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RESEARCH ARTICLE

Remote muscle contraction enhances spinal reflexes in multiple lower-limb muscles elicited by transcutaneous spinal cord stimulation

Yohei Masugi[1,2] · Atsushi Sasaki[1] · Naotsugu Kaneko[1] · Kimitaka Nakazawa[1]

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Abstract
Transcutaneous spinal cord stimulation (tSCS) is a useful technique for the clinical assessment of neurological disorders. However, the characteristics of the spinal cord circuits activated by tSCS are not yet fully understood. In this study, we examined whether remote muscle contraction enhances the spinal reflexes evoked by tSCS in multiple lower-limb muscles. Eight healthy men participated in the current experiment, which required them to grip a dynamometer as fast as possible after the presentation of an auditory cue. Spinal reflexes were evoked in multiple lower-limb muscles with different time intervals (50–400 ms) after the auditory signals. The amplitudes of the spinal reflexes in all the recorded leg muscles significantly increased at 50–250 ms after remote muscle activation onset. This suggests that remote muscle contraction simultaneously facilitates the spinal reflexes in multiple lower-limb muscles. In addition, eight healthy men performed five different tasks (i.e., rest, hand grip, pinch grip, elbow flexion, and shoulder flexion). Compared to control values recorded just before each task, the spinal reflexes evoked at 250 ms after the auditory signals were significantly enhanced by the above tasks except for the rest task. This indicates that such facilitatory effects are also induced by remote muscle contractions in different upper-limb areas. The present results demonstrate the existence of a neural interaction between remote upper-limb muscles and spinal reflex circuits activated by tSCS in multiple lower-limb muscles. The combination of tSCS and remote muscle contraction may be useful for the neurological examination of spinal cord circuits.

Keywords Jendrássik maneuver · Remote muscle · Spinal reflex · Transcutaneous spinal cord stimulation

Abbreviations

APB	Abductor pollicis brevis
AD	Anterior deltoid
BB	Biceps brachii
BF	Biceps femoris
EMG	Electromyographic
Exp	Experiment
ECR	Extensor carpi radialis
FDI	First dorsal interosseous
FCR	Flexor carpi radialis
H-reflex	Hoffmann reflex
JM	Jendrássik maneuver
MVC	Maximum voluntary contraction
MVF	Maximum voluntary force
MG	Medial gastrocnemius
RMS	Root mean square
SOL	Soleus
T-reflex	Tendon-reflex
TA	Tibialis anterior
tSCS	Transcutaneous spinal cord stimulation
VM	Vastus medialis

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✉ Kimitaka Nakazawa
nakazawa@idaten.c.u-tokyo.ac.jp

[1] Department of Life Sciences, Graduate School of Arts and Sciences, The University of Tokyo, 3-8-1 Komaba, Meguro-ku, Tokyo 153-8902, Japan

[2] Institute of Sports Medicine and Science, Tokyo International University, 2509 Matoba, Kawagoe-shi, Saitama 350-1198, Japan

Introduction

Spinal reflex tests, including the tendon jerk, are widely utilized as a diagnostic tool to examine the dysfunction of the upper- and lower-motor neurons in patients with neurological disorders. To reinforce the reflex response in patients who do not show a spinal reflex response, voluntary contraction of the muscles distant from the examination site (hereinafter referred to as remote muscles) is often simultaneously performed. This procedure is named as the Jendrássik maneuver (JM) and is clinically used by both neurologists and physical therapists. This phenomenon suggests the existence of a neural interaction between the remote muscles and the monosynaptic reflex circuits of other muscles in the cortical and/or subcortical sites (Boroojerdi et al. 2000).

The mechanisms behind the JM-induced facilitation of the monosynaptic reflex have been well studied through various electrophysiological techniques (Tazoe and Komiyama 2014). For example, basic studies using the Hoffmann (H-)reflex, which can be elicited by electrical stimulation of the Ia afferents in mixed peripheral nerves, reported the H-reflex amplitudes in the soleus (SOL) muscle to be enhanced by both wrist flexion (Tazoe et al. 2005) and teeth clenching (Miyahara et al. 1996). The JM-induced facilitation of the monosynaptic reflex was thought to be related to an increased muscle spindle sensitivity (Landau and Clare 1964), a reduction in presynaptic inhibition to the Ia terminals on the motoneurons (Dowman and Wolpaw 1988; Zehr and Stein 1999) and a direct postsynaptic facilitation of motoneurons (Boroojerdi et al. 2000; Furubayashi et al. 2003). These studies provided scientific evidence of the mechanisms underlying the JM-induced facilitation of the monosynaptic reflex in a few lower-limb muscles in which the H-reflex and/or Tendon-reflex (T-reflex) can be easily elicited. However, whether and to what extent the remote muscle contraction facilitates the spinal reflexes in other lower-limb muscles remains unknown.

Recently, transcutaneous spinal cord stimulation (tSCS) at the lumbar spinal cord was used to elicit the spinal reflexes in multiple lower-limb muscles (Courtine et al. 2007; Minassian et al. 2007; Kitano and Koceja 2009; Roy et al. 2012; Danner et al. 2016). While the H-reflex can be provoked in a single muscle such as the SOL and the flexor carpi radialis (FCR) (Zehr 2002), the tSCS technique can simultaneously evoke spinal reflexes in multiple lower-limb muscles (Courtine et al. 2007). Therefore, tSCS which can assess the spinal reflexes in multiple muscles is a useful technique for basic neurophysiological studies (Courtine et al. 2007; Minassian et al. 2007; Roy et al. 2014) and neurological examinations (Andriyanova 2010; Dy et al. 2010; Gerasimenko et al. 2015).

The facilitatory effects of remote muscle contraction (e.g., arm muscle contraction and teeth clenching) on the H-reflex have been reported for individual muscles such as tibialis anterior (TA) (Takada et al. 2000), SOL (Miyahara et al. 1996, Tazoe et al. 2005), quadriceps (Burke et al. 1996) and FCR (Sugawara and Kasai 2002). However, there are no studies examining the effects of remote muscle contraction in multiple muscles under the same stimulation and recording conditions. In the present study, using the tSCS technique, we investigated the effects of remote muscle contraction on the spinal reflexes in multiple lower-limb muscles. We hypothesized that the JM-induced facilitation of the reflex responses would occur with a similar time course in multiple lower-limb muscles and that such facilitatory effects would also be induced by remote muscle contractions in different upper-limb areas. To test these hypotheses, the present study investigated both the time course of the JM-induced facilitation and the facilitatory effects of the different upper-limb tasks (i.e., pinch grip, hand grip, elbow flexion and shoulder flexion) on the spinal reflexes evoked by tSCS in multiple lower-limb muscles.

Materials and methods

Participants

Eight healthy men participated in experiment 1 (Exp 1: $n = 8$, 23–32 years of age). Additional two healthy men were recruited for experiment 2, and six men participated in both experiments (Exp 2: $n = 8$, 23–31 years of age). All the participants were right handed. This study was conducted in accordance with the tenets of the Declaration of Helsinki (1964) and with the approval of the human ethic committee of The University of Tokyo (2017, no. 533). Each participant provided a signed informed consent to participate in the experimental procedures.

Electromyographic (EMG) recording

In Exp 1, surface EMG signals were recorded from the TA, SOL, medial gastrocnemius (MG), vastus medialis (VM), biceps femoris (BF), FCR, extensor carpi radialis (ECR), and first dorsal interosseous (FDI) of the dominant (right) side. In Exp 2, surface EMG signals were recorded from the TA, SOL, MG, VM, BF, anterior deltoid (AD), biceps brachii (BB), FCR, ECR, abductor pollicis brevis (APB), and FDI of the dominant (right) side. Bipolar Ag/AgCl surface electrodes (Vitrode F-150S, Nihon Kohden, Japan) were placed over the muscle belly with an interelectrode distance of 20 mm after reducing skin impedance using alcohol. Using a bioamplifer system (MEG-6108, Nihon Kohden, Japan), the EMG signals were amplified ($\times 1000$) and bandpass filtered between 15 Hz and 3 kHz.

Transcutaneous spinal cord stimulation (tSCS)

To evaluate the excitability of the spinal reflexes in multiple lower-limb muscles during remote muscle contraction, a constant current electrical stimulator with a pulse width set to 1 ms (DS7A, Digitimer, UK) was used with both a cathode (50×50 mm), placed on the midline on the skin between the spinous process of the higher lumbar vertebrae, and an anode (100 × 75 mm), located over the abdomen (Roy et al. 2012). Prior to the experiment, the location where the larger responses were produced in all the recorded lower-limb muscles was selected as the optimal site for the cathodal electrode (Exp 1: T12/L1 $n = 1$, L1/L2 $n = 6$, L2/L3 $n = 1$, Exp 2: L1/L2 $n = 6$, L2/L3 $n = 2$). The electrodes were fixed with adhesive tape to prevent their movement during the experiment. Successively, the recruitment curves of the responses of all the recorded lower-limb muscles were obtained to determine the stimulus intensity in each participant. Based on these recruitment curves, the stimulus intensity was adjusted so as to simultaneously evoke pronounced responses in all the recorded lower-limb muscles and to evoke below-plateau responses in most muscles. The stimulus intensity was kept constant during each experiment [Exp 1: 35–77 mA, Exp 2: 45–70 mA, range (min–max)]. Subsequently, a double-pulse stimulation (50 ms interval) was applied to confirm whether the response was initiated in the afferent fibers and the reflex was present at the beginning of the experiments, as the EMG responses could potentially be contaminated with the direct M wave evoked in the efferent fibers within the anterior roots. This test was utilized in previous studies (Courtine et al. 2007; Minassian et al. 2007; Dy et al. 2010; Roy et al. 2012).

General procedures

Participants were asked to maintain the supine position and to relax their whole body, while the tested (right) leg was fixed with an ankle foot orthosis and with an elastic band to prevent its movement during experiment (Fig. 1a). This study consisted of two experiments aiming at elucidating the characteristic of remote effects on the spinal reflexes in multiple lower-limb muscles. Exp 1 focused on the relationship between the timing of remote muscle contraction (i.e., hand grip) and the amplitude of the reflex responses in multiple lower-limb muscles. Exp 2 investigated the effects of the other tasks of remote muscle contraction (i.e., pinch grip, elbow flexion and shoulder flexion) on spinal reflexes in multiple lower-limb muscles. Exp 1 and 2 were performed on different days.

Experiment 1

Prior to the experiment, the maximum voluntary force (MVF) values of the hand grip (Grip) of the dominant (right) hand were measured with a hand grip dynamometer (GB, Takei, Tokyo, Japan). The procedures of Exp 1 were partially based on a previous study, which used the H-reflex technique (Tazoe et al. 2005). Realtime visual feedback for the hand grip force was given to each participant using an oscilloscope (TDS2012C, Tektronix Inc., USA) placed

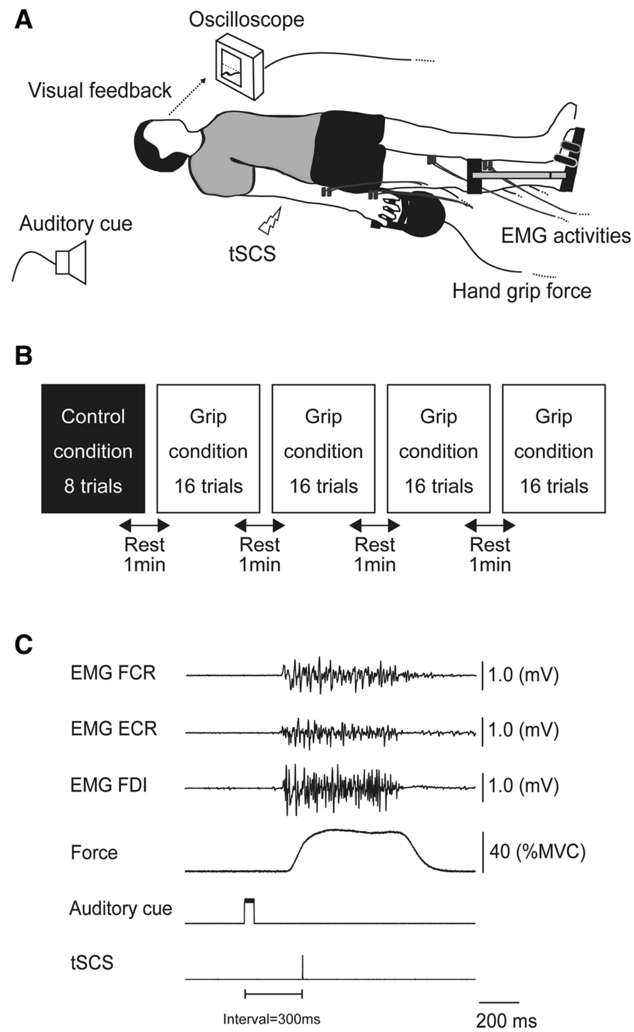

Fig. 1 Experimental setup and protocol of experiment 1: **a** participants were in the supine position. Real-time visual feedback for the hand grip force was provided to each participant using an oscilloscope. **b** Exp 1 consisted of five blocks; specifically, the first block represented the Control condition, whereas the second to the fifth block were the Grip conditions. **c** Typical example of hand grip force and EMG data in one trial (tSCS timing: 300 ms after the auditory cue)

approximately 30 cm in front of the participants (Fig. 1a). In a previous study, the target force level was set to 50% MVF (Tazoe et al. 2005). In this study, in consideration of muscle fatigue caused by muscle contraction, the target force level was set to 40% MVF. Specifically, one horizontal cursor on the oscilloscope display indicated the target force level (40% MVF), whereas the other horizontal cursor presented the hand grip force and it could be moved to the upper-forward section with the hand grip contraction. tSCS was delivered at 50–400 ms (50 ms step) after the presentation of the auditory cue (50 ms duration), which is controlled by a custom written LabVIEW program (National Instruments, USA) and delivered in a pseudo-random order. Exp 1 consisted of five blocks (Fig. 1b). The first block (8 trials for this block) represented the Control condition, in which participants were instructed to relax their whole body and to ignore the auditory cue. The next four blocks (16 trials for each block) represented the Grip condition, in which participants were asked to grip the hand dynamometer with their dominant (right) hand as fast as possible after hearing the auditory cue and to match the forces to a 40% MVF level for 1 s. In each trial, a single stimulus was delivered, and the spinal reflexes were simultaneously evoked in multiple lower-limb muscles. In total, 72 trials were conducted with each participant (Control condition: 1 block × 8 trials = total of 8 trials, Grip condition: 4 blocks × 16 trials = total of 64 trials; with each of the 8 stimulation delays tested two times per block). Figure 1c shows typical examples of EMG signals of the FCR, ECR and FDI muscles and the hand grip force signals from one trial (tSCS timing: 300 ms after auditory cue). The time interval between the trials was set to approximately 10 s, whereas the time interval between blocks was set to approximately 1 min.

Experiment 2

Exp 2 required each participant to perform five different tasks, including rest (Rest), pinch grip (Pinch), hand grip (Grip), elbow flexion (Elbow), and shoulder flexion (Shoulder). Both the pinch and the hand grip force in the dominant (right) hand were measured with a pinch and grip dynamometer, respectively (GB, Takei, Tokyo, Japan). In contrast, the dominant (right) elbow and shoulder flexion forces were assessed using a strain gauge sensor (LCB03K025L, A&D Company Limited, Japan), which was fixed on metal frames. In the Elbow and Shoulder tasks, the sensor was located on the distal part of the forearm and upper arm, respectively. In the Rest task, participants were instructed to relax their body and not to move while recording. In the Pinch task, participants were instructed to pinch with the first and second finger of their dominant (right) hand, whereas they had to grip with all the fingers of their dominant (right) hand in the Grip task. In the Elbow task, participants were instructed to exert a flexion torque of their dominant (right) elbow in isometric mode with the elbow fully extended and with the forearm supinated. In contrast, participants had to exert a shoulder flexion of their dominant (right) shoulder in the Shoulder task in isometric mode with the shoulder in a neutral position, the elbow fully extended, and the forearm pronated. These four remote contraction tasks were chosen to investigate the effects of remote muscle contraction in the different upper-limb areas (i.e., hand, forearm, and upper arm) on the spinal reflexes provoked by tSCS.

Figure 2a shows the experimental protocol of Exp 2. The Rest task was conducted in the C1, C2, C3, and C4 blocks as a Control condition for each task (Pinch, Grip, Elbow and Shoulder tasks). Additionally, the Pinch, Grip, Elbow, and Shoulder tasks were performed as test conditions in the T1, T2, T3, and T4 blocks, in a randomized order across the participants. The Rest task was also conducted in both the C5 and T5 blocks. Each block included 8 trials. In each trial, a single stimulus was delivered, and the spinal reflexes were evoked simultaneously in multiple lower-limb muscles. Prior to the C1, C2, C3, and C4 blocks, MVF values for each task were measured with each force sensor. Successively to the recording of the MVF values, each participant practiced at least 5 trials of each task before the beginning of data collection (before C1, C2, C3, and C4 blocks).

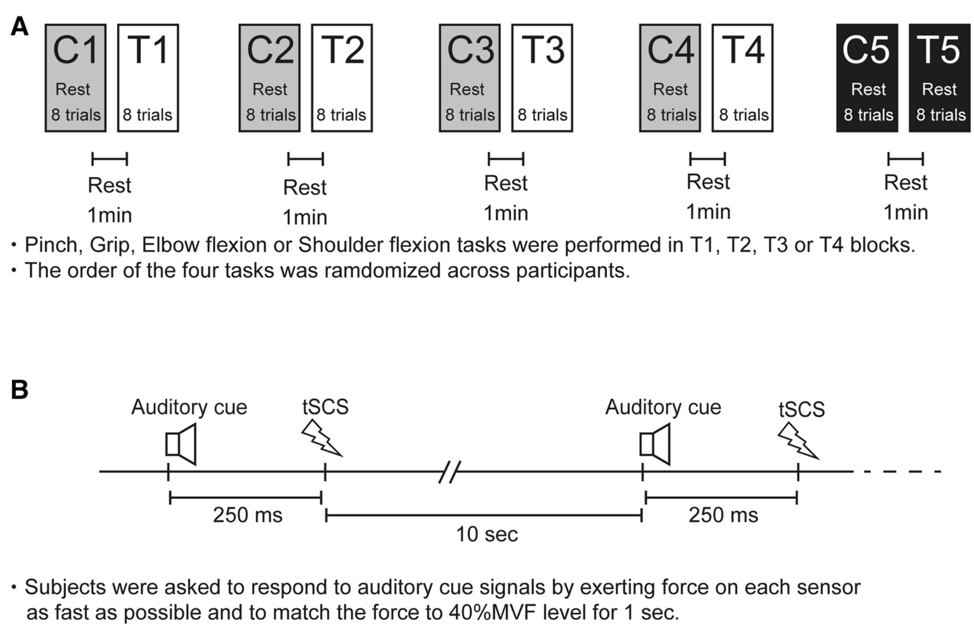

Fig. 2 Experimental setup and protocol of experiment 2: **a** Participants performed one of five different tasks (i.e., Rest, Pinch, Grip, Elbow and Shoulder) in each block. **b** tSCS was delivered at 250 ms after the auditory cue in all tasks. The time interval between trials was set to approximately 10 s

Additionally, tSCS was delivered at 250 ms after the auditory cue (50 ms duration) in all the five tasks, including the Rest task. The time interval between the auditory cue and the tSCS was determined based on the results of Exp 1 which reported the spinal reflexes to be strongly enhanced in all the recorded muscles with this interval (250 ms). The time interval between trials was set to approximately 10 s (Fig. 2b). The visual feedback in Exp 2 followed the same procedure of Exp 1. Specifically, in the Pinch, Grip, Elbow, and Shoulder tasks, each participant was asked to produce the required force with their dominant (right) hand as fast as possible after hearing the auditory cue and to match the forces to a 40% MVF level for 1 s. With regard to the EMG recording of the upper-limb muscles, the EMG signals were only recorded from the prime mover muscles of each task (Pinch: FDI, APB; Grip: FCR, ECR; Elbow: BB; Shoulder: AD) given the limited number of EMG channels.

Data acquisition and data analysis

All the EMG and force data were digitized at a sampling rate of 4 kHz using an A/D converter (Power Lab, AD Instruments, Australia) and stored on a computer for off-line analysis with Matlab 2018a (Mathworks, USA). Peak-to-peak amplitude of the spinal reflex was measured in each lower-limb muscle. The background EMG activity was calculated in each lower-limb muscle as the root mean square (RMS) values of the EMG signals for 50 ms prior to stimulation.

In the Grip condition during Exp 1, FCR EMG onset of each trial was determined as the time point at which the rectified FCR EMG signal visually reached three standard deviations from the baseline signal after the auditory cue. We measured the time interval between the time of EMG onset and the time of tSCS (i.e., the time of tSCS minus the time of EMG onset). A positive value of the time interval meant that tSCS was delivered after FCR EMG onset. Successively, each time interval was classified into eight phases (Phase 1: −99 to −50 ms, Phase 2: −49 to 0 ms, Phase 3: 1–50 ms, Phase 4: 51–100 ms, Phase 5: 101–150 ms, Phase 6: 151–200 ms, Phase 7: 201–250 ms, Phase 8: 251–300 ms after FCR EMG onset). Peak-to-peak amplitudes of the spinal reflexes and the background EMG activities were averaged in each phase of the Grip condition and in the Control condition. Additionally, the averaged values of the peak-to-peak amplitudes of spinal reflexes in each phase of the Grip condition were normalized to those in the Control condition.

In Exp 2, both the peak-to peak amplitudes of the spinal reflexes and the background EMG activities were averaged in each block (10 blocks: C1–C5, T1–T5). Thereafter, these values were statistically analyzed between Control and Test conditions for each task.

Statistical analysis

With regard to the double-pulse stimulation test of both experiments (Exp 1 and Exp 2), the Wilcoxon signed-rank test was used to compare the peak-to-peak amplitudes between the first and the second response.

In Exp 1, comparisons of the peak-to-peak amplitudes (%Control) of spinal reflexes and the background EMG activities (μV) between the control and the eight different phases (Control and Phases 1–8) were conducted using the Friedman test. When the Friedman test showed significance, multiple comparisons using the Wilcoxon signed-rank test were performed to compare between the control and the eight different phases (Control and Phases 1–8).

In Exp 2, comparisons of the peak-to-peak amplitudes (mV) of spinal reflexes and the background EMG activities (μV) between the Control and Test conditions were conducted using the Wilcoxon signed-rank test.

Non-parametric tests (i.e., the Friedman test and the Wilcoxon signed-rank test) were used in this study given that some measures were not normally distributed and the sample size ($n = 8$) was small. Due to the small number of participants, a conservative Bonferroni correction for multiple comparisons was not performed. The descriptive statistics are represented as either the mean or the median (25–75%). $p < 0.05$ was set as the significance level. These statistical analyses were performed using the SPSS software version 24.0 (SPSS, Chicago, IL, USA).

Results

Double-pulse stimulation test

The second responses elicited by the second stimulation were completely suppressed by the first stimulation (Exp 1: Supplementary Figure 1A, Exp 2: Supplementary Figure 2A). Additionally, the Wilcoxon signed-rank test showed that the peak-to-peak amplitudes of the second response were significantly smaller than those of the first response in all the recorded lower-limb muscles (all, $p < 0.05$, Exp 1: Supplementary Figure 1B, Exp 2: Supplementary Figure 2B).

Experiment 1

Figure 3 illustrates the averaged waveforms obtained from a single subject. The amplitudes of responses evoked by tSCS were enhanced in the Grip condition (Phase 4) compared with those in the Control condition. Figure 4 shows the relationship between the peak-to-peak amplitudes (%Control) of spinal reflexes and the eight different phases. In each muscle, the individual data (unfilled circles) represent similar trends

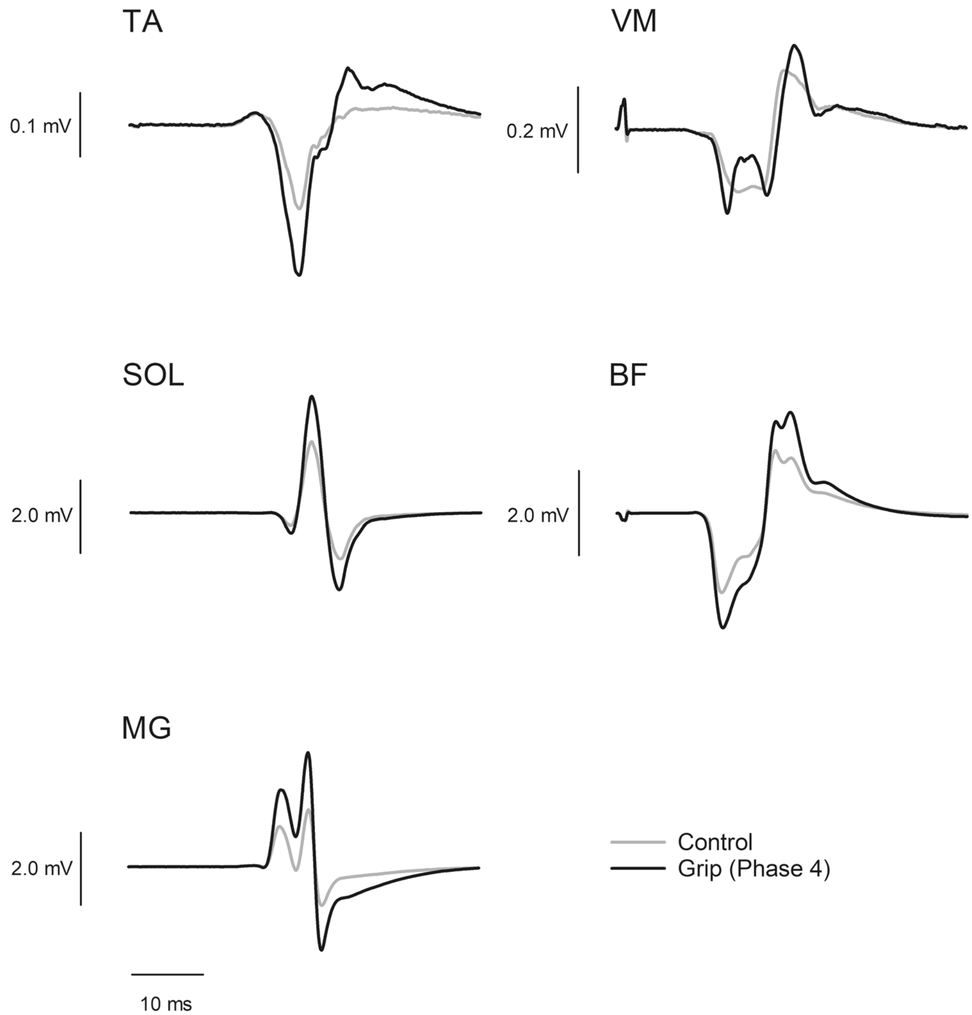

Fig. 3 Representative data from experiment 1 obtained from a single subject. Each waveform represents the average waveform of the responses evoked by tSCS in each muscle from Control condition (gray lines) or Grip condition (black lines)

across the participants (Fig. 4a). The Friedman test reported statistical significance in the spinal reflex amplitudes of all the muscles [TA: $\chi^2(8)=51.667$, $p<0.001$, SOL: $\chi^2(8)=52.700$, $p<0.001$, MG: $\chi^2(8)=46.340$, $p<0.001$, VM: $\chi^2(8)=54.000$, $p<0.001$, BF: $\chi^2(8)=39.033$, $p<0.001$]. Subsequently, the spinal reflex responses of TA, SOL and MG were significantly enhanced in Phases 4, 5, 6, 7 and 8 compared with the control values (100%) ($p<0.05$, Wilcoxon signed-rank test). The spinal reflex responses of VM were significantly enhanced in Phases 3, 4, 5, 6, 7 and 8 compared with the control values (100%) ($p<0.05$, Wilcoxon signed-rank test). The spinal reflex responses of BF were significantly enhanced in Phases 4, 5, 6 and 7 compared with the control values (100%) ($p<0.05$, Wilcoxon signed-rank test). Figure 4b shows a qualitatively similar time course of the onset of enhancement of spinal reflex amplitudes in the five lower-limb muscles. With regard to the background EMG activities (μV), the Friedman test did not show any statistical significance in the lower-limb muscles [TA: $\chi^2(8)=10.267$, $p=0.247$, SOL: $\chi^2(8)=15.067$, $p=0.058$, MG: $\chi^2(8)=4.600$, $p=0.799$, VM: $\chi^2(8)=9.767$, $p=0.282$, BF: $\chi^2(8)=6.833$, $p=0.555$, Supplementary Figure 3]. The average time interval between the auditory cue and EMG onset was 149.95 ± 21.70 ms (mean ± SD).

Experiment 2

Figure 5 illustrates the peak-to-peak amplitudes (mV) of the spinal reflexes in each condition. The Wilcoxon signed-rank test showed statistical difference between Control and Test conditions for each task (i.e., Pinch, Grip, Elbow and Shoulder) except for the Rest task in all the muscles ($p<0.05$). With regard to the background EMG activities (μV) in the lower-limb muscles, the Wilcoxon signed-rank test showed statistical significances between Control and Test conditions for BF in Shoulder condition and SOL in Elbow condition ($p<0.05$, Supplementary Figure 4).

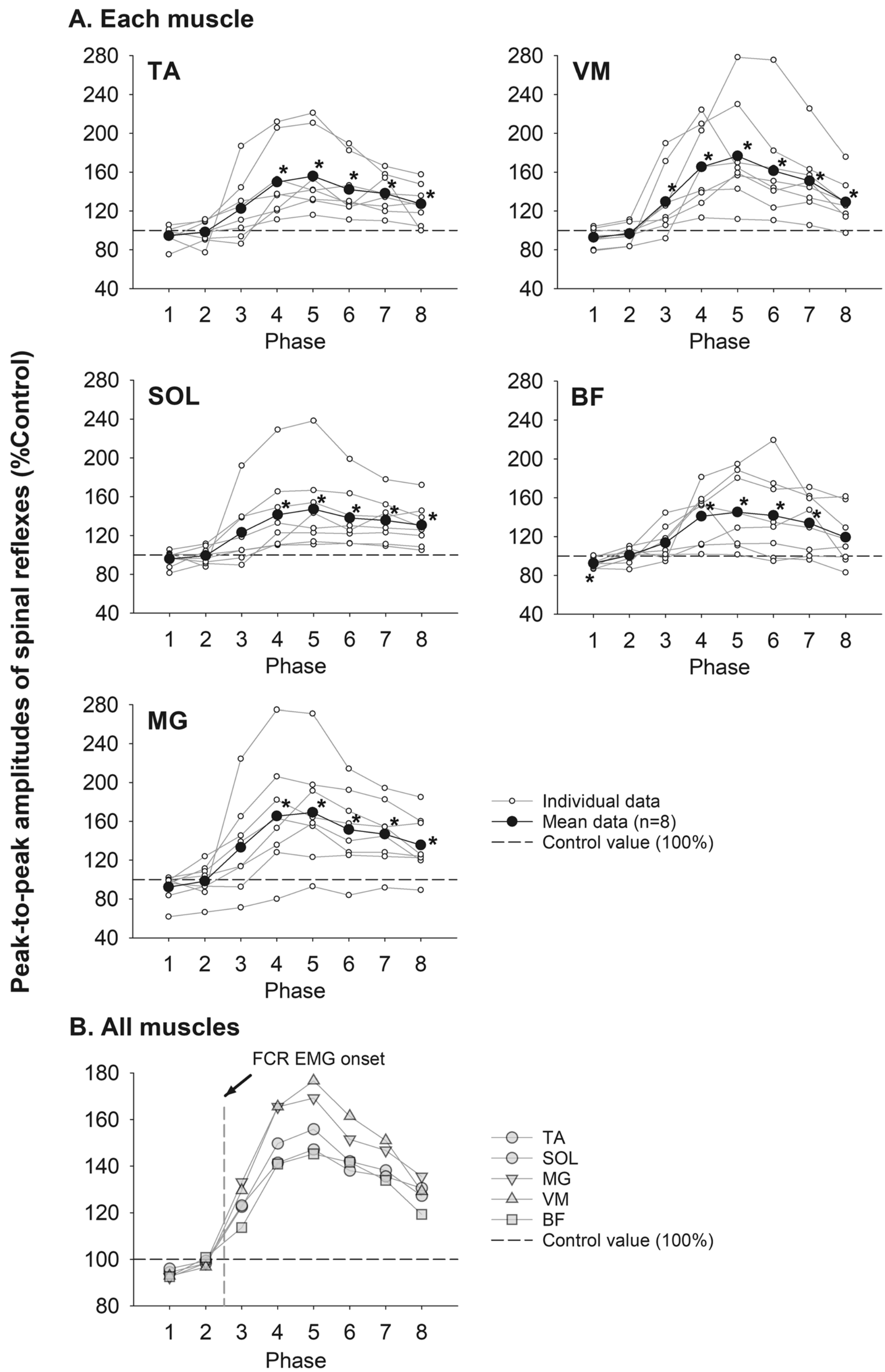

Fig. 4 Time course of the facilitation of the spinal reflexes by tSCS: **a** the averaged peak-to-peak amplitude of the spinal reflex responses in multiple lower-limb muscles was significantly enhanced from Phase 4 to Phase 7 (51–250 ms after EMG activation onset) when compared to the Control (Rest) condition. The filled and unfilled circles represent the mean data ($n = 8$) and individual data, respectively. **b** The time course of the average peak-to-peak amplitudes of all the muscles is overlaid in **b**. *$p < 0.05$

Discussion

The aim of this study was to determine whether remote muscle contraction would affect spinal reflexes in multiple lower-limb muscles evoked by tSCS. The results from Exp 1 clearly showed that the spinal reflex amplitudes in multiple lower-limb muscles were significantly enhanced from Phase 4 to Phase 7 (51–250 ms after FCR EMG onset) in Grip condition when compared to the Control condition. In addition, the results from Exp 2 showed that the other tasks of remote muscle contraction also significantly increased the spinal reflex amplitudes in multiple lower-limb muscles similarly

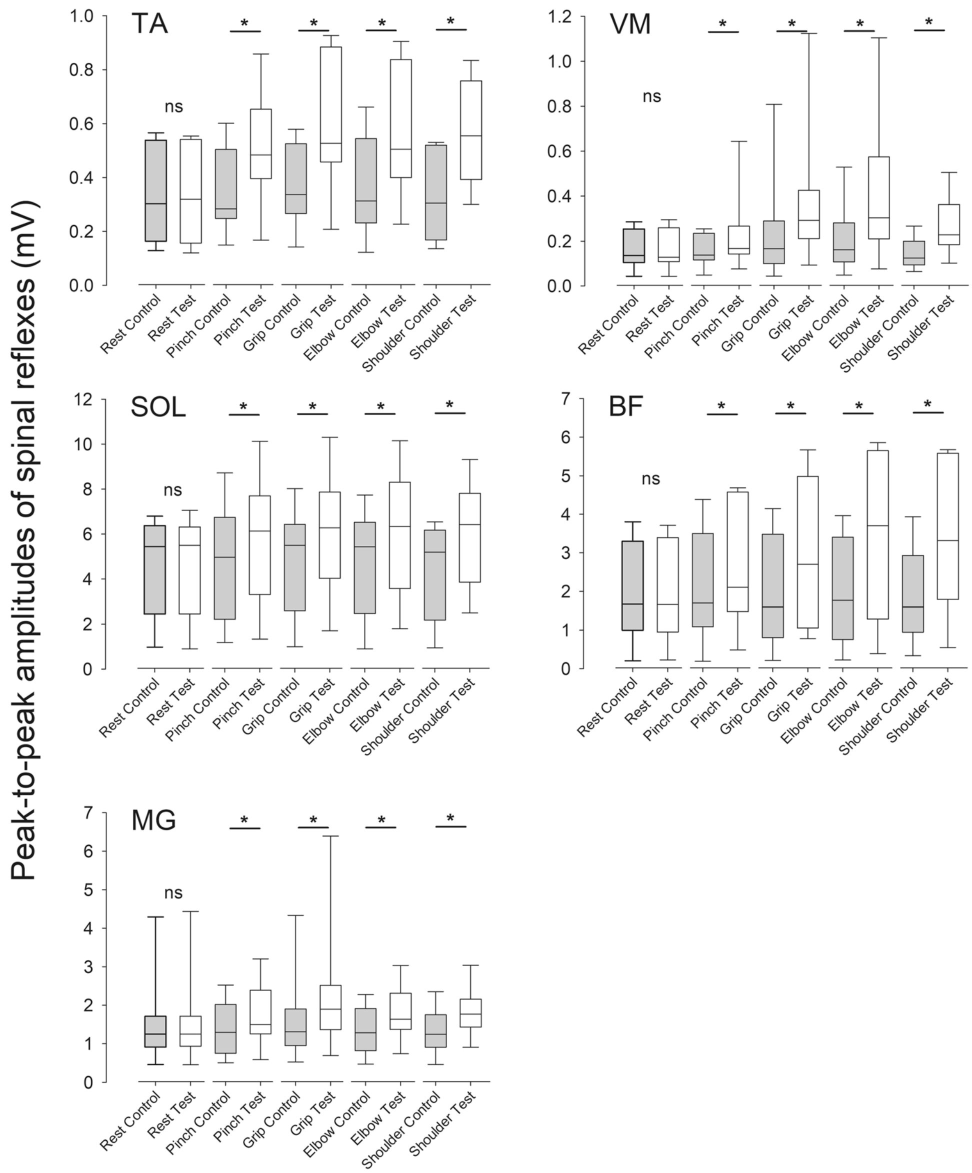

Fig. 5 Comparison of the spinal reflex amplitudes between Control and Test conditions. The Wilcoxon signed-rank test showed statistical differences between Control (gray boxes) and Test (white boxes*)* conditions in each task (i.e., Pinch, Grip, Elbow and Shoulder) except for Rest in all the muscles studied ($p<0.05$). The lines in the box plots indicate median values. The ends of the boxes represent the 25th and 75th percentiles. The whiskers on the boxplot illustrate the 10th and 90th percentiles. *ns* non-significant ($p>0.05$); *$P<0.05$

like the Grip task. The present study shows for the first time that the remote contraction of the upper-limb muscles simultaneously facilitates the excitability of spinal reflexes evoked by tSCS in multiple lower-limb muscles.

Responses evoked by transcutaneous spinal cord stimulation

In the current study, tSCS to the upper lumbar vertebrae levels evoked responses in the five lower-limb muscles in which the EMG signals were recorded. The responses by tSCS are thought to be a result of (1) sensory fibers (posterior root), (2) motor fibers (anterior root), and (3) mixed sensory-motor fiber stimulation (Danner et al. 2016). A computer simulation study demonstrated that posterior root fibers can be mainly activated by tSCS with low stimulation intensity (Danner et al. 2011). In addition, experimental studies using the double-pulse stimulation paradigm also showed that the response by tSCS mainly results from the activation of sensory fibers (Courtine et al. 2007; Minassian et al. 2007; Roy et al. 2012; Sayenko et al. 2015). In the present study, the second responses elicited by the second stimulation were suppressed by the first stimulation (Exp 1: Supplementary Figure 1, Exp 2: Supplementary Figure 2). A previous study assumed that post-attenuation by homosynaptic depression and heteronymous inhibitory circuits could impede a spinal reflex response (Roy et al. 2012). However, if the tSCS mainly activated the motor fibers, the second response should not be suppressed by the first stimulation. Therefore, the present results indicated that the responses by tSCS are not motor but trans-synaptic responses instead (i.e., spinal reflex responses). Furthermore, previous studies described the neural structures involved in the responses elicited by tSCS to be dependent on electrode location (Troni et al. 2011; Roy et al. 2012; Krenn et al. 2015; Masugi et al. 2017), stimulus intensity (Danner et al. 2011, 2016; Sayenko et al. 2015) and testing posture (Danner et al. 2016). In the present study, such conditions were considered appropriate for evoking spinal reflex responses by tSCS. The spinal reflexes evoked by tSCS show some characteristics similar to the H-reflex response (Courtine et al. 2007). However, compared with the H-reflex technique, the multiple posterior root activation caused by tSCS might introduce some complexity in these evoked responses. While the H-reflex response mainly results from homonymous Ia excitation, the tSCS-evoked responses can result from both homonymous and heteronymous Ia excitation. This is because tSCS simultaneously activates multiple posterior root fibers, including Ia afferent fibers from various lower-limb muscles. Therefore, it is reasonable to conclude that the spinal reflexes evoked by tSCS reflect the excitability of the spinal reflexes activated by both homonymous and heteronymous Ia afferents.

Time course of the facilitation of the spinal reflexes by tSCS

In both the H-reflex and T-reflex studies, the JM-induced facilitation was reported to be affected by several factors, including remote muscle contraction strength (Miyahara et al. 1996; Takada et al. 2000; Tazoe et al. 2005) and type of contraction (i.e., ballistic contractions or slowly progressive contractions) (Delwaide and Toulouse 1981). In addition, the timing is also recognized as a prominent factor in the JM-induced facilitation (Kawamura and Watanabe 1975). Although several studies reported the relationship between the timing of remote muscle contraction and the amplitudes of the responses (Kawamura and Watanabe 1975; Delwaide and Toulouse 1980, 1981; Miyahara et al. 1996; Takada et al. 2000; Tazoe et al. 2005), the results of one of these studies by Tazoe et al. (2005) are comparable to our results. In fact, their study indicated the JM-induced facilitation in the SOL H-reflex to be present from around 60 to 150 ms following EMG onset of the FCR muscle. In the present study, the spinal reflex in SOL evoked by tSCS was significantly facilitated from Phase 4 to Phase 8 (51–300 ms following EMG onset of the FCR). Considering that the latency of the spinal reflex evoked by tSCS is approximately 10 ms shorter than that of the H-reflex response (Kitano and Koceja 2009), the time course in this study (using the tSCS method) was very similar to that in the H-reflex study. Interestingly, the spinal reflexes in all the recorded muscles were enhanced as indicated by the qualitatively similar time course (Fig. 4b). So far, the facilitatory effects of remote muscle contraction on the H-reflex have been reported for individual muscles such as the TA (Takada et al. 2000), SOL (Miyahara et al. 1996; Tazoe et al. 2005), quadriceps (Burke et al. 1996), and FCR (Sugawara and Kasai 2002). However, there are no studies examining the effects in multiple muscles with same stimulation and recording conditions. To our knowledge, the present study was the first to show that the spinal reflexes in multiple lower-limb muscles were simultaneously facilitated by remote forearm muscle contraction using the tSCS method.

Although this study was not designed to clarify such mechanisms, our results could provide some insight into the mechanisms behind the JM-induced facilitation. Previous studies reported the JM-induced facilitation of the monosynaptic reflex to result from (1) an increase muscle spindle sensitivity (Landau and Clare 1964), (2) a reduction of presynaptic inhibition to the Ia terminals on the motoneurons (Dowman and Wolpaw 1988; Zehr and Stein 1999), and (3) a direct postsynaptic facilitation of motoneurons (Boroojerdi et al. 2000; Furubayashi et al. 2003). To date, several studies have excluded the possibility of an increase in muscle spindle sensitivity to play a role in the JM-induced facilitation (Hagbarth et al. 1975; Bussel et al. 1978). For example, Hagbarth et al. (1975) performed the microneurographic recordings of the Ia afferents during the JM and indicated that the JM did not affect the muscle spindle activity. Other candidates, specifically a reduction of presynaptic inhibition and a postsynaptic facilitation of motoneurons, would contribute to the JM-induced facilitation of monosynaptic spinal reflex (Dowman and Wolpaw 1988; Zehr and Stein 1999; Boroojerdi et al. 2000; Furubayashi et al. 2003). Although the site where the JM-induced facilitation occurs could not be determined by this study, upper-limb voluntary contraction would simultaneously influence the premotoneuronal mechanisms (i.e., reduction of presynaptic inhibition to the homonymous and heteronymous Ia terminals on the motoneurons) and/or the spinal motoneurons in multiple lower-limb muscles.

Jendrássik facilitation in the spinal reflexes by tSCS induced by the different upper-limb tasks

We investigated in Exp 2 whether other tasks (i.e., Pinch, Elbow and Shoulder) also facilitated the spinal reflex responses similarly like the Grip task when compared with the Rest. As a result, all remote muscle contraction tasks enhanced the spinal reflex responses in multiple lower-limb muscles when compared with the Rest task. These are unsurprising results given that the H-reflex in SOL has been shown to be enhanced not only by wrist flexion but also by teeth clenching (Tazoe et al. 2005; Miyahara et al. 1996; Takada et al. 2000). Overall, remote muscle contraction might facilitate the spinal reflexes in multiple lower-limb muscles, irrespective of the remote contraction tasks.

Functional significance

The present study showed the Jendrássik-induced facilitation of the spinal reflexes to be present in all the recorded muscles, regardless of both the distal/proximal muscles and agonist/antagonist muscles. These results were partially consistent with the study reported by Takada et al. (2000), considering that they reported the voluntary teeth clenching to facilitate the H-reflex, not only in the SOL, but also in the TA muscles. Therefore, the facilitation of the H-reflex occurred irrespective of the agonist/antagonist muscles. Based on such results, they proposed that voluntary teeth clenching would not contribute to smoothness of movement but to the stabilization of the postural stance instead (Takada et al. 2000). In concordance, the present study supports their ideas and also proposed the remote muscle contraction to enhance the stability of the whole distal/proximal and agonist/antagonist leg muscles.

Clinical implications for the diagnosis

tSCS is a novel method used to evoke spinal reflexes in multiple lower-limb muscles innervated by different segments of the spinal cord. To examine the precise pathological condition of patients with spinal cord disorders, the evaluation of the spinal reflex arcs in multiple muscles is crucial. In fact, the spinal reflexes by tSCS were previously used for the evaluation of spinal cord disorders (Andriyanova 2010; Dy et al. 2010; Gerasimenko et al. 2015). When spinal reflexes are observed, the spinal reflex arcs, including the afferent neurons and motor neurons, are considered to be preserved. Given that the somatosensory inputs are thought to be key elements for both the activation of the spinal locomotor circuits (Kojima et al. 1998, 1999; Dietz et al. 2002) and the recovery of locomotor function after spinal cord injury (Hubli and Dietz 2013; Takeoka et al. 2014), the evaluation of the reflex arcs in multiple lower-limb muscles is important for the prediction of functional improvements following spinal cord injury. The JMs reported in the present study were helpful for such an evaluation of the spinal reflexes by tSCS. With regard to the limitations of this study, we investigated the effects of remote muscle contraction on the intact spinal cord circuits only, considering that healthy participants were recruited. Future studies on spinal cord-injured patients should be conducted.

In conclusion, our results showed the remote muscle contraction (through the hand grip task) to have facilitatory effects on the spinal reflexes evoked in multiple lower-limb muscles. Furthermore, the time courses of the spinal reflex amplitudes were similar between different lower-limb muscles. In addition, other remote muscle contractions (i.e., pinch grip, elbow flexion and shoulder flexion) also facilitated the spinal reflexes in multiple lower-limb muscles, suggesting that these effects are induced by remote muscle contraction in different upper-limb regions. Overall, a spinal reflex test employing tSCS combined with the JMs would be helpful for the neurological examination of spinal cord-injured patients.

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Compliance with ethical standards

Conflict of interest The authors declare that no conflict of interests exist.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

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