. *Petrides M.* Deficits in non-spatial conditional associative learning after periarcuate lesions in the monkey. Behav Brain Res 1985; 16(2−3): 95−101.
35. *Halsband U, Freund HJ.* Premotor cortex and conditional motor learning in man. Brain 1990; 113(Pt 1): 207–22.
36. *Huang Y, Edwards MJ, Rounis E, Bhatia KP, Rothwell JC.* Theta burst stimulation of the human motor cortex. Neuron 2005; 45(2): 201−6.
37. *Carson RG.* Neural pathways mediating bilateral interactions between the upper limbs. Brain Res Brain Res Rev 2005; 49(3): 641−62.
38. *Lenzi D, Conte A, Mainero C, Frasca V, Fubelli F, Totaro P,* et al. Effect of corpus callosum damage on ipsilateral motor activation in patients with multiple sclerosis: A functional and anatomical study. Hum Brain Mapp 2007; 28(7): 636−44.
39. *Grefkes C, Eickhoff SB, Nowak DA, Dafotakis M, Fink GR.* Dynamic intra- and interhemispheric interactions during unilateral and bilateral hand movements assessed with fMRI and DCM. Neuroimage 2008; 41(4): 1382−94.
40. *Fling BW, Benson BL, Seidler RD.* Transcallosal sensorimotor fiber tract structure-function relationships. Hum Brain Mapp 2013; 34(2): 384−95.
41. *Di Lazzaro V, Pilato F, Dileone M, Profice P, Oliviero A, Mazzone P,* et al. The physiological basis of the effects of intermittent theta burst stimulation of the human motor cortex. J Physiol (Lond) 2008; 586(Pt 16): 3871−9.
42. *Kinsbourne M.* Mechanisms of hemispheric interaction in man. In: *Kinsbourne M, Smith W*, editors. Hemispheric disconnection and cerebral function. Springfield, *IL:* Thomas*;* 1974. p. 260−85.
43. *Neva JL, Singh AM, Vesia M, Staines WR.* Selective modulation of left primary motor cortex excitability after continuous theta burst stimulation to right primary motor cortex and bimanual training. Behav Brain Res 2014; 269: 138−46.

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